Lekarz Anna Fedorczak

Analiza stężeń sirtuiny 1 u dzieci zdrowych oraz u dzieci z niedoborem wzrostu o różnej etiologii.

Rozprawa na stopień doktora w dziedzinie nauk medycznych i nauk o zdrowiu w dyscyplinie nauki medyczne.

Łódź, 2024r.

Promotor: dr hab. n. med. Renata Stawerska

Klinika Endokrynologii i Chorób Metabolicznych

Instytut Centrum Zdrowia Matki Polki w Łodzi

Szczególnie dziękuję

Pani Profesor Renacie Stawerskiej, za inspirację, pomoc merytoryczną,

a przy tym niekończący się entuzjazm i życzliwość,

Wszystkim Współautorom, za zaangażowanie i dobre rady

oraz

Mężowi Michałowi, za nieocenione wsparcie.

Synku Franciszku, dedykuję Ci tę pracę, jesteś moją największą motywacją.

Spis treści

1. Wykaz skrótów

BMI – ang. body mass index – wskaźnik masy ciała

CA – ang. calendar age – wiek kalendarzowy

GH – ang. growth hormone – hormon wzrostu

GHD – ang. growth hormone deficiency – niedobór hormonu wzrostu

HA – ang. height age – wiek wzrostowy

IGF-1 – ang. insulin-like growth factor 1 – insulinopodobny czynnik wzrostowy 1

IGFBP-3 – ang. insulin-like growth factor binding protein-3 – białko wiążące IGF, typu 3

IGF-1/IGFBP-3 m.r. – ang. molar ratio – stosunek molowy IGF-1/IGFBP-3

ISS – ang. idiopatic short stature – idiopatyczny niedobór wzrostu

pGHD – ang. partial growth hormone deficiency – częściowy niedobór hormonu wzrostu

sGHD – ang. severe growth hormone deficiency – ciężki niedobór hormonu wzrostu

SDS – ang. standard deviation score – wskaźnik odchylenia standardowego

SIRT1– sirtuina 1

SNP – somatotropinowa niedoczynność przysadki

2. Streszczenie w języku polskim

Przedmiotem niniejszej rozprawy doktorskiej jest cykl publikacji dotyczących analizy stężeń sirtuiny 1 (SIRT1) w surowicy dzieci zdrowych oraz dzieci z niedoborem wzrostu o różnej etiologii.

Artykuł 1

Pierwszą publikacją w cyklu był przegląd literatury dotyczący udziału SIRT1 w transdukcji sygnału hormonu wzrostu (GH)/insulinopodobnego czynnika wzrostu 1 (IGF-1) oraz jej wpływu na proces wzrastania. Powszechnie wiadomo, że regulacja wzrastania u dzieci zależy przede wszystkim od syntezy GH oraz IGF-1 – który jest głównym, obwodowym mediatorem działania GH. W ostatnich latach wykazano, że SIRT1 hamuje wewnątrzkomórkowy przekaz sygnału GH dla syntezy IGF-1 poprzez blokadę szlaku sygnałowego JAK/STAT. Dowiedziono również, że SIRT1 uczestniczy w podwzgórzowej sygnalizacji związanej z aktywacją receptora dla GH (GHR). Przedstawiono również dowody wskazujące, że SIRT1 wpływa na chondrogenezę płytki wzrostowej oraz wzrost kości na długość. Wydaje się, że te działania SIRT1 mogą mieć potencjalny wpływ na proces wzrastania u dzieci, jednak jak dotąd prac oryginalnych na ten temat jest niewiele.

W artykule omówiono również udział SIRT1 w innych procesach zachodzących wewnątrzkomórkowo, uwzględniając przede wszystkim jej znaczenie w regulacji metabolizmu, cyklu komórkowego, apoptozy, naprawie DNA czy odpowiedzi na stres oksydacyjny.

Artykuł 2

Drugi artykuł był pracą oryginalną, która miała na celu ocenę stężenia SIRT1 w surowicy na czczo u zdrowych dzieci oraz analizę wpływu wieku, płci, dojrzewania, masy ciała, wzrostu oraz diety na jej stężenie. Do badania zostało włączonych 47 zdrowych dzieci w wieku 4-14 lat z prawidłową masą ciała i wzrostem, nie chorujących przewlekle oraz niebędących w trakcie ostrej infekcji. Stężenie SIRT1 w surowicy na czczo oceniano za pomocą testu immunoenzymatycznego (ELISA). Wyniki wykazały, że stężenie SIRT1 w surowicy zdrowych dzieci nie różniło się w zależności od płci, wieku, wzrostu, masy ciała czy okresu dojrzewania. Natomiast okazało się, że częstsze spożycie owoców, warzyw i produktów mlecznych było związane z wyższym

stężeniem SIRT1 w surowicy. Co więcej, dzieci, które częściej spożywały owoce i warzywa były szczuplejsze i miały niższe stężenia IGF-1. Ocena stężenia SIRT1 w surowicy w kontekście zdrowia dzieci może być pomocna w rozumieniu procesów wzrastania i dojrzewania, ale także zaburzeń metabolicznych czy zaburzeń odżywiania.

Artykuł 3

Trzecia publikacja nosząca tytuł „Związek między stężeniem SIRT1 w surowicy a stężeniem GH/IGF-1 u dzieci z niedoborem hormonu wzrostu (GHD) oraz niskorosłością idiopatyczną (ISS)" była również pracą oryginalną. Celem tego badania było porównanie stężeń SIRT1 w surowicy dzieci z niedoborem GH (GHD) w stosunku do dzieci z idiopatycznym niedoborem wzrostu (ISS, bez niedoboru GH) oraz określenie możliwego wpływu zmian stężeń SIRT1 w surowicy na wydzielanie GH i IGF-1. Grupa badana obejmowała 100 dzieci z niedoborem wzrostu: 38 z GHD i 62 z ISS (maksymalne wydzielanie GH w dwóch testach stymulacji <10 i ≥ 10 ng/ml, odpowiednio). Stężenie SIRT1 nie różniło się istotnie między grupami GHD i ISS (średnia±SD: 0,89 ng/ml±0,45 dla ISS oraz 1,24 ng/ml±0,86 dla GHD), ale w obu tych grupach było istotnie wyższe niż w grupie dzieci zdrowych, prezentowanych w pracy nr 2 (0,29±0,21 ng/ml, $p<0.0001$,). Stwierdzono istotne ujemne korelacje między stężeniem SIRT1 a: SDS wzrostu, stężeniem IGF-1 oraz wartością IGF-1/IGFBP-3 molar ratio. Na tej podstawie wyciągnięty został wniosek, iż zmiany stężenia SIRT1 mogą stanowić jeden z mechanizmów, poprzez który wydzielanie IGF-1 jest zmniejszone u dzieci z niskim wzrostem.

3. Streszczenie w języku angielskim

The subject of this dissertation is a series of publications focusing on analysis of sirtuin 1 (SIRT1) concentrations in the serum of healthy children and children with short stature of various etiologies as well as the involvement of SIRT1 in growth hormone (GH) signal transduction.

Articule 1

The first publication was a review entitled "Involvement of sirtuin 1 in the growth hormone/insulin-like growth factor 1 signal transduction and its impact on growth processes in children". It is known that the regulation of growth processes depends on the synthesis of growth hormone (GH) and insulin-like growth factor 1 (IGF-1). IGF-1,

which is mainly secreted in liver in response to GH, is the major peripheral mediator of GH action. Sirtuin 1 (SIRT1) inhibits GH intracellular signaling for the IGF-1 synthesis via the janus kinase (JAK)/signal transducer and activator of transcription proteins (STATs) pathway. In addition, SIRT1 acts as a hypothalamic mediator of the GHR signaling. SIRT1 is also suggested to impact the growth plate chondrogenesis and longitudinal bone growth as it has a positive effect on the epiphyseal growth plate. Besides, SIRT1 is involved in various cellular processes, including energy metabolism, cell cycle regulation, apoptosis, DNA repair and oxidative stress response. This review focused on the influence of SIRT1 on GH signal transduction and the implications that may arise for growth processes in children.

Article 2

The second, original article concerned SIRT1 serum concentration in healthy children. This study aimed to evaluate fasting serum SIRT1 levels in healthy children, and to analyse the influence of age, sex, puberty, body weight, height, and diet on its concentration. 47 healthy children aged 4-14 with normal weight and height and no chronic disease were included into the study. Fasting serum SIRT1 concentrations were estimated by Enzyme Linked Immunosorbent Assay (ELISA). Results showed that serum SIRT1 concentrations in healthy children did not differ with respect to sex, age, height, weight and puberty. Whereas, it appeared that a higher frequency of fruits, vegetables and dairy products consumption was associated with an increase in serum SIRT1 levels. Studying SIRT1 in the context of children's health may have implications for a broader understanding of growth processes, pubertal development, metabolic disorders and nutrition.

Article 3

The third, also original, article was entitled "Relationship between serum sirtuin 1 and growth hormone/insulin-like growth factor 1 concentrations in children with growth hormone deficiency and idiopathic short stature". The aim of this study was to compare SIRT1 concentrations in children with GH deficiency (GHD) and idiopathic short stature (ISS, non-GH deficient), in order to determine the possible impact of changes in serum SIRT1 concentrations on the GH and IGF-1 secretion . The study group included 100 children with short stature: 38 with GHD and 62 with ISS (maxGH in two

stimulation tests <10 and ≥10 ng/ml, respectively). For each child, the concentrations of SIRT1, IGF-1 and insulin-like growth factor-binding protein 3 (IGFBP-3) were determined and the IGF-1/IGFBP-3 molar ratio was calculated. There were no differences in SIRT1 levels between children with GHD and ISS (mean ± SD: 0.89 ± 0.45 ng/ml for ISS; 1.24 ± 0.86 ng/ml for GHD), but in both those groups SIRT1 concentration were significantly higher than in healthy children (0.29 ± 0.21 ng/ml, $p<0.0001$, Article 2). A significant negative correlation was found between SIRT1 concentration and: height SDS, concentration of IGF-1 and IGF-1/IGFBP-3 molar ratio, but not between concentration of SIRT1 and maxGH. Elevated SIRT1 levels may serve as one of the mechanisms through which the secretion of IGF-1 is reduced in children with short stature; however, further research is required to confirm this issue.

4. Wykaz publikacji

W skład cyklu publikacji będącego podstawą niniejszej rozprawy doktorskiej wchodzą 3 artykuły o łącznym wskaźniku oddziaływania (impact factor, IF) - **14.7** i łącznej punktacji MNiSW **- 340** punktów.

1. Fedorczak A, Lewiński A, Stawerska R. Involvement of sirtuin 1 in the growth hormone/insulin-like growth factor 1 signal transduction and its impact on growth processes in children. Int J Mol Sci. 2023; 24(20):15406. doi:10.3390/ijms242015406

Praca przeglądowa, IF **5.6**, punktacja MNiSW **140**

2. Fedorczak A, Lewiński A, Stawerska R. Sirtuin 1 serum concentration in healthy children - dependence on sex, age, stage of puberty, body weight and diet. Front Endocrinol (Lausanne), 2024;15:1356612. doi:10.3389/fendo.2024.1356612

Praca oryginalna, IF **5.2**, punktacja MNiSW **100**

3. Fedorczak A, Kowalik D, Kopciuch J, Głowacka E, Mikołajczyk K, Tkaczyk M, Lewiński A, Stawerska R.. Relationship between serum sirtuin 1 and growth I hormone/insulin-like growth factor 1 concentrations in children with growth hormone deficiency and idiopathic short stature. Biomedicines, 2024;12(7):1433. doi:10.3390/biomedicines12071433

Praca oryginalna, IF **3.9**, punktacja MNiSW **100**

International Journal of
Molecular Sciences

MDPI

Review

Involvement of Sirtuin 1 in the Growth Hormone/Insulin-like Growth Factor 1 Signal Transduction and Its Impact on Growth Processes in Children

Anna Fedorczak [1], Andrzej Lewin´ski [1,2] and Renata Stawerska [1,3,*]

[1] Department of Endocrinology and Metabolic Diseases, Polish Mother's Memorial Hospital—Research Institute, 93-338 Lodz, Poland; anna.fedorczak@outlook.com (A.F.); andrzej.lewinski@umed.lodz.pl (A.L.)
[2] Department of Endocrinology and Metabolic Diseases, Medical University of Lodz, 93-338 Lodz, Poland
[3] Department of Paediatric Endocrinology, Medical University of Lodz, 93-338 Lodz, Poland
* Correspondence: renata.stawerska@icloud.com

Abstract: The regulation of growth processes in children depends on the synthesis of growth hormone (GH) and insulin-like growth factor 1 (IGF-1). Insulin-like growth factor 1, which is mainly secreted in the liver in response to GH, is the main peripheral mediator of GH action. Newly discovered factors regulating GH secretion and its effects are being studied recently. One of them is sirtuin 1 (SIRT1). This NAD^+-dependent deacetylase, by modulating the JAK2/STAT pathway, is involved in the transduction of the GH signal in hepatocytes, leading to the synthesis of IGF-1. In addition, it participates in the regulation of the synthesis of GHRH in the hypothalamus and GH in the somatotropic cells. SIRT1 is suggested to be involved in growth plate chondrogenesis and longitudinal bone growth as it has a positive effect on the epiphyseal growth plate. SIRT1 is also implicated in various cellular processes, including metabolism, cell cycle regulation, apoptosis, oxidative stress response, and DNA repair. Thus, its expression varies depending on the different metabolic states. During malnutrition, SIRT1 blocks GH signal transduction in hepatocytes to reduce the IGF-1 secretion and prevent hypoglycemia (i.e., it causes transient GH resistance). In this review, we focused on the influence of SIRT1 on GH signal transduction and the implications that may arise for growth processes in children.

Keywords: sirtuin 1; JAK2-STAT pathway; growth hormone; IGF-1; short stature children

Citation: Fedorczak, A.; Lewin´ski, A.; Stawerska, R. Involvement of Sirtuin 1 in the Growth Hormone/Insulin-like Growth Factor 1 Signal Transduction and Its Impact on Growth Processes in Children. *Int. J. Mol. Sci.* **2023**, *24*, 15406. https://doi.org/10.3390/ijms242015406

Academic Editor: Jacques J. Tremblay

Received: 7 September 2023
Revised: 1 October 2023
Accepted: 19 October 2023
Published: 20 October 2023

1. Introduction

The regulation of growth processes in children depends on the synthesis, secretion, and action of growth hormone (GH) and insulin-like growth factor 1 (IGF-1). Insulin-like growth factor 1 IGF-1, which is mainly secreted in the liver in response to GH, is the main peripheral mediator of GH action. It is well known that GH is synthesized, stored, and secreted by somatotropic cells of the anterior pituitary gland. It is secreted in a circadian rhythm under the influence of the hypothalamic hormones: the stimulating effect of a GH-releasing hormone (GHRH, somatoliberin) and the inhibitory effect of a GH-inhibiting hormone (GHIH, somatostatin). GHRH and GHIH secretion is modulated by various hormones and factors [1,2]. One of them is ghrelin, which acts both indirectly, via the hypothalamus, where it modulates the release of the above mentioned hormones, and directly, via somatotrophs, where it stimulates GH secretion [3]. In addition, ghrelin also affects the orexigenic centre in the hypothalamus, stimulating food intake [4]. Somatoliberin, somatostatin, ghrelin, and their respective cascades of action, are important agents in the network of factors regulating GH secretion [5]. GH affects numerous cells, tissues, and organs, but its main mediator promoting longitudinal growth is IGF-1, synthesized mainly in the liver [6].

Recently, newly defined factors have been emerging that may contribute to the regulation of GH secretion and its effects, one of them being sirtuin 1 (SIRT1) [7,8]. This NAD+-dependent deacetylase, by modulating the Janus kinase 2 (JAK2)/Signal Transducers and Activators of Transcription (STAT) pathway, is involved in the transduction of the GH signal in hepatocytes, leading to the synthesis of IGF-1, but also in the regulation of GHRH synthesis in hypothalamic neurons with GH receptor activity [9,10]. It has been suggested that SIRT1 is involved in growth plate chondrogenesis and longitudinal bone growth [11]. It is also engaged in various cellular processes, including metabolism, cell cycle regulation, apoptosis, oxidative stress response, DNA repair, inflammation processes, as well as the regulation of hunger and satiety [12,13]. Thus, its expression changes according to different metabolic states to help maintain homeostasis. In this review, we have attempted to present the current knowledge concerning the involvement of SIRT1 in GH/IGF-1 signal transduction and to discuss its possible impact on growth processes in children.

2. GH Intracellular Signal Transduction—GHR-JAK2-STAT Pathway

The binding of GH to its cell surface receptor (GH receptor—GHR) leads to the activation of several intracellular signalling pathways. With respect to the growth processes of children, the main pathway is the JAK2/STAT5β pathway in the liver because it is responsible for IGF-1 synthesis [9].

The GHR is a member of the class 1 haematopoietic cytokine receptor family. Although this receptor is found on the cells of many tissues, the liver is the richest in it [10]. It is a cell surface glycoprotein receptor, and consists of a large extracellular domain containing the GH binding site, an intracellular domain and a single transmembrane domain (TD). The GH molecule binds on the cell surface to the binding site of one GH receptor and then to the other one. As a result, a complex is formed containing two GHRs (GHR dimer) in combination with one GH molecule. The dimerization of the GHR is critical for GHR signalling and GH action, because it results in a conformational change in GHR and—subsequently—the repositioning of the proximal part of the intracellular domain—the proline-rich region (Box1) associated with JAK2. This starts a JAK2-mediated tyrosine phosphorylation cascade involving JAK2 itself, the GHR and STATs [10]. The main STATs are 1, 3, and 5; the most important—for further consideration—are STAT3 and STAT5β. After their phosphorylation, STATs translocate from the cytoplasm to the nucleus and bind to the promoter region of DNA which results in the promotion the transcription of IGF-1.

Also, the IGF-2, IGF-binding protein 3 (IGFBP-3) and acid labile subunit (ALS) genes are promoted in the same mechanism [14]. This pathway is therefore responsible for the activation of the *IGF-1* gene and the synthesis of IGF-1, and also ensures the subsequent stability of IGF-1 in the serum, as it is involved in the production of the elements used to form the IGF-1–ALS–IGFBP-3 tertiary complex. It has been proven that SIRT1 is one of the modulators of the activity of the JAK2/STAT pathway [15].

The activation of JAK-STAT signalling occurs within minutes after GH stimulation, but is transient due to the tight regulation of the signalling termination by the suppressors of cytokine signalling (SOCS), protein tyrosine phosphatases (PTPs), protein inhibitors of activated states (PIAS), and GHR internalisation [10].

GH also activates other pathways, including the mitogen-activated protein kinase (MAPK), extracellular signal-regulated kinase (ERK) 1 and 2, as well as the insulin-receptor substrates (IRS) 1 and 2 and phosphatidyl inositol-3 kinase (PI3K) pathways. They clearly contribute to the metabolic effects of GH; however, their role in the growth of bones remains unclear [16–18]. Considering the cardinal importance of the GH–IGF-1 axis in linear human growth, the defects or modifications at various points of this axis may result in an impaired growth rate, leading to short stature [19,20].

3. IGF-1—The Main Mediator of GH Activity

It is well known that GH in peripheral tissues acts through IGF-1 [6]. IGF-1 is responsible for stimulating the growth of all cell types and causes significant metabolic effects [21,22]. As was mentioned above, IGF-1 is produced mainly in hepatocytes, but other cell types are also able to synthesise and secrete IGF-1 [6]. Besides GH, a number of other factors influence the synthesis of IGF-1, such as nutrients and nutritional status, some components of the immune system, as well as other hormones. IGF-1 inhibits hypothalamic GHRH release and directly inhibits GH production in pituitary somatotrophs by the negative feedback mechanism [6,22,23].

The IGF-1 bioavailability and stability in circulation depend on binding to specific proteins, IGFBP-3 in particular. IGFBPs both extend the half-life of IGFs and modulate IGFs' availability and activity [24,25]. IGF-1/IGFBP-3 then binds ALS and thus forms a large (150 kDa) ternary complex, which is a reservoir of IGF-1 [6]. The IGF-1 and IGF-2 are liberated from this complex by pappalysin 2 (PAPPA2), which enables them to pass through the capillary epithelium and enter the interstitium. As it was mentioned above, IGF-2, IGFBP-3, and ALS synthesis and secretion are also dependent on the JAK2/STAT pathway and its modulator—SIRT1. In the target tissues, IGF-1 binds to two receptors: IGF-1R and IGF-2R, which exhibit intrinsic enzymatic activity. The growth-promoting effects of IGF-1 are mediated by IGF-1R, which is structurally homologous to the insulin receptor and can bind both IGF-1 and insulin, but has a greater affinity for IGF-1. In turn, IGF-1 can also bind to and activate the insulin receptor [5]. Thus, the effect of IGF-1 on glucose metabolism is similar to insulin (insulin-like effect) and—in contrast to GH—promotes hypoglycaemia. GH increases glucose production in the liver by enhancing glycogenolysis and gluconeogenesis [26,27], and in a state of energy deficiency, GH is an important signal for mobilizing body fat and glycogen in order to maintain normal blood glucose levels. This explains why a mechanism of GH resistance with respect to IGF-1 secretion can be triggered to maintain normal glucose levels and prevent hypoglycemia. There are many conditions in which IGF-1 secretion is reduced, even though GH secretion is normal or even elevated: e.g., liver disease, kidney dysfunction, sepsis, cancer, systemic autoimmune disease, conditions after severe surgical trauma, and other massive activations of the immune system (due to interleukin 1 and 6 activity) [28]. It has been proven that in patients with untreated celiac disease or other (even oligosymptomatic) gastrointestinal tract diseases, the concentration of IGF-1 is reduced, despite a normal GH secretion, confirmed in stimulating tests [29]. By the same mechanism, low IGF-1 levels with GH resistance are observed in malnourished children or children and adolescents with anorexia nervosa [30]. In all those cases, where these conditions are prolonged, the children's growth rate slows down and improves after this unfavourable phenomenon disappears [29].

It seems that SIRT1, which regulates the JAK2/STAT pathway, may be involved in peripheral GH resistance and be responsible for the reduced IGF-1 concentration in some conditions (e.g., malnutrition) that require increased glucose production to prevent hypoglycaemia. In such cases, the rate of growth may be temporarily impaired.

4. SIRT1—The Key Player in Metabolism

Sirtuins (silent information regulator 2 proteins) constitute a family of highly conserved NAD^+-dependent deacetylases. They use NAD^+ as an essential cofactor to remove acetyl groups from various proteins and alter their function. Seven types of sirtuins, SIRT 1–7, have been detected in mammals [7]. Sirtuins play key roles in responding to nutritional and environmental perturbations and are important in regulating a broad variety of cellular processes, e.g., metabolism, mitochondria homeostasis, autophagy, DNA repair, apoptosis, oxidative stress, and senescence [12,13,31]. Among the mammalian sirtuins, SIRT1 is the most extensively researched one. SIRT1 is the closest homolog to yeast Sir2p. It has been detected in multiple organs and tissues, including the liver, pancreas, brain, heart, muscle, and adipose tissue [32]. Although SIRT1 is mainly located in the nucleus, in certain cell types (e.g., pancreatic β cells) it can be localized in the cytoplasm [33]. SIRT1 interacts

with and regulates a number of histone and non-histone protein substrates. Similar to other sirtuins, it is involved in cell cycle regulation, apoptosis, autophagy, oxidative stress response, DNA repair, and inflammation [32,34–40], as well as in the regulation of cellular senescence and aging [12,13,31,39] (Figure 1). Moreover, SIRT1 also modulates important cellular processes in the cardiovascular system [41]. SIRT1 acts in endothelial and smooth muscle cells to protect the vasculature [42–44].

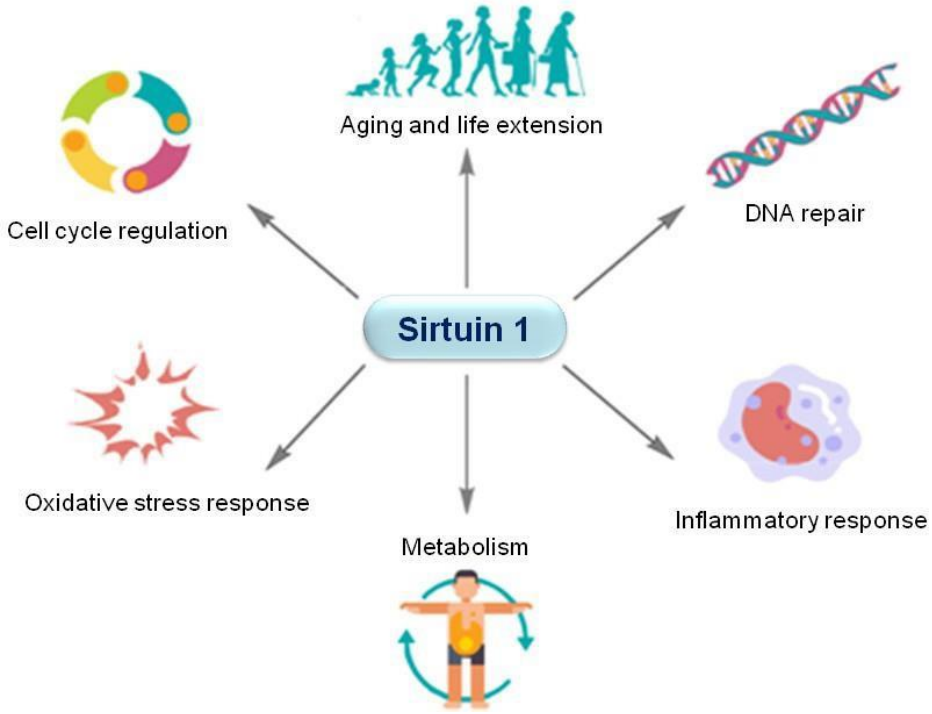

Figure 1. SIRT1 involvement in biological processes.

The amount of SIRT1 depends on the availability and type of nutrients [45], because this nutrient-responsive protein is involved in responding to metabolic imbalances that are triggered by fasting, caloric restriction, and malnutrition. In a fasting state, caloric restriction, or malnutrition, it intensifies catabolic processes and inhibits anabolic processes to maintain homeostasis [32] (Figure 2). In the liver, SIRT1 promotes fatty acids' oxidation via the activation of the peroxisome proliferator-activated receptor alpha (PPARα)/peroxisome proliferator-activated receptor-gamma coactivator (PGC)-1α signalling pathway [46], as well as inhibits fatty acids' synthesis by attenuating the transcriptional activity of sterol regulatory element-binding protein (SREBP)-1c [47,48]. In skeletal muscles, SIRT1 correspondingly increases fatty acids' oxidation via PGC-1α deacetylation [49,50]. In white adipose tissue, SIRT1 enhances lipolysis and fat mobilization from adipocytes, as well as reduces adipogenesis by inhibiting PPARγ [49,51]. SIRT1 drives white fat browning to disperse stored energy as heat [52]. Regarding glucose metabolism, SIRT1 stimulates gluconeogenesis in the liver by inhibiting STAT3, while activating PGC-1α and Forkhead box protein 1 (FOXO1), as well as inhibits glycolysis by deacetylating PGC-1α and by repressing the glycolytic enzyme PGAM-1 [53–56]. In pancreatic β-cells, SIRT1 enhances insulin release in response to glucose [33]. SIRT1 also regulates the production and secretion of insulin-sensitizing factors, such as adiponectin and fibroblast growth factor 21 (FGF21) through the regulation of FOXO1 and PPARγ [57]. Additionally, SIRT1 improves the insulin sensitivity of adipose tissue, skeletal muscle, and the liver [58]. What is particularly interesting is that SIRT1 inhibits GHR intracellular signal transduction for *IGF-1* gene expression to reduce IGF-1 synthesis and—in this way—maintains glucose levels [15,59].

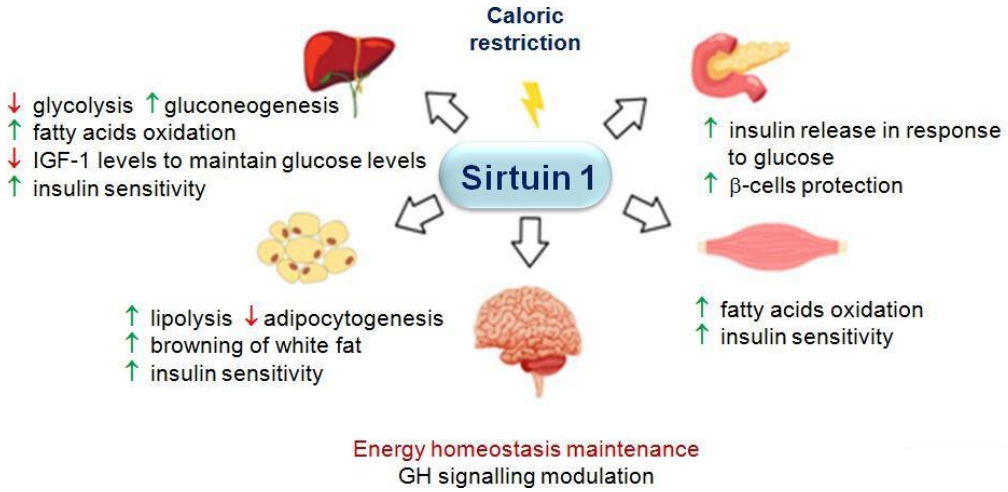

Figure 2. Summary of the effect of SIRT1 on various metabolic processes during caloric restriction. The red arrows indicates a reduction and the green arrows indicates an intensification of the processes in question.

5. Regulation of SIRT1 Activity

The classic activation of sirtuins is caused by an increase in the NAD+/NADH ratio. Thus, as mentioned, SIRT1 activation occurs under caloric restriction and starvation to enhance its catabolic effects. Also, physical activity increases the expression levels of SIRT1 [60]. However, there are other factors that regulate SIRT1 activity. A known SIRT1 activator is resveratrol—a polyphenol which can be found in red wine and some dietary products [61–63]. Quercetin, one of the major flavonoids which is widely expressed in fruits (grapes and peaches) and vegetables (onions and garlic) regulates cellular senescence and multiple aging-related cellular processes via SIRT1 [64]. The synthetic SIRT1-activating compounds (STACs), i.e., SRT1720, SRT2183, and SRT1460, have also been reported to activate SIRT1 in rodents and improve lipid profiles, glucose tolerance, and health span [12,65]. In turn, nicotinamide, a form of vitamin B3, was first introduced as a sirtuin inhibitor [66]. Several specific inhibitors of SIRT1 action have also been described, including sirtinol, cabol, tenovin-1 and tenovin-6, salermide, EX527, and others [67,68].

It seems that activation of SIRT1 by genetic or pharmacological methods can bring many metabolic benefits [69]. Sirtuin triggers are being considered for the treatment of obesity, and their beneficial effect could result from the intensification of catabolic processes [70–72].

Furthermore, it is important to mention the post-transcriptional factors that are involved in SIRT1 mRNA regulation. This post-transcriptional regulation is mediated by microRNAs (miRNAs) and RNA-binding proteins (RBPs) [42]. MicroRNAs are short non-coding RNAs that modulate target gene expression and, as a result, impact important processes including proliferation, apoptosis, and senescence. Various miRNAs have been proven to regulate SIRT1 expression and activity [42]. For example, miR-34a induces endothelial senescence and apoptosis through the inhibition of SIRT1 [73]. Whereas, RBPs are proteins that bind to RNA to form a ribonucleoprotein complex and play an important role in the post-transcriptional control of RNAs. Hu antigen R (HuR), one of the RBPs, promotes SIRT1 expression by directly binding to SIRT1 mRNA, as well as through indirect pathways via microRNA [74]. HuR and TIA1/TIAL1 are involved in the regulation of alternative splicing of SIRT1 pre-mRNA [75]. Moreover, adenosine deaminase, acting on RNA 1 (ADAR1), a RNA editing enzyme, regulates SIRT1 expression by affecting its RNA stability through HuR. Whereas, ADAR1 suppression promotes senescence via the downregulation of *SIRT1* mRNA by a decrease in HuR binding [76].

To sum up, the epigenetic regulation of SIRT1 may have impact on various pathways, concerning metabolic diseases, cardiovascular diseases, cancer, and senescence.

6. SIRT1 as a Negative Regulator of JAK2/STAT5β Pathway in the Liver and Its Impact on Growth

In a condition of caloric restriction, a number of adaptive processes to survive and provide energy for vital organs are activated. Most studies have shown that caloric restriction improves glucose metabolism, increases mitochondrial activity, and even extends life span [77]. In terms of growing, fasting induces GH resistance in the liver, leading to a decrease in the serum IGF-1 levels as one of the adaptive mechanisms for malnutrition [78]. Under fasting conditions, elevated GH levels are important to prevent hypoglycaemia and mobilize free fatty acids from adipose tissue, while reduced IGF-1 impairs growth and preserves energy for vital life processes [79]. It has been described that in poorly nourished children with short stature, despite increased ghrelin synthesis, low IGF-1 concentrations are observed [80].

As it has been explained above in detail, the GH intracellular signal transduction from the GH receptor leads through the JAK2/STAT5β pathway and results in IGF-1 synthesis. The GH signal transduction via the JAK2/STAT5β pathway may be impaired at different stages and due to various reasons. Besides the mutations that may occur in the *GHR*, *JAK2*, or *STAT5β* genes and result in a severe primary IGF-1 deficiency and short stature [81–83], there are some malfunctions of the GHR-JAK2-STAT pathway which may be caused by many modulators, some of which may be linked to eating habits or result from the action of other modifiable factors, one of them being SIRT1 [5]. Yamamoto et al. [15] proved SIRT1's role in that adaptation, concluding that SIRT1 regulates GH-induced IGF-1 production in the liver, thereby reducing the serum concentration of IGF-1. Also, SIRT1 negatively regulates GH-induced IGF-1 mRNA production, which was confirmed in a human hepatocellular carcinoma cell line (HepG2) and rat primary hepatocytes, with the use of SIRT1 inhibitors (sirtinol and nicotinamide) and stimulators (resveratrol and NAD) [15].

In the fed condition, the Src Homology 2 (SH2) domain of STAT5 recognizes and binds to tyr-phosphorylated GHR, causing JAK2 to phosphorylate and activate STAT5. In the fasting condition, activated SIRT1 interacts with STAT5 by deacetylating the lys residues adjacent to the SH2 domain of STAT5. As a result, the activation of STAT5 is inhibited by the impaired ability to bind tyr-phosphorylated GHR. In turn, fasting-induced GH resistance, as well as tyr-phosphorylation of STAT5 are restored by treatment with a SIRT1 inhibitor—nicotinamide [15].

SIRT1 also exerts an inhibitory effect on STAT3 activity [53]. It was demonstrated that treating the immobilized mouse hepatocytes of a SV40 cell line with nicotinamide increased the level of acetylation and phosphorylation of STAT3, and its activity was independent of JAK2 [53]. The levels of STAT3 acetylation and phosphorylation are constitutively higher in the SIRT1 knockout mice embryonic fibroblasts (MEFs) than in the wild-type ones. The cell culture treatment with a SIRT1 inhibitor (EX527) increases, while SIRT1 activator (resveratrol) decreases, STAT3 acetylation and phosphorylation only in wild-type MEFs, but not in SIRT1 knockout mice MEFs. Thus, deacetylation of STAT3 also depends on SIRT1 [53].

Summing up, it has been found that under fasting conditions, SIRT1 can inhibit GH signalling by interacting with STAT3 and/or STAT5, and in this way negatively regulates GH-induced IGF-1 production. SIRT1-dependent negative regulation of GH-induced IGF-1 production seems to be an adaptive mechanism under fasting conditions and malnutrition. However, the role of SIRT1 in the regulation of the GH–IGF-1 axis occurs not only at the level of the liver, but also in the central nervous system.

7. SIRT1 in the Hypothalamic–Pituitary Axis

In addition to the metabolic functions of SIRT1 discussed above and its involvement in GH-induced IGF-1 synthesis in hepatocytes, SIRT1 also plays an important role in the regulatory mechanisms in the hypothalamus and in the hypothalamic–pituitary axis [8].

SIRT1 mRNA is widely expressed throughout the central nervous system. It shows nuclear localization in the neurons of the hypothalamus, hippocampus, and extranuclear localization in the neurons of the dorsal regions of cerebral cortex. In the hypothalamus, SIRT1 is expressed in the arcuate (ARC), ventromedial (VMH), dorsomedial (DMH), lateral (LH), and paraventricular (PVN) nuclei, brain regions that all play critical roles in the central regulation of the adaptive responses to food availability, energy expenditure, thermoregulation, as well as the synthesis of GHRH and GHIH. SIRT1 is also expressed in the suprachiasmatic nucleus (SCN), a region important for the central regulation of circadian rhythms [84,85].

For over a dozen years, intensive research has been conducted (mainly on an animal model) on the role of SIRT1 in the regulation of the hunger and satiety centres, as well as on the impact of its deficiency and excess in this region on behaviour regarding food intake and physical activity, as well as on energy expenditure. As a result of these studies, SIRT1 appears to mediate several processes controlled by the hypothalamus. Caloric restriction and fasting have been found to increase SIRT1 expression and activity, not only in hepatocytes but also in the hypothalamus [84–87]. It has already been proven that SIRT1's effects on the energy balance are mediated through melanocortin signalling. The hypothalamic part of the central melanocortin system is located in the ARC and engaged in the regulation of food intake, energy expenditure, and metabolism [88]. SIRT1 regulates the expression of both agouti-related protein (AgRP) and proopiomelanocortin (POMC) neurons, which produce neuropeptides that are responsible for appetite stimulation (the oryxogenic effect) and food intake reduction (anorexigenic effect), respectively [86,87,89–91]. During fasting, SIRT1 induces food intake (orexigenic effect via AgRP activity). The inhibition of hypothalamic SIRT1 activity resulted in decreased AgRP and increased POMC expressions [86,90]. In the feeding state, SIRT1 decreased the expression of the orexigenic neuropeptide agouti-related peptide and reduced food intake [87].

The overexpression of SIRT1 in POMC neurons was associated with increased sympathetic activity in adipose tissue, leading to increased energy expenditure and a lean phenotype [89]. Surprisingly, the overexpression of SIRT1 in AgRP neurons also suppressed food intake [85,89].

SIRT1 also plays an important role in the neurobehavioral adaptation to energy limitation and in maintaining homeostasis. The overexpression of brain-specific SIRT1 in transgenic mice resulted in the increased activation of neurons in the DMH and LH, maintenance of a higher body temperature range, and increased physical activity in response to various dietary restrictive conditions [92]. In contrast, SIRT1-deficient mice exhibited defects in neurobehavioral adaptation to diet-restricting conditions [85].

It seems that hypothalamic SIRT1 acts as a modulator of GH signalling. SIRT1 brain-specific knockout mice were dwarfed and had reduced somatotropic signalling—both GH and IGF-1 were decreased [93]. Although the central SIRT1/p53 pathway mediates the orexigenic action of ghrelin, blocking this pathway does not modify ghrelin-induced GH secretion [94]. The injection of a SIRT1 inhibitor blunted the effect of ghrelin on food intake, whereas it did not change ghrelin-induced increase in plasma GH levels [94]. This data suggest that at the central level, SIRT1 may play a role in the regulation of GH signalling in other mechanisms.

Growth hormone modulates the neuroendocrine responses to food deprivation via AgRP neurons [95]. AgRP-specific GHR ablation mitigates the fasting-induced activation of AgRP/NPY neurons and neuroendocrine energy-saving adaptations to caloric restriction [95]. SIRT1 expression is present in the AgRP neurons that express GHR [96]. In the fasted state, SIRT1 expression increases in AgRP neurons, whereas, in animals with GHR deletion in AgRP neurons, this response is attenuated. Thus, SIRT1 appears to act as a hypothalamic mediator of the GHR signaling in the adaptive responses to fasting [96].

SIRT1 also mediates the effects of the supraphysiological GH levels. SIRT1 levels were elevated not only in caloric-restricted mice but also in mice overexpressing GH. However,

in GH receptor knockout (GHRKO) mice, despite low IGF-1, the SIRT1 protein levels were not increased [97].

On the other hand, SIRT1 has been shown to inhibit the transcription factor cAMP response element-binding protein (CREB) and negatively regulate pituitary GH synthesis [98].

In conclusion, the presented data indicate that the region-specific expression of SIRT1 plays a significant role in the regulation of appetite, energy expenditure, general metabolism, and growth. SIRT1 has been found to modulate the hypothalamus–pituitary axis.

8. SIRT1 in IUGR and in Short Stature Children

The negative relation between SIRT1 and IGF-1 in the context of growth was observed in intrauterine growth restriction (IUGR) cases. Chriett et al. [99] examined the impact of IUGR on hormones and gene expression in pig skeletal muscle. The gene expression trajectories of the sirtuins and metabolic genes were altered in IUGR and correlated to IGF-1 dysregulation. Higher *SIRT1* gene expression and lower IGF-1 levels were observed in the pig IUGR group [99]. The catch-up growth model in children born small for gestational age (SGA) has been discussed by Griffin [59]. If the fetus is malnourished during pregnancy, the secretion of ghrelin increases, which stimulates the hunger centre, as well as the secretion of GH. This should increase the IGF-1 pool. However, at the same time, caloric restriction induces the synthesis of SIRT1 and FGF21, which block GH signal transduction in the liver by obstructing the JAK/STAT pathway, thus limiting IGF-1 production. Thus, despite a high level of GH, intrauterine growth retardation is observed. Postpartum, as nutrients become available, the SIRT1 and FGF21 levels decline, the liver sensitivity to GH returns to normal, and IGF-1 production resumes. This thesis is confirmed by the fact that in prepubertal children with SGA who have undergone the catch-up phenomenon (with currently normal height), the concentrations of both ghrelin and IGF-1 are significantly higher than in children with SGA without the catch-up phenomenon (with permanent short stature) [100].

So far, there have been few studies on the SIRT1 levels in short stature children. Interestingly, in the study performed by Kaplan et al. [101] on short stature children, both the SIRT1 and IGF-1 serum levels were decreased in the untreated with GH short stature children compared to the GH treatment children. However, it is not clear whether among the children with short stature there were children with GHD or ISS, and the BMI SDS was lower in the study groups.

It should also be taken into account that the serum levels of SIRT1 may not reflect its activity in different tissues and organs. One of these tissues is the epiphyseal growth plate, because the effect of SIRT1 on the expression and production of IGF-1 in chondrocytes will be important for the longitudinal growth of children.

9. SIRT1 Activity in Chondrocytes

Chondrocytes in the growth plate are influenced by various regulatory factors and hormones that together determine the rate of proliferation and maturation until late puberty when the growth plate fuses [102]. Among many others (i.e., estrogens, androgens, or PTHrP), both the GH receptors and the IGF-1 receptors are expressed on human growth plate chondrocytes [86]. The data suggest that SIRT1 may have a positive impact on the growth plate in chondrocytes. Gabay et al. [103] established that SIRT1 enzymatic activity is needed for cartilage homeostasis—they demonstrated that mice with defective SIRT1 also had defective cartilage, with elevated rates of cartilage degradation with age [103]. Shtaif et al. investigated the role of SIRT1 in modulating the response of the epiphyseal growth plate to nutritional manipulation [11]. In collagen type II-specific SIRT1 knockout mice the epiphyseal growth plate was less organized and catch-up growth was less efficient [11]. SIRT1 appears to be important in the proper regulation of chondrogenesis. In the study presented by Jin et al., SIRT1 ablation inhibited growth plate chondrogenesis and contributed to growth retardation through the hyperactivation of mammalian target of rapamycin complex 1 (mTORC1) signaling [104]. Whereas, the findings reported by Kang

et al. indicate that SIRT1 deacetylates protein kinase-like endoplasmic reticulum kinase (PERK) and attenuates the PERK–Eukaryotic Initiation Factor 2 (eIF-2)–C/EBP-homologous protein (CHOP) pathway in chondrocytes and thus promotes growth plate chondrogenesis and longitudinal bone growth [105]. Authors confirmed that SIRT1 expression in growth plate facilitates chondrocyte proliferation and hypertrophy as well as prevents apoptosis [104,105]. Conversely, it was found that DNA damage-inducible transcript 3 (DDIT3)/CHOP interfere with SIRT1 to stimulate autophagy [106] as well as SIRT1 directly triggers autophagy in chondrocytes [107]. Moreover, in the condition of endoplasmic reticulum stress, DDIT3/CHOP upregulated SIRT1 and thereby had an inhibitory effect on chondrocyte differentiation and matrix synthesis.

Interestingly, studies have demonstrated the direct link between epiphyseal growth plate micro RNA and SIRT1. Cheng et al. [108] showed that SIRT1 exerts a protective effect on growth plate chondrocytes under dexamethasone stimulation. MiR-211-5p downregulated SIRT1 in growth plate chondrocytes treated with dexamethasone. The inhibition of miR-211-5p led to an elevation in SIRT1 expression, subsequently restoring the function of chondrocytes [108]. Whereas, Pando et al. proved that nutrition restriction decreased miR-140 levels leading to the elevation of SIRT1 [109]. Authors have concluded that SIRT1 upregulation may inactivate hypoxia inducible factor 1α (HIF-1α), which is necessary for chondrocyte survival [109–111].

Overall, the data suggest that SIRT1 is involved in growth plate chondrogenesis and longitudinal bone growth as well as chondrocyte homeostasis; however, the exact mechanisms responsible for this process remain to be clarified.

10. Conclusions

In conclusion, SIRT1 is an important factor in the regulation of various biological processes, with particular emphasis on maintaining homeostasis. This review indicates that SIRT1 may modulate growth processes in children, acting differently depending on the cell type and conditions in a tissue- and context-specific manner (Table 1).

Table 1. SIRT1 actions with respect to growth and food supply.

Condition	AgRP Neurons	POMC Neurons	GHR Neurons	Hepatocytes
Feeding	Stabilized SIRT1 expression → decreased AgRP activity → decreased food intake	SIRT1 activity in POMC neurons is required for normal energy expenditure adaptations	SIRT1 is needed for GH synthesis or secretion	Preserved GH signalling
Caloric restriction	High SIRT1 expression → increased AgRP activity → increased food intake	Decreased POMC activity → reduced energy expenditure	High SIRT1 expression → stimulation to increase GH synthesis (for hyperglycaemic and other metabolic effects)	High SIRT1 expression → inhibition of GH signal transduction to decrease IGF-1 synthesis (to reduce hypoglycaemic and growth effects)
Knock-out/inhibition of SIRT1	Decreased AgRP activity → decreased food intake	Increased POMC activity → reduced food intake, increased energy expenditure	Impaired GH signalling → decreased GH and IGF-1	Enhanced GH-induced increase in serum IGF-1
Overexpression of SIRT1	Food intake suppression	Increased energy expenditure	Increased GH synthesis?	Suppressed GH-induced IGF-1 production

It seems that SIRT1 modifies the GH/IGF-1 axis by affecting the transmission of signals in the hypothalamic neurons releasing GHRH, AgRP, and POMC, as well as by

modulating ghrelin action. It promotes the secretion of GH from the pituitary, but during caloric restriction, it inhibits the peripheral action of GH by decreasing the expression and secretion of IGF-1. The purpose of this modulation is to maintain a balance between stimulating and inhibiting growth processes, preserving the energy necessary for important metabolic processes in maintaining homeostasis.

Thus, it appears that in children with malnutrition, the level of SIRT1 is increased to inhibit peripheral IGF-1 production and maintain homeostasis. This, in turn, slows down the growth rate of children and inhibits the maturation and ossification of their epiphyseal cartilages. This mechanism allows for a potential improvement in the final height if the current unfavourable conditions are improved. Considering that SIRT1 activity can be modulated by various factors, it should be noted that sirtuin triggers and inhibitors may be utilized in the future to develop new treatments for children with growth disorders.

11. Methodology

We conducted a search of the literature from reputable databases, including PubMed and Google Scholar. Articles published up to December 2022 were taken into consideration, with later updates in September 2023. The search was limited to English-language articles. Combinations of the following MeSH terms were searched: growth, children, growth hormone, GH, sirtuin 1, SIRT1, sirtuins, insulin-growth like factor 1, IGF-1, STAT5, hypothalamus, hepatocytes, liver, chondrocytes, caloric restriction, fasting, and malnutrition. The initial search was followed by the removal of duplicate articles. The whole paper was read only if the abstract indicated relevant content. We incorporated peer-reviewed articles that directly addressed our research topic. In addition, a reference search was performed to find other important manuscripts. Finally, 111 articles were included in this review. Findings were summarized and discussed in the subsequent sections of this review.

Author Contributions: Conceptualization, A.F. and R.S.; methodology, A.F. and R.S.; validation, R.S.; investigation, A.F. and R.S.; resources, A.F.; writing—original draft preparation, A.F.; writing—review and editing, R.S. and A.L.; visualization, A.F.; supervision, R.S. and A.L. All authors have read and agreed to the published version of the manuscript.

Funding: This study was funded by statutory funds from the Medical University of Lodz, Lodz, Poland (503/1-107-03/503-11-001).

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Ranke, M.B.; Wit, J.M. Growth Hormone—Past, Present and Future. *Nat. Rev. Endocrinol.* **2018**, *14*, 285–300. [CrossRef] [PubMed]
2. Bonnefont, X.; Lacampagne, A.; Sanchez-Hormigo, A.; Fino, E.; Creff, A.; Mathieu, M.-N.; Smallwood, S.; Carmignac, D.; Fontanaud, P.; Travo, P.; et al. Revealing the Large-Scale Network Organization of Growth Hormone-Secreting Cells. *Proc. Natl. Acad. Sci. USA* **2005**, *102*, 16880–16885. [CrossRef] [PubMed]
3. Kojima, M.; Hosoda, H.; Date, Y.; Nakazato, M.; Matsuo, H.; Kangawa, K. Ghrelin Is a Growth-Hormone-Releasing Acylated Peptide from Stomach. *Nature* **1999**, *402*, 656–660. [CrossRef] [PubMed]
4. Nakazato, M.; Murakami, N.; Date, Y.; Kojima, M.; Matsuo, H.; Kangawa, K.; Matsukura, S. A Role for Ghrelin in the Central Regulation of Feeding. *Nature* **2001**, *409*, 194–198. [CrossRef] [PubMed]
5. Lewin´ski, A.; Karbownik-Lewin´ska, M.; Wieczorek-Szukała, K.; Stasiak, M.; Stawerska, R. Contribution of Ghrelin to the Pathogenesis of Growth Hormone Deficiency. *Int. J. Mol. Sci.* **2021**, *22*, 9066. [CrossRef] [PubMed]
6. Juul, A. Serum Levels of Insulin-like Growth Factor I and Its Binding Proteins in Health and Disease. *Growth Horm. IGF Res.* **2003**, *13*, 113–170. [CrossRef] [PubMed]
7. Haigis, M.C.; Sinclair, D.A. Mammalian Sirtuins: Biological Insights and Disease Relevance. *Annu. Rev. Pathol. Mech. Dis.* **2010**, *5*, 253–295. [CrossRef] [PubMed]
8. Yamamoto, M.; Takahashi, Y. The Essential Role of SIRT1 in Hypothalamic-Pituitary Axis. *Front. Endocrinol.* **2018**, *9*, 605. [CrossRef]

9. Boguszewski, C.L.; Barbosa, E.J.L.; Svensson, P.A.; Johannsson, G.; Glad, C.A.M. Mechanisms in Endocrinology: Clinical and Pharmacogenetic Aspects of the Growth Hormone Receptor Polymorphism. *Eur. J. Endocrinol.* **2017**, *177*, R309–R321. [CrossRef]
10. Waters, M.J.; Brooks, A.J. JAK2 Activation by Growth Hormone and Other Cytokines. *Biochem. J.* **2015**, *466*, 1–11. [CrossRef]
11. Shtaif, B.; Bar-Maisels, M.; Gabet, Y.; Hiram-Bab, S.; Yackobovitch-Gavan, M.; Phillip, M.; Gat-Yablonski, G. Cartilage -Specific Knockout of Sirt1 Significantly Reduces Bone Quality and Catch-up Growth Efficiency. *Bone* **2020**, *138*, 115468. [CrossRef] [PubMed]
12. Chen, C.; Zhou, M.; Ge, Y.; Wang, X. SIRT1 and Aging Related Signaling Pathways. *Mech. Ageing Dev.* **2020**, *187*, 111215. [CrossRef] [PubMed]
13. Lee, S.-H.; Lee, J.-H.; Lee, H.-Y.; Min, K.-J. Sirtuin Signaling in Cellular Senescence and Aging. *BMB Rep.* **2019**, *52*, 24–34. [CrossRef]
14. Yakar, S.; Liu, J.-L.; Stannard, B.; Butler, A.; Accili, D.; Sauer, B.; LeRoith, D. Normal Growth and Development in the Absence of Hepatic Insulin-like Growth Factor I. *Proc. Natl. Acad. Sci. USA* **1999**, *96*, 7324–7329. [CrossRef] [PubMed]
15. Yamamoto, M.; Iguchi, G.; Fukuoka, H.; Suda, K.; Bando, H.; Takahashi, M.; Nishizawa, H.; Seino, S.; Takahashi, Y. SIRT1 Regulates Adaptive Response of the Growth Hormone–Insulin-like Growth Factor-I Axis under Fasting Conditions in Liver. *Proc. Natl. Acad. Sci. USA* **2013**, *110*, 14948–14953. [CrossRef]
16. Dehkhoda, F.; Lee, C.M.M.; Medina, J.; Brooks, A.J. The Growth Hormone Receptor: Mechanism of Receptor Activation, Cell Signaling, and Physiological Aspects. *Front. Endocrinol.* **2018**, *9*, 35. [CrossRef] [PubMed]
17. Rosenfeld, R.G.; Hwa, V. Biology of the Somatotroph Axis (after the Pituitary). *Ann. Endocrinol.* **2017**, *78*, 80–82. [CrossRef] [PubMed]
18. Wójcik, M.; Krawczyn´ska, A.; Antushevich, H.; Herman, A.P. Post-Receptor Inhibitors of the GHR-JAK2-STAT Pathway in the Growth Hormone Signal Transduction. *Int. J. Mol. Sci.* **2018**, *19*, 1843. [CrossRef]
19. Savage, M.O.; Burren, C.P.; Rosenfeld, R.G. The Continuum of Growth Hormone–IGF-I Axis Defects Causing Short Stature: Diagnostic and Therapeutic Challenges. *Clin. Endocrinol.* **2010**, *72*, 721–728. [CrossRef]
20. Smyczyn´ska, J.; Smyczyn´ska, U.; Hilczer, M.; Stawerska, R.; Lewin´ski, A. Significance of Direct Confirmation of Growth Hormone Insensitivity for the Diagnosis of Primary IGF-I Deficiency. *J. Clin. Med.* **2020**, *9*, 240. [CrossRef]
21. Clemmons, D.R. Metabolic Actions of Insulin-like Growth Factor-I in Normal Physiology and Diabetes. *Endocrinol. Metab. Clin. N. Am.* **2012**, *41*, 425–443. [CrossRef] [PubMed]
22. Al-Samerria, S.; Radovick, S. The Role of Insulin-like Growth Factor-1 (IGF-1) in the Control of Neuroendocrine Regulation of Growth. *Cells* **2021**, *10*, 2664. [CrossRef] [PubMed]
23. Romero, C.J.; Ng, Y.; Luque, R.M.; Kineman, R.D.; Koch, L.; Bruning, J.C.; Radovick, S. Targeted Deletion of Somatotroph Insulin-Like Growth Factor-I Signaling in a Cell-Specific Knockout Mouse Model. *Mol. Endocrinol.* **2010**, *24*, 1077–1089. [CrossRef] [PubMed]
24. Hwa, V.; Oh, Y.; Rosenfeld, R.G. The Insulin-like Growth Factor-Binding Protein (IGFBP) Superfamily. *Endocr. Rev.* **1999**, *20*, 761–787. [CrossRef] [PubMed]
25. Jones, J.I.; Clemmons, D.R. Insulin-like Growth Factors and Their Binding Proteins: Biological Actions. *Endocr. Rev.* **1995**, *16*, 3–34. [CrossRef] [PubMed]
26. Møller, N.; Jørgensen, J.O.L. Effects of Growth Hormone on Glucose, Lipid, and Protein Metabolism in Human Subjects. *Endocr. Rev.* **2009**, *30*, 152–177. [CrossRef] [PubMed]
27. Vijayakumar, A.; Novosyadlyy, R.; Wu, Y.; Yakar, S.; LeRoith, D. Biological Effects of Growth Hormone on Carbohydrate and Lipid Metabolism. *Growth Horm. IGF Res.* **2010**, *20*, 1–7. [CrossRef]
28. Blum, W.F.; Albertsson-Wikland, K.; Rosberg, S.; Ranke, M.B. Serum Levels of Insulin-like Growth Factor I (IGF-I) and IGF Binding Protein 3 Reflect Spontaneous Growth Hormone Secretion. *J. Clin. Endocrinol. Metab.* **1993**, *76*, 1610–1616. [CrossRef]
29. Stawerska, R.; Kolasa-Kicin´ska, M.; Kolejwa, M.; Smyczyn´ska, J.; Hilczer, M.; Czkwianianc, E.; Lewin´ski, A. Frequency of Oligosymptomatic Gastrointestinal Tract Diseases and Its Relation to Insulin-like Growth Factor I in Idiopathic (Non-GH-Deficient) Short Stature Children. *Arch. Med. Sci.* **2021**, *17*, 1663–1671. [CrossRef]
30. Misra, M.; Miller, K.K.; Bjornson, J.; Hackman, A.; Aggarwal, A.; Chung, J.; Ott, M.; Herzog, D.B.; Johnson, M.L.; Klibanski, A. Alterations in Growth Hormone Secretory Dynamics in Adolescent Girls with Anorexia Nervosa and Effects on Bone Metabolism. *J. Clin. Endocrinol. Metab.* **2003**, *88*, 5615–5623. [CrossRef]
31. Imai, S.; Guarente, L. NAD+ and Sirtuins in Aging and Disease. *Trends Cell Biol.* **2014**, *24*, 464–471. [CrossRef] [PubMed]
32. Nogueiras, R.; Habegger, K.M.; Chaudhary, N.; Finan, B.; Banks, A.S.; Dietrich, M.O.; Horvath, T.L.; Sinclair, D.A.; Pfluger, P.T.; Tschöop, M.H. Sirtuin 1 and Sirtuin 3: Physiological Modulators of Metabolism. *Physiol. Rev.* **2012**, *92*, 1479–1514. [CrossRef]
33. Moynihan, K.A.; Grimm, A.A.; Plueger, M.M.; Bernal-Mizrachi, E.; Ford, E.; Cras-Méneur, C.; Permutt, M.A.; Imai, S. Increased Dosage of Mammalian Sir2 in Pancreatic β Cells Enhances Glucose-Stimulated Insulin Secretion in Mice. *Cell Metab.* **2005**, *2*, 105–117. [CrossRef] [PubMed]
34. Xie, L.; Huang, R.; Liu, S.; Wu, W.; Su, A.; Li, R.; Liu, X.; Lei, Y.; Sun, H.; Liu, X.; et al. A Positive Feedback Loop of SIRT1 and miR17HG Promotes the Repair of DNA Double-Stranded Breaks. *Cell Cycle* **2019**, *18*, 2110–2123. [CrossRef] [PubMed]
35. Alves-Fernandes, D.K.; Jasiulionis, M.G. The Role of SIRT1 on DNA Damage Response and Epigenetic Alterations in Cancer. *Int. J. Mol. Sci.* **2019**, *20*, 3153. [CrossRef] [PubMed]

36. Kauppinen, A.; Suuronen, T.; Ojala, J.; Kaarniranta, K.; Salminen, A. Antagonistic Crosstalk between NF-κB and SIRT1 in the Regulation of Inflammation and Metabolic Disorders. *Cell. Signal.* **2013**, *25*, 1939–1948. [CrossRef] [PubMed]
37. Pang, J.; Xiong, H.; Ou, Y.; Yang, H.; Xu, Y.; Chen, S.; Lai, L.; Ye, Y.; Su, Z.; Lin, H.; et al. SIRT1 Protects Cochlear Hair Cell and Delays Age-Related Hearing Loss via Autophagy. *Neurobiol. Aging* **2019**, *80*, 127–137. [CrossRef] [PubMed]
38. Hori, Y.S.; Kuno, A.; Hosoda, R.; Horio, Y. Regulation of FOXOs and P53 by SIRT1 Modulators under Oxidative Stress. *PLoS ONE* **2013**, *8*, e73875. [CrossRef]
39. Mercken, E.M.; Mitchell, S.J.; Martin-Montalvo, A.; Minor, R.K.; Almeida, M.; Gomes, A.P.; Scheibye-Knudsen, M.; Palacios, H.H.; Licata, J.J.; Zhang, Y.; et al. SRT2104 Extends Survival of Male Mice on a Standard Diet and Preserves Bone and Muscle Mass. *Aging Cell* **2014**, *13*, 787–796. [CrossRef]
40. Hwang, J.; Yao, H.; Caito, S.; Sundar, I.K.; Rahman, I. Redox Regulation of SIRT1 in Inflammation and Cellular Senescence. *Free Radic. Biol. Med.* **2013**, *61*, 95–110. [CrossRef]
41. Chan, S.-H.; Hung, C.-H.; Shih, J.-Y.; Chu, P.-M.; Cheng, Y.-H.; Lin, H.-C.; Hsieh, P.-L.; Tsai, K.-L. Exercise Intervention Attenuates Hyperhomocysteinemia-Induced Aortic Endothelial Oxidative Injury by Regulating SIRT1 through Mitigating NADPH Oxidase/LOX-1 Signaling. *Redox Biol.* **2018**, *14*, 116–125. [CrossRef]
42. Yamakuchi, M. MicroRNA Regulation of SIRT1. *Front. Physiol.* **2012**, *3*, 68. [CrossRef] [PubMed]
43. Du, S.; Shen, S.; Ding, S.; Wang, L. Suppression of microRNA-323-3p Restrains Vascular Endothelial Cell Apoptosis via Promoting Sirtuin-1 Expression in Coronary Heart Disease. *Life Sci.* **2021**, *270*, 119065. [CrossRef] [PubMed]
44. Chan, S.-H.; Hung, C.-H.; Shih, J.-Y.; Chu, P.-M.; Cheng, Y.-H.; Lin, H.-C.; Tsai, K.-L. SIRT1 Inhibition Causes Oxidative Stress and Inflammation in Patients with Coronary Artery Disease. *Redox Biol.* **2017**, *13*, 301–309. [CrossRef] [PubMed]
45. Chang, H.-C.; Guarente, L. SIRT1 and Other Sirtuins in Metabolism. *Trends Endocrinol. Metab.* **2014**, *25*, 138–145. [CrossRef] [PubMed]
46. Purushotham, A.; Schug, T.T.; Xu, Q.; Surapureddi, S.; Guo, X.; Li, X. Hepatocyte-Specific Deletion of SIRT1 Alters Fatty Acid Metabolism and Results in Hepatic Steatosis and Inflammation. *Cell Metab.* **2009**, *9*, 327–338. [CrossRef] [PubMed]
47. Walker, A.K.; Yang, F.; Jiang, K.; Ji, J.Y.; Watts, J.L.; Purushotham, A.; Boss, O.; Hirsch, M.L.; Ribich, S.; Smith, J.J.; et al. Conserved Role of SIRT1 Orthologs in Fasting-Dependent Inhibition of the Lipid/Cholesterol Regulator SREBP. *Genes. Dev.* **2010**, *24*, 1403–1417. [CrossRef] [PubMed]
48. Ding, R.-B.; Bao, J.; Deng, C.-X. Emerging Roles of SIRT1 in Fatty Liver Diseases. *Int. J. Biol. Sci.* **2017**, *13*, 852–867. [CrossRef] [PubMed]
49. Lomb, D.J.; Laurent, G.; Haigis, M.C. Sirtuins Regulate Key Aspects of Lipid Metabolism. *Biochim. Biophys. Acta* **2010**, *1804*, 1652–1657. [CrossRef]
50. Gerhart-Hines, Z.; Rodgers, J.T.; Bare, O.; Lerin, C.; Kim, S.H.; Mostoslavsky, R.; Alt, F.W.; Wu, Z.; Puigserver, P. Metabolic Control of Muscle Mitochondrial Function and Fatty Acid Oxidation through SIRT1/PGC-1α. *EMBO J.* **2007**, *26*, 1913–1923. [CrossRef]
51. Picard, F.; Kurtev, M.; Chung, N.; Topark-Ngarm, A.; Senawong, T.; Machado De Oliveira, R.; Leid, M.; McBurney, M.W.; Guarente, L. Sirt1 Promotes Fat Mobilization in White Adipocytes by Repressing PPAR-γ. *Nature* **2004**, *429*, 771–776. [CrossRef]
52. Qiang, L.; Wang, L.; Kon, N.; Zhao, W.; Lee, S.; Zhang, Y.; Rosenbaum, M.; Zhao, Y.; Gu, W.; Farmer, S.R.; et al. Brown Remodeling of White Adipose Tissue by SirT1-Dependent Deacetylation of Pparγ. *Cell* **2012**, *150*, 620–632. [CrossRef] [PubMed]
53. Nie, Y.; Erion, D.M.; Yuan, Z.; Dietrich, M.; Shulman, G.I.; Horvath, T.L.; Gao, Q. STAT3 Inhibition of Gluconeogenesis Is Downregulated by SirT1. *Nat. Cell Biol.* **2009**, *11*, 492–500. [CrossRef] [PubMed]
54. Nemoto, S.; Fergusson, M.M.; Finkel, T. SIRT1 Functionally Interacts with the Metabolic Regulator and Transcriptional Coactivator PGC-1α. *J. Biol. Chem.* **2005**, *280*, 16456–16460. [CrossRef] [PubMed]
55. Rodgers, J.T.; Lerin, C.; Haas, W.; Gygi, S.P.; Spiegelman, B.M.; Puigserver, P. Nutrient Control of Glucose Homeostasis through a Complex of PGC-1α and SIRT1. *Nature* **2005**, *434*, 113–118. [CrossRef] [PubMed]
56. Hallows, W.C.; Yu, W.; Denu, J.M. Regulation of Glycolytic Enzyme Phosphoglycerate Mutase-1 by Sirt1 Protein-Mediated Deacetylation. *J. Biol. Chem.* **2012**, *287*, 3850–3858. [CrossRef] [PubMed]
57. Qiao, L.; Shao, J. SIRT1 Regulates Adiponectin Gene Expression through Foxo1-C/Enhancer-Binding Protein α Transcriptional Complex. *J. Biol. Chem.* **2006**, *281*, 39915–39924. [CrossRef] [PubMed]
58. Cao, Y.; Jiang, X.; Ma, H.; Wang, Y.; Xue, P.; Liu, Y. SIRT1 and Insulin Resistance. *J. Diabetes Its Complicat.* **2016**, *30*, 178–183. [CrossRef]
59. Griffin, I.J. Catch-Up Growth: Basic Mechanisms. *Nestle Nutr. Inst. Workshop Ser.* **2015**, *81*, 87–97. [CrossRef]
60. Pacifici, F.; Di Cola, D.; Pastore, D.; Abete, P.; Guadagni, F.; Donadel, G.; Bellia, A.; Esposito, E.; Salimei, C.; Sinibaldi Salimei, P.; et al. Proposed Tandem Effect of Physical Activity and Sirtuin 1 and 3 Activation in Regulating Glucose Homeostasis. *Int. J. Mol. Sci.* **2019**, *20*, 4748. [CrossRef]
61. Howitz, K.T.; Bitterman, K.J.; Cohen, H.Y.; Lamming, D.W.; Lavu, S.; Wood, J.G.; Zipkin, R.E.; Chung, P.; Kisielewski, A.; Zhang, L.-L.; et al. Small Molecule Activators of Sirtuins Extend Saccharomyces Cerevisiae Lifespan. *Nature* **2003**, *425*, 191–196. [CrossRef] [PubMed]
62. Malaguarnera, L. Influence of Resveratrol on the Immune Response. *Nutrients* **2019**, *11*, 946. [CrossRef] [PubMed]
63. Akan, O.D.; Qin, D.; Guo, T.; Lin, Q.; Luo, F. Sirtfoods: New Concept Foods, Functions, and Mechanisms. *Foods* **2022**, *11*, 2955. [CrossRef] [PubMed]
64. Meydani, M.; Hasan, S.T. Dietary Polyphenols and Obesity. *Nutrients* **2010**, *2*, 737–751. [CrossRef] [PubMed]

65. Dai, H.; Sinclair, D.A.; Ellis, J.L.; Steegborn, C. Sirtuin Activators and Inhibitors: Promises, Achievements, and Challenges. *Pharmacol. Ther.* **2018**, *188*, 140–154. [CrossRef] [PubMed]
66. Hwang, E.S.; Song, S.B. Nicotinamide Is an Inhibitor of SIRT1 in Vitro, but Can Be a Stimulator in Cells. *Cell. Mol. Life Sci.* **2017**, *74*, 3347–3362. [CrossRef] [PubMed]
67. Villalba, J.M.; Alcaín, F.J. Sirtuin Activators and Inhibitors. *Biofactors* **2012**, *38*, 349–359. [CrossRef]
68. Alcaín, F.J.; Villalba, J.M. Sirtuin Inhibitors. *Expert Opin. Ther. Pat.* **2009**, *19*, 283–294. [CrossRef]
69. Sokolowska, K.E.; Maciejewska-Markiewicz, D.; Bińkowski, J.; Palma, J.; Taryma-Les´niak, O.; Kozlowska-Petriczko, K.; Borowski, K.; Bas´kiewicz-Hałasa, M.; Hawryłkowicz, V.; Załęcka, P.; et al. Identified in Blood Diet-Related Methylation Changes Stratify Liver Biopsies of NAFLD Patients According to Fibrosis Grade. *Clin. Epigenetics* **2022**, *14*, 157. [CrossRef]
70. Li, X. SIRT1 and Energy Metabolism. *Acta Biochim. Biophys. Sin.* **2013**, *45*, 51–60. [CrossRef]
71. Zhao, Y.; Chen, B.; Shen, J.; Wan, L.; Zhu, Y.; Yi, T.; Xiao, Z. The Beneficial Effects of Quercetin, Curcumin, and Resveratrol in Obesity. *Oxid. Med. Cell. Longev.* **2017**, *2017*, 1459497. [CrossRef] [PubMed]
72. Hofer, S.J.; Carmona-Gutierrez, D.; Mueller, M.I.; Madeo, F. The Ups and Downs of Caloric Restriction and Fasting: From Molecular Effects to Clinical Application. *EMBO Mol. Med.* **2022**, *14*, e14418. [CrossRef] [PubMed]
73. Tabuchi, T.; Satoh, M.; Itoh, T.; Nakamura, M. MicroRNA-34a Regulates the Longevity-Associated Protein SIRT1 in Coronary Artery Disease: Effect of Statins on SIRT1 and microRNA-34a Expression. *Clin. Sci.* **2012**, *123*, 161–171. [CrossRef]
74. Abdelmohsen, K.; Pullmann, R.; Lal, A.; Kim, H.H.; Galban, S.; Yang, X.; Blethrow, J.D.; Walker, M.; Shubert, J.; Gillespie, D.A.; et al. Phosphorylation of HuR by Chk2 Regulates SIRT1 Expression. *Mol. Cell* **2007**, *25*, 543–557. [CrossRef] [PubMed]
75. Zhao, S.; Li, N.; Zhu, Y.; Straub, L.; Zhang, Z.; Wang, M.-Y.; Zhu, Q.; Kusminski, C.M.; Elmquist, J.K.; Scherer, P.E. Partial Leptin Deficiency Confers Resistance to Diet-Induced Obesity in Mice. *Mol. Metab.* **2020**, *37*, 100995. [CrossRef] [PubMed]
76. Hao, X.; Shiromoto, Y.; Sakurai, M.; Towers, M.; Zhang, Q.; Wu, S.; Havas, A.; Wang, L.; Berger, S.; Adams, P.D.; et al. ADAR1 Downregulation by Autophagy Drives Senescence Independently of RNA Editing by Enhancing p16INK4a Levels. *Nat. Cell Biol.* **2022**, *24*, 1202–1210. [CrossRef] [PubMed]
77. Wilhelmi de Toledo, F.; Grundler, F.; Sirtori, C.R.; Ruscica, M. Unravelling the Health Effects of Fasting: A Long Road from Obesity Treatment to Healthy Life Span Increase and Improved Cognition. *Ann. Med.* **2020**, *52*, 147–161. [CrossRef] [PubMed]
78. Thissen, J.P.; Pucilowska, J.B.; Underwood, L.E. Differential Regulation of Insulin-like Growth Factor I (IGF-I) and IGF Binding Protein-1 Messenger Ribonucleic Acids by Amino Acid Availability and Growth Hormone in Rat Hepatocyte Primary Culture. *Endocrinology* **1994**, *134*, 1570–1576. [CrossRef]
79. Fazeli, P.K.; Klibanski, A. Determinants of GH Resistance in Malnutrition. *J. Endocrinol.* **2014**, *220*, R57–R65. [CrossRef]
80. Stawerska, R.; Czkwianianc, E.; Smyczyn´ska, J.; Hilczer, M.; Lewin´ski, A. Nutritional Status in Short Stature Children Is Related to Both Ghrelin and Insulin-like Growth Factor I Concentrations. *J. Pediatr. Gastroenterol. Nutr.* **2017**, *64*, 812–817. [CrossRef]
81. Hwa, V. Human Growth Disorders Associated with Impaired GH Action: Defects in STAT5B and JAK2. *Mol. Cell. Endocrinol.* **2021**, *519*, 111063. [CrossRef]
82. Kofoed, E.M.; Hwa, V.; Little, B.; Woods, K.A.; Buckway, C.K.; Tsubaki, J.; Pratt, K.L.; Bezrodnik, L.; Jasper, H.; Tepper, A.; et al. Growth Hormone Insensitivity Associated with a STAT5b Mutation. *N. Engl. J. Med.* **2003**, *349*, 1139–1147. [CrossRef] [PubMed]
83. Lin, S.; Li, C.; Li, C.; Zhang, X. Growth Hormone Receptor Mutations Related to Individual Dwarfism. *Int. J. Mol. Sci.* **2018**, *19*, 1433. [CrossRef] [PubMed]
84. Ramadori, G.; Lee, C.E.; Bookout, A.L.; Lee, S.; Williams, K.W.; Anderson, J.; Elmquist, J.K.; Coppari, R. Brain SIRT1: Anatomical Distribution and Regulation by Energy Availability. *J. Neurosci.* **2008**, *28*, 9989–9996. [CrossRef] [PubMed]
85. Satoh, A.; Brace, C.S.; Ben-Josef, G.; West, T.; Wozniak, D.F.; Holtzman, D.M.; Herzog, E.D.; Imai, S.-i. SIRT1 Promotes the Central Adaptive Response to Diet Restriction through Activation of the Dorsomedial and Lateral Nuclei of the Hypothalamus. *J. Neurosci.* **2010**, *30*, 10220–10232. [CrossRef]
86. Cakir, I.; Perello, M.; Lansari, O.; Messier, N.J.; Vaslet, C.A.; Nillni, E.A. Hypothalamic Sirt1 Regulates Food Intake in a Rodent Model System. *PLoS ONE* **2009**, *4*, e8322. [CrossRef]
87. Sasaki, T.; Kim, H.-J.; Kobayashi, M.; Kitamura, Y.-I.; Yokota-Hashimoto, H.; Shiuchi, T.; Minokoshi, Y.; Kitamura, T. Induction of Hypothalamic Sirt1 Leads to Cessation of Feeding via Agouti-Related Peptide. *Endocrinology* **2010**, *151*, 2556–2566. [CrossRef]
88. Cone, R.D. Studies on the Physiological Functions of the Melanocortin System. *Endocr. Rev.* **2006**, *27*, 736–749. [CrossRef] [PubMed]
89. Sasaki, T.; Kikuchi, O.; Shimpuku, M.; Susanti, V.Y.; Yokota-Hashimoto, H.; Taguchi, R.; Shibusawa, N.; Sato, T.; Tang, L.; Amano, K.; et al. Hypothalamic SIRT1 Prevents Age-Associated Weight Gain by Improving Leptin Sensitivity in Mice. *Diabetologia* **2014**, *57*, 819–831. [CrossRef]
90. Dietrich, M.O.; Antunes, C.; Geliang, G.; Liu, Z.-W.; Borok, E.; Nie, Y.; Xu, A.W.; Souza, D.O.; Gao, Q.; Diano, S.; et al. Agrp Neurons Mediate Sirt1^{t}s Action on the Melanocortin System and Energy Balance: Roles for Sirt1 in Neuronal Firing and Synaptic Plasticity. *J. Neurosci.* **2010**, *30*, 11815–11825. [CrossRef]
91. Ramadori, G.; Fujikawa, T.; Fukuda, M.; Anderson, J.; Morgan, D.A.; Mostoslavsky, R.; Stuart, R.C.; Perello, M.; Vianna, C.R.; Nillni, E.A.; et al. SIRT1 Deacetylase in POMC Neurons Is Required for Homeostatic Defenses against Diet-Induced Obesity. *Cell Metab.* **2010**, *12*, 78–87. [CrossRef] [PubMed]

92. Satoh, A.; Brace, C.S.; Rensing, N.; Clifton, P.; Wozniak, D.F.; Herzog, E.D.; Yamada, K.A.; Imai, S. Sirt1 Extends Life Span and Delays Aging in Mice through the Regulation of Nk2 Homeobox 1 in the DMH and LH. *Cell Metab.* **2013**, *18*, 416–430. [CrossRef] [PubMed]
93. Cohen, D.E.; Supinski, A.M.; Bonkowski, M.S.; Donmez, G.; Guarente, L.P. Neuronal SIRT1 Regulates Endocrine and Behavioral Responses to Calorie Restriction. *Genes Dev.* **2009**, *23*, 2812–2817. [CrossRef] [PubMed]
94. Velásquez, D.A.; Martínez, G.; Romero, A.; Vázquez, M.J.; Boit, K.D.; Dopeso-Reyes, I.G.; López, M.; Vidal, A.; Nogueiras, R.; Diéguez, C. The Central Sirtuin 1/P53 Pathway Is Essential for the Orexigenic Action of Ghrelin. *Diabetes* **2011**, *60*, 1177–1185. [CrossRef]
95. Furigo, I.C.; Teixeira, P.D.S.; de Souza, G.O.; Couto, G.C.L.; Romero, G.G.; Perelló, M.; Frazão, R.; Elias, L.L.; Metzger, M.; List, E.O.; et al. Growth Hormone Regulates Neuroendocrine Responses to Weight Loss via AgRP Neurons. *Nat. Commun.* **2019**, *10*, 662. [CrossRef] [PubMed]
96. de Lima, J.B.M.; Ubah, C.; Debarba, L.K.; Ayyar, I.; Didyuk, O.; Sadagurski, M. Hypothalamic GHR-SIRT1 Axis in Fasting. *Cells* **2021**, *10*, 891. [CrossRef] [PubMed]
97. Al-Regaiey, K.A.; Masternak, M.M.; Bonkowski, M.; Sun, L.; Bartke, A. Long-Lived Growth Hormone Receptor Knockout Mice: Interaction of Reduced Insulin-like Growth Factor i/Insulin Signaling and Caloric Restriction. *Endocrinology* **2005**, *146*, 851–860. [CrossRef]
98. Monteserin-Garcia, J.; Al-Massadi, O.; Seoane, L.M.; Alvarez, C.V.; Shan, B.; Stalla, J.; Paez-Pereda, M.; Casanueva, F.F.; Stalla, G.K.; Theodoropoulou, M. Sirt1 Inhibits the Transcription Factor CREB to Regulate Pituitary Growth Hormone Synthesis. *FASEB J.* **2013**, *27*, 1561–1571. [CrossRef]
99. Chriett, S.; Le Huërou-Luron, I.; Vidal, H.; Pirola, L. Dysregulation of Sirtuins and Key Metabolic Genes in Skeletal Muscle of Pigs with Spontaneous Intrauterine Growth Restriction Is Associated with Alterations of Circulating IGF-1. *Gen. Comp. Endocrinol.* **2016**, *232*, 76–85. [CrossRef]
100. Stawerska, R.; Szałapska, M.; Hilczer, M.; Lewin´ ski, A. Ghrelin, Insulin-like Growth Factor I and Adipocytokines Concentrations in Born Small for Gestational Age Prepubertal Children after the Catch-up Growth. *J. Pediatr. Endocrinol. Metab.* **2016**, *29*, 939–945. [CrossRef]
101. Kaplan, D.S.; Canak, A.; Isık, E.; Orkmez, M.; Kumru, B. Relationship of Fibroblast Growth Factor 21, Sirtuin 1, Visfatin, and Regulators in Children with Short Stature. *Growth Factors* **2018**, *36*, 172–177. [CrossRef]
102. Emons, J.; Chagin, A.S.; Sävendahl, L.; Karperien, M.; Wit, J.M. Mechanisms of Growth Plate Maturation and Epiphyseal Fusion. *Horm. Res. Paediatr.* **2011**, *75*, 383–391. [CrossRef] [PubMed]
103. Gabay, O.; Sanchez, C.; Dvir-Ginzberg, M.; Gagarina, V.; Zaal, K.J.; Song, Y.; He, X.H.; McBurney, M.W. Sirtuin 1 Enzymatic Activity Is Required for Cartilage Homeostasis in Vivo in a Mouse Model. *Arthritis Rheum.* **2013**, *65*, 159–166. [CrossRef] [PubMed]
104. Jin, X.; Kang, X.; Zhao, L.; Xu, M.; Xie, T.; Li, H.; Li, F.; Qian, Z.; Ma, Z.; Zhang, Y.; et al. Cartilage Ablation of Sirt1 Causes Inhibition of Growth Plate Chondrogenesis by Hyperactivation of mTORC1 Signaling. *Endocrinology* **2019**, *160*, 3001–3017. [CrossRef] [PubMed]
105. Kang, X.; Yang, W.; Wang, R.; Xie, T.; Li, H.; Feng, D.; Jin, X.; Sun, H.; Wu, S. Sirtuin-1 (SIRT1) Stimulates Growth-Plate Chondrogenesis by Attenuating the PERK-eIF-2α-CHOP Pathway in the Unfolded Protein Response. *J. Biol. Chem.* **2018**, *293*, 8614–8625. [CrossRef] [PubMed]
106. Yang, C.; Xu, X.; Dong, X.; Yang, B.; Dong, W.; Luo, Y.; Liu, X.; Wu, Y.; Wang, J. DDIT3/CHOP Promotes Autophagy in Chondrocytes via SIRT1-AKT Pathway. *Biochim. Biophys. Acta Mol. Cell Res.* **2021**, *1868*, 119074. [CrossRef] [PubMed]
107. Sacitharan, P.K.; Bou-Gharios, G.; Edwards, J.R. SIRT1 Directly Activates Autophagy in Human Chondrocytes. *Cell Death Discov.* **2020**, *6*, 41. [CrossRef] [PubMed]
108. Cheng, D.; Zhang, L.; Liang, X. SIRT1 Targeted by miR-211-5p Regulated Proliferation and Apoptosis of Dex-Treated Growth Plate Chondrocytes via Mediating SOX2. *Clin. Exp. Pharmacol. Physiol.* **2023**, *50*, 50–58. [CrossRef]
109. Pando, R.; Even-Zohar, N.; Shtaif, B.; Edry, L.; Shomron, N.; Phillip, M.; Gat-Yablonski, G. MicroRNAs in the Growth Plate Are Responsive to Nutritional Cues: Association between miR-140 and SIRT1. *J. Nutr. Biochem.* **2012**, *23*, 1474–1481. [CrossRef]
110. Lim, J.-H.; Lee, Y.-M.; Chun, Y.-S.; Chen, J.; Kim, J.-E.; Park, J.-W. Sirtuin 1 Modulates Cellular Responses to Hypoxia by Deacetylating Hypoxia-Inducible Factor 1α. *Mol. Cell* **2010**, *38*, 864–878. [CrossRef]
111. Stegen, S.; Laperre, K.; Eelen, G.; Rinaldi, G.; Fraisl, P.; Torrekens, S.; Van Looveren, R.; Loopmans, S.; Bultynck, G.; Vinckier, S.; et al. HIF-1α Metabolically Controls Collagen Synthesis and Modification in Chondrocytes. *Nature* **2019**, *565*, 511–515. [CrossRef]

frontiers | Frontiers in Endocrinology

TYPE Original Research
PUBLISHED 11 March 2024
DOI 10.3389/fendo.2024.1356612

OPEN ACCESS

EDITED BY
Giulio Maltoni,
IRCCS University Hospital of Bologna Sant Orsola Polyclinic, Italy

REVIEWED BY
Gerdi Tuli,
Regina Margherita Hospital, Italy
Agnieszka Domin,
University of Rzeszow, Poland

*CORRESPONDENCE
Renata Stawerska
renata.stawerska@icloud.com

RECEIVED 15 December 2023
ACCEPTED 21 February 2024
PUBLISHED 11 March 2024

CITATION
Fedorczak A, Lewin´ ski A and Stawerska R (2024) Sirtuin 1 serum concentration in healthy children - dependence on sex, age, stage of puberty, body weight and diet. *Front. Endocrinol.* 15:1356612. doi: 10.3389/fendo.2024.1356612

Sirtuin 1 serum concentration in healthy children - dependence on sex, age, stage of puberty, body weight and diet

Anna Fedorczak[1], Andrzej Lewin´ ski[1,2] and Renata Stawerska[1,2]*

[1]Department of Endocrinology and Metabolic Diseases, Polish Mother's Memorial Hospital – Research Institute, Lodz, Poland, [2]Department of Paediatric Endocrinology, Medical University of Lodz, Lodz, Poland

Introduction: Sirtuin 1 (SIRT1) is known to be involved in sensing cellular energy levels and regulating energy metabolism. This study aimed to evaluate fasting serum SIRT1 levels in healthy children, and to analyse the influence of age, sex, puberty, body weight, height, and diet on its concentration.

Methods: 47 healthy children aged 4-14 with weight and height within normal range and no chronic disease were included into the study. Fasting serum SIRT1 concentrations were estimated by Enzyme Linked Immunosorbent Assay (ELISA).

Results: Results showed that serum SIRT1 concentrations in healthy children did not differ with respect to sex, age, height, weight and puberty. Whereas, it appeared that a higher frequency of fruits, vegetables and dairy products consumption was associated with an increase in serum SIRT1 levels.

Discussion: Studying SIRT1 in the context of children's health may have implications for a broader understanding of growth processes, pubertal development, metabolic disorders and nutrition.

KEYWORDS

sirtuin 1, growth, puberty, IGF-1, diet, healthy children

1 Introduction

The sirtuins are a family of nicotinamide adenine dinucleotide (NAD+)-dependent deacylases that regulate many cellular processes (1). Among the seven currently known types of sirtuins, sirtuin 1 (SIRT1) is involved in the cellular signal transduction and metabolism, DNA repair, inflammation as well as regulation of cellular senescence and aging (2–11). Despite numerous experimental studies on the importance of SIRT1 in the human body, there is very limited data on the level of blood concentration of this protein in humans. It should be noted that SIRT1 is directly engaged in intracellular GH signal

transmission for IGF-1 secretion via modulation of JAK2/STAT pathway, also in the regulation of GH release in the central nervous system, as well as growth-plate chondrogenesis and longitudinal bone growth (12–15). Thus, it can be assumed that the concentration of SIRT1 should vary in dependence on age and growth rate in children.

On the other hand, it is known that changes in SIRT1 concentration affects the function of the hunger and satiety center, and the use of sirtuin activators promotes weight loss in obese individuals (16). Whereas in a fasting state, caloric restriction or malnutrition, SIRT1 intensifies catabolic processes and inhibits anabolic processes to maintain homeostasis (17–19). SIRT1 is also involved in regulating puberty (20). Moreover, there are some natural substances contained in daily consumed foods that act as sirtuin triggers (21–24). Thus, its concentration also probably varies depending on the body mass, stage of puberty, as well as diet.

The aim of this study was to evaluate fasting serum SIRT1 concentration in healthy children, and to analyse the influence of age, sex, body height, body mass and stage of puberty, as well as dietary habits and type and amount of nutrients intake.

2 Materials and methods

2.1 Study group

From among the children admitted to the Polish Mother Memorial Hospital - Research Institute in Lodz, Poland, the study group included those children who did not have any known healthy problems and who did meet the inclusion criteria and did not meet the exclusion criteria.

Inclusion criteria:

aged: 4-16 years, height and weight in the reference range (3rd-97th percentile for age and sex based on local percentile charts), completing a nutritional questionnaire, written consent of the legal representative to participate in the study.

Exclusion criteria:

chronic health problems which may influence the results (e.g. chronic diseases of the gastrointestinal tract, respiratory system, circulatory system, endocrine system, anorexia nervosa, undernutrition, obesity, short stature, excessive height, genetic disorders and syndromes), acute illness, no written consent of the legal representative to participate in the study.

Finally, 47 children were enrolled into the study group.

2.2 Auxological assessment

In each child: a detailed medical history was collected and a physical examination was performed. Height and weight measurements were taken in the morning on the day of hospital admission by the clinicians involved in this study (AF or RS). Children's height was measured with an accuracy of 1 mm using a Harpenden stadiometer. Children were measured without shoes, with their heads in the Frankfort plane and their feet together. The measurement was performed three times and the average value was taken. Body weight was measured using an electronic scale, with an accuracy of 100 grams. During the measurement, the child was in underwear. Based on results of body height and weight, the standard deviation scores (SDS) in relation to the reference values for age and sex were calculated: for height - height standard deviation score (height SDS) and for body mass - weight SDS. Also, the body mass index (BMI) was calculated and expressed as BMI SDS with respect to the reference values for age and sex for Polish population (25, 26). The puberty stage was assessed according to Tanner scale (27).

2.3 Laboratory methods

The blood samples were taken in fasting state in the morning, to measure the routinely determined basic biochemical parameters as well as the concentrations of IGF-1 and IGFBP-3. An additional blood sample (2.4 ml) was taken in fasting state in the morning, for the determination of SIRT1 serum concentration. After collection, the blood was centrifuged to obtain serum. The serum with no signs of haemolysis was then frozen and stored at the appropriate temperature (see below), as required by the test manufacturer, until SIRT1 analysis.

All measurements were performed at the Centre for Medical Laboratory Diagnostics and Screening of the Polish Mother's Memorial Hospital – Research Institute in Lodz, Poland.

SIRT1 concentration was determined by double-binding immunoenzymatic assay (ELISA) using 2 Human NAD-dependent deacetylase Sirtuin-1 (SIRT1/SIR2L1) ELISA Kits (Cusabio, Houston, TX, USA), according to the manufacturer's instructions (User Manual; catalogue number: CSB-E15058h). The concentration of each sample was measured in duplicate. The sensitivity of the assay is 0.03 ng/ml, while the manufacturer's guaranteed detection range of the assay is 0.15 ng/ml - 10 ng/ml, with an intra-assay coefficient of variation of less than 8% and an inter-assay coefficient of variation of less than 10%.

IGF-1 and IGFBP-3 concentrations were assessed using Immulite, DPC assays. For IGF-1, the WHO NIBSC 1st IRR 87/518 standard was used, with an analytical sensitivity of 20 ng/mL, a calibration ranges up to 1600 ng/mL, an intra-assay coefficient of variation: 3.1-4.3% and inter-assay coefficient of variation CV: 5,8-8,4%. The assay to assess IGFBP-3 was calibrated to the WHO NIBSC Reagent 93/560 standard, with an analytical sensitivity of 0.02 mg/mL, a calibration ranges up to 426 mg/mL, an intra-assay coefficient of variation of 3.5-5.6%, and an inter-assay coefficient of variation of 7.5-9.9%. IGF-1 concentration was expressed in terms of standard deviation for sex and age (IGF-I SDS), according to reference data (28). The molar ratio of IGF-1/IGFBP-3 was calculated assuming a molecular weight for IGF-1 of 7.5kDa and for IGFBP-3 of 42.0 kDa. The molar ratio of IGF-1 to IGFBP-3 is considered an indicator of the bioavailability of IGF-1 (29).

2.4 Assessment of children's dietary habits and type and frequency of nutrients intake

The assessment of the children's dietary habits was carried out through dietary survey with parent, wherein the frequency of their offspring's consumption of particular food categories over the preceding month was examined. The survey was administered by a physician-researcher. The survey was developed based on the CoCu Questionnaire validated on a population of german children (30). After obtaining permission from the authors to use the questionnaire, it was translated bilaterally, and then both versions were checked for compatibility. The survey consisted of two parts. The first part contained 14 questions about the composition of the diet and the second part of the questionnaire contained questions about eating habits and food culture. The parent was asked to rate how many servings of various foods the child consumes per day (fruits or vegetables, unsweetened dairy products, sweetened dairy products, sweetened beverages, whole wheat bread, white bread) or per week (meat, fish, french fries, potatoes, rice or pasta, ready meals, pastries, sweet or salty snacks). Reference portions were described in the text or illustrated with photos. The selection of food items was largely based on the Food Frequency Questionnaire FFQ and the healthy eating pyramid (31, 32). The second part of the survey included questions about eating habits (i.e. whether they follow any specific diet, number of meals they have per day). The questionnaire was included in the Supplementary Materials.

2.5 Statistical analysis

Statistical analysis of the collected data was performed using STATISTICA ver. 13.3 software (Statsoft, Poland). The Shapiro-Wilk test was used to assess normality of distribution, and the Levene's test was used to assess equality of variance. Comparative analysis was performed using non-parametric tests for independent variables. Non-parametric The Kruskal–Wallis test by ranks and Mann-Whitney U test were used for intergroup comparisons of quantitative continuous variables. Intergroup comparisons of nominal/qualitative variables were performed using the Chi-square test. In addition, a correlation analysis of the variables was performed (Pearson's correlation coefficient). Continuous variables were presented median and interquartile ranges (median (Q1-Q3)) and range, categorical variables by N (%)). Statistically significant differences were taken as p-values below 0.05.

2.6 Ethics approval

Approval was obtained from the Bioethics Committee at the Polish Mother's Memorial Hospital – Research Institute in Lodz (Opinion No. 47/2020).

2.7 Informed consent statement

The legal representatives of all patients gave their informed written consent to participate in the study prior to their inclusion in the study.

3 Results

3.1 Study group characteristics

There were 47 healthy children included in the study. Mean age of children 10.35 ± 2.6 years, 57,5% were male. Study group characteristics is presented in Table 1.

3.2 The analysis of serum SIRT1 concentration in healthy children in dependence on age, sex and stage of puberty

Serum SIRT1 concentration in healthy children ranged from 0.04 to 0.96 ng/ml. The mean SIRT1 concentration in healthy children was 0.29 ± 0.21 ng/ml (mean ± SD), while the median value (Q1-Q3) was 0.26 (0.14-0.38) ng/ml. The normality of the distribution of SIRT1 concentration was assessed - no normal distribution was found (Shapiro - Wilk test, p=0.0002, Figure 1).

There was no significant correlation between SIRT1 concentration and age of children (r=0.16), SIRT1 concentration and weight of children (r=0.11), SIRT1 concentration and weight SDS values (r=-0.05), SIRT1 concentration and height of children (r=0.13), as well as SIRT1 concentration and height SDS values (r=-0.01), SIRT1 concentration and their BMI (r=0.05), SIRT1 concentration and BMI SDS values (r=-0.04). We also did not find correlations between SIRT1 and IGF-1 concentrations (r=0.11), SIRT1 concentration and IGF-1 SDS value (r=0.08), SIRT1 and IGFBP-3 concentrations (r=0.12), as well as between SIRT1 concentration and IGF-1/IGFBP-3 molar ratio (r=0.06). The mentioned results are presented in Figure 2 and Supplementary Figure 1.

We also compared the SIRT1 concentration in individual subgroups of children, differentiated by gender (girls vs boys), age (younger than 10 years vs older or equal to 10 years old), stage of puberty (prepubertal vs pubertal), body weight and height (BMI SDS/hSDS greater or equal to 0 vs less than 0) and IGF1 concentration (IGF-1 SDS above or equal to 0 vs below 0). There were no statistical differences between SIRT 1 levels in the analysed subgroups (Table 2).

3.3 Dependence of SIRT1 concentration on the frequency of consumption of particular types of food

Based on the declared daily fruits and vegatables intake estimated in the overview survey described in Materials and methods, we found that children who consumed at least 4-5 portions of fruits or vegetables per day had significantly higher levels of SIRT1 than children who consumed less (0-3 portions per day) [0.41 (0.23 – 0.6) ng/ml vs 0.2 (0.1 – 0.34) ng/ml, p=0.02, Figure 3]. A tendency towards higher SIRT1 concentration was also found in the group of children eating at least 2-3 or more servings of fruits and vegetables, compared to those eating 0 or a maximum of 1

TABLE 1 Study group characteristics.

Variable	Mean ± SD Median (Q1 – Q3)	Range
age [years] M F	10. 4 ± 2.6 10.6 (8.37 – 12.78) 10.8 ± 2.7 9.7 ± 2.4	4.2 – 14.4 4.2 – 14.4 5.3 – 13.9
sex M; N (%) F; N (%)	27 (57.5%) 20 (42.5%)	
Tanner stage 1; N (M, F) ≥2; N (M, F)	24 (15, 9) 23 (12, 11)	
height SDS	0.52 ± 1.02 0.48 (-0.4 – 1.1)	-1.07 – 2.9
weight SDS	0.48 ± 1.3 0.26 (-0.4 – 1.4)	-2.08 – 3.5
BMI SDS	0.18 ± 1.3 -0.05 (-0.8 –1.1)	-2.25 – 2.8
IGF-1 [ng/ml]	270.5 ± 183.4 203.9 (119.5 – 404.2)	36.6 – 679.4
IGF-1 SDS	-0.39 ± 1.14 -0.16 (-1.33 – 0.47)	-3.66 – 1.41
IGFBP-3 [ng/ml]	4316.6 ± 1434.9 4721 (2905 – 5438)	1542 – 6336
IGF1/IGFBP-3 m.r.	0.32 ± 0.16 0.24 (0.19 – 0.43)	0.11 – 0.7

M, male; F, female; SDS, standard deviation score; BMI, body mass index; IGF-1, insulin like growth factor 1; IGFBP-3, IGF-binding protein 3; m.r., molar ratio.

serving per day [0.29 (0.18 – 0.44) ng/ml vs 0.17 (0.14 – 0.3) ng/ml, p=0.068].

Moreover, SIRT1 concentration increased with declared frequency of fruits and vegetables consumption (Figure 4).

Although no correlation was observed between SIRT1 and IGF-1 levels in the whole analysed group, after dividing children into groups according to the amount of vegetables and fruits consumed, we found that children eating more fruits and vegetables (at least 2-3 servings per day), despite higher levels of SIRT1, had also significantly lower IGF-1 concentration ([149.6 (87.4 – 349.8) ng/ml vs 379 (156.2 – 502.3) ng/ml, p=0.01, Figure 5], and IGF-1 SDS values [-0.69 (-1.59 – 0.2) vs 0.37 (-0.4 – 0.68), p=0.006, Figure 5], as well as decreased IGF-1/IGFBP-3 molar ratio [0.22 (0.18 – 0.38) vs 0.4 (0.22 – 0.5), p=0.02]. Furthermore, those children were found to have lower BMI [16.64 (15 – 18.67) vs 18.24 (16.22 – 21.2), p=0.056, Figure 5] and BMI SDS [-0.39 (-0.92 – 0.31) vs 0.3 (-0.34 – 1.6), p=0.088, Figure 5]. Nevertheless, they did not differ with respect to height, gender and age.

SIRT1 concentration was also slightly but significantly higher in the group of children consuming unsweetened dairy products more frequently; that is, with consumption at least 2-3 times a day compared to consumption of 0-1 once a day [0.34 (0.2 – 0.54) ng/ml vs 0.18 (0.14 – 0.29) ng/ml, p=0.018, Figure 6]. There were no significant differences in height, weight and IGF-1 concentration with respect to the frequency of the consumption of dairy products.

No differences in SIRT1 levels were detected with respect to the consumption frequency of bread, rice, pasta, fish, meat, sweets and sugary drinks, nor in relation to a specific diet or a number of daily meals. Data on those individual food groups that were compared in the survey are included in the Supplementary Materials.

4 Discussion

SIRT1 is a protein that is primarily localized intracellularly (in a nucleus of liver, muscle, and white adipose tissue and in a cytoplasm of pancreatic and endothelial cells) (3, 5). It has been detected in the human adult serum (33) and its concentration appears to be altered in various disease states. SIRT1 serum reduction was shown in Alzheimer's disease and mild cognitive impairment (33), obesity (34) and lung diseases (35), suggesting that serum SIRT1 may be a potential biomarker for various aging-associated diseases. On the

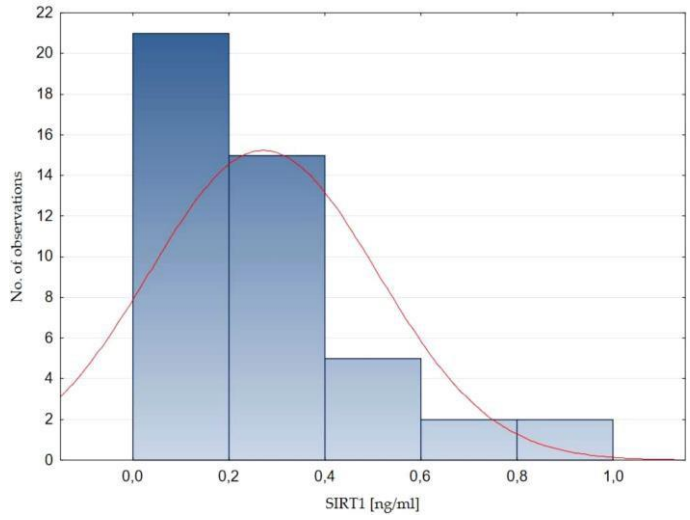

FIGURE 1
Distribution of sirtuin 1 concentration in a group of healthy children. SIRT1, sirtuin 1.

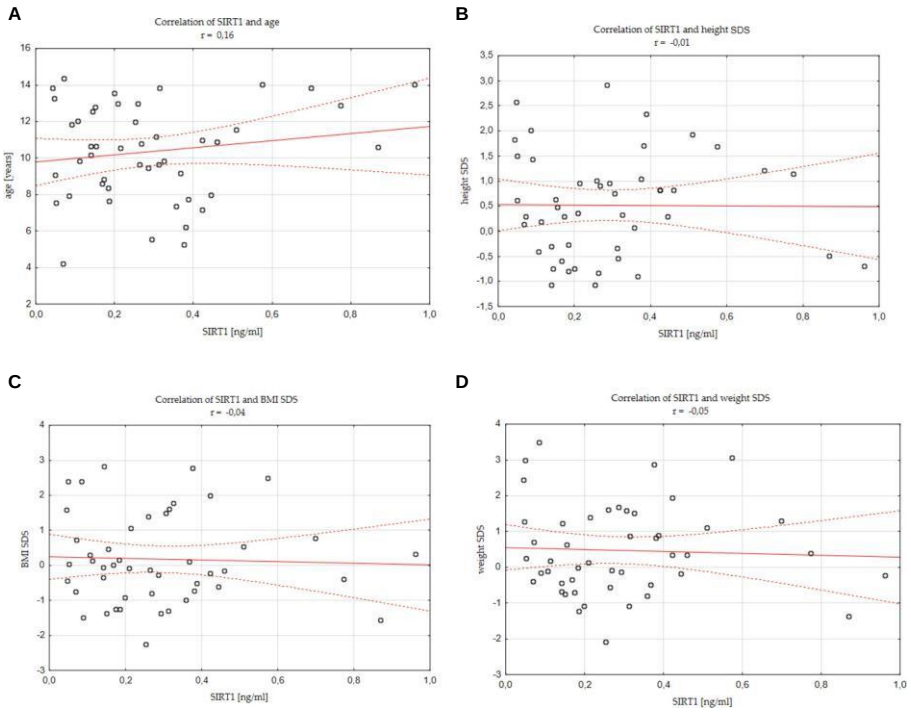

FIGURE 2
Correlations between SIRT1 concentration and individual analysed parameters (A) age, (B) height SDS, (C) BMI SDS, (D) weight SDS, standard deviation score; BMI, body mass index.

other hand, increased serum SIRT1 levels were noticed in acute ischemic stroke (36), asthma (37) systemic lupus erythematosus (38) and frailty (39). To quantify SIRT1 levels in serum samples enzyme-linked immunosorbent assays (ELISA) technique was used most frequently.

To our knowledge, this study represents the first attempt to assess the serum concentration of SIRT1 in children and to explore the potential influencing factors. When compared to studies conducted in the adult population, the levels of SIRT1 observed in children within our study exhibited slightly lower concentration (35, 37, 40–43). There was one earlier study (44) focusing on SIRT1 levels in children in the context of growth. In this study SIRT1 levels were notably higher than the values reported in our observation. However, the authors of the cited report presented a small sample size with only male children and did not refer to other potential factors that may influence SIRT1 concentration in a population of

TABLE 2 SIRT1 concentration in healthy patients with respect to various parameters.

Variable	Variable	No	SIRT1 [ng/ml]	p
Sex	female	21	0.24 (0.15 – 0.38)	0.9
	male	26	0.26 (0.14 – 0.37)	
Age [years]	<10	21	0.28 (0.17 – 0.37)	0.84
	≥10	26	0.23 (0.14 – 0.46)	
Puberty [Tanner stage]	=1	24	0.29 (0.14 – 0.38)	0.52
	≥2	23	0.21 (0.14 – 0.37)	
BMI SDS	<0	24	0.26 (0.16 – 0.38)	0.55
	≥0	23	0.20 (0.10 – 0.38)	
height SDS	<0	15	0.20 (0.14 – 0.31)	0.9
	≥0	32	0.27 (0.1 – 0.41)	
IGF-1 SDS	<0	26	0.28 (0.14 – 0.37)	0.43
	≥0	21	0.26 (0.14 – 0.44)	

BMI, body mass index; SDS, standard deviation score; IGF-1, Insulin-like growth factor 1.

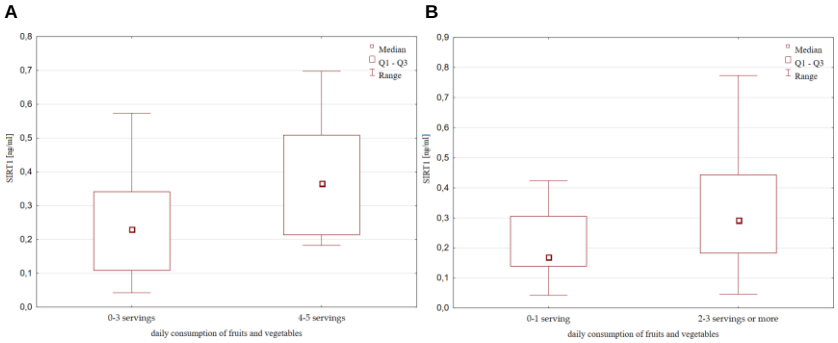

FIGURE 3
SIRT1 concentration with respect to the daily fruits and vegetable consumption (A) 0-3 servings vs 4-5 servings per day; (B) 0-1 serving vs 2-3 servings or more per day;.

healthy children (44). Nevertheless, the observed differences in serum concentration could be attributed to variations in the assay sources or the laboratory techniques (35, 37, 40–43).

It was proven that, SIRT1 play an important role in detection of cellular energy levels and regulation of energy metabolism and increase in response to fasting, caloric restriction and malnutrition (3). Several reports have shown that serum SIRT1 levels correlated negatively with BMI and were elevated in patients with anorexia nervosa (1, 45). It is well known that caloric restriction contributes to a state of growth hormone resistance and is associated with a decrease in serum IGF-1 levels, which is a main mediator of growth hormone (GH) action in peripheral tissues. SIRT1 is known to influence the process of intracellular GH signal transduction for IGF-1 synthesis (46). In situations of fasting or nutrient deficiencies, SIRT1 has been found to decrease the release of IGF-1 from the liver through the STAT5 pathway and enhance resistance to GH by promoting the release of GH from the pituitary gland (13, 47, 48). Furthermore, the presence of SIRT1 in the hypothalamus, particularly in neurons expressing the growth hormone receptor (GHR), and in chondrocytes, suggests potential relevance to the context of growth regulation (12, 14, 15, 49). It has been also suggested that SIRT1 regulate kisspeptin expression in the hypothalamus, affecting the timing of puberty onset (20). Therefore, it seemed to us that SIRT1 levels might vary depending on child height, or pubertal stage. However, we did not observe any significant differences in serum SIRT1 levels in relation to, age, height and IGF-1 levels as well as pubertal development. Although it has been found that SIRT1 levels decrease with age (50), in our paediatric cohort with a limited age range these differences may not be apparent.

As our results did not reflect associations between SIRT1 serum levels and height, weight or puberty, the intracellular function of

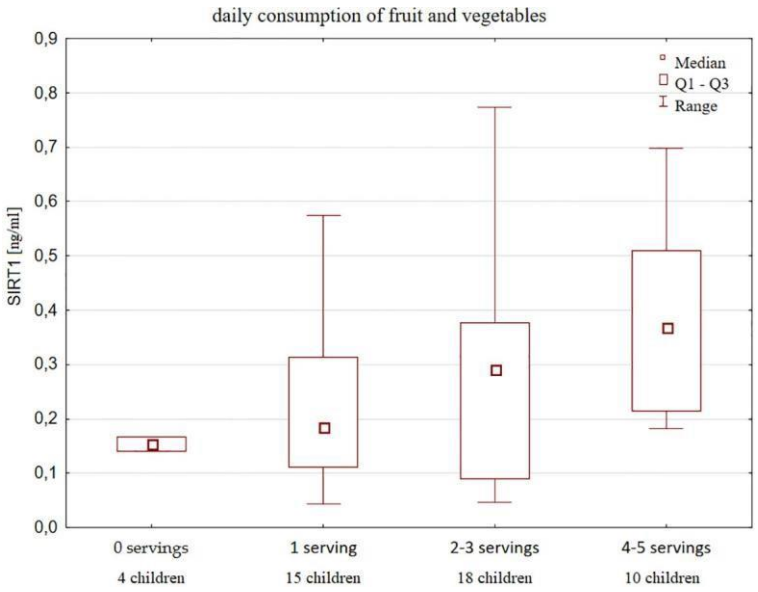

FIGURE 4
Serum SIRT1 concentration according to the declared frequency of fruits and vegetables intake in healthy children and a number of children in each group.

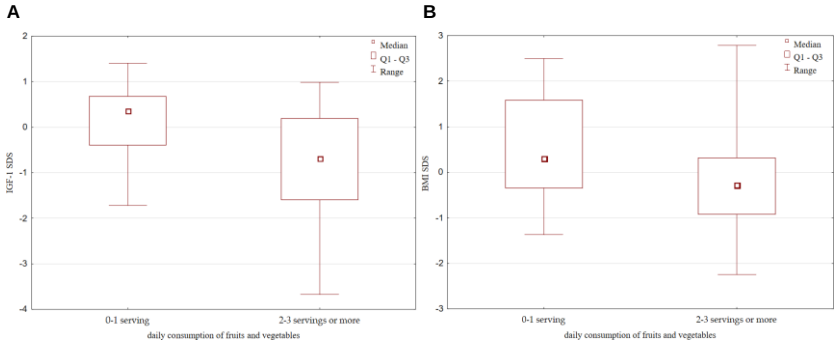

FIGURE 5
Differences in: (A) IGF-1 SDS; (B) BMI SDS with respect to the daily fruits and vegetables consumption. IGFF-1, Insulin-like growth factor 1; IGFBP-3, insulin-like growth factor binding protein-3.

SIRT1 may be defined by factors other than blood concentration. However, taking into account data from the literature, further investigation regarding SIRT1 involvement in growth disorders as well as weight and pubertal disturbances is warranted.

As SIRT1 is engaged in responding to metabolic imbalances, the amount of SIRT1 depends on the availability and type of nutrients (51). We assessed that SIRT1 serum levels were higher in children that consume more fruits and vegetables (at least 2-3 portions per day). Interestingly, those children were thinner and had lower IGF-1 serum levels, whereas their height was not affected the reduced IGF-1 serum levels may be related to sirtuins (46), but too many factors affect IGF-1 values to draw conclusions on this topic. Our outcomes are consistent with studies showing that sirtuin 1 may be activated by certain polyphenols, a class of naturally-occurring phytochemicals, which are compounds of some dietary products - fruits and vegetables in particular. The best known SIRT1 activator, resveratrol can be found in grapes, blueberries and grape products such as red wine (21, 23, 51–53). Piceatannol is a metabolite of resveratrol detected in grapes, passion fruit and white tea (23, 24). Quercetin, flavonoid polyphenol is present in fruits (peaches), vegetables (onions, garlic) and nuts (22, 54). Other dietary polyphenols that activates SIRT1 is fisetin, which can be found in apples, kiwi, dactyls, strawberries and blueberries among others (22, 53, 55). Some data suggest that besides polyphenols, dairy components may also serve as SIRT1 activators. Regarding our results, children with high consumption of dairy products had significantly increased SIRT1 serum concentration. Correspondingly, *in vitro* study in muscle and adipose cells indicate that systemic effects of high dairy feeding resulted in increased SIRT1 gene expression and activity in those cells (56). In addition, leucine, which is present in dairy food was proven to increase SIRT1 expression (57). Despite studies indicate that both natural and synthetic sirtuin activating compounds increase SIRT1 activity *in vivo*, the precise mechanism by which they activate SIRT1 remains unclear (58, 59). According to data from the literature resveratrol and others sirtuin 1 activators have demonstrated promising outcomes in a wide range of age-related diseases including obesity, diabetes, inflammation, cardiovascular disease, among others (60–64). Huang et al. summarized the current experience regarding resveratrol treatment in people with obesity, finding a significant improvement in metabolic complications and body weight reduction (65). A favourable effect of resveratrol has also been shown in certain diseases in children, such as ADHD or muscular dystrophies (66, 67).

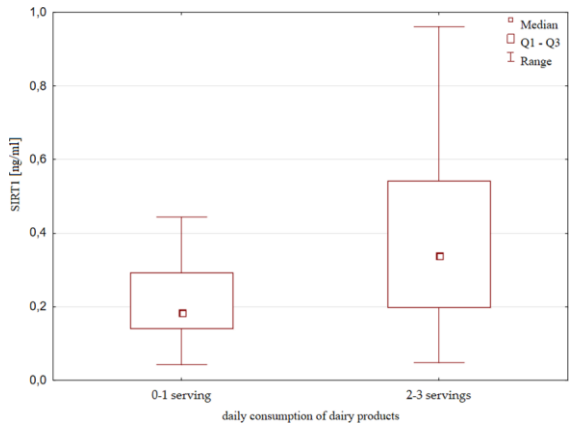

FIGURE 6
Serum SIRT1 concentration in terms of dairy products consumption.

Sirtfood is a novel food concept, according to which compounds from diet can affect sirtuins (53). Sirtfoods are said to induce a calorie restriction state and reduce nutrient consumption without causing malnutrition (68). The vast majority of data show that foods containing sirtuins (Sirtfoods) may produce pleiotropic, beneficial effects on health, alleviating metabolic disorders (53). Formulating a dietary regimen that integrates sirtuin-activating components derived from both the Asian and Mediterranean diets presents a potentially efficacious strategy in the prevention of chronic diseases. This approach holds promise for fostering health and supporting healthy aging (69). However, low bioavailability and rapid metabolism of polyphenols are issues that need to be scientifically addressed (70, 71). Further research is needed to evaluate dietary impact on sirtuin 1 and Sirtfoods-associated clinical implications.

Despite our study is limited by a small sample size, it is important to highlight that it represents the first investigation focusing on assessing SIRT1 serum levels in children which may offer valuable insights into the role of SIRT1 in pediatric physiology and its potential relevance to various aspects of child health. The

preliminary findings presented here may serve as a basis for further research in this area and contribute to our understanding of SIRT1 implications in childhood development and disease, with an emphasis on growth and nutrition. Future studies with larger sample sizes and a wider age range are warranted to validate and expand upon the findings of this pioneering investigation.

5 Conclusions/highlights

1. Serum sirtuin 1 concentrations in healthy children did not differ with respect to sex, age, pubertal development, axiological parameters and IGF-1 levels.
2. Higher frequency of fruits, vegetables and dairy products consumption appeared to increase serum sirtuin 1 levels.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Ethics statement

The studies involving humans were approved by Bioethics Committee at the Polish Mother's Memorial Hospital – Research Institute in Lodz (Opinion No. 47/2020). The studies were conducted in accordance with the local legislation and institutional requirements. Written informed consent for participation in this study was provided by the participants' legal guardians/next of kin.

Author contributions

AF: Conceptualization, Data curation, Funding acquisition, Investigation, Methodology, Project administration, Resources, Software, Validation, Visualization, Writing – original draft, Writing – review & editing. AL: Supervision, Writing – review & editing. RS: Conceptualization, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Supervision, Validation, Visualization, Writing – review & editing.

Funding

The author(s) declare financial support was received for the research, authorship, and/or publication of this article. Study was funded by the Polish Mother's Memorial Hospital – Research Institute Internal Grant No. 15GW/2021 financed by the Ministry of Science and Higher Education subsidy.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fendo.2024.1356612/full#supplementary-material

References

1. Haigis MC, Sinclair DA. Mammalian sirtuins: b iological insights and disease relevance. *Annu Rev Pathol: Mech Dis*. (2010) 5:253–95. doi: 10.1146/annurev.pathol.4.110807.092250

2. Bonkowski MS, Sinclair DA. Slowing ageing by design: the rise of NAD+ and sirtuin-activating compounds. *Nat Rev Mol Cell Biol*. (2016) 17:679–90. doi: 10.1038/nrm.2016.93

3. Nogueiras R, Habegger KM, Chaudhary N, Finan B, Banks AS, Dietrich MO, et al. SIRTUIN 1 AND SIRTUIN 3: PHYSIOLOGICAL MODULATORS OF METABOLISM. *Physiol Rev*. (2012) 92:1479–514. doi: 10.1152/physrev.00022.2011

4. Hwang Jw, Yao H, Caito S, Sundar IK, Rahman I. Redox regulation of SIRT1 in inflammation and cellular senescence. *Free Radic Biol Med*. (2013) 61:95–110. doi: 10.1016/j.freeradbiomed.2013.03.015

5. Xie L, Huang R, Liu S, Wu W, Su A, Li R, et al. A positive feedback loop of SIRT1 and miR17HG promotes the repair of DNA double-stranded breaks. *Cell Cycle*. (2019) 18:2110–23. doi: 10.1080/15384101.2019.1641388

6. Alves-Fernandes DK, Jasiulionis MG. The role of SIRT1 on DNA damage response and epigenetic alterations in cancer. *Int J Mol Sci*. (2019) 20:3153. doi: 10.3390/ijms20133153

7. Kauppinen A, Suuronen T, Ojala J, Kaarniranta K, Salminen A. Antagonistic crosstalk between NF-kB and SIRT1 in the regulation of inflammation and metabolic disorders. *Cell Signal*. (2013) 25:1939–48. doi: 10.1016/j.cellsig.2013.06.007

8. Pang J, Xiong H, Ou Y, Yang H, Xu Y, Chen S, et al. SIRT1 protects cochlear hair cell and delays age-related hearing loss via autophagy. *Neurobiol Aging*. (2019) 80:127–37. doi: 10.1016/j.neurobiolaging.2019.04.003

9. Zhou S, Tang X, Chen HZ. Sirtuins and insulin resistance. *Front Endocrinol (Lausanne)*. (2018) 9:748. doi: 10.3389/fendo.2018.00748

10. Yang Y, Liu Y, Wang Y, Chao Y, Zhang J, Jia Y, et al. Regulation of SIRT1 and its roles in inflammation. *Front Immunol*. (2022) 13:831168. doi: 10.3389/fimmu.2022.831168

11. Kitada M, Ogura Y, Monno I, Koya D. Sirtuins and type 2 diabetes: role in inflammation, oxidative stress, and mitochondrial function. *Front Endocrinol (Lausanne)*. (2019) 10:187. doi: 10.3389/fendo.2019.00187

12. Kang X, Yang W, Wang R, Xie T, Li H, Feng D, et al. Sirtuin-1 (SIRT1) stimulates growth-plate chondrogenesis by attenuating the PERK-eIF-2a-CHOP pathway in the unfolded protein response. *J Biol Chem*. (2018) 293:8614–25. doi: 10.1074/jbc.M117.809822

13. Yamamoto M, Iguchi G, Fukuoka H, Suda K, Bando H, Takahashi M, et al. SIRT1 regulates adaptive response of the growth hormone–insulin-like growth factor-I axis under fasting conditions in liver. *Proc Natl Acad Sci USA*. (2013) 110:14948–53. doi: 10.1073/pnas.1220606110

14. de Lima JBM, Ubah C, Debarba LK, Ayyar I, Didyuk O, Sadagurski M. Hypothalamic GHR-SIRT1 axis in fasting. *Cells*. (2021) 10:891. doi: 10.3390/cells10040891

15. Satoh A, Brace CS, Ben-Josef G, West T, Wozniak DF, Holtzman DM, et al. SIRT1 promotes the central adaptive response to diet restriction through activation of the dorsomedial and lateral nuclei of the hypothalamus. *J Neurosci*. (2010) 30:10220–32. doi: 10.1523/JNEUROSCI.1385-10.2010

16. Ramadori G, Fujikawa T, Fukuda M, Anderson J, Morgan DA, Mostoslavsky R, et al. SIRT1 deacetylase in POMC neurons is required for homeostatic defenses against diet-induced obesity. *Cell Metab*. (2010) 12:78–87. doi: 10.1016/j.cmet.2010.05.010

17. Opstad TB, Sundfør T, Tonstad S, Seljeflot I. Effect of intermittent and continuous caloric restriction on Sirtuin1 concentration depends on sex and body mass index. *Nutr Metab Cardiovasc Dis*. (2021) 31:1871–8. doi: 10.1016/j.numecd.2021.03.005

18. Gillum MP, Erion DM, Shulman GI. Sirtuin-1 regulation of mammalian metabolism. *Trends Mol Med*. (2011) 17:8–13. doi: 10.1016/j.molmed.2010.09.005

19. Bordone L, Guarente L. Calorie restriction, SIRT1 and metabolism: understanding longevity. *Nat Rev Mol Cell Biol*. (2005) 6:298–305. doi: 10.1038/nrm1616

20. Vazquez MJ, Toro CA, Castellano JM, Ruiz-Pino F, Roa J, Beiroa D, et al. SIRT1 mediates obesity- and nutrient-dependent perturbation of pubertal timing by epigenetically controlling Kiss1 expression. *Nat Commun*. (2018) 9:4194. doi: 10.1038/s41467-018-06459-9

21. Gertz M, Nguyen GTT, Fischer F, Suenkel B, Schlicker C, Fränzel B, et al. A molecular mechanism for direct sirtuin activation by resveratrol. *PloS One*. (2012) 7:e49761. doi: 10.1371/journal.pone.0049761

22. Iside C, Scafuro M, Nebbioso A, Altucci L. SIRT1 activation by natural phytochemicals: an overview. *Front Pharmacol*. (2020) 11:1225. doi: 10.3389/fphar.2020.01225

23. Dai H, Sinclair DA, Ellis JL, Steegborn C. Sirtuin activators and inhibitors: Promises, achievements, and challenges. *Pharmacol Ther*. (2018) 188:140–54. doi: 10.1016/j.pharmthera.2018.03.004

24. Piotrowska H, Kucinska M, Murias M. Biological activity of piceatannol: Leaving the shadow of resveratrol. *Mutat Res/Reviews Mutat Res*. (2012) 750:60–82. doi: 10.1016/j.mrrev.2011.11.001

25. Kułaga Z, Grajda A, Gurzkowska B, Góźdź M, Wojtyło M, Swiader A, et al. Polish 2012 growth references for preschool children. *Eur J Pediatr*. (2013) 172:753–61. doi: 10.1007/s00431-013-1954-2

26. Kułaga Z, Litwin M, Tkaczyk M, Palczewska I, Zajączkowska M, Zwolińska D, et al. Polish 2010 growth references for school-aged children and adolescents. *Eur J Pediatr*. (2011) 170:599–609. doi: 10.1007/s00431-010-1329-x

27. Tanner JM, Whitehouse RH. Clinical longitudinal standards for height, weight, height velocity, weight velocity, and stages of puberty. *Arch Dis Child*. (1976) 51:170–9. doi: 10.1136/adc.51.3.170

28. Elmlinger MW, Kühnel W, Weber MM, Ranke MB. Reference ranges for two automated chemiluminescent assays for serum insulin-like growth factor I (IGF-I) and IGF-binding protein 3 (IGFBP-3). *Clin Chem Lab Med*. (2004) 42:654–64. doi: 10.1515/CCLM.2004.112

29. Haj-Ahmad LM, Mahmoud MM, Sweis NWG, Bsisu I, Alghrabli AM, Ibrahim AM, et al. Serum IGF-1 to IGFBP-3 molar ratio: A promising diagnostic tool for growth hormone deficiency in children. *J Clin Endocrinol Metab*. (2023) 108(4):986–94. doi: 10.1210/clinem/dgac609

30. Poulain T, Spielau U, Vogel M, Körner A, Kiess W. CoCu: A new short questionnaire to evaluate diet composition and culture of eating in children and adolescents. *Clin Nutr*. (2019) 38:2858–65. doi: 10.1016/j.clnu.2018.12.020

31. Jarosz M. Nowa Piramida Zdrowego Żywienia i Stylu Życia Dzieci i Młodzieży. *Żyw Człow Meta*. (2019) 46(1):13–4. Available at: https://ncez.pzh.gov.pl/dzieci-i-mlodziez/piramida-zdrowego-zywienia-i-stylu-zycia-dzieci-i-mlodziezy-2/.

32. Kowalkowska J, Wadolowska L, Hamulka J, Wojtas N, Czlapka-Matyasik M, Kozirok W, et al. Reproducibility of a short-form, multicomponent dietary questionnaire to assess food frequency consumption, nutrition knowledge, and lifestyle (SF-FFQ4PolishChildren) in Polish children and adolescents. *Nutrients*. (2019) 11:2929. doi: 10.3390/nu11122929

33. Kumar R, Chaterjee P, Sharma PK, Singh AK, Gupta A, Gill K, et al. Sirtuin1: A promising serum protein marker for early detection of Alzheimer's disease. *PloS One*. (2013) 8:e61560. doi: 10.1371/journal.pone.0061560

34. Mariani S, Fiore D, Persichetti A, Basciani S, Lubrano C, Poggiogalle E, et al. Circulating SIRT1 increases after intragastric balloon fat loss in obese patients. *Obes Surg*. (2016) 26:1215–20. doi: 10.1007/s11695-015-1859-4

35. Yanagisawa S, Papaioannou AI, Papaporfyriou A, Baker JR, Vuppusetty C, Loukides S, et al. Decreased serum sirtuin-1 in COPD. *Chest*. (2017) 152:343–52. doi: 10.1016/j.chest.2017.05.004

36. Liu Y, Jia S, Liang X, Dong M, Xu X, Lu C, et al. Prognostic value of Sirtuin1 in acute ischemic stroke and its correlation with functional outcomes. *Med (Baltimore)*. (2018) 97:e12959. doi: 10.1097/MD.0000000000012959

37. Wang Y, Li D, Ma G, Li W, Wu J, Lai T, et al. Increases in peripheral SIRT1: a new biological characteristic of asthma. *Respirol*. (2015) 20:1066–72. doi: 10.1111/resp.12558

38. Yang C, Li R, Xu WD, Huang AF. Increased levels of sirtuin-1 in systemic lupus erythematosus. *Int J Rheumatic Dis*. (2022) 25:869–76. doi: 10.1111/1756-185X.14360

39. Ma L, Niu H, Sha G, Zhang Y, Liu P, Li Y. Serum SIRT1 is associated with frailty and adipokines in older adults. *J Nutrition Health Aging*. (2019) 23:246–50. doi: 10.1007/s12603-018-1149-7

40. Gonçalinho GHF, Nascimento JR de O, Mioto BM, Amato RV, Moretti MA, Strunz CMC, et al. Effects of coffee on sirtuin-1, homocysteine, and cholesterol of healthy adults: does the coffee powder matter? *J Clin Med*. (2022) 11:2985. doi: 10.3390/jcm11112985

41. Charązka B, Siejka A. Correlations between serum sirtuin levels and cardiovascular risk factors in women with polycystic ovary syndrome. *Adv Med Sci*. (2022) 67:123–8. doi: 10.1016/j.advms.2022.01.004

42. Ozkan Kurtgoz P, Karakose S, Cetinkaya CD, Erkus E, Guney I. Evaluation of sirtuin 1 (SIRT1) levels in autosomal dominant polycystic kidney disease. *Int Urol Nephrol*. (2022) 54:131–5. doi: 10.1007/s11255-021-02862-2

43. Mansur AP, Roggerio A, Goes MFS, Avakian SD, Leal DP, Maranhão RC, et al. Serum concentrations and gene expression of sirtuin 1 in healthy and slightly overweight subjects after caloric restriction or resveratrol supplementation: A randomized trial. *Int J Cardiol*. (2017) 227:788–94. doi: 10.1016/j.ijcard.2016.10.058

44. Kaplan DS, Canak A, Isık E, Orkmez M, Kumru B. Relationship of fibroblast growth factor 21, sirtuin 1, visfatin, and regulators in children with short stature. *Growth Factors*. (2018) 36:172–7. doi: 10.1080/08977194.2018.1513504

45. Amerio A, Escelsior A, Martino E, Strangio A, Aguglia A, Marcatili M, et al. The association between blood SIRT1 and ghrelin, leptin, and antibody anti-hypothalamus: A comparison in normal weight and anorexia nervosa. *J Pers Med*. (2023) 13:928. doi: 10.3390/jpm13060928

46. Fedorczak A, Lewiński A, Stawerska R. Involvement of sirtuin 1 in the growth hormone/insulin-like growth factor 1 signal transduction and its impact on growth processes in children. *Int J Mol Sci*. (2023) 24:15406. doi: 10.3390/ijms242015406

47. Fazeli PK, Klibanski A. Determinants of GH resistance in malnutrition. *J Endocrinol*. (2014) 220:R57–65. doi: 10.1530/JOE-13-0477

48. Wójcik M, Krawczyńska A, Antushevich H, Herman AP. Post-receptor inhibitors of the GHR-JAK2-STAT pathway in the growth hormone signal transduction. *Int J Mol Sci*. (2018) 19(7):1843. doi: 10.3390/ijms19071843

49. Yamamoto M, Takahashi Y. The essential role of SIRT1 in hypothalamic-pituitary axis. *Front Endocrinol (Lausanne)*. (2018) 9:605. doi: 10.3389/fendo.2018.00605

50. Zhong Y, Chen AF, Zhao J, Gu YJ, Fu GX. Serum levels of cathepsin D, sirtuin1, and endothelial nitric oxide synthase are correlatively reduced in elderly healthy people. *Aging Clin Exp Res*. (2016) 28:641–5. doi: 10.1007/s40520-015-0472-7

51. Howitz KT, Bitterman KJ, Cohen HY, Lamming DW, Lavu S, Wood JG, et al. Small molecule activators of sirtuins extend Saccharomyces cerevisiae lifespan. *Nature*. (2003) 425:191–6. doi: 10.1038/nature01960

52. Malaguarnera L. Influence of resveratrol on the immune response. *Nutrients*. (2019) 11:946. doi: 10.3390/nu11050946

53. Akan OD, Qin D, Guo T, Lin Q, Luo F. Sirtfoods: new concept foods, functions, and mechanisms. *Foods*. (2022) 11:2955. doi: 10.3390/foods11192955

54. Cui Z, Zhao X, Amevor FK, Du X, Wang Y, Li D, et al. Therapeutic application of quercetin in aging-related diseases: SIRT1 as a potential mechanism. *Front Immunol*. (2022) 13:943321. doi: 10.3389/fimmu.2022.943321

55. Khan N, Syed DN, Ahmad N, Mukhtar H. Fisetin: a dietary antioxidant for health promotion. *Antioxid Redox Signal*. (2013) 19:151–62. doi: 10.1089/ars.2012.4901

56. Bruckbauer A, Zemel MB. Effects of dairy consumption on SIRT1 and mitochondrial biogenesis in adipocytes and muscle cells. *Nutr Metab (Lond)*. (2011) 8:91. doi: 10.1186/1743-7075-8-91

57. Li H, Xu M, Lee J, He C, Xie Z. Leucine supplementation increases SIRT1 expression and prevents mitochondrial dysfunction and metabolic disorders in high-fat diet-induced obese mice. *Am J Physiol Endocrinol Metab*. (2012) 303:E1234–44. doi: 10.1152/ajpendo.00198.2012

58. Hubbard BP, Sinclair DA. Small molecule SIRT1 activators for the treatment of aging and age-related diseases. *Trends Pharmacol Sci*. (2014) 35:146–54. doi: 10.1016/j.tips.2013.12.004

59. Huang Y, Lu J, Zhan L, Wang M, Shi R, Yuan X, et al. Resveratrol-induced Sirt1 phosphorylation by LKB1 mediates mitochondrial metabolism. *J Biol Chem*. (2021) 297:100929. doi: 10.1016/j.jbc.2021.100929

60. Milne JC, Lambert PD, Schenk S, Carney DP, Smith JJ, Gagne DJ, et al. Small molecule activators of SIRT1 as therapeutics for the treatment of type 2 diabetes. *Nature*. (2007) 450:712–6. doi: 10.1038/nature06261

61. García-Martínez BI, Ruiz-Ramos M, Pedraza-Chaverri J, Santiago-Osorio E, Mendoza-Núñez VM. Effect of resveratrol on markers of oxidative stress and sirtuin 1 in elderly adults with type 2 diabetes. *Int J Mol Sci*. (2023) 24:7422. doi: 10.3390/ijms24087422

62. Li H, Xia N, Hasselwander S, Daiber A. Resveratrol and vascular function. *Int J Mol Sci*. (2019) 20:2155. doi: 10.3390/ijms20092155

63. Najjar RS, Feresin RG. Protective role of polyphenols in heart failure: molecular targets and cellular mechanisms underlying their therapeutic potential. *Int J Mol Sci*. (2021) 22:1668. doi: 10.3390/ijms22041668

64. Chen J, Lou R, Zhou F, Li D, Peng C, Lin L. Sirtuins: Key players in obesity-associated adipose tissue remodeling. *Front Immunol*. (2022) 13:1068986. doi: 10.3389/fimmu.2022.1068986

65. Huang H, Chen G, Liao D, Zhu Y, Pu R, Xue X. The effects of resveratrol intervention on risk markers of cardiovascular health in overweight and obese subjects: a pooled analysis of randomized controlled trials. *Obes Rev*. (2016) 17:1329–40. doi: 10.1111/obr.12458

66. Kawamura K, Fukumura S, Nikaido K, Tachi N, Kozuka N, Seino T, et al. Resveratrol improves motor function in patients with muscular dystrophies: an open-label, single-arm, phase IIa study. *Sci Rep*. (2020) 10:20585. doi: 10.1038/s41598-020-77197-6

67. Rafeiy-Torghabeh M, Ashraf-Ganjouei A, Moradi K, Bagheri S, Mohammadi MR, Akhondzadeh S. Resveratrol adjunct to methylphenidate improves symptoms of attention-deficit/hyperactivity disorder: a randomized, double-blinded, placebo-controlled clinical trial. *Eur Child Adolesc Psychiatry*. (2021) 30:799–807. doi: 10.1007/s00787-020-01562-z

68. Lilja S, Bäck H, Duszka K, Hippe B, Suarez L, Höfinger I, et al. Fasting and fasting mimetic supplementation address sirtuin expression, miRNA and microbiota composition. *Funct Foods Health Dis*. (2020) 10:439. doi: 10.31989/ffhd.v10i10.752

69. Pallauf K, Giller K, Huebbe P, Rimbach G. Nutrition and healthy ageing: calorie restriction or polyphenol-rich 'MediterrAsian' diet? *Oxid Med Cell Longev*. (2013) 2013:707421. doi: 10.1155/2013/707421

70. Cueva C, Silva M, Pinillos I, Bartolomé B, Moreno-Arribas MV. Interplay between dietary polyphenols and oral and gut microbiota in the development of colorectal cancer. *Nutrients*. (2020) 12:625. doi: 10.3390/nu12030625

71. van Dorsten FA, Grün CH, van Velzen EJJ, Jacobs DM, Draijer R, van Duynhoven JPM. The metabolic fate of red wine and grape juice polyphenols in humans assessed by metabolomics. *Mol Nutr Food Res*. (2010) 54:897–908. doi: 10.1002/mnfr.200900212

biomedicines

MDPI

Article

Relationship between Serum Sirtuin 1 and Growth Hormone/Insulin-like Growth Factor 1 Concentrations in Children with Growth Hormone Deficiency and Idiopathic Short Stature

Anna Fedorczak [1], Dorota Kowalik [1], Justyna Kopciuch [2], Ewa Głowacka [2], Katarzyna Mikołajczyk [3,4], Marcin Tkaczyk [3,4], Andrzej Lewin´ ski [1,5] and Renata Stawerska [1,5,*]

1 Department of Endocrinology and Metabolic Diseases, Polish Mother's Memorial Hospital—Research Institute, 93-338 Lodz, Poland; anna.fedorczak@outlook.com (A.F.); dorota.kowalik96@gmail.com (D.K.); alewin@csk.umed.lodz.pl (A.L.)
2 Center of Medical Laboratory Diagnostics and Screening, Polish Mother's Memorial Hospital—Research Institute, 93-338 Lodz, Poland; justyna.kopciuch@iczmp.edu.pl (J.K.); ewa.biol@interia.pl (E.G.)
3 Department of Pediatrics, Immunology and Nephrology, Polish Mother's Memorial Hospital—Research Institute, 93-338 Lodz, Poland; kasia.pazdziora@gmail.com (K.M.); marcin.tkaczyk@umed.lodz.pl (M.T.)
4 Department of Pediatrics, Nephrology and Immunology, Medical University of Lodz, 93-338 Lodz, Poland
5 Department of Pediatric Endocrinology, Medical University of Lodz, 93-338 Lodz, Poland
* Correspondence: renata.stawerska@icloud.com; Tel.: +48-058-83354

Abstract: Sirtuin 1 (SIRT1) inhibits growth hormone (GH) intracellular signaling for the insulin-like growth factor 1 (IGF-1) synthesis via the janus kinase (JAK)/signal transducer and activator of transcription proteins (STATs) pathway. The aim of this study was to compare SIRT1 concentrations in children with GH deficiency (GHD) and so-called idiopathic short stature (ISS, non-GH deficient), in order to determine the possible impact of changes in serum SIRT1 concentrations on the GH-IGF-1 axis. The study group included 100 short-stature children: 38 with GHD and 62 with ISS (maxGH in two stimulation tests <10 and ≥10 ng/mL, respectively). The control group consisted of 47 healthy, normal-height children. For each child, the concentrations of SIRT1, IGF-1 and insulin-like growth factor-binding protein 3 (IGFBP-3) were determined and the IGF-1/IGFBP-3 molar ratio was calculated. The level of SIRT1 was significantly higher in both groups of short children than in the controls ($p < 0.0001$), but there were no differences between GHD and ISS (mean ± SD: 0.89 ± 0.45 for ISS; 1.24 ± 0, 86 for GHD; and 0.29 ± 0.21 for controls). A significant negative correlation was found between SIRT1 and height standard deviation score (SDS), IGF-1 and IGF-1/IGFBP-3, but not between SIRT1 and maxGH. Elevated SIRT1 levels may serve as one of the mechanisms through which the secretion of IGF-1 is reduced in children with short stature; however, further research is required to confirm this issue.

Keywords: growth hormone; growth hormone deficiency; non-GH-deficient children; short stature; JAK/STATs pathway; insulin-like growth factor 1; IGF-1/IGFBP-3 molar ratio; sirtuin 1

check for updates

Citation: Fedorczak, A.; Kowalik, D.; Kopciuch, J.; Głowacka, E.; Mikołajczyk, K.; Tkaczyk, M.; Lewin´ski, A.; Stawerska, R. Relationship between Serum Sirtuin 1 and Growth Hormone/Insulin-like Growth Factor 1 Concentrations in Children with Growth Hormone Deficiency and Idiopathic Short Stature. *Biomedicines* **2024**, *12*, 1433. https://doi.org/10.3390/biomedicines12071433

Academic Editor: Thomas Kietzmann

Received: 4 June 2024
Revised: 19 June 2024
Accepted: 24 June 2024
Published: 27 June 2024

1. Introduction

The regulation of growth processes in children is dependent on the actions of both the growth hormone (GH) and growth insulin-like growth factor 1 (IGF-1), wherein IGF-1 is the main mediator of GH activity for promoting longitudinal growth [1]. The activation of the Janus kinase 2 (JAK2)/Signal Transducers and Activators of Transcription (STATs) pathway is responsible for the synthesis of IGF-1 in hepatocytes in response to GH binding to its cell surface receptor [2]. Also, the IGF-binding protein 3 (IGFBP-3) gene is promoted through the same pathway [3]. The molar ratio of IGF-1 to IGFBP-3 is considered to be the indicator that best reflects GH activity in children [4,5].

Recently, it has been proven that sirtuin 1 (SIRT1), which is a NAD+-dependent deacetylase, is one of the modulators of JAK2/STATs pathway activity in hepatocytes [6]. SIRT1 is also involved in the cell cycle, apoptosis, response to oxidative stress, DNA repair, inflammatory processes and metabolism, as well as in the regulation of hunger and satiety [7–10]. Changes in SIRT1 expression depend on various factors and are intended to help maintain homeostasis. Generally, SIRT1 downregulates IGF-1 mRNA synthesis induced by GH [6]. Although SIRT1 acts primarily intracellularly, the serum SIRT1 concentration can also be determined. In our earlier study, we assessed the SIRT1 concentration in healthy children and analyzed the relationship between its concentration and gender, age and stage of puberty; we did not find correspondence between any of them [11]. Therefore, the assessment of the SIRT1 concentration in short-stature children seems interesting, as changes in its concentration may play an important role in growth disorders in children—being the cause or effect of certain abnormalities or adaptation mechanism. So far, reports on this issue are scarce [6]. Thus, the aim of the present study was to assess the serum SIRT1 concentrations in children with GH deficiency (GHD) (characterized by low GH concentration and low IGF-1 concentration) and in those with non-GH-deficient short stature (idiopathic short stature, ISS) (characterized by normal GH and low/normal IGF-1 concentration) in order to find a possible relationship between SIRT1 and GH levels (the latter assessed during stimulation tests), as well as between SIRT1 levels and IGF-1 and the IGF-1/IGFBP-3 molar ratio, depending on the diagnosis.

2. Materials and Methods

For this study, we enrolled 110 consecutive children with short stature who were admitted to the Department of Endocrinology and Metabolic Diseases of the Polish Mother's Memorial Hospital—Research Institute (PMMH-RI) in Lodz, from January 2021 to April 2023, and whose parents consented to their participation in this study.

The inclusion criteria included the following: age, ≥2.0 and ≤17.0 years; patient's height, less than −2.0 SDS with respect to age and sex; no identifiable organic or genetic causes of short stature; born at term (≥37 weeks of pregnancy) and with normal body length and weight (appropriate for gestational age, AGA); and the written consent of the legal representative to participate in this study. The height and weight of each child were measured using a Harpenden stadiometer and body eight scale, followed by the calculation of the BMI. The standard deviation score (SDS) for height, body weight and BMI were calculated according to the reference values for Polish children and adolescents [12,13]. As it was an analysis of children with short stature, in order to objectify the BMI SDS results, the height age (HA) of the children was calculated (as the age corresponding to the 50th percentile for a given child's height), and the BMI SDS for the HA value was determined. From a clinical examination, the stage of pubertal development was assessed according to the Tanner scale [14]. The patients' histories were taken, including their gestational age, birth weight and length, chronic diseases and other endocrine diseases, as well as parental height and family history. For the children with visible dysmorphic features, a karyotype assessment was performed. Children with other possible causes of short stature (e.g., celiac diseases (n = 1), hypothyroidism (n = 3), born small for gestational age (n = 6) and Turner syndrome (n = 1)) were excluded from this study (all together 11 children). Parental height data (measured at a visit or reported) were also collected and each patient's target height (TH) was calculated, using the following formula: (mother's height + father's height)/2 + 6.5 cm for boys and (mother's height + father's height)/2 − 6.5 cm for girls. Next, the TH SDS value was also calculated using the same national reference values. Among the children with ISS, based on these data and the formula provided by Ranke et al., a group of patients with familial short stature (FSS) was distinguished—when height SDS > TH SDS − 1.28—and a group of non-FSS—when height SDS < TH SDS − 1.28 [15]. The patients with GHD underwent a pituitary MRI, which excluded organic causes of pituitary dysfunction and allowed for the diagnosis of idiopathic somatotropic hypopituitarism. The bone age (BA) was estimated according to

Greulich–Pyle (G&P) evaluation standards, based on radiographs of the wrist and hand of the non-dominant hand.

During the hospital stay, two GH stimulation tests were conducted. The first test was performed after oral clonidine administration (at a dose of 0.15 mg/m^2 of body surface area, with GH serum level measurements at the 0, 30th, 60th, 90th and 120th minute of the test). The second one was performed after an intramuscular glucagon injection (at a dose of 30 µg/kg of body mass, limited to a maximum of 1 mg), with the measurement of GH serum levels at the 0, 90th, 120th, 150th and 180th minute). For each child, before the first stimulation test, blood samples for SIRT1, IGF-1 and IGFBP-3 measurements were taken after nocturnal fasting, at 6:00 in the morning, approximately 10–12 h after the last meal.

The control group, presented in our previous study [11], consisted of 47 healthy children, matched by age and gender to the study group, with normal height and weight, hospitalized at PMMH-RI for follow-up examinations, but no abnormalities were found in them. For this group, the height, weight and puberty stage according to the Tanner scale, as well as the SIRT1, IGF-1 and IGFBP-3 concentrations were assessed.

All the analyses were conducted at the Centre for Medical Laboratory Diagnostics and Screening of the PMMH-RI. The methodology for the IGF-1, IGFBP-3 and GH determinations in the samples was described in great detail by our team in a previously published article [16]. The blood samples, in a volume of 2.6 mL for the SIRT1 evaluation, were drawn into S-monovette Serum Gel Cat tubes (SARSTEDT AG & Co. KG, Nümbrecht, Germany), and then the tubes were left at room temperature for 2 h to enable the samples to clot. Once the time had elapsed, the samples were centrifuged for 15 min at 1000× *g*, following the manufacturer's recommendations for the kit that was used to examine the SIRT1 levels. The serum samples were visually assessed for hemolysis, and were then removed immediately and aliquoted into labeled Eppendorf 1.5 mL tubes. The samples were stored at −80 °C until the date of the assay. The SIRT1 concentration was determined by the double-binding ELISA method. For this purpose, 2 Human NAD-dependent deacetylase Sirtuin-1 (SIRT1/SIR2L1) ELISA Kits (Cusabio, Houston, TX, USA) were used. The steps were followed according to the manufacturer's instructions (User Manual; catalogue number: CSB-E15058h). For the procedure, the initial step was the preparation of the samples and standards, and then applying them to the plate. The provided plate was pre-coated with SIRT1/SIR2L1-specific monoclonal antibodies. The preparatory step was followed by an incubation period of two hours at 37 °C. Upon the completion of this time, the wells were thoroughly and carefully drained of the fluid contents and without a washing procedure, biotin-conjugated antibodies were added. This was followed by another incubation period of an hour under the same conditions. As per the manufacturer's instructions, three washing steps in total were applied, and blotting with a clean absorbent surface was also implemented. For the next step, streptavidin–horseradish peroxidase conjugate (HRP-avidin) was used and distributed into the wells and allowed to set for another incubation period of one hour at 37 °C. After that incubation ended, five wash steps in total were conducted. Following that action was the addition of chromogenic substrate 3,3′,5,5′-tetramethylbenzidine (TMB) and the incubation of the plate in the same temperature conditions for 20 min. The reaction was stopped with sulfuric acid, provided as a Stop Solution by the manufacturer. The last step was to read the absorbance using an ELISA plate reader (Bio-Rad iMark; Hercules, CA, USA) at 450 nm. The manufacturer ensures in the attached IFU that the sensitivity of the assay is 0.039 ng/mL, while the guaranteed detection range of the assay is 0.156 ng/mL–10 ng/mL, with an intra-assay coefficient of variation of less than 8%, and an inter-assay coefficient of variation of less than 10%.

The collected data underwent statistical analysis using STATISTICA ver. 13.3 software (Statsoft, Cracow, Poland). The normality of the distribution was assessed using the Shapiro–Wilk test, and the equality of variance was evaluated using Levene's test. Non-parametric tests were used for intergroup comparisons of the quantitative continuous variables: the Kruskal–Wallis rank ANOVA and Mann–Whitney U test. Intergroup com-

parisons of the nominal/qualitative variables were performed using the chi-square test. A correlation analysis of the variables studied (Pearson correlation coefficient) was also conducted. The continuous variables were displayed as the mean and standard deviation (mean ± SD), and the categorical variables by N (%). Statistically significant differences were defined as *p*-values less than 0.05. Approval was obtained from the Bioethics Committee at the Polish Mother's Memorial Hospital—Research Institute in Lodz (Opinion No. 47/2020).

3. Results

3.1. Study Group Characteristics

Finally, one hundred (100) children with short stature were enrolled in this study. GHD was diagnosed when the maximum GH secretion in both stimulation tests was found to be below 10 ng/mL, while ISS was diagnosed when the maximal GH secretion in at least one stimulation test was higher than or equal to 10 ng/mL. With respect to the results of the GH stimulation tests, the patients were divided as follow: 38 children were diagnosed with GHD and 62 children with ISS. The control group consisted of 47 healthy individuals with normal height. In the whole group, 86 (58.5%) children were boys, 61 (41.5%) were girls and their mean age was 10.45 ± 2.72 years. There were no statistically significant differences between the individual groups regarding sex or age. The characteristics of the study groups with respect to the diagnosis are presented in Table 1.

Table 1. Study groups characteristics with respect to diagnosis (numerical variables are given as means ± SD and min; max range).

Variable		**ISS, n = 62**	**GHD, n = 38**	**ISS vs. GHD, *p*<**	**Controls, n = 47**	***p*<**
age [years]		10.4 ± 2.75 5.02; 15.98	10.75 ± 2.88 3.01; 15.26	0.4754	10.35 ± 2.6 4.21; 14.35	0.6780
sex, N (%)	female male	28 (45.16%) 34 (54.84%)	13 (34.21%) 25 (65.7%)	0.2798	20 (42.5%) 27 (57.5%)	0.5500
height [cm]		127.93 ± 14.17 94; 150.1	130.97 ± 15.39 89; 154.4	0.2233	146.46 ± 17.89 105; 181	**0.0001 ***
height SDS		−2.62 ± 0.51 −4.85; −2	−2.40 ± 0.3 −3.77; −2	0.1586	0.52 ± 1.02 −1.07; 2.91	**0.0001 ***
body mass [kg]		26.56 ± 7.86 12.2; 44.6	31.17 ± 10.54 12.5; 54.2	**0.0285 ***	39.40 ± 14.25 16; 78	**0.0001 ***
body mass [SDS]		−1.99 ± 0.62 −3.41; −0.60	−1.40 ± 1.05 −3.05; −1.77	**0.0050 ***	0.48 ± 1.26 −2.08; 3.5	**0.0001 ***
BMI [kg/m^2]		15.82 ± 2.13 12.53; 22.87	17.61 ± 3.22 12.93; 25.89	**0.0060 ***	17.69 ± 2.73 13.83; 24.07	**0.0004 ***
BMI SDS for CA		−0.87 ± 1.08 −2.60; 2.27	−0.01 ± 1.61 −2.69; 2.46	**0.0066 ***	0.18 ± 1.27 −2.25; 2.83	**0.0001 ***
height age, HA [years]		7.77 ± 2.35 2.79; 12.81	8.33 ± 2.45 2.96; 11.82	0.2062	x	x
BMI SDS for HA		−0.34 ± 1.36 −2.70; 4.60	0.69 ± 1.83 −1.98; 5.03	0.0057 *	x	x

* $p < 0.05$ is bolded. ISS—idiopathic short stature; GHD—growth hormone deficiency; CA—calendar age; SDS—standard deviation score; BMI—body mass index; HA—height age.

3.2. Results of Serum Tests

Since the division of the short-stature patients was based on the results of the GH peak in the stimulation tests, the statistical analysis obviously showed a significant difference ($p < 0.0001$) between the GH peak secretion in the GHD (GH peak < 10 ng/mL)

and ISS (GH peak ≥ 10 ng/mL) groups (no stimulation tests were performed on the controls). In both (GHD and ISS) groups, the IGF-1 concentrations [(137.17 ng/mL ± 59.49) vs. (155.48 ng/mL ± 86.92) vs. (270.56 ng/mL ± 183.39), $p < 0.0001$)], and the IGF-1 SDS [(−1.67 ± 0.98) vs. (−1.28 ± 0.84) vs. (−0.39 ± 1.14), $p < 0.0001$)] were significantly reduced compared to the controls, and the IGF-1 SDS was significantly lower in the GHD than in the ISS group ($p < 0.0386$). Similarly, the IGFBP-3 levels [(3481 ng/mL ± 979) and (3482 ng/mL ± 979) vs. (4317 ng/mL ± 1435), $p < 0.0017$)] and the IGF-1/IGFBP-3 molar ratio [(0.22 ng/mL ± 0.06) and (0.23 ng/mL ± 0.09) vs. (0.32 ng/mL ± 0.16), $p < 0.0088$)] were found to be significantly lower in both groups of short children than in the control group, while they did not differ between the individual groups of short children (GHD and ISS), despite the completely different GH secretion and—therefore—final diagnosis. The results of the serum tests performed on the short-stature children and control group are presented in Table 2.

Table 2. The results of the serum tests performed on the study groups (GHD and ISS) and on the control group (numerical variables are presented as the means ± SD and min; max range).

Variable	ISS, n = 62	GHD, n = 38	ISS vs. GHD, *p*<	Control Group, n = 47	*p*<
Max GH after clonidine [ng/mL]	13.98 ± 4.42 3.30; 26.12	6.05 ± 2.64 0.90; 9.83	**0.0001 ***	x	x
Max GH after glucagon [ng/mL]	9.94 ± 5.48 1.88; 27.25	4.91 ± 2.66 0.28; 9.85	**0.0001 ***	x	x
IGF-1 [ng/mL]	155.48 ± 86.92 40.00; 510.90	137.17 ± 59.49 19.10; 303.90	0.6443	270.56 ± 183.39 36.60; 679.40	**0.0001 ***
IGF-1 SDS	−1.28 ± 0.84 −2.92; 0.83	−1.67 ± 0.98 −3.69; 0.29	**0.0386 ***	−0.39 ± 1.14 −3.67; 1.41	**0.0001 ***
IGFBP-3 [ng/mL]	3482 ± 979 2100; 5521	3481 ± 979 1458; 5703	0.7493	4317 ± 1435 1542; 6336	**0.0017 ***
IGF-1/IGFBP-3 molar ratio	0.23 ± 0.09 0.08; 0.51	0.22 ± 0.06 0.06; 0.42	0.5820	0.32 ± 0.16 0.11; 0.71	**0.0088 ***
SIRT1 [ng/mL]	0.89 ± 0.45 0.15; 2.14	1.24 ± 0.86 0.16; 3.33	0.090	0.29 ± 0.21 0.04; 0.96	**0.0001 ***

* $p < 0.05$ is bolded. GH—growth hormone; ISS—idiopathic short stature; GHD—growth hormone deficiency; CA—calendar age; IGF-1—insulin-like growth factor 1; SDS—standard deviation score; IGFBP-3—IGF-binding protein 3; m.r.—molar ratio; SIRT1—sirtuin 1, x—test was not performed.

The SIRT1 concentration was significantly higher in both groups of short-stature children (GHD and ISS) than in the control group [(1.24 ng/mL ± 0.86) and (0.89 ng/mL ± 0.45) vs. (0.29 ng/mL ± 0.21), $p < 0.0001$)], but similarly to the IGF-1 and IGFBP-3 concentrations and the IGF-1/IGFBP-3 molar ratio value, it did not differ between the two groups of short children (GHD and ISS). The SIRT1 concentrations are displayed in Figure 1 to illustrate the wide range of results with respect to the mean in the GHD group.

3.3. Correlations of SIRT1 with Height, Body Mass and IGF-1

In the whole analyzed group (short stature and control group), a significant negative correlation was found between SIRT1 and each of the listed parameters: height (r = −0.29, $p < 0.0008$), height SDS (r = −0.43, $p < 0.0001$), IGF-1 (r =−0.21, $p < 0.0097$), and IGF-1/IGFBP-3 molar ratio (r = −0.18, $p < 0.0325$). Notably, when we took a SIRT1 concentration of 1.5 ng/mL as the cut-off point, all the children whose SIRT1 concentration exceeded this value had a height SDS below <−2.0 and IGF-1 lower than 200 ng/mL (Figure 2).

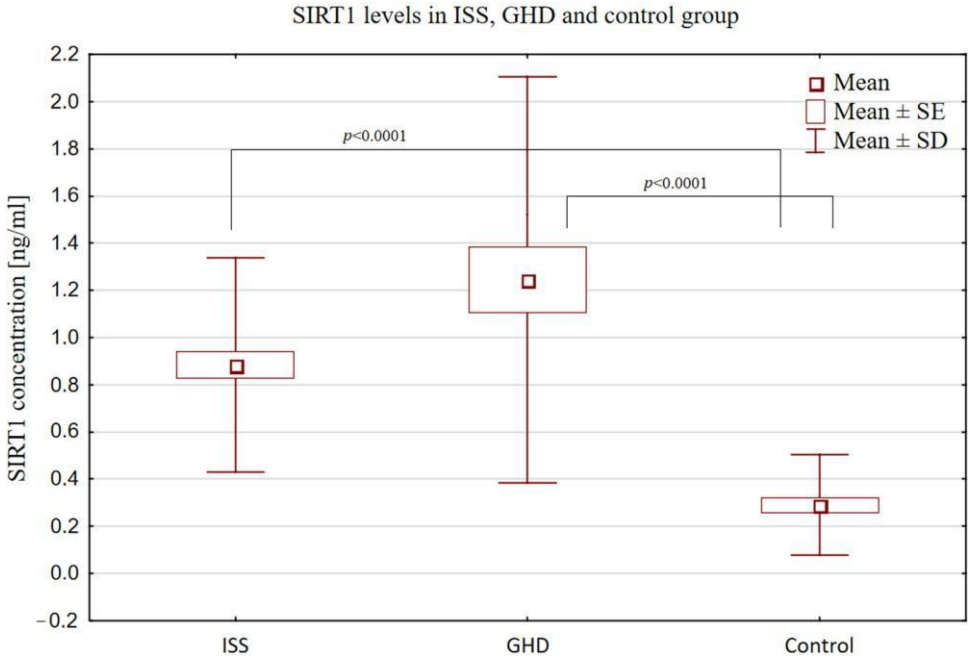

Figure 1. Comparison of SIRT1 concentrations in short children with ISS (GH secretion ≥ 10 ng/mL) and GHD (GH secretion < 10 ng/mL) and in the control group.

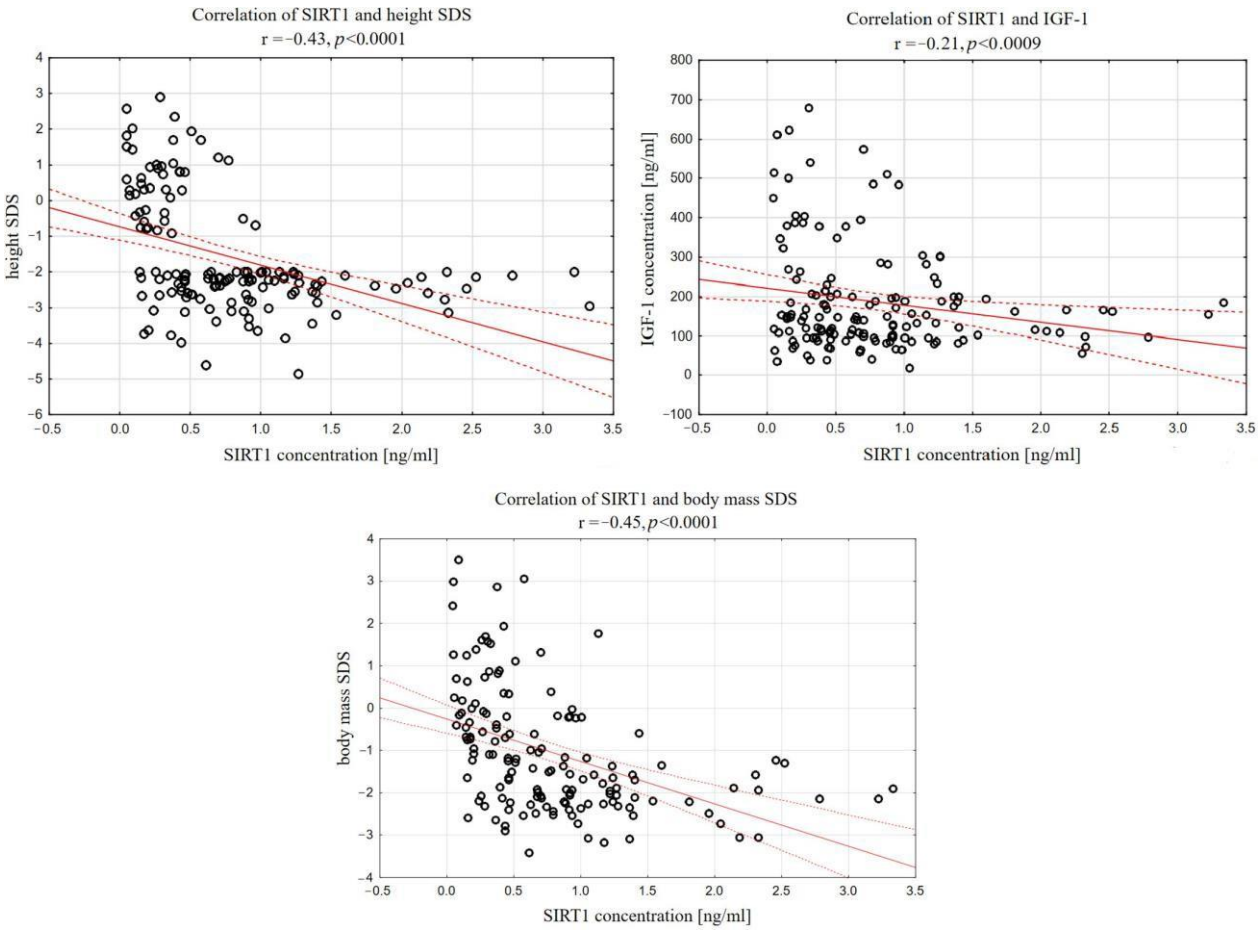

Figure 2. Correlations between sirtuin 1 and height SDS, IGF-1 and IGF-1/IGFBP-3 molar ratio in whole analyzed group of children.

Moreover, serum SIRT1 correlated negatively with body mass (r = −0.29, $p < 0.0005$), body mass SDS (r = −0.42, $p < 0.0001$), as well as BMI (r = −0.24, $p < 0.0060$) and BMI SDS (r = −0.26, $p < 0.0019$), as shown in Figure 3.

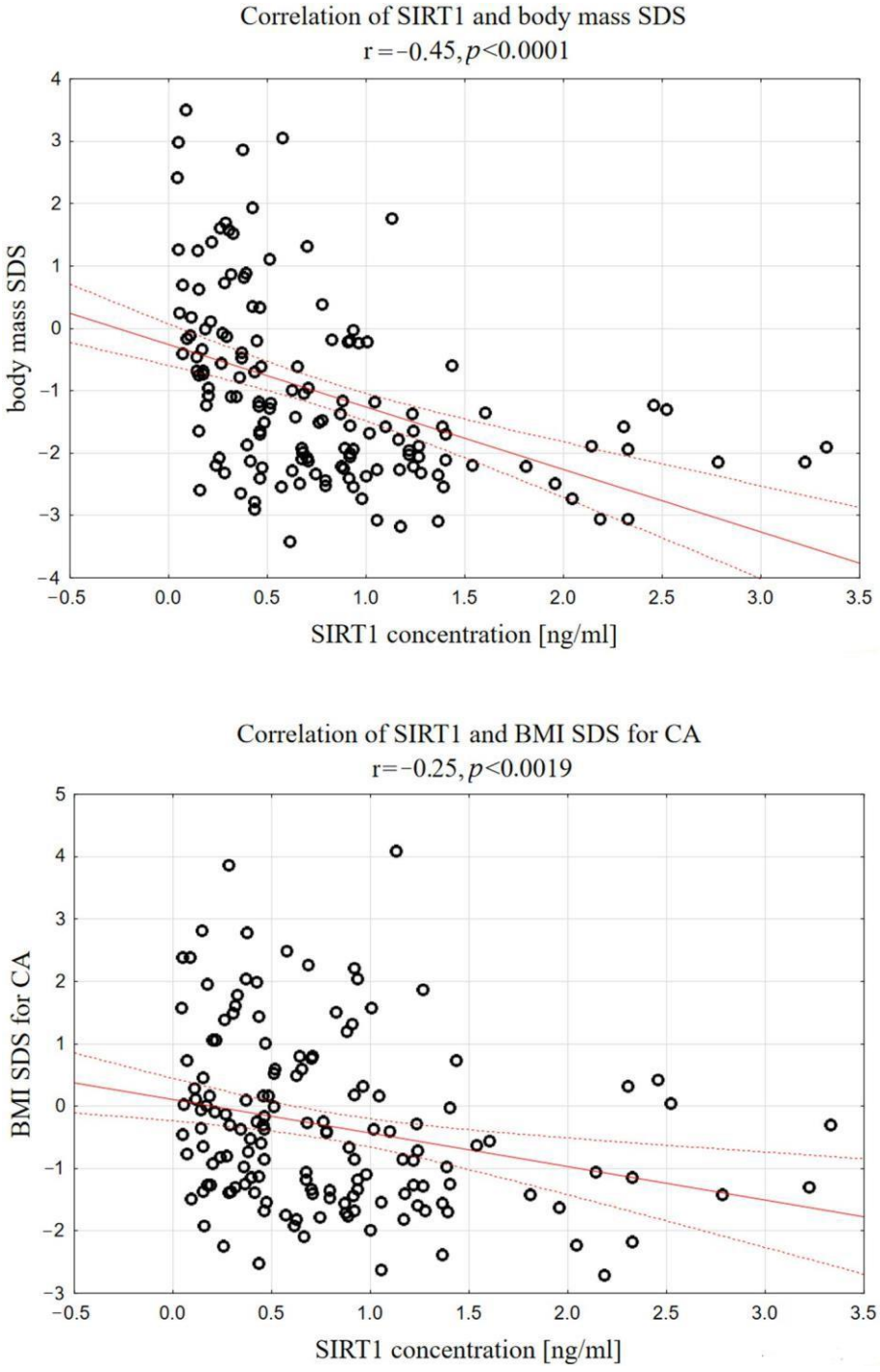

Figure 3. Correlations between serum SIRT1 concentration and body mass SDS, as well as serum SIRT1 and BMI SDS in analyzed group of short-stature children.

As SIRT1 does not have a normal distribution, log-transformed SIRT1 values were also calculated, revealing significant correlations between the LogSIRT1 and the following parameters: height (r = −0.35, $p < 0.0001$), height SDS (r = −0.57, $p < 0.0001$), IGF-1 (r = −0.27, $p < 0.0009$), IGF-1/IGFBP-3 molar ratio (r = −0.26, $p < 0.0025$), body mass

(r = −0.36, $p < 0.0001$), body mass SDS (r = −0.59, $p < 0.0001$), as well as BMI (r = −0.25, $p < 0.0027$) and BMI SDS (r = −0.28, $p < 0.0007$).

3.4. Sirtuin 1 Levels with Respect to the Severity of GHD

In the whole group of short-stature children, no correlations between SIRT1 and the serum GH peak in any of the stimulation tests were observed. However, after dividing them into subgroups, SIRT1 was positively correlated with the max GH secretion in the stimulation tests only in GHD group (r = 0.37, $p < 0.05$). Moreover, in the GHD group, the BMI SDS was correlated negatively with the max GH secretion (r = −0.43, $p < 0.05$). So, thinner children with GHD had higher GH secretion and higher sirtuin 1 levels.

Subsequently, the short-stature children were divided into three subgroups with respect to their maximal GH secretion in both stimulation tests: severe GHD (sGHD), when maxGH was <7 ng/mL (n = 16); partial GHD (pGHD), when maxGH was in the range of 7–10 ng/mL (n = 22); and ISS, when GH max was ≥10 ng/mL (n = 62). There were only four children with very low GH secretion (<3 ng/mL), so they were classified as sGHD. The auxological parameters and serum test results of the above-subdivided groups are presented in Tables 3 and 4.

Table 3. Comparison of auxological parameters in ISS, pGHD and sGHD groups (numerical variables are presented as mean ± SD and min; max range).

Variable		ISS, n = 62	pGHD, n = 22	sGHD, n = 16	*p*<
age [years]		10.4 ± 2.75 5.02; 15.98	10.34 ± 2.84 5.35; 13.85	11.32 ± 2.92 3.01; 15.26	0.6780
sex, N (%)	female male	28 (45.16%) 34 (54.84%)	9 (40.91%) 13 (59.09%)	12 (75%) 5 (25%)	0.5500
height [cm]		127.93 ± 14.17 94; 150.1	128.59 ± 15.05 101.20; 148.50	134.25 ± 15.73 89; 154.40	0.2134
height SDS		−2.62 ± 0.64 −4.85; −2	−2.46 ± 0.56 −3.77; −2	−2.32 ± 0.48 −2.61; −2.09	0.3321
BMI [kg/m^2]		15.82 ± 2.13 **a,b** 12.53; 22.87	16.36 ± 2.25 **a,c** 12.93; 21.48	19.33 ± 3.60 **b,c** 13.89; 25,89	**0.0011 ***
BMI SDS for CA		−0.87 ± 1.08 **a** −2.60; 2.27	−0.59 ± 1.09 **b** −2.17; 1.52	0.79 ± 1.09 **a,b** 2.17; 1.52	**0.0020 ***
height age, HA [years]		7.77 ± 2.35 2.83; 12.81	7.97 ± 2.44 3.75; 11.5	8.83 ± 2.45 2.16; 12	0.2288
BMI SDS for HA		−0.34 ± 1.36 **a** −2.70; 4.60	−0.01± 1.26 −1.98; 2.53	1.65 ± 2.08 **a** −1.87; 5.03	**0.0018 ***

* The values in the columns marked with the same bold letters (a, b, c) differ significantly; $p < 0.05$. GH—growth hormone; ISS—idiopathic short stature; pGHD—partial growth hormone deficiency; sGHD—severe growth hormone deficiency; CA—calendar age; SDS—standard deviation score; BMI—body mass index; HA—height age.

The SIRT1 levels were significantly higher in the patients with pGHD than in the patients with ISS [(1.51 ng/mL ± 0.98) vs. (0.89 ng/mL ± 0.45), $p < 0.0391$, Figure 4]. Children with sGHD formed a very small group (n = 16), but it became visible that the SIRT1 levels were the lowest in them (0.87 ng/mL ± 0.49), and did not differ from the SIRT1 levels observed in the ISS group. In turn, the ISS and pGHD groups were similar in terms of their BMI SDS (−0.87 ± 1.08 and −0.59 ± 1.09) and IGF-1 SDS (−1.28 ± 0.84 and −1.44 ± 0.89), and differed significantly from the sGHD group (BMI SDS 0.79 ± 1.09, $p < 0.0020$; IGF-1 SDS −1.99 ± 1.04, $p < 0.0395$) in this respect.

We also compared the children with ISS according to a diagnosis of familial short stature (FSS) or non-FSS, but we found no correspondence with SIRT1 levels.

Table 4. Sirtuin 1 levels in short-stature subjects in groups with respect to maximal growth hormone secretion in stimulation tests.

Variable	ISS, n = 62	pGHD, n = 22	sGHD, n = 16	*p*<
GH peak [ng/mL]	15.23 ± 4.28 **a,b** 10; 27.25	8.52 ± 0.98 **a,c** 7.09; 9.85	4.3 ± 2.19 **b,c** 0.90; 6.9	**0.0001 ***
IGF-1 [ng/mL]	155.48 ± 86.92 40.00; 510.90	137.72 ± 56.35 55.40; 287.20	136.43 ± 65.45 19.10; 303.90	0.8942
IGF-1 SDS	−1.28 ± 0.84 **a** −2.92; 0.83	−1.44 ± 0.89 3.63; 0.29	−1.99 ± 1.04 **a** −2.61; −1.40	**0.0395 ***
IGFBP-3 [ng/mL]	3482 ± 979 2100; 5521	3501 ± 935 2180; 5703	3454 ± 1069 1458; 5628	0.9379
IGF-1/IGFBP-3	0.23 ± 0.09 0.08; 0.51	0.22 ± 0.07 0.11; 0.42	0.21 ± 0.06 0.06; 0.30	0.8570
SIRT1 [ng/mL]	0.89 ± 0.45 **a**	1.51 ± 0.98 **a**	0.87 ± 0.49	**0.0391 ***

* The values in the columns marked with the same bold letters (a, b, c) differ significantly; $p < 0.05$. GH—growth hormone; ISS—idiopathic short stature; pGHD—partial growth hormone deficiency; sGHD—severe growth hormone deficiency; CA—calendar age; SDS—standard deviation score; BMI—body mass index; HA—height age.

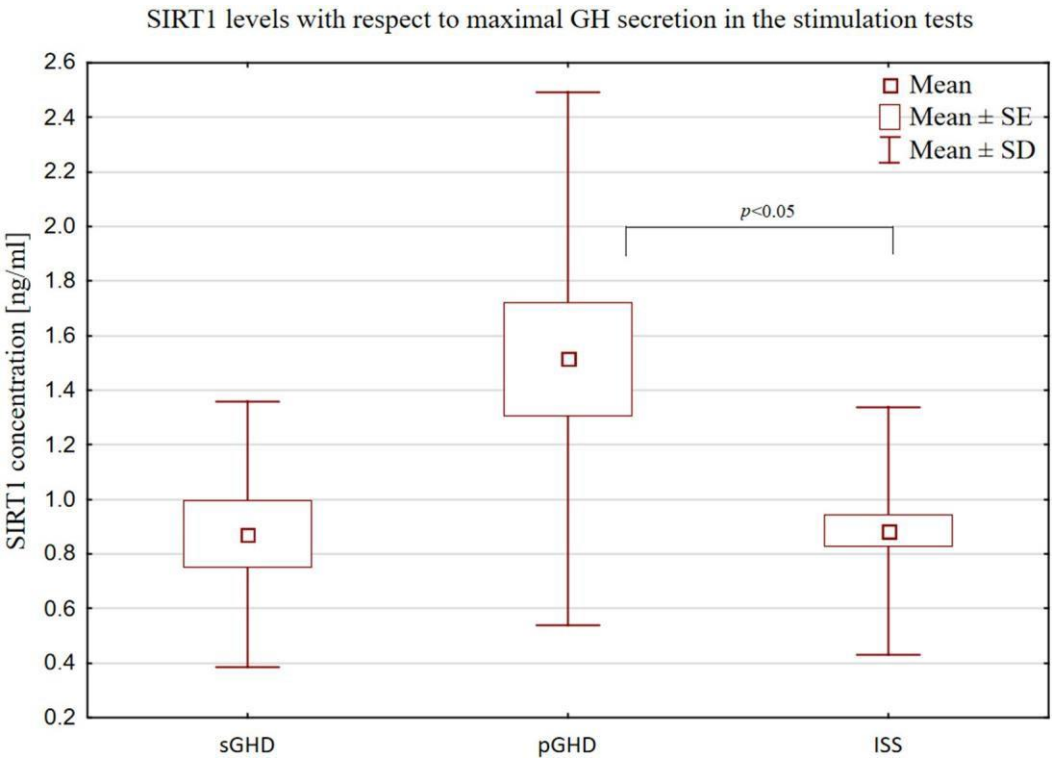

Figure 4. Comparison of SIRT1 levels in ISS (GH secretion ≥ 10 ng/mL), pGHD (GH secretion ≥ 7 and <10 ng/mL) and sGHD (GH secretion < 7 ng/mL) groups.

4. Discussion

Based on the results of our research, we found that the concentration of SIRT1 in serum was significantly higher in both groups of short-stature children (ISS and GHD) than in the control group. Thus, it seems that SIRT1 may play a role in growth disorders in children. However, the mechanisms involved in these relationships are not clear.

As is known, short stature and a slow growth rate in children depend primarily on a low concentration or availability of IGF-1. A reduced concentration of IGF-1 may result from a GH deficiency, since the GH is the most important stimulator of its secretion [17]. However, many other causes of insufficient IGF-1 secretion are also known. These include

chronic diseases (e.g., of the respiratory or circulatory system), liver dysfunction, malabsorption, malnutrition and other endocrine diseases, such as hypothyroidism [18]. In our research, at the initial stage, we eliminated cases of children suffering from chronic diseases, complaining of gastrointestinal problems, and children with untreated hypothyroidism. A history was taken of the patients, which did not point to undernutrition as a clinically evident cause of short stature. However, we did not exclude patients with low BMI from this study (as this was the majority of them). Therefore, it was difficult to determine whether the children who were poor eaters and whose caloric balance was negative, for example, in relation to proteins, were not qualified for the study group. Nevertheless, in the group of children we analyzed, the causes of IGF-1 decrease with normal GH levels remained uncertain. We believe that at least some of them may be related to excessive SIRT1 levels.

Our study shows significant inverse relationships between the SIRT1 concentration and the IGF-1 levels, the IGF-1/IGFBP-3 molar ratio and the degree of growth deficiency. It appears that in certain adverse conditions, increased SIRT1 levels may serve as a factor leading to the inhibition of IGF-1 secretion to conserve energy and maintain homeostasis. It should be emphasized that SIRT1 is involved in reacting to metabolic imbalances caused by caloric restriction and malnutrition [11,19]. We found that SIRT1 levels were related to a child's nutritional status. Within our investigation, SIRT1 levels were negatively correlated with the weight SDS and BMI SDS. Correspondingly, *SIRT1* gene expressions were also upregulated in response to calorie restriction and weight loss, as well as negatively correlated with BMI [20]. It is known that GH has a hyperglycemic effect by releasing glucose reserves from stores, whereas IGF-1 has a hypoglycemic effect similar to insulin [21–23]. Therefore, in cases of malnutrition conditions, an increased secretion of GH from the pituitary and decreased secretion of IGF-1 from the liver at the same time (as, for example, in anorexia nervosa) is beneficial for the body (to maintain glucose levels) [24]. It seems that an increasing concentration of SIRT1 contributes to this, as SIRT1 has been found to negatively regulate GH-induced IGF-1 production by modulating JAK2/STATs pathway activity in hepatocytes [6,25]. Whether such a prolonged status could cause growth impairment (in response to an insufficient IGF-1 concentration, with normal or even high GH levels) is a subject for further research. Similarly, an inverse association between SIRT1 and IGF-1 has also been observed in cases of intrauterine growth restriction (IUGR). Chriet et al. [20] found that the trajectories of gene expression for sirtuins and metabolic genes were perturbed in pigs with IUGR, showing a correspondence with IGF-1 dysregulation. There was a notable increase in *SIRT1* gene expression accompanied by decreased levels of IGF-1.

On the other hand, the reason why SIRT1 levels were higher in the children with GHD (both pGHD and sGHD) than in the control group is not fully understood. Our results also reveal that SIRT1 was positively correlated with the maximum GH secretion in the stimulation tests only in the GHD group. This might be explained in the following way. In children with GHD and a normal BMI, there is no limitation to providing exogenous nutrients (food), but because of the primarily low secretion of GH, there is an impairment in the catabolic actions of GH—providing glucose and free fatty acids from tissues into the bloodstream. Such a constellation is supposed to trigger signals similar to the caloric restriction state (activating ghrelin) and—in this way—activate SIRT1 in some tissues, e.g., the hypothalamus. Thus, we speculate that in the case of patients with GHD, SIRT1 may increase in response to decreased GH (as an attempt to promote GH secretion in a positive feedback mechanism). Actually, SIRT1 is supposed to influence GH signal transduction at various levels [26]. SIRT1 is widely expressed in the hypothalamus (in particular in the arcuate nucleus) [27]. Moreover, SIRT1 activity in the brain is probably involved in the regulation of the somatotropic axis in response to energy supply [28]. It has been proven that in the arcuate nucleus (ARC), the majority of the GH receptor-expressing neurons also express SIRT1, and respond to fasting by upregulating *SIRT1* expression [28,29]. It is also worth mentioning ghrelin, which specifically triggers a central SIRT1/p53 pathway—essential for its orexigenic action [30].

Although the statistical analysis did not show a significant difference between the SIRT1 concentration in the GHD and ISS groups ($p < 0.09$), it is worth emphasizing that

among the children in the GHD group, there were cases of very high SIRT1 concentrations (up to 3.33 ng/mL), and the dispersion around the mean was large, which indicates the heterogeneity of this group. The division of patients into sGHD (GH secretion < 7 ng/mL) and pGHD (GH secretion 7–10 ng/mL) also sheds some light on this issue. Children with pGHD presented with similar features to the ISS group—they were similarly thin and had comparably decreased levels of IGF-1 (despite different GH secretion), and significantly differed from the patients with severe GHD in those respects.

This leads to the question of whether patients with pGHD overlap or form the same group as patients with ISS. It is often mentioned in medical reports that children with pGHD may constitute a group that does not require rhGH treatment and does not differ significantly from children with ISS [31,32]. In fact, distinguishing between GHD and ISS raises difficulties, as there is a continuum between normal GH secretion and GH deficiency [33]. To diagnose GHD, the maximum secretion of GH in stimulation tests has been arbitrarily assumed to be less than 10 ng/mL (or less than 7 depending on the recommendations) [34]. Patients with higher GH secretion are diagnosed with ISS. Such an arbitrary division is questionable, especially given the poor reproducibility and reliability of the stimulation tests. Although at present there are no better tools for diagnosing GHD than the stimulation tests, this topic should be the subject of further research and discussion. Nevertheless, the levels of SIRT1 differed significantly between these two groups (pGHD and ISS). At present, it is difficult to determine the reason for this observation; one can only speculate that the higher levels of SIRT1 in pGHD may primarily result from decreased GH secretion—and that differences in GH secretion levels do matter.

It should also be noted that although SIRT1 is an intracellular enzyme, it is also found in the serum and its extracellular pool appears to play a role in the human body [11]. However, the receptors for SIRT1 have not been described, and how SIRT1 is transported in the blood has not been determined yet. To date, only one study has assessed SIRT1 serum levels in terms of growth. It did not reveal higher SIRT1 levels in the short-stature boys than in the controls, but the GH secretion status in those children was not reported, and one group had been already treated with rhGH [35]. We believe that the results of our study will contribute to the knowledge on this topic—as we have shown that the serum concentration of SIRT1 may reflect its intracellular biological function [6].

Increased SIRT1 levels in children with short stature may lead to favorable metabolic effects on the human body [36,37]. Moreover, in numerous studies, SIRT1 activation has been shown to regulate pathways with beneficial effects on aging and metabolic disorders, inflammatory processes, DNA damage and oxidative stress [38–42]. Our analysis, which is one of the first attempts to assess serum SIRT1 concentrations in children, displays a link between short stature, GH and SIRT1. The involvement of SIRT1 in the regulatory mechanisms concerning growth, metabolism and lifespan provides an interesting perspective for further research.

5. Conclusions

In conclusion, in short-stature children, regardless of GH secretion, SIRT1 serum concentrations were increased. Considering the significant negative correlations between SIRT1 concentration and the levels of IGF-1, the IGF-1/IGFBP-3 molar ratio and the severity of the growth deficiency, elevated SIRT1 levels may serve as one of the mechanisms through which the secretion of IGF-1 is reduced in children with short stature; however, further research is required to confirm this issue.

Author Contributions: Conceptualization, A.F. and R.S.; methodology, A.F. and R.S.; software, A.F.; validation, A.F. and R.S.; formal analysis, R.S.; investigation, A.F., J.K. and E.G.; resources, A.F., K.M., E.G. and J.K.; data curation, A.F. and K.M.; writing—original draft preparation, A.F.; writing—review and editing, A.F., D.K. and R.S.; visualization, A.F. and D.K.; supervision, R.S., M.T. and A.L.; project administration, A.F.; funding acquisition, A.F. and R.S. All authors have read and agreed to the published version of the manuscript.

Funding: This study was financially supported by the Polish Mother's Memorial Hospital—Research Institute Internal Grant No. 15GW/2021, which was financed by a Ministry of Science and Higher Education subsidy.

Institutional Review Board Statement: Approval was obtained from the Bioethics Committee at the Polish Mother's Memorial Hospital—Research Institute in Lodz (Opinion No. 47/2020).

Informed Consent Statement: The legal representatives of all the patients gave their informed written consent to participate in this study prior to their inclusion in this study.

Data Availability Statement: The data presented in this study are available on request from the corresponding author.

Conflicts of Interest: The authors declare no conflicts of interest.

References

1. Juul, A. Serum Levels of Insulin-like Growth Factor I and Its Binding Proteins in Health and Disease. *Growth Horm. IGF Res.* **2003**, *13*, 113–170. [CrossRef]
2. Boguszewski, C.L.; Barbosa, E.J.L.; Svensson, P.A.; Johannsson, G.; Glad, C.A.M. Mechanisms in Endocrinology: Clinical and Pharmacogenetic Aspects of the Growth Hormone Receptor Polymorphism. *Eur. J. Endocrinol.* **2017**, *177*, R309–R321. [CrossRef] [PubMed]
3. Yakar, S.; Liu, J.-L.; Stannard, B.; Butler, A.; Accili, D.; Sauer, B.; LeRoith, D. Normal Growth and Development in the Absence of Hepatic Insulin-like Growth Factor I. *Proc. Natl. Acad. Sci. USA* **1999**, *96*, 7324–7329. [CrossRef] [PubMed]
4. Juul, A.; Dalgaard, P.; Blum, W.F.; Bang, P.; Hall, K.; Michaelsen, K.F.; Müller, J.; Skakkebaek, N.E. Serum Levels of Insulin-like Growth Factor (IGF)-Binding Protein-3 (IGFBP-3) in Healthy Infants, Children, and Adolescents: The Relation to IGF-I, IGF-II, IGFBP-1, IGFBP-2, Age, Sex, Body Mass Index, and Pubertal Maturation. *J. Clin. Endocrinol. Metab.* **1995**, *80*, 2534–2542. [CrossRef] [PubMed]
5. Rosenfeld, R.G. Biochemical Diagnostic Strategies in the Evaluation of Short Stature: The Diagnosis of Insulin-like Growth Factor Deficiency. *Horm. Res.* **1996**, *46*, 170–173. [CrossRef] [PubMed]
6. Yamamoto, M.; Iguchi, G.; Fukuoka, H.; Suda, K.; Bando, H.; Takahashi, M.; Nishizawa, H.; Seino, S.; Takahashi, Y. SIRT1 Regulates Adaptive Response of the Growth Hormone--Insulin-like Growth Factor-I Axis under Fasting Conditions in Liver. *Proc. Natl. Acad. Sci. USA* **2013**, *110*, 14948–14953. [CrossRef] [PubMed]
7. Lee, S.-H.; Lee, J.-H.; Lee, H.-Y.; Min, K.-J. Sirtuin Signaling in Cellular Senescence and Aging. *BMB Rep.* **2019**, *52*, 24–34. [CrossRef] [PubMed]
8. Chen, C.; Zhou, M.; Ge, Y.; Wang, X. SIRT1 and Aging Related Signaling Pathways. *Mech. Ageing Dev.* **2020**, *187*, 111215. [CrossRef] [PubMed]
9. Sasaki, T.; Kitamura, T. Roles of FoxO1 and Sirt1 in the Central Regulation of Food Intake. *Endocr. J.* **2010**, *57*, 939–946. [CrossRef]
10. Sipos, F.; Mu˝zes, G. Sirtuins Affect Cancer Stem Cells via Epigenetic Regulation of Autophagy. *Biomedicines* **2024**, *12*, 386. [CrossRef]
11. Fedorczak, A.; Lewin´ski, A.; Stawerska, R. Sirtuin 1 Serum Concentration in Healthy Children—Dependence on Sex, Age, Stage of Puberty, Body Weight and Diet. *Front. Endocrinol.* **2024**, *15*, 1356612. [CrossRef]
12. Kułaga, Z.; Grajda, A.; Gurzkowska, B.; Góz´dz´, M.; Wojtyło, M.; Swia˛der, A.; Róz˙dz˙yn´ska-S´wia˛tkowska, A.; Litwin, M. Polish 2012 Growth References for Preschool Children. *Eur. J. Pediatr.* **2013**, *172*, 753–761. [CrossRef]
13. Kułaga, Z.; Litwin, M.; Tkaczyk, M.; Palczewska, I.; Zaja˛czkowska, M.; Zwolin´ska, D.; Krynicki, T.; Wasilewska, A.; Moczulska, A.; Morawiec-Knysak, A.; et al. Polish 2010 Growth References for School-Aged Children and Adolescents. *Eur. J. Pediatr.* **2011**, *170*, 599–609. [CrossRef]
14. Marshall, W.A.; Tanner, J.M. Variations in the Pattern of Pubertal Changes in Boys. *Arch. Dis. Child.* **1970**, *45*, 13–23. [CrossRef]
15. Ranke, M.B.; Lindberg, A. Growth Hormone Treatment of Idiopathic Short Stature: Analysis of the Database from KIGS, the Kabi Pharmacia International Growth Study. *Acta Paediatr. Suppl.* **1994**, *406*, 18–23, discussion 24. [CrossRef]
16. Adamczewska, K.; Adamczewski, Z.; Łupin´ska, A.; Lewin´ski, A.; Stawerska, R. Strong Positive Correlation between TSH and Ghrelin in Euthyroid Non-Growth Hormone-Deficient Children with Short Stature. *Molecules* **2020**, *25*, 3912. [CrossRef]
17. Ranke, M.B.; Wit, J.M. Growth Hormone—Past, Present and Future. *Nat. Rev. Endocrinol.* **2018**, *14*, 285–300. [CrossRef]
18. Cohen, L.E. Idiopathic Short Stature: A Clinical Review. *JAMA* **2014**, *311*, 1787–1796. [CrossRef] [PubMed]
19. Howitz, K.T.; Bitterman, K.J.; Cohen, H.Y.; Lamming, D.W.; Lavu, S.; Wood, J.G.; Zipkin, R.E.; Chung, P.; Kisielewski, A.; Zhang, L.-L.; et al. Small Molecule Activators of Sirtuins Extend Saccharomyces Cerevisiae Lifespan. *Nature* **2003**, *425*, 191–196. [CrossRef] [PubMed]
20. Arab Sadeghabadi, Z.; Nourbakhsh, M.; Pasalar, P.; Emamgholipour, S.; Golestani, A.; Larijani, B.; Razzaghy-Azar, M. Reduced Gene Expression of Sirtuins and Active AMPK Levels in Children and Adolescents with Obesity and Insulin Resistance. *Obes. Res. Clin. Pract.* **2018**, *12*, 167–173. [CrossRef] [PubMed]
21. Donato, J.; Wasinski, F.; Furigo, I.C.; Metzger, M.; Frazão, R. Central Regulation of Metabolism by Growth Hormone. *Cells* **2021**, *10*, 129. [CrossRef] [PubMed]

22. Huang, Z.; Huang, L.; Waters, M.J.; Chen, C. Insulin and Growth Hormone Balance: Implications for Obesity. *Trends Endocrinol. Metab.* **2020**, *31*, 642–654. [CrossRef]
23. Holt, R.I.G.; Sönksen, P.H. Growth Hormone, IGF-I and Insulin and Their Abuse in Sport. *Br. J. Pharmacol.* **2008**, *154*, 542–556. [CrossRef]
24. Fazeli, P.K.; Klibanski, A. Determinants of GH Resistance in Malnutrition. *J. Endocrinol.* **2014**, *220*, R57–R65. [CrossRef]
25. Wójcik, M.; Krawczyńska, A.; Antushevich, H.; Herman, A.P. Post-Receptor Inhibitors of the GHR-JAK2-STAT Pathway in the Growth Hormone Signal Transduction. *Int. J. Mol. Sci.* **2018**, *19*, 1843. [CrossRef] [PubMed]
26. Fedorczak, A.; Lewin´ski, A.; Stawerska, R. Involvement of Sirtuin 1 in the Growth Hormone/Insulin-like Growth Factor 1 Signal Transduction and Its Impact on Growth Processes in Children. *Int. J. Mol. Sci.* **2023**, *24*, 15406. [CrossRef] [PubMed]
27. Satoh, A.; Brace, C.S.; Ben-Josef, G.; West, T.; Wozniak, D.F.; Holtzman, D.M.; Herzog, E.D.; Imai, S.-i. SIRT1 Promotes the Central Adaptive Response to Diet Restriction through Activation of the Dorsomedial and Lateral Nuclei of the Hypothalamus. *J. Neurosci.* **2010**, *30*, 10220–10232. [CrossRef]
28. De Lima, J.B.M.; Ubah, C.; Debarba, L.K.; Ayyar, I.; Didyuk, O.; Sadagurski, M. Hypothalamic GHR-SIRT1 Axis in Fasting. *Cells* **2021**, *10*, 891. [CrossRef]
29. Cohen, D.E.; Supinski, A.M.; Bonkowski, M.S.; Donmez, G.; Guarente, L.P. Neuronal SIRT1 Regulates Endocrine and Behavioral Responses to Calorie Restriction. *Genes. Dev.* **2009**, *23*, 2812–2817. [CrossRef]
30. Velásquez, D.A.; Martínez, G.; Romero, A.; Vázquez, M.J.; Boit, K.D.; Dopeso-Reyes, I.G.; López, M.; Vidal, A.; Nogueiras, R.; Diéguez, C. The Central Sirtuin 1/P53 Pathway Is Essential for the Orexigenic Action of Ghrelin. *Diabetes* **2011**, *60*, 1177–1185. [CrossRef]
31. Krukowska-Andrzejczyk, B.; Kalina, M.; Kalina-Faska, B.; Małecka-Tendera, E. Growth Hormone Therapy in Children with Partial Growth Hormone Deficiency. Are We Treating the Right Patients? *Pediatr. Endocrinol. Diabetes Metab.* **2020**, *26*, 65–72. [CrossRef] [PubMed]
32. Smyczyn´ska, J.; Lewin´ski, A.; Hilczer, M.; Stawerska, R.; Karasek, M. Partial Growth Hormone Deficiency (GHD) in Children Has More Similarities to Idiopathic Short Stature than to Severe GHD. *Endokrynol. Pol.* **2007**, *58*, 182–187.
33. Savage, M.O.; Burren, C.P.; Rosenfeld, R.G. The Continuum of Growth Hormone–IGF-I Axis Defects Causing Short Stature: Diagnostic and Therapeutic Challenges. *Clin. Endocrinol.* **2010**, *72*, 721–728. [CrossRef]
34. Murray, P.G.; Dattani, M.T.; Clayton, P.E. Controversies in the Diagnosis and Management of Growth Hormone Deficiency in Childhood and Adolescence. *Arch. Dis. Child.* **2016**, *101*, 96–100. [CrossRef]
35. Kaplan, D.S.; Canak, A.; Isık, E.; Orkmez, M.; Kumru, B. Relationship of Fibroblast Growth Factor 21, Sirtuin 1, Visfatin, and Regulators in Children with Short Stature. *Growth Factors* **2018**, *36*, 172–177. [CrossRef]
36. Yamamoto, M.; Bando, H. A New Insight into GH Regulation and Its Disturbance from Nutrition and Autoimmune Perspectives. *Endocr. J.* **2023**, *70*, 867–874. [CrossRef]
37. Yamamoto, M.; Takahashi, Y. The Essential Role of SIRT1 in Hypothalamic-Pituitary Axis. *Front. Endocrinol.* **2018**, *9*, 605. [CrossRef]
38. Piao, S.; Lee, I.; Jin, S.-A.; Kim, S.; Nagar, H.; Choi, S.; Jeon, B.H.; Kim, C.-S. SIRT1 Activation Attenuates the Cardiac Dysfunction Induced by Endothelial Cell-Specific Deletion of CRIF1. *Biomedicines* **2021**, *9*, 52. [CrossRef]
39. Chojdak-Łukasiewicz, J.; Bizon´, A.; Waliszewska-Prosół, M.; Piwowar, A.; Budrewicz, S.; Pokryszko-Dragan, A. Role of Sirtuins in Physiology and Diseases of the Central Nervous System. *Biomedicines* **2022**, *10*, 2434. [CrossRef]
40. Pardo, P.S.; Boriek, A.M. SIRT1 Regulation in Ageing and Obesity. *Mech. Ageing Dev.* **2020**, *188*, 111249. [CrossRef]
41. Alves-Fernandes, D.K.; Jasiulionis, M.G. The Role of SIRT1 on DNA Damage Response and Epigenetic Alterations in Cancer. *Int. J. Mol. Sci.* **2019**, *20*, 3153. [CrossRef]
42. Chen, J.; Lou, R.; Zhou, F.; Li, D.; Peng, C.; Lin, L. Sirtuins: Key Players in Obesity-Associated Adipose Tissue Remodeling. *Front. Immunol.* **2022**, *13*, 1068986. [CrossRef]

5. Komentarz autora

5.1. Wstęp

Niski wzrost dziecka jest jedną z najczęstszych przyczyn zgłaszania się dzieci do endokrynologa. W procesie diagnostycznym należy uwzględnić szeroki wachlarz możliwych chorób zarówno ogólnosystemowych, jak i bezpośrednio związanych z procesem wzrastania. Należą do nich choroby przewlekłe (np. układu oddechowego lub krążenia), dysfunkcja wątroby, zaburzenia wchłaniania, niedożywienie oraz choroby endokrynologiczne, takie jak niedoczynność tarczycy czy zespół/choroba Cushinga. U części pacjentów stwierdza się niedobór hormonu wzrostu (GHD), nazywany też somatotropinową niedoczynnością przysadki (SNP) [1]. W tych przypadkach obniżone stężenie GH powoduje niedostateczną stymulację receptora dla GH w komórkach hepatocytów i – w rezultacie – zbyt niskie wydzielanie IGF-1. Należy przy tym podkreślić, że proces wzrastania jest w dużej mierze uwarunkowany właśnie działaniem IGF-1. Znane są jeszcze inne przyczyny niedoboru IGF-1, nie związane bezpośrednio z niedoborem GH, np. oporność czy niewrażliwość na GH lub wtórny niedobór IGF-1 towarzyszący chorobom wątroby, celiakii czy niedoczynności tarczycy. Natomiast w znaczącej grupie diagnozowanych dzieci nie udaje się znaleźć przyczyny niskorosłości, nawet jeśli stężenie IGF-1 jest obniżone. U tych pacjentów stwierdza się tak zwany idiopatyczny niedobór wzrostu (ISS). Niestety, większość z tych dzieci nie osiąga przewidywanego prawidłowego wzrostu ostatecznego.

Ostatnio odkryto, że sirtuina 1 (SIRT1) jest zaangażowana w transdukcję sygnału GH w hepatocytach, gdzie hamuje syntezę IGF-1 poprzez modulację szlaku JAK2/STAT [2]. Co więcej SIRT1 uczestniczy w podwzgórzowej sygnalizacji związanej z działaniem GH na swój receptor w OUN. Sugeruje się również, że SIRT1 wpływa na chondrogenezę płytki wzrostowej [3]. Poza tym SIRT1 jest zaangażowana w wiele procesów komórkowych, związanych z regulacją metabolizmu, cyklu komórkowego, odpowiedzi na stres oksydacyjny, naprawą DNA, czy regulacją głodu i sytości [4]. Ilość SIRT1 zależy od dostępności i rodzaju składników odżywczych, ponieważ bierze ona udział w odpowiedzi na zaburzenia równowagi metabolicznej, które są wywoływane przez ograniczenie kalorii i niedożywienie. Należy jednak podkreślić, że SIRT1 jest enzymem wewnątrzkomórkowym, niewiele wiadomo o tym w jaki sposób stężenie wewnątrzkomórkowe tego białka przekłada się na jego stężenie w

surowicy, nie są również znane receptory dla SIRT1. Natomiast coraz więcej jest danych dotyczących substancji będących wyzwalaczami sirtuin, które obejmują m.in. produkty codziennego spożycia. Wobec tego ocena stężenia SIRT1 u dzieci wydaje się być interesująca, ze szczególnym uwzględnieniem dzieci z niskim wzrostem. Lepsze poznanie mechanizmów biorących udział w regulacji procesu wzrastania pozwoli na wdrożenie nowych opcji terapeutycznych dla niskorosłych pacjentów.

5.2. Cele pracy

1. Przegląd literatury dotyczącej SIRT1 z uwzględnieniem jej udziału w transdukcji sygnału wzrostowego oraz potencjalnym wpływie na proces wzrastania u dzieci.

2. Analiza wpływu wybranych parametrów (wiek, płeć, stadium dojrzewania płciowego, wzrost, masa ciała) na stężenie SIRT1 u zdrowych dzieci.

3. Próba oceny zależności stężenia SIRT1 w surowicy od sposobu odżywiania dzieci zdrowych.

4. Porównanie stężenia SIRT1 w surowicy krwi u dzieci zdrowych oraz u dzieci z niskorosłością z grup: z niedoborem hormonu wzrostu (GHD) i z idiopatycznym niedoborem wzrostu (ISS).

5. Poszukiwanie korelacji pomiędzy nasileniem niedoboru wzrostu a stężeniem SIRT1 w całej analizowanej grupie oraz w poszczególnych podgrupach dzieci niskorosłych (GHD i ISS).

6. Analiza wzajemnych relacji pomiędzy stężeniem SIRT1 w surowicy a stężeniem GH (ocenianym w testach stymulacyjnych), IGF-1 i IGFBP-3 oraz stanem odżywienia wyrażonym wartością BMI w badanej grupie pacjentów.

5.3. Materiały i metody

Dzieci do grupy badanej były rekrutowane spośród pacjentów hospitalizowanych w Instytucie Centrum Zdrowia Matki Polki w Łodzi w latach 2021-2023.

Kryteria włączenia do grupy dzieci zdrowych:

- dzieci z prawidłowym wzrostem i masą ciała (3 – 97 centyl dla wieku i płci);

- brak istotnych zaburzeń hormonalnych i chorób przewlekłych;
- pisemna zgoda przedstawiciela ustawowego na udział w badaniu.

Kryteria włączenia do grupy dzieci niskorosłych:

Dzieci z idiopatycznym niedoborem wzrostu (ISS)

- wzrost pacjenta równy lub niższy od -2.0 odchyleń standardowych w odniesieniu do wieku i płci;
- brak uchwytnej przyczyny organicznej, endokrynologicznej, żywieniowej czy genetycznej niskorosłości;
- pisemna zgoda przedstawiciela ustawowego na udział w badaniu.

Dzieci niedoborem hormonu wzrostu (GHD):

- wzrost pacjenta równy lub niższy niż -2.0 odchylenia standardowe w odniesieniu do wieku i płci;
- wolne tempo wzrastania;
- zbyt niskie wydzielanie GH w każdym z 2 testów stymulacyjnych;
- pisemna zgoda przedstawiciela ustawowego na udział w badaniu.

Kryteria wyłączenia z badania:

- brak pisemnej zgody przedstawiciela ustawowego na udział w badaniu;
- przewlekłe problemy zdrowotne mogące mieć wpływ na wystąpienie zaburzeń hormonalnych (m.in. przewlekle choroby przewodu pokarmowego, układu oddechowego, układu krążenia, układu wydalniczego, jadłowstręt psychiczny, niewyrównana niedoczynność tarczycy, zaburzenia i zespoły genetyczne);
- ostra choroba infekcyjna;
- dzieci urodzone przedwcześnie oraz dzieci urodzone jako to za małe w stosunku do wieku płodowego - SGA (small for gestational age);
- wielohormonalna niedoczynność przysadki lub wywiad w kierunku zmian organicznych w ośrodkowym układzie nerwowym.

Przebieg badania:

U każdego dziecka przeprowadzono badanie podmiotowe i przedmiotowe. Szczegółowy wywiad obejmował wiek ciążowy, masę i długość urodzeniową, choroby przewlekłe i inne choroby endokrynologiczne, a także wzrost rodziców i wywiad rodzinny. Pomiary wzrostu i masy ciała zostały wykonane przy użyciu stadiometru Harpenden i wagi elektronicznej. Na podstawie wysokości i masy ciała określono wskaźnik odchylenia standardowego wzrostu (SDS wzrostu) i masy ciała (SDS masy ciała) w stosunku do wartości referencyjnych dla wieku i płci [5] oraz obliczono wskaźnik masy ciała (BMI) oraz BMI SDS. Dla dzieci z niskorosłością, w celu obiektywizacji wyników BMI SDS, obliczono wiek wzrostowy (HA) dzieci (jako wiek odpowiadający 50. centylowi dla danego wzrostu dziecka) i wyznaczono BMI SDS dla wartości HA. Na podstawie badania przedmiotowego oceniano stadium dojrzewania płciowego według skali Tannera [6]. Zebrano również dane dotyczące wzrostu rodziców i obliczono docelowy wzrost (TH) każdego pacjenta, stosując następujący wzór: (wzrost matki + wzrost ojca)/2 + 6,5 cm dla chłopców i (wzrost matki + wzrost ojca)/2 - 6,5 cm dla dziewcząt. U dzieci z cechami dysmorfii przeprowadzono ocenę kariotypu. Wiek kostny został oceniony u dzieci niskorosłych na podstawie badania rentgenowskiego nadgarstka i ręki niedominującej przy użyciu metody Greulich–Pyle

U wszystkich dzieci próbki krwi pobierano rano na czczo w celu pomiaru rutynowo oznaczanych podstawowych parametrów biochemicznych, a także stężeń IGF-1 i IGFBP-3. Dodatkowa próbka krwi (2,4 ml) została pobrana rano na czczo w celu oznaczenia stężenia SIRT1 w surowicy. Po pobraniu krew odwirowywano w celu uzyskania surowicy. Surowica bez oznak hemolizy była następnie zamrażana i przechowywana w odpowiedniej temperaturze, zgodnie z wymaganiami producenta testu, do czasu analizy SIRT1.

U dzieci z niedoborem wzrostu podczas hospitalizacji dokonano oceny wydzielania GH w 2 różnych testach stymulacyjnych. Pierwszy test przeprowadzono po doustnym podaniu klonidyny (w dawce 0,15 mg/m^2 powierzchni ciała, z pomiarami poziomu GH w surowicy w 0, 30, 60, 90 i 120 minucie testu). Drugi test przeprowadzono po domięśniowym podaniu glukagonu (w dawce 30 µg/kg masy ciała, maksymalnie 1 mg, z pomiarem poziomu GH w surowicy w 0, 90, 120, 150 i 180 minucie testu).

Ocena nawyków żywieniowych dzieci została przeprowadzona poprzez ankietę dietetyczną z rodzicem, w której pytano o częstość spożywania przez dzieci poszczególnych rodzajów żywności w poprzednim miesiącu.

Metody laboratoryjne

Wszystkie oznaczenia zostały wykonane w Centrum Medycznej Diagnostyki Laboratoryjnej i Badań Przesiewowych Instytutu Centrum Zdrowia Matki Polki w Łodzi.

Stężenie SIRT1 oznaczono metodą immunoenzymatyczną podwójnego wiązania (test ELISA), wykorzystując 2 zestawy *Human NAD-dependent deacetylase Sirtuin-1 (SIRT1/SIR2L1) ELISA Kit* (Cusabio, Houston, TX, USA), zgodnie z zaleceniami producenta (*User Manual*; numer katalogowy: CSB-E15058h). Stężenie w każdej próbce zmierzono dwukrotnie. Czułość testu wynosi 0,039 ng/ml, natomiast gwarantowany przez producenta zakres wykrywalności testu to 0,156 ng/ml – 10 ng/ml, przy współczynniku zmienności wewnątrzoznaczeniowej poniżej 8% oraz współczynniku zmienności międzyoznaczeniowej poniżej 10%.

Stężenia IGF-1 i IGFBP-3 oceniano za pomocą testów Immulite, DPC. Dla IGF-1 zastosowano standard WHO NIBSC 1st IRR 87/518, o czułości analitycznej 20 ng/mL, zakresie kalibracji do 1600 ng/mL, współczynnik zmienności wewnątrzoznaczeniowej: 3,1-4,3% i współczynnik zmienności międzyoznaczeniowej CV: 5,8-8,4%. Test do oceny IGFBP-3 był skalibrowany do standardu WHO NIBSC Reagent 93/560, z czułością analityczną 0,02 µg/mL, zakresem kalibracji do 426 µg/mL, współczynnik zmienności wewnątrzoznaczeniowej 3,5-5,6%, a współczynnik zmienności międzyoznaczeniowej 7,5-9,9%. Stężenia IGF-1 wyrażono wartością odchylenia standardowego dla płci i wieku (IGF-1 SDS). Stosunek molowy stężeń IGF-1/IGFBP-3 obliczono przyjmując masę cząsteczkową dla IGF-1 – 7,5kDa, a dla IGFBP-3 – 42,0 kDa. Stosunek molowy IGF-1 do IGFBP-3 jest uważany za wskaźnik biodostępności IGF-1.

Analiza statystyczna

Analizę statystyczną zebranych danych przeprowadzono przy użyciu oprogramowania STATISTICA ver. 13.3 software (Statsoft, Polska). Do oceny normalności rozkładu zastosowano test Shapiro-Wilka, a do oceny równości wariancji

test Levene'a. Analiza porównawcza została przeprowadzona za pomocą testów nieparametrycznych dla zmiennych niezależnych. W celu międzygrupowych porównań ilościowych zmiennych ciągłych wykorzystano testy nieparametryczne: analizę wariancji ANOVA rang Kruskala – Wallisa oraz test U Manna-Whitneya. Międzygrupowe porównania zmiennych nominalnych/jakościowych przeprowadzono za pomocą testu Chi-kwadrat. Ponadto przeprowadzono analizę korelacji zmiennych (współczynnik korelacji Pearsona). Zmienne ciągłe zostały przedstawione za pomocą średniej i odchylenia standardowego (mean ± SD) i/lub mediany (Q1 – Q3), zmienne kategorialne za pomocą N (%). Za różnice istotne statystycznie zostały przyjęte wartości $p < 0,05$.

Na prowadzenie badanie pozyskano zgodę Komisji Bioetycznej przy ICZMP w Łodzi. Przedstawiciele ustawowi wszystkich pacjentów wyrazili świadomą, pisemną zgodę na udział w badaniu, po uprzednim zapoznaniu się z informacją na temat prowadzanego badania naukowego.

5.4. Wyniki

Publikacja nr 1

Publikacja nr 1 zatytułowana "Involvement of sirtuin 1 in the growth hormone/insulin-like growth factor 1 signal transduction and its impact on growth processes in children" stanowi pracę przeglądową i została opisana w Dyskusji (Rozdział 6.5.).

Publikacja nr 2

Grupę dzieci zdrowych stanowiło 47 dzieci z prawidłowym wzrostem. Średni wiek dzieci wynosił 10,35 ± 2,6 lat, 57,5% stanowili chłopcy. Stężenia SIRT1 w surowicy u zdrowych dzieci mieściły się w zakresie od 0,04 do 0,96 ng/ml. Średnie stężenie SIRT1 u zdrowych dzieci wynosiło 0,29 ± 0,21 ng/ml (średnia ± SD), podczas gdy mediana (Q1 – Q3) wynosiła 0,26 (0,14 – 0,38) ng/ml. Nie stwierdzono istotnej korelacji pomiędzy stężeniem SIRT1 a: wiekiem dzieci ($r = 0,16$), masą ciała ($r = 0,11$) i SDS masy ciała ($r = -0,05$), wzrostem ($r = 0,13$), SDS wzrostu ($r = -0,01$), BMI ($r = 0,05$), BMI SDS ($r = -0,04$). Nie wykazano również korelacji między stężeniami SIRT1 a:

IGF-1 (r = 0,11), IGF-1 SDS (r = 0,08), IGFBP-3 (r = 0,12), IGF-1/IGFBP-3 molar ratio (r = 0,06). Stężenia SIRT1 nie różniły się istotnie w podgrupach dzieci, podzielonych ze względu na: płeć, wiek (≥10 vs <10 lat), stadium dojrzewania (przedpokwitaniowe vs będące w okresie pokwitania), BMI (BMI SDS ≥0 vs <0), wzrost (SDS wzrostu ≥0 vs <0) czy stężenie IGF-1 (IGF-1 SDS ≥0 vs <0).

W badaniu wykazano natomiast, że dzieci, które spożywały co najmniej 4-5 porcji owoców lub warzyw dziennie miały istotnie wyższe poziomy SIRT1 niż dzieci, które spożywały mniej porcji (0-3 dziennie): 0,41 (0,23 – 0,6) ng/ml vs 0,2 (0,1 – 0,34) ng/ml, p<0,02, odpowiednio. Stwierdzono również, że dzieci spożywające więcej owoców i warzyw (co najmniej 2-3 porcje dziennie) miały istotnie niższe wartości IGF-1 SDS [-0,69 (-1,59 – -0,2) ng/ml vs 0,37 (-0,4 – 0,68) ng/ml, p<0,006], a także były szczuplejsze [BMI 16,64 (15 – 18,67) vs 18,24 (16,22 – 21,2), p<0,056 oraz BMI SDS -0,39 (-0,92 – 0,31) vs 0,3 (-0,34 – 1,6), p<0,088]. Stężenie SIRT1 było również nieznacznie, ale istotnie wyższe w grupie dzieci spożywających produkty mleczne częściej, tj. przy spożyciu co najmniej 2-3 razy dziennie w porównaniu do spożycia 0-1 raz dziennie: 0,34 (0,2 – 0,54) ng/ml vs 0,18 (0,14 – 0,29) ng/ml, p<0,018.

Publikacja nr 3

Do badania zakwalifikowanych zostało 100 dzieci z niedoborem wzrostu. Niedobór hormonu wzrostu (GHD) rozpoznawano w przypadku stwierdzenia maksymalnego wydzielania GH w obu testach stymulacyjnych poniżej 10 ng/ml. Idiopatyczny niedobór wzrostu (ISS) rozpoznawano w przypadku stwierdzenia maksymalnego wydzielania GH w co najmniej jednym teście stymulacyjnych powyżej 10 ng/ml. W odniesieniu do wyników testów stymulacji GH, pacjenci zostali podzieleni w następujący sposób: u 38 dzieci zdiagnozowano GHD, a u 62 dzieci ISS.

Stężenie SIRT1 było istotnie wyższe w obu grupach dzieci niskorosłych (GHD i ISS) niż w grupie dzieci zdrowych (odpowiednio 1,24 ± 0,86 ng/ml oraz 0,89 ± 0,45 ng/ml vs 0,29 ± 0,21 ng/ml, p<0,0001), ale podobnie jak stężenia IGF-1 i IGFBP-3 oraz IGF-1/IGFBP-3 molar ratio, nie różniło się między dwiema grupami dzieci niskorosłych (GHD i ISS).

W całej analizowanej grupie stwierdzono istotną ujemną korelację między SIRT1 a każdym z wymienionych parametrów: wzrost (r = -0,29, p<0,0008), SDS wzrostu (r = -

0,43, p<0,0001), IGF-1 (r = -0,21, p<0,0097), IGF1/IGFBP-3 molar ratio (r = -0,18, p<0,0325), a także z masą ciała (r = -0,29, p<0,0005), SDS masy ciała (r = -0,42, p<0,0001), BMI (r = -0,24, p<0,006) oraz BMI SDS dla HA (r = -0,26, p<0,0019).

Biorąc pod uwagę, że wydzielanie GH stanowi swojego rodzaju continuum i zalecenia co do punktu odcięcia pomiędzy GHD a ISS różnią się na przestrzeni lat w zależności od kraju i doświadczeń naukowców, pacjentów z GHD podzielono dodatkowo na dwie podgrupy: z ciężkim GHD (severe GHD, sGHD) i częściowym GHD (partial GHD, pGHD). Stężenia SIRT1 w surowicy były istotnie wyższe w grupie pacjentów z pGHD niż ISS (1,51 ± 0,98 ng/ml vs. 0,89 ± 0,45 ng/ml, p<0,0391). Z kolei dzieci z sGHD, choć stanowiły bardzo małą grupę (n=16), to było widoczne, że poziomy SIRT1 były u nich również niższe niż w grupie pGHD (0,87 ± 0,49 ng/ml). Z kolei grupy ISS i pGHD były podobne pod względem BMI SDS (-0,87 ± 1,08 i -0,59 ± 1,09) oraz IGF-1 SDS (-1,28 ± 0,84 i -1,44 ± 0,89) i różniły się istotnie od grupy sGHD (BMI SDS 0,79 ± 1,09, p<0,002; IGF-1 SDS -1,99 ± 1,04, p<0,0395) w tej kwestii.

5.5. Dyskusja

Pierwsza publikacja w cyklu zatytułowana "Involvement of Sirtuin 1 in the Growth Hormone/Insulin-like Growth Factor 1 Signal Transduction and Its Impact on Growth Processes in Children" stanowiła analizą piśmiennictwa dotyczącego SIRT1 oraz jej udziału w transdukcji sygnału GH oraz w procesie wzrastania u dzieci. Przegląd literatury obejmuje 111 pozycji. Uwzględniono artykuły opublikowane w języku angielskim w renomowanych bazach danych, w tym PubMed i Google Scholar do grudnia 2022 roku, z późniejszymi aktualizacjami we wrześniu 2023 roku. Wyszukiwano kombinacje następujących terminów MeSH: growth, children, growth hormone, GH, sirtuin 1, SIRT1, sirtuins, insulin-like growth factor 1, IGF-1, STAT5, hypothalamus, hepatocytes, liver, chondrocytes, caloric restriction, fasting, and malnutrition.

W powyższej pracy przeglądowej opisano czynniki wpływające na proces wzrastania u dzieci. Regulacja wzrastania zależy przede wszystkim od działania GH wydzielanego przez przysadkę oraz IGF-1, wydzielanego głównie w hepatocytach [8]. Działanie GH jest regulowane zarówno przez hormony podwzgórzowe, jak i inne

hormony i czynniki [9]. IGF-1 jest natomiast głównym mediatorem aktywności GH w promowaniu wzrostu kości na długość [10]. W odpowiedzi na wiązanie GH z jego receptorem na powierzchni komórek aktywowanych zostaje kilka szlaków sygnałowych. W odniesieniu do procesu wzrastania, głównym jest szlak JAK2/STAT5β w hepatocytach, ponieważ jest on odpowiedzialny za syntezę IGF-1 [11].

W dalszej części pracy opisano Sirtuinę 1 (SIRT1), białko kodowane przez gen *SIRT1* należące do rodziny sirtuin, czyli NAD+ zależnych deacetylaz, które reguluje wiele procesów biologicznych [12]. U ssaków wykryto siedem rodzajów sirtuin [4]. SIRT1 odgrywa ważną rolę w różnych procesach komórkowych m. in. w regulacji metabolizmu, cyklu komórkowego, apoptozy, stresu oksydacyjnego, naprawy DNA oraz starzenia [13,14]. SIRT1 jest aktywowana w odpowiedzi na zaburzenia równowagi metabolicznej, które są wywoływane przez post i deficyt kaloryczny [15]. Wpływa na procesy metaboliczne takie jak glukoneogeneza, β-oksydacja kwasów tłuszczowych czy wrażliwość tkanek na insulinę [16,17].

W dotychczasowych badaniach stwierdzono, że SIRT1 może hamować sygnalizację GH poprzez interakcję z STAT3 i/lub STAT5, a tym samym negatywnie regulować produkcję IGF-1 indukowaną przez GH [2]. Ponadto SIRT1 uczestniczy w podwzgórzowej sygnalizacji związanej z GHR [18]. Sugeruje się również, że SIRT1 wpływa na chondrogenezę płytki wzrostowej oraz wzrost kości na długość [19]. Wydaje się, że SIRT1 modyfikuje działanie GH i IGF-1 poprzez wpływ na przekazywanie sygnałów w neuronach podwzgórza wydzielających GHRH, AgRP i POMC, a także poprzez modulowanie działania greliny [20]. Promuje wydzielanie GH z przysadki, ale podczas deficytu kalorycznego hamuje obwodowe działanie GH poprzez zmniejszenie ekspresji i wydzielania IGF-1. Celem tej regulacji jest utrzymanie równowagi między stymulowaniem i hamowaniem wzrastania, zachowując energię niezbędną dla kluczowych procesów metabolicznych. Wydaje się więc, że u dzieci z niedożywieniem poziom SIRT1 jest zwiększony w celu zahamowania obwodowej produkcji IGF-1 i utrzymania homeostazy. To z kolei spowalnia tempo wzrostu dzieci, hamuje dojrzewanie i kostnienie chrząstek nasadowych. Mechanizm ten pozwala na potencjalną poprawę ostatecznego wzrostu, jeżeli niekorzystne warunki ulegną poprawie. Biorąc pod uwagę, że aktywność SIRT1 może być modulowana przez różne

czynniki, wyzwalacze i inhibitory sirtuin w przyszłości mogłyby zostać wykorzystane do opracowania nowych metod leczenia dzieci z zaburzeniami wzrastania [21].

Podsumowując, SIRT1 jest ważnym czynnikiem w regulacji różnych procesów biologicznych. Niniejszy przegląd literatury sugeruje, że SIRT1 może modulować proces wzrastania u dzieci.

Druga publikacja zatytułowana "Sirtuin 1 serum concentration in healthy children – dependence on sex, age, stage of puberty, body weight and diet" dotyczyła stężenia SIRT1 we krwi u zdrowych dzieci [22]. Pomimo wielu badań na temat roli SIRT1 w organizmie człowieka, dotychczas w niewielu pracach analizowano stężenie sirtuin 1 w surowicy krwi [23–25]. Niniejsze badanie to miało na celu ocenę stężenia SIRT1 w surowicy u zdrowych dzieci oraz analizę potencjalnych czynników wpływających na jej stężenie.

Dotychczas udowodniono, że SIRT1 odgrywa ważną rolę w wykrywaniu poziomu energii komórkowej oraz wzrasta w odpowiedzi na ograniczenie kalorii i niedożywienie. SIRT1 uczestniczy w transdukcji sygnału wzrostowego na różnych poziomach – podwzgórze, wątroba, chrząstka wzrostowa - co zostało opisane w Publikacji nr 1 [26]. Wykazano również, że SIRT1 reguluje ekspresję kisspeptyny w podwzgórzu, wpływając na czas rozpoczęcia dojrzewania [27]. Zostało także opisane, że poziomy SIRT1 zmniejszają się wraz z wiekiem [28]. Dlatego naszą hipotezą było założenie, że stężenia SIRT1 we krwi mogą różnić się w zależności wzrostu, masy ciała dziecka, wieku czy etapu dojrzewania. Nie zaobserwowaliśmy jednak żadnych znaczących różnic w stężeniach SIRT1 w surowicy w odniesieniu do wieku, wzrostu i poziomów IGF-1, a także płci czy pokwitania.

SIRT1 uczestniczy w reagowaniu na zaburzenia równowagi metabolicznej, a jej ilość zależy od dostępności i rodzaju składników odżywczych. W naszym badaniu zostało wykazane, że poziom SIRT1 w surowicy był wyższy u dzieci spożywających więcej owoców i warzyw (co najmniej 2-3 porcje dziennie). Co ciekawe, dzieci te były również szczuplejsze i miały niższy poziom IGF-1 w surowicy, natomiast ich wzrost się nie różnił. Obniżony poziom IGF-1 w surowicy może być związany z sirtuinami, ale zbyt wiele czynników wpływa na wartości IGF-1, aby wyciągnąć wnioski na ten temat. Wyniki naszej pracy są zgodne z badaniami wykazującymi, że SIRT1 może być

aktywowana przez polifenole, występujące w szczególności w niektórych owocach i warzywach. Najlepiej poznanym polifenolem wyzwalającym SIRT1 jest resweratrol, ale także piceatannol, kwercetyna czy fisetyna, mogą aktywować SIRT1 [29,30]. Polifenole te można znaleźć między innymi w winogronach i produktach z winogron takich jak czerwone wino oraz jabłkach, kiwi, daktylach, truskawkach, jagodach, winogronach, cebuli, czosnku, czy w białej herbacie. Niektóre dane sugerują, że poza polifenolami, także składniki nabiału (np. leucyna) mogą służyć jako aktywatory SIRT1 [31]. W naszym badaniu dzieci z wysokim spożyciem produktów mlecznych również miały istotnie zwiększone stężenie SIRT1 w surowicy. Wyniki prezentowanej analizy potwierdzają, że dieta ma wpływ na stężenie SIRT1 u dzieci, co może mieć implikacje terapeutyczne. Według danych z literatury resweratrol i inne aktywatory SIRT1 wykazują korzystne efekty w chorobach związanych z wiekiem, w tym otyłości, cukrzycy, stanach zapalnych czy chorobach sercowo-naczyniowych, ale także ADHD czy dystrofii mięśniowej [32–35]. W ostatnich latach pojawiła się koncepcja „Sirtfood" jako diety opartej na produktach będących wyzwalaczami sirtuin, która może wykazywać działanie prozdrowotne, wpływając korzystnie na metabolizm oraz zapobiegając chorobom przewlekłym [36].

Pomimo, że to nasze badanie było ograniczone małą liczebnością próby, należy podkreślić, że stanowi ono pierwsze badanie skupiające się na wpływie różnych czynników na stężenie SIRT1 w surowicy u dzieci, co może przyczynić się do poznania roli SIRT1 w fizjologii pediatrycznej i jej potencjalnego znaczenia dla różnych aspektów zdrowia dzieci, ze szczególnym uwzględnieniem wzrastania i odżywiania.

Trzecia publikacja dotyczyła analizy stężeń SIRT1 u pacjentów z niedoborem wzrostu o różnej etiologii. W badaniu, zostało wykazane, że stężenie SIRT1 w surowicy nie różniło się między grupami GHD i ISS, ale w obu tych grupach było istotnie wyższe niż w grupie dzieci zdrowych. Przypuszcza się, że SIRT1 może więc odgrywać rolę w zaburzeniach wzrastania u dzieci. Powszechnie wiadomo, że niski wzrost i wolne tempo wzrastania dzieci zależą przede wszystkim od niskiego stężenia lub dostępności IGF-1, który jest głównym mediatorem działania GH w tkankach obwodowych [37]. Z powyższego badania zostały wykluczone dzieci, które mogły mieć obniżone IGF-1 ze znanych przyczyn, innych niż niedobór GH, czyli m.in. dzieci chore przewlekle, z zaburzeniami wchłaniania, niedożywione czy z nieleczoną niedoczynnością tarczycy.

Wobec tego w analizowanej grupie dzieci (ISS) powody obniżonego IGF-1 przy prawidłowym poziomie GH pozostawały nieznane. Wydaje się, że przynajmniej niektóre z nich mogą być związane z nadmiernym poziomem SIRT1.

W badaniu zostało wykazane, że SIRT1 koreluje ujemne ze stężeniem IGF-1, stosunkiem molowym IGF-1/IGFBP-3 oraz stopniem niedoboru wzrostu. W pewnych niekorzystnych warunkach zwiększony poziom SIRT1 może służyć jako czynnik prowadzący do zahamowania wydzielania IGF-1 w celu oszczędzania energii i utrzymania homeostazy. Należy podkreślić, że SIRT1 reaguje na zaburzenia równowagi metabolicznej spowodowane ograniczeniem kalorii i niedożywieniem. W badaniu wykazano, że poziomy SIRT1 były związane ze stanem odżywienia dziecka. Stężenia SIRT1 były ujemnie skorelowane z SDS masy ciała i BMI SDS. Wiadomo, że GH ma działanie hiperglikemiczne poprzez uwalnianie rezerw glukozy z tkanek, podczas gdy IGF-1 ma działanie hipoglikemiczne podobne do insuliny [38,39]. Dlatego w przypadku niedożywienia, zwiększone wydzielanie GH z przysadki i zmniejszone wydzielanie IGF-1 z wątroby w tym samym czasie (jak na przykład w jadłowstręcie psychicznym) jest korzystne dla organizmu (w celu utrzymania prawidłowego poziomu glukozy we krwi) [40]. Wydaje się, że przyczynia się do tego rosnące stężenie SIRT1, ponieważ zostało wykazane, że SIRT1 negatywnie reguluje indukowaną przez GH produkcję IGF-1 poprzez modulowanie aktywności szlaku JAK2/STAT w hepatocytach [2,41].

Z drugiej strony, nie jest w pełni zrozumiale, dlaczego poziomy SIRT1 były wyższe również u dzieci z GHD (zarówno pGHD, jak i sGHD) niż w grupie dzieci zdrowych. Nasze wyniki pokazały, że SIRT1 dodatnio korelowała z maksymalnym wydzielaniem GH w testach stymulacyjnych tylko w grupie dzieci z GHD. Można to wyjaśnić w następujący sposób: u dzieci z GHD i prawidłowym BMI nie ma ograniczeń w dostarczaniu egzogennych składników odżywczych, ale z powodu niskiego wydzielania GH dochodzi do upośledzenia katabolicznego działania GH - dostarczania glukozy i wolnych kwasów tłuszczowych z tkanek do krwiobiegu. Taka konstelacja może wyzwalać sygnały podobne do stanu deficytu kalorycznego (aktywacja greliny) i w ten sposób aktywować SIRT1 w niektórych tkankach, np. w podwzgórzu. Dlatego podejrzewamy, że w przypadku pacjentów z GHD, SIRT1 może wzrastać w odpowiedzi na obniżony poziom GH (w celu promowania wydzielania GH w mechanizmie dodatniego sprzężenia zwrotnego). W dotychczasowych badaniach stwierdzono

bowiem, że SIRT1 ulega szerokiej ekspresji w podwzgórzu (w szczególności w jądrze łukowatym) [42]. Aktywność SIRT1 w mózgu jest prawdopodobnie zaangażowana w regulację osi somatotropowej w odpowiedzi na podaż energii [18,43].

Chociaż analiza statystyczna nie wykazała istotnej różnicy między stężeniem SIRT1 w grupach GHD i ISS, warto podkreślić, że wśród dzieci z grupy GHD zdarzały się przypadki bardzo wysokich stężeń SIRT1, a dyspersja wokół średniej była duża, co sugeruje heterogeniczność tej grupy. Podzielono więc pacjentów pod względem maksymalnego wydzielania GH w testach stymulacyjnych na sGHD (maxGH < 7 ng/ml) i pGHD (maxGH 7 – 10 ng/ml). Okazało się, że dzieci z pGHD wykazywały podobne cechy do dzieci z grupy ISS - były podobnie szczupłe i miały podobnie obniżone stężenia IGF-1 (pomimo innego wydzielania GH) oraz znacznie różniły się pod tymi względami od pacjentów z ciężką postacią GHD. W doniesieniach medycznych często wspomina się, że dzieci z pGHD mogą stanowić grupę, która nie wymaga leczenia rhGH i nie różni się znacząco od dzieci z ISS [44]. Diagnostyka różnicowa między GHD a ISS nastręcza trudności, ponieważ pomiędzy prawidłowym wydzielaniem GH a niedoborem GH występuje *continuum* a rozpoznanie GHD, w zależności od maksymalnego wydzielania GH w testach stymulacyjnych poniżej 10 ng/ml (lub mniej niż 7, w zależności od rekomendacji) zostało ustalone w sposób arbitralny [45]. Niemniej jednak, stężenia SIRT1 różniły się znacząco między tymi dwiema grupami (pGHD i ISS). Obecnie trudno jest określić przyczynę tej obserwacji; można jedynie spekulować, że wyższe poziomy SIRT1 w pGHD wynikają ze zmniejszonego wydzielania GH, a różnice w wydzielaniu GH mają znaczenie.

Podsumowując, powyższa analiza, która jest jedną z pierwszych prób oceny stężenia SIRT1 w surowicy u dzieci, wykazuje związek między niskim wzrostem, GH/IGF-1 i SIRT1. Zaangażowanie SIRT1 w mechanizmy regulacyjne dotyczące wzrastania, metabolizmu i starzenia stanowi interesującą perspektywę dla dalszych badań.

5.6. Wnioski

1. Stężenia SIRT1 w surowicy u zdrowych dzieci nie różnią się w zależności od płci, wieku, stadium dojrzewania, parametrów auksologicznych i poziomów IGF-1.

2. Większa częstość spożywania owoców, warzyw oraz produktów mlecznych wiąże się ze zwiększonym stężeniem SIRT1 w surowicy u dzieci zdrowych.

3. U niskorosłych dzieci, niezależnie od wydzielania GH, stężenie SIRT1 w surowicy jest podwyższone.

4. Istnieje istotna ujemna korelacja pomiędzy SIRT1 a: poziomem IGF-1, stosunkiem molowym IGF-1/IGFBP-3 oraz nasileniem niedoboru wzrostu. SIRT1 może stanowić jeden z mechanizmów, poprzez które wydzielanie IGF-1 jest zmniejszone u niskorosłych dzieci.

5.7. Piśmiennictwo

1. Boguszewski, M.C.S. Growth Hormone Deficiency and Replacement in Children. *Rev Endocr Metab Disord* **2021**, *22*, 101–108, doi:10.1007/s11154-020-09604-2.
2. Yamamoto, M.; Iguchi, G.; Fukuoka, H.; Suda, K.; Bando, H.; Takahashi, M.; Nishizawa, H.; Seino, S.; Takahashi, Y. SIRT1 Regulates Adaptive Response of the Growth Hormone--Insulin-like Growth Factor-I Axis under Fasting Conditions in Liver. *Proc Natl Acad Sci U S A* **2013**, *110*, 14948–14953, doi:10.1073/pnas.1220606110.
3. Kang, X.; Yang, W.; Wang, R.; Xie, T.; Li, H.; Feng, D.; Jin, X.; Sun, H.; Wu, S. Sirtuin-1 (SIRT1) Stimulates Growth-Plate Chondrogenesis by Attenuating the PERK-eIF-2α-CHOP Pathway in the Unfolded Protein Response. *J Biol Chem* **2018**, *293*, 8614–8625, doi:10.1074/jbc.M117.809822.
4. Haigis, M.C.; Sinclair, D.A. Mammalian Sirtuins: Biological Insights and Disease Relevance. *Annual Review of Pathology: Mechanisms of Disease* **2010**, *5*, 253–295, doi:10.1146/annurev.pathol.4.110807.092250.
5. Kułaga, Z.; Litwin, M.; Tkaczyk, M.; Palczewska, I.; Zajączkowska, M.; Zwolińska, D.; Krynicki, T.; Wasilewska, A.; Moczulska, A.; Morawiec-Knysak, A.; et al. Polish 2010 Growth References for School-Aged Children and Adolescents. *Eur J Pediatr* **2011**, *170*, 599–609, doi:10.1007/s00431-010-1329-x.
6. Tanner, J.M.; Goldstein, H.; Whitehouse, R.H. Standards for Children's Height at Age 2 to 9 Years Allowing for Height of Parents. *Arch Dis Child* **1970**, *45*, 819.
7. Walker, D.G. Radiographic Atlas of Skeletal Development of the Hand and Wrist. William Walter Greulich and S. Idell Pyle. Stanford University Press, Stanford, Calif.; Oxford University Press, London, Ed. 2, 1959. Xvi + 256 Pp. Illus. $15. *Science* **1959**, doi:10.1126/science.130.3369.215.a.
8. Ranke, M.B.; Wit, J.M. Growth Hormone - Past, Present and Future. *Nat Rev Endocrinol* **2018**, *14*, 285–300, doi:10.1038/nrendo.2018.22.
9. Lewiński, A.; Karbownik-Lewińska, M.; Wieczorek-Szukała, K.; Stasiak, M.; Stawerska, R. Contribution of Ghrelin to the Pathogenesis of Growth Hormone Deficiency. *Int J Mol Sci* **2021**, *22*, 9066, doi:10.3390/ijms22169066.
10. Juul, A. Serum Levels of Insulin-like Growth Factor I and Its Binding Proteins in Health and Disease. *Growth Hormone & IGF Research* **2003**, *13*, 113–170, doi:10.1016/S1096-6374(03)00038-8.

11. Boguszewski, C.L.; Barbosa, E.J.L.; Svensson, P.A.; Johannsson, G.; Glad, C.A.M. Mechanisms in Endocrinology: Clinical and Pharmacogenetic Aspects of the Growth Hormone Receptor Polymorphism. *European Journal of Endocrinology* **2017**, *177*, undefined-undefined, doi:10.1530/EJE-17-0549.
12. Pardo, P.S.; Boriek, A.M. SIRT1 Regulation in Ageing and Obesity. *Mech Ageing Dev* **2020**, *188*, 111249, doi:10.1016/j.mad.2020.111249.
13. Chen, C.; Zhou, M.; Ge, Y.; Wang, X. SIRT1 and Aging Related Signaling Pathways. *Mechanisms of Ageing and Development* **2020**, *187*, 111215, doi:10.1016/j.mad.2020.111215.
14. Yang, Y.; Liu, Y.; Wang, Y.; Chao, Y.; Zhang, J.; Jia, Y.; Tie, J.; Hu, D. Regulation of SIRT1 and Its Roles in Inflammation. *Frontiers in Immunology* **2022**, *13*.
15. de Lima, J.B.M.; Ubah, C.; Debarba, L.K.; Ayyar, I.; Didyuk, O.; Sadagurski, M. Hypothalamic GHR-SIRT1 Axis in Fasting. *Cells* **2021**, *10*, 891, doi:10.3390/cells10040891.
16. Cao, Y.; Jiang, X.; Ma, H.; Wang, Y.; Xue, P.; Liu, Y. SIRT1 and Insulin Resistance. *Journal of Diabetes and its Complications* **2016**, *30*, 178–183, doi:10.1016/j.jdiacomp.2015.08.022.
17. Lee, S.-H.; Lee, J.-H.; Lee, H.-Y.; Min, K.-J. Sirtuin Signaling in Cellular Senescence and Aging. *BMB Rep* **2019**, *52*, 24–34, doi:10.5483/BMBRep.2019.52.1.290.
18. Yamamoto, M.; Takahashi, Y. The Essential Role of SIRT1 in Hypothalamic-Pituitary Axis. *Front Endocrinol (Lausanne)* **2018**, *9*, 605, doi:10.3389/fendo.2018.00605.
19. Waters, M.J.; Brooks, A.J. JAK2 Activation by Growth Hormone and Other Cytokines. *Biochem J* **2015**, *466*, 1–11, doi:10.1042/BJ20141293.
20. Velásquez, D.A.; Martínez, G.; Romero, A.; Vázquez, M.J.; Boit, K.D.; Dopeso-Reyes, I.G.; López, M.; Vidal, A.; Nogueiras, R.; Diéguez, C. The Central Sirtuin 1/P53 Pathway Is Essential for the Orexigenic Action of Ghrelin. *Diabetes* **2011**, *60*, 1177–1185, doi:10.2337/db10-0802.
21. Dai, H.; Sinclair, D.A.; Ellis, J.L.; Steegborn, C. Sirtuin Activators and Inhibitors: Promises, Achievements, and Challenges. *Pharmacol Ther* **2018**, *188*, 140–154, doi:10.1016/j.pharmthera.2018.03.004.
22. Fedorczak, A.; Lewiński, A.; Stawerska, R. Sirtuin 1 Serum Concentration in Healthy Children - Dependence on Sex, Age, Stage of Puberty, Body Weight and Diet. *Front Endocrinol (Lausanne)* **2024**, *15*, 1356612, doi:10.3389/fendo.2024.1356612.
23. Gonçalinho, G.H.F.; Nascimento, J.R. de O.; Mioto, B.M.; Amato, R.V.; Moretti, M.A.; Strunz, C.M.C.; César, L.A.M.; Mansur, A. de P. Effects of Coffee on Sirtuin-1, Homocysteine, and Cholesterol of Healthy Adults: Does the Coffee Powder Matter? *J Clin Med* **2022**, *11*, 2985, doi:10.3390/jcm11112985.
24. Charążka, B.; Siejka, A. Correlations between Serum Sirtuin Levels and Cardiovascular Risk Factors in Women with Polycystic Ovary Syndrome. *Adv Med Sci* **2022**, *67*, 123–128, doi:10.1016/j.advms.2022.01.004.
25. Mariani, S.; Fiore, D.; Persichetti, A.; Basciani, S.; Lubrano, C.; Poggiogalle, E.; Genco, A.; Donini, L.M.; Gnessi, L. Circulating SIRT1 Increases After Intragastric Balloon Fat Loss in Obese Patients. *Obes Surg* **2016**, *26*, 1215–1220, doi:10.1007/s11695-015-1859-4.
26. Fedorczak, A.; Lewiński, A.; Stawerska, R. Involvement of Sirtuin 1 in the Growth Hormone/Insulin-like Growth Factor 1 Signal Transduction and Its Impact on

Growth Processes in Children. *Int J Mol Sci* **2023**, *24*, 15406, doi:10.3390/ijms242015406.
27. Vazquez, M.J.; Toro, C.A.; Castellano, J.M.; Ruiz-Pino, F.; Roa, J.; Beiroa, D.; Heras, V.; Velasco, I.; Dieguez, C.; Pinilla, L.; et al. SIRT1 Mediates Obesity- and Nutrient-Dependent Perturbation of Pubertal Timing by Epigenetically Controlling Kiss1 Expression. *Nat Commun* **2018**, *9*, 4194, doi:10.1038/s41467-018-06459-9.
28. Zhong, Y.; Chen, A.F.; Zhao, J.; Gu, Y.-J.; Fu, G.-X. Serum Levels of Cathepsin D, Sirtuin1, and Endothelial Nitric Oxide Synthase Are Correlatively Reduced in Elderly Healthy People. *Aging Clin Exp Res* **2016**, *28*, 641–645, doi:10.1007/s40520-015-0472-7.
29. Cui, Z.; Zhao, X.; Amevor, F.K.; Du, X.; Wang, Y.; Li, D.; Shu, G.; Tian, Y.; Zhao, X. Therapeutic Application of Quercetin in Aging-Related Diseases: SIRT1 as a Potential Mechanism. *Front Immunol* **2022**, *13*, 943321, doi:10.3389/fimmu.2022.943321.
30. Mahjabeen, W.; Khan, D.A.; Mirza, S.A. Role of Resveratrol Supplementation in Regulation of Glucose Hemostasis, Inflammation and Oxidative Stress in Patients with Diabetes Mellitus Type 2: A Randomized, Placebo-Controlled Trial. *Complement Ther Med* **2022**, *66*, 102819, doi:10.1016/j.ctim.2022.102819.
31. Li, H.; Xu, M.; Lee, J.; He, C.; Xie, Z. Leucine Supplementation Increases SIRT1 Expression and Prevents Mitochondrial Dysfunction and Metabolic Disorders in High-Fat Diet-Induced Obese Mice. *Am J Physiol Endocrinol Metab* **2012**, *303*, E1234–E1244, doi:10.1152/ajpendo.00198.2012.
32. Chen, J.; Lou, R.; Zhou, F.; Li, D.; Peng, C.; Lin, L. Sirtuins: Key Players in Obesity-Associated Adipose Tissue Remodeling. *Front Immunol* **2022**, *13*, 1068986, doi:10.3389/fimmu.2022.1068986.
33. García-Martínez, B.I.; Ruiz-Ramos, M.; Pedraza-Chaverri, J.; Santiago-Osorio, E.; Mendoza-Núñez, V.M. Effect of Resveratrol on Markers of Oxidative Stress and Sirtuin 1 in Elderly Adults with Type 2 Diabetes. *Int J Mol Sci* **2023**, *24*, 7422, doi:10.3390/ijms24087422.
34. Najjar, R.S.; Feresin, R.G. Protective Role of Polyphenols in Heart Failure: Molecular Targets and Cellular Mechanisms Underlying Their Therapeutic Potential. *International Journal of Molecular Sciences* **2021**, *22*, 1668, doi:10.3390/ijms22041668.
35. Kawamura, K.; Fukumura, S.; Nikaido, K.; Tachi, N.; Kozuka, N.; Seino, T.; Hatakeyama, K.; Mori, M.; Ito, Y.M.; Takami, A.; et al. Resveratrol Improves Motor Function in Patients with Muscular Dystrophies: An Open-Label, Single-Arm, Phase IIa Study. *Sci Rep* **2020**, *10*, 20585, doi:10.1038/s41598-020-77197-6.
36. Akan, O.D.; Qin, D.; Guo, T.; Lin, Q.; Luo, F. Sirtfoods: New Concept Foods, Functions, and Mechanisms. *Foods* **2022**, *11*, 2955, doi:10.3390/foods11192955.
37. Smyczyńska, J.; Stawerska, R.; Lewiński, A.; Hilczer, M. Do IGF-I Concentrations Better Reflect Growth Hormone (GH) Action in Children with Short Stature than the Results of GH Stimulating Tests? Evidence from the Simultaneous Assessment of Thyroid Function. *Thyroid Res* **2011**, *4*, 6, doi:10.1186/1756-6614-4-6.
38. Donato, J.; Wasinski, F.; Furigo, I.C.; Metzger, M.; Frazão, R. Central Regulation of Metabolism by Growth Hormone. *Cells* **2021**, *10*, 129, doi:10.3390/cells10010129.
39. Huang, Z.; Huang, L.; Waters, M.J.; Chen, C. Insulin and Growth Hormone Balance: Implications for Obesity. *Trends Endocrinol Metab* **2020**, *31*, 642–654, doi:10.1016/j.tem.2020.04.005.

40. Fazeli, P.K.; Klibanski, A. Determinants of GH Resistance in Malnutrition. *J Endocrinol* **2014**, *220*, R57-65, doi:10.1530/JOE-13-0477.
41. Wójcik, M.; Krawczyńska, A.; Antushevich, H.; Herman, A.P. Post-Receptor Inhibitors of the GHR-JAK2-STAT Pathway in the Growth Hormone Signal Transduction. *Int J Mol Sci* **2018**, *19*, E1843, doi:10.3390/ijms19071843.
42. Satoh, A.; Brace, C.S.; Ben-Josef, G.; West, T.; Wozniak, D.F.; Holtzman, D.M.; Herzog, E.D.; Imai, S. -i. SIRT1 Promotes the Central Adaptive Response to Diet Restriction through Activation of the Dorsomedial and Lateral Nuclei of the Hypothalamus. *Journal of Neuroscience* **2010**, *30*, 10220–10232, doi:10.1523/JNEUROSCI.1385-10.2010.
43. de Lima, J.B.M.; Ubah, C.; Debarba, L.K.; Ayyar, I.; Didyuk, O.; Sadagurski, M. Hypothalamic GHR-SIRT1 Axis in Fasting. *Cells* **2021**, *10*, 891, doi:10.3390/cells10040891.
44. Krukowska-Andrzejczyk, B.; Kalina, M.; Kalina-Faska, B.; Małecka-Tendera, E. Growth Hormone Therapy in Children with Partial Growth Hormone Deficiency. Are We Treating the Right Patients? *Pediatr Endocrinol Diabetes Metab* **2020**, *26*, 65–72, doi:10.5114/pedm.2020.95624.
45. Murray, P.G.; Dattani, M.T.; Clayton, P.E. Controversies in the Diagnosis and Management of Growth Hormone Deficiency in Childhood and Adolescence. *Arch Dis Child* **2016**, *101*, 96–100, doi:10.1136/archdischild-2014-307228.

6. Zgoda komisji bioetycznej

Uzyskano zgodę Komisji Bioetycznej przy Instytucie Centrum Zdrowia Matki Polki w Łodzi (opinia nr 47/2020).

Komisja Bioetyczna
przy Instytucie Centrum Zdrowia Matki Polki
93-338 Łódź, Rzgowska 281/289 tel. (42) 271 15 97
e-mail pnserca@iczmp.edu.pl

Łódź, dnia 15 września 2020 r.

Dr hab. n. med. Renata Stawerska
Klinika Endokrynologii i Chorób Metabolicznych
Instytutu Centrum Zdrowia Matki Polki w Łodzi

Komisja Bioetyczna przy Instytucie Centrum Zdrowia Matki Polki działając zgodnie z zasadami Dobrej Praktyki Klinicznej na posiedzeniu w dniu 15 września 2020 r. rozpatrywała wniosek dotyczący pracy:

„Analiza zależności między SIRT1, FGF21 i wisfatyną a stężeniem IGF-1 u dzieci z idiopatycznym niedoborem wzrostu."

Zespół badaczy:
1. Dr hab. n. med. Renata Stawerska
2. Lek. Anna Fedorczak
3. Lek. Anna Łupińska
4. Lek. Małgorzata Kolasa-Kicińska
5. Prof. dr hab. n. med. Andrzej Lewiński

Opinia Nr 47/2020

Komisja Bioetyczna przy Instytucie Centrum Zdrowia Matki Polki zapoznała się z ww projektem eksperymentu medycznego, przeanalizowała wniosek, wysłuchała opinii recenzenta o przedstawionym projekcie i wyniku przeprowadzonej dyskusji oraz tajnego głosowania, po rozważeniu kryteriów etycznych oraz celowości i wykonalności projektu pozytywnie zaopiniowała projekt eksperymentu medycznego.

Uchwałę podjęto jednogłośnie.
Uchwałę podjęto przy sprzeciwie .....

Przewodnicząca:
Dr hab. med. Iwona Maroszyńska, prof. instytutu .....

Zastępca Przewodniczącej:
Prof. dr hab. n. farm. Daria Orszulak-Michalak .....

Członkowie:
Mec. Michał Araszkiewicz .....
Prof. dr hab. n. med. Tadeusz Biegański .....
Dr n. med. Paweł Czekalski .....
Dr hab. n. med. Piotr Grzelak, prof. instytutu .....
Mgr Grażyna Korybut .....
Dr n. med. Michał Krekora .....
Prof. dr hab. med. Jacek Rysz .....
Dr n. filozofii Wojciech Sztombka .....
Ks. dr hab. Jan Wolski .....
Dr hab. n. med. Marek Zadrożny, prof. instytutu .....
Prof. dr hab. n. med. Krzysztof Zeman .....

7. Finansowanie badania

Badania zostały zrealizowane dzięki środkom pozyskanym z grantu wewnętrznego Instytutu Centrum Zdrowia Matki Polki nr 15GW/2021, który był finansowany z budżetu Ministerstwa Nauki i Szkolnictwa Wyższego.

Instytut „Centrum Zdrowia Matki Polki"
93-338 Łódź, ul. Rzgowska 281/289
tel.: 42-271-11-23, 42-271-11-29, 42-271-16-10; e-mail: nauka@iczmp.edu.pl

Łódź, 25.05.2021 r.

Anna Fedorczak
Klinika Endokrynologii i Chorób Metabolicznych
Instytutu „Centrum Zdrowia Matki Polki"

Szanowna Pani,

Uprzejmie informuję, że zgodnie z rekomendacją Komisji ds. grantów wewnętrznych i decyzją Dyrektora ICZMP projekt badawczy, którego jest Pani kierownikiem, został przyjęty do realizacji i uzyskał finansowanie w wysokości **20 000 zł** w związku z tym proszę o restrukturyzację budżetu **do dnia 30.05.2021 r.,** gdyż jest to warunek konieczny do uruchomienia projektu.

- Tytuł projektu: *Analiza zależności między SIRT1, FGF-21 i wisfatyną, a stężeniem IGF-1 u dzieci z idiopatycznym niedoborem wzrostu*
- Nr projektu: 15GW/2021

Przypominam również, że zgodnie z zaleceniami zawartymi w sprawozdaniu z audytu zewnętrznego działalności statutowej wymagane jest monitorowanie stanu realizacji projektów w ciągu roku. Brak sprawozdania może skutkować wstrzymaniem finansowania lub zakończeniem projektu. W związku z powyższym, po zakończeniu (drugiego półrocza 2021 roku), proszę o złożenie w Dziale Współpracy Naukowej i Badawczej (pawilon A, poziom I, pok. nr 13) krótkiego sprawozdania odnoszącego się do harmonogramu realizowanego projektu.

Zwracam się z prośbą by każdy zakup usług i produktów finansowany z budżetu grantów wewnętrznych był wcześniej konsultowany z opiekunem projektu (Julia Sauter, tel. 42-271-16-10), w jakim trybie zakupu powinien zostać nabyty. Nieprzestrzeganie zapisów zamówień publicznych może skutkować brakiem możliwości finansowania zakupów ze środków Instytutu.

Z poważaniem,

Dr hab. Dariusz Trzmielak
Przewodniczący Komisji ds. grantów wewnętrznych

Do wiadomości opiekuna naukowego:
dr hab. n. med. Renata Stawerska

Łódź, 28.08.2024r.

lek. Anna Fedorczak

Klinika Endokrynologii i Chorób Metabolicznych

Instytut Centrum Zdrowia Matki Polki w Łodzi

OŚWIADCZENIE

Jako pierwszy autor publikacji:

"Involvement of sirtuin 1 in the growth hormone/insulin-like growth factor 1 signal transduction and its impact on growth processes in children."

oświadczam, iż w wyżej wymienionej pracy mój wkład w powstanie publikacji polegał na: stworzeniu konceptu pracy, wykonaniu przeglądu literatury, wyboru publikacji, interpretacji wyników, napisaniu artykułu. Mój udział w realizacji pracy szacuję na 60%.

Jednocześnie wyrażam zgodę na wykorzystanie w/w pracy jako części cyklu publikacji do przeprowadzenia przewodu doktorskiego.

Anna Fedorczak

Podpis

Łódź, 28.08.2024r.

Prof. dr hab. n. med. Andrzej Lewiński
Klinika Endokrynologii i Chorób Metabolicznych
Instytut Centrum Zdrowia Matki Polki w Łodzi

OŚWIADCZENIE

Jako współautor publikacji:

„Involvement of sirtuin 1 in the growth hormone/insulin-like growth factor 1 signal transduction and its impact on growth processes in children."

oświadczam, iż w wyżej wymienionej pracy mój wkład w powstanie publikacji polegał na: krytycznej ocenie treści artykułu, zaakceptowaniu ostatecznej treści artykułu. Mój udział w realizacji pracy szacuję na 10%.

Jednocześnie wyrażam zgodę na wykorzystanie w/w pracy przedłożonej przez lek. Annę Fedorczak, jako części cyklu publikacji do przeprowadzenia przewodu doktorskiego.

.....
Podpis

Łódź, 28.08.2024r.

Prof. dr hab. n. med. Renata Stawerska

Klinika Endokrynologii i Chorób Metabolicznych

Instytut Centrum Zdrowia Matki Polki w Łodzi

OŚWIADCZENIE

Jako współautor publikacji:

„Involvement of sirtuin 1 in the growth hormone/insulin-like growth factor 1 signal transduction and its impact on growth processes in children."

oświadczam, iż w wyżej wymienionej pracy mój wkład w powstanie publikacji polegał na: stworzeniu konceptu pracy, interpretacji wyników, poprawianiu artykułu, krytycznej ocenie treści artykułu, zaakceptowaniu ostatecznej treści artykułu. Mój udział w realizacji pracy szacuję na 30%.

Jednocześnie wyrażam zgodę na wykorzystanie w/w pracy przedłożonej przez lek. Annę Fedorczak, jako części cyklu publikacji do przeprowadzenia przewodu doktorskiego.

.....

Podpis

Łódź, 28.08.2024r.

.

lek. Anna Fedorczak

Klinika Endokrynologii i Chorób Metabolicznych

Instytut Centrum Zdrowia Matki Polki w Łodzi

OŚWIADCZENIE

Jako pierwszy autor publikacji:

"Sirtuin 1 serum concentration in healthy children - dependence on sex, age, stage of puberty, body weight and diet."

oświadczam, iż w wyżej wymienionej pracy mój wkład w powstanie publikacji polegał na: stworzeniu konceptu pracy, pozyskaniu finansowania, zebraniu materiału badawczego, zakupie odczynników, analizie statystycznej, interpretacji wyników, napisaniu artykułu. Mój udział w realizacji pracy szacuję na 60%.

Jednocześnie wyrażam zgodę na wykorzystanie w/w pracy jako części cyklu publikacji do przeprowadzenia przewodu doktorskiego.

Anna Fedorczak

Podpis

Łódź, 28.08.2024r.

Prof. dr hab. n. med. Andrzej Lewiński
Klinika Endokrynologii i Chorób Metabolicznych
Instytut Centrum Zdrowia Matki Polki w Łodzi

OŚWIADCZENIE

Jako współautor publikacji:

„Sirtuin 1 serum concentration in healthy children - dependence on sex, age, stage of puberty, body weight and diet."

oświadczam, iż w wyżej wymienionej pracy mój wkład w powstanie publikacji polegał na: krytycznej ocenie treści artykułu, zaakceptowaniu ostatecznej treści artykułu. Mój udział w realizacji pracy szacuję na 10%.

Jednocześnie wyrażam zgodę na wykorzystanie w/w pracy przedłożonej przez lek. Annę Fedorczak, jako części cyklu publikacji do przeprowadzenia przewodu doktorskiego.

.....
Podpis

Łódź, 28.08.2024r.

Prof. dr hab. n. med. Renata Stawerska
Klinika Endokrynologii i Chorób Metabolicznych
Instytut Centrum Zdrowia Matki Polki w Łodzi

OŚWIADCZENIE

Jako współautor publikacji:

"Sirtuin 1 serum concentration in healthy children - dependence on sex, age, stage of puberty, body weight and diet."

oświadczam, iż w wyżej wymienionej pracy mój wkład w powstanie publikacji polegał na: stworzeniu konceptu pracy, analizie statystycznej, interpretacji wyników, krytycznej ocenie treści artykułu, zaakceptowaniu ostatecznej treści artykułu. Mój udział w realizacji pracy szacuję na 30%.

Jednocześnie wyrażam zgodę na wykorzystanie w/w pracy przedłożonej przez lek. Annę Fedorczak, jako części cyklu publikacji do przeprowadzenia przewodu doktorskiego.

.....
Podpis

Łódź, 28.08.2024r.

lek. Anna Fedorczak

Klinika Endokrynologii i Chorób Metabolicznych

Instytut Centrum Zdrowia Matki Polki w Łodzi

OŚWIADCZENIE

Jako pierwszy autor publikacji:

"Relationship between serum sirtuin 1 and growth I hormone/insulin-like growth factor 1 concentrations in children with growth hormone deficiency and idiopathic short stature."

oświadczam, iż w wyżej wymienionej pracy mój wkład w powstanie publikacji polegał na: stworzeniu konceptu pracy, zebraniu materiału badawczego, analizie statystycznnej, interpretacji wyników, napisaniu artykułu. Mój udział w realizacji pracy szacuję na 40%.

Jednocześnie wyrażam zgodę na wykorzystanie w/w pracy jako części cyklu publikacji do przeprowadzenia przewodu doktorskiego.

Anna Fedorczak

Podpis

Łódź, 28.08.2024r.

lek. Dorota Kowalik

Klinika Endokrynologii i Chorób Metabolicznych

Instytut Centrum Zdrowia Matki Polki w Łodzi

OŚWIADCZENIE

Jako współautor publikacji:

„Relationship between serum sirtuin 1 and growth I hormone/insulin-like growth factor 1 concentrations in children with growth hormone deficiency and idiopathic short stature."

oświadczam, iż w wyżej wymienionej pracy mój wkład w powstanie publikacji polegał na: zebraniu materiału badawczego, interpretacji wyników. Mój udział w realizacji pracy szacuję na 5%.

Jednocześnie wyrażam zgodę na wykorzystanie w/w pracy przedłożonej przez lek. Annę Fedorczak, jako części cyklu publikacji do przeprowadzenia przewodu doktorskiego.

Dorota Kowalik

.....

Podpis

Łódź, 28.08.2024r.

lic. Justyna Kopciuch

Centrum Medycznej Diagnostyki Laboratoryjnej i Badań Przesiewowych

Instytut Centrum Zdrowia Matki Polki w Łodzi

OŚWIADCZENIE

Jako współautor publikacji:

„Relationship between serum sirtuin 1 and growth I hormone/insulin-like growth factor 1 concentrations in children with growth hormone deficiency and idiopathic short stature."

oświadczam, iż w wyżej wymienionej pracy mój wkład w powstanie publikacji polegał na: wykonaniu analiz. Mój udział w realizacji pracy szacuję na 10%.

Jednocześnie wyrażam zgodę na wykorzystanie w/w pracy przedłożonej przez lek. Annę Fedorczak, jako części cyklu publikacji do przeprowadzenia przewodu doktorskiego.

Justyna Kopciuch

.....
Podpis

Łódź, 28.08.2024r.

.

dr n. medycznych Ewa Głowacka

Centrum Medycznej Diagnostyki Laboratoryjnej i Badań Przesiewowych

Instytut Centrum Zdrowia Matki Polki w Łodzi

OŚWIADCZENIE

Jako współautor publikacji:

„Relationship between serum sirtuin 1 and growth I hormone/insulin-like growth factor 1 concentrations in children with growth hormone deficiency and idiopathic short stature."

oświadczam, iż w wyżej wymienionej pracy mój wkład w powstanie publikacji polegał na: wykonaniu analiz. Mój udział w realizacji pracy szacuję na 10%.

Jednocześnie wyrażam zgodę na wykorzystanie w/w pracy przedłożonej przez lek. Annę Fedorczak, jako części cyklu publikacji do przeprowadzenia przewodu doktorskiego.

.....

Podpis

Łódź, 28.08.2024r.

lek. Katarzyna Mikołajczyk
Klinika Pediatrii, Immunologii i Nefrologii
Instytut Centrum Zdrowia Matki Polki w Łodzi

OŚWIADCZENIE

Jako współautor publikacji:

„Relationship between serum sirtuin 1 and growth I hormone/insulin-like growth factor 1 concentrations in children with growth hormone deficiency and idiopathic short stature."

oświadczam, iż w wyżej wymienionej pracy mój wkład w powstanie publikacji polegał na: zebraniu materiału badawczego. Mój udział w realizacji pracy szacuję na 5%.

Jednocześnie wyrażam zgodę na wykorzystanie w/w pracy przedłożonej przez lek. Annę Fedorczak, jako części cyklu publikacji do przeprowadzenia przewodu doktorskiego.

Katarzyna Mikołajczyk
.....
Podpis

Łódź, 28.08.2024r.

Prof. dr hab. n. med. Marcin Tkaczyk
Klinika Pediatrii, Immunologii i Nefrologii
Instytut Centrum Zdrowia Matki Polki w Łodzi

OŚWIADCZENIE

Jako współautor publikacji:

„Relationship between serum sirtuin 1 and growth I hormone/insulin-like growth factor 1 concentrations in children with growth hormone deficiency and idiopathic short stature."

oświadczam, iż w wyżej wymienionej pracy mój wkład w powstanie publikacji polegał na: krytycznej ocenie treści artykułu. Mój udział w realizacji pracy szacuję na 5%.

Jednocześnie wyrażam zgodę na wykorzystanie w/w pracy przedłożonej przez lek. Annę Fedorczak, jako części cyklu publikacji do przeprowadzenia przewodu doktorskiego.

.....
Podpis

Łódź, 28.08.2024r.

Prof. dr hab. n. med. Andrzej Lewiński
Klinika Endokrynologii i Chorób Metabolicznych
Instytut Centrum Zdrowia Matki Polki w Łodzi

OŚWIADCZENIE

Jako współautor publikacji:

„Relationship between serum sirtuin 1 and growth I hormone/insulin-like growth factor 1 concentrations in children with growth hormone deficiency and idiopathic short stature."

oświadczam, iż w wyżej wymienionej pracy mój wkład w powstanie publikacji polegał na: krytycznej ocenie treści artykułu, zaakceptowaniu ostatecznej treści artykułu. Mój udział w realizacji pracy szacuję na 10%.

Jednocześnie wyrażam zgodę na wykorzystanie w/w pracy przedłożonej przez lek. Annę Fedorczak, jako części cyklu publikacji do przeprowadzenia przewodu doktorskiego.

.....
Podpis

Łódź, 28.08.2024r.

Prof. dr hab. n. med. Renata Stawerska
Klinika Endokrynologii i Chorób Metabolicznych
Instytut Centrum Zdrowia Matki Polki w Łodzi

OŚWIADCZENIE

Jako współautor publikacji:

"Relationship between serum sirtuin 1 and growth I hormone/insulin-like growth factor 1 concentrations in children with growth hormone deficiency and idiopathic short stature."

oświadczam, iż w wyżej wymienionej pracy mój wkład w powstanie publikacji polegał na: stworzeniu konceptu pracy, obliczeniu statystycznym, interpretacji wyników, krytycznej ocenie treści artykułu, zaakceptowaniu ostatecznej treści artykułu. Mój udział w realizacji pracy szacuję na 15%.

Jednocześnie wyrażam zgodę na wykorzystanie w/w pracy przedłożonej przez lek. Annę Fedorczak, jako części cyklu publikacji do przeprowadzenia przewodu doktorskiego.

.....
Podpis