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# Longitudinal changes in preschoolers' adiposity indicators according to compliance with 24-hour movement behavior guidelines: results from the ToyBox-study

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## Abstract

**Background** There is a lack of studies evaluating longitudinal changes in adiposity indicators and the association with 24-hour movement behavior guidelines in preschoolers. Therefore, this study aimed to investigate associations between changes in Body Mass Index (BMI) z-scores, waist circumference and waist-to-height ratio, and compliance with the 24-hour movement behavior guidelines in three- to six-year-old children from a European sample.

**Methods** In total, data from a European sample of 719 preschoolers (50.9% boys,  $4.76 \pm 0.45$  years) were analyzed at baseline and one year later. Physical activity (steps/day) was objectively measured using Omron pedometers. Screen time and sleep duration were subjectively measured using a parental questionnaire. Height, weight and waist circumference were measured by well-trained researchers, using standardized procedures and equipment, and were used to calculate BMI z-score and waist-to-height ratio. Linear mixed-effects models were used to investigate associations between changes in adiposity indicators (BMI z-score, waist circumference and waist-to-height ratio) and compliance with 24-hour movement behavior guidelines.

**Results** BMI z-scores changed differently from baseline to follow-up according to compliance with the integrated guidelines, both for weekdays (effect estimate =  $-0.15$ ,  $p=0.001$ ) and weekend days (effect estimate =  $-0.12$ ,  $p=0.046$ ). Preschoolers who did not comply with the integrated guidelines showed a small, significant increase in BMI z-scores compared to compliers having no significant change in BMI z-score. No significant difference between both groups over time was found for waist circumference and waist-to-height ratio neither for weekdays nor weekend days.

**Conclusions** The change in BMI z-scores after one year is less optimal for preschoolers who do not comply with the 24-hour movement behavior guidelines compared to preschoolers who comply with the integrated guidelines, although not clinically relevant. Future studies should incorporate longer follow-up periods to observe the effects of compliance to the integrated guidelines on adiposity indicators in preschool children.

**Keywords** 24-hour movement behaviors, Physical activity, Screen time, Sleep, Adiposity, Preschooler, BMI z-score, Waist circumference, Longitudinal

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## Background

Overweight and obesity are a public health concern and already occur at a young age. A systematic review and meta-regression by [1] showed that 17.9% of European 2- to 7-year olds were overweight and 5.3% were obese between 2006 and 2016 [1]. Childhood overweight and obesity is associated with an increased risk of for example type 2 diabetes mellitus, metabolic syndrome and cardiovascular risk factors [1]. In addition, overweight and obesity track from childhood to adulthood emphasizing the need for early prevention including a healthy lifestyle [2].

High levels of physical activity, low levels of sedentary behavior and a sufficient amount of sleep are important for health (including weight status) [3]. In the past, physical activity, sedentary behavior and sleep have been studied in isolation in preschoolers [4–7], and this was reflected in separate physical activity guidelines (i.e., 180 min of total physical activity per day), screen time guidelines (i.e., no more than 1 h of screen time per day), and sleep guidelines (i.e., sleep for 10 to 13 h per night, including nap times) for 3- to 5-year-old children [8–12]. A recent shift in research emphasizes that physical activity, sedentary behavior and sleep are co-dependent, which means that spending time on one or more behaviors affects the duration of the other behaviors within a 24-hour day, which has a consequence for health [13–16]. This integrated 24-hour approach has led to the establishment of new World Health Organization (WHO) guidelines which combined and updated the pre-existing guidelines in this age group. Following the WHO-guidelines, launched in 2019, a healthy 24-hour day for preschoolers consists of (1) at least 180 min of physical activity of which 60 min are spent in energetic play, (2) a maximum of one hour of sedentary screen time and not being restrained/sedentary for more than one hour at a time, and (3) 10 to 13 h of good quality sleep [17].

Since the launch of the 24-hour movement behavior guidelines, several studies have investigated preschoolers' compliance which were recently summarized in a systematic review showing that only 11.26% of all preschoolers worldwide comply with the 24-hour movement behavior guidelines [18]. In addition, a systematic review of [16] reported that up until then, four studies already studied the compliance with 24-hour movement behavior guidelines and adiposity indicators in preschoolers, being used as an indicator for health [3, 19–22]. Only the study of [21] found that meeting one (physical activity or sleep) or two (physical activity and sleep) of the three guidelines was associated with lower waist circumference and lower Body Mass Index (BMI) in Finnish preschool children (mean age 4.7 years old) [21]. Since the review of [16], new studies were published looking at the compliance with 24-hour movement behavior guidelines

and adiposity indicators in preschool children. However, most of these studies were cross-sectional which means that no statement can be made regarding causality. For example, are preschoolers with a higher BMI z-score more likely to not comply with the 24-hour movement behavior guidelines, or are preschoolers that do not comply with the 24-hour movement behavior guidelines more likely to have a higher BMI z-score [23]. The study of [24] looked at movement behaviors between one and five years of age and whether this can predict body composition at five years of age using dual-energy x-ray absorptiometry scans. They found that compositional time use was not associated with BMI z-score at five years of age [24]. One recent study of [25] looked at longitudinal associations between 24-hour movement behaviors and body composition in a small Swedish sample ( $n=231$  at follow-up) using air displacement plethysmography to measure body fatness from four years of age until nine years of age [25]. They found that movement behaviors at four years of age were not associated with body fatness at nine years of age.

Due to the lack of longitudinal data in larger samples, the aim of the current study was to investigate associations between compliance with 24-hour movement behavior guidelines and changes in adiposity indicators (Body Mass Index (BMI) z-scores, waist circumference and waist-to-height ratio) using longitudinal data from the European ToyBox-study. As behaviors might differ between weekdays and weekend days [26, 27], compliance with the integrated movement behavior guidelines and associations with changes in adiposity indicators will be explored for weekdays and weekend days separately. Results from this study will enable us to make causal statements regarding compliance with the 24-hour movement behavior guidelines and the development of adiposity as a key indicator of preschoolers' health.

## Methods

### Study protocol

The ToyBox-study aimed to develop, implement and evaluate a kindergarten-based, family-involved intervention in six European countries to prevent overweight and obesity among four- to six-year-old preschool children ([www.toybox-study.eu](http://www.toybox-study.eu)) [28]. The intervention was evaluated using a cluster randomized pre-test post-test design across the following six European countries: Belgium, Bulgaria, Germany, Greece, Poland, and Spain. Only data from the control group was included in the data analysis of this study.

Preschoolers and their parents/caregivers were recruited at kindergartens, daycare centers or preschool settings, depending on the country regulations and legislation. In Germany, Bulgaria, Spain and Poland, children

and their families were recruited from kindergartens, in Greece from kindergartens and daycare centers, and in Belgium from preschool settings. To avoid confusion for the reader, all these settings (kindergartens, daycare centers, preschool settings) will be referred to as “kindergartens” in this paper. The ToyBox-study was approved by the Ethical Committees in all European countries, in line with national regulations. The ToyBox-study is registered with the clinical trials registry [clinicaltrials.gov](https://clinicaltrials.gov), ID: NCT02116296 (registration date: 02/04/2014).

Kindergartens from different socio-economic status (SES) backgrounds were randomly selected in the provinces of East- and West-Flanders in Belgium, Varna in Bulgaria, Bavaria in Germany, Attica in Greece, Mazowiecki in Poland, and Zaragoza in Spain. Within all selected provinces, lists of all municipalities were constructed. To create tertiles based on SES, information on mean years of education for the population aged 25–55 years or annual income was collected. After creating these tertiles, five municipalities were randomly selected from each tertile in each country, resulting in five municipalities for low SES, five for medium SES, and five for high SES. Within these randomly chosen municipalities, 906 kindergartens were randomly selected (with the exclusion of the lowest 20% of kindergartens with the smallest number of children) after which the kindergarten staff was informed about the ToyBox-study by means of a personal visit. Across the five countries, 297 kindergartens (32.7%) decided to contribute in the study. All four- to six-year-old children born in 2007 or 2008 (age at baseline was 3.5–5.5 years old) received an information letter to inform preschoolers’ parents/caregivers about the purpose of the study. Sample size calculations and the flow of kindergartens through the study can be found elsewhere [29, 30]. Eventually, 8,374 parents/caregivers agreed that their child participated in the study, and 7,554 participants provided complete data at baseline.

Baseline measurements (T0) were conducted on weekdays from May to June 2012. Researchers then visited all kindergartens and fitted preschoolers with a written informed consent from their parents/caregivers with a pedometer and handed out a questionnaire to be filled in at home by parents/caregivers (i.e., Primary Caregivers’ Questionnaire). After recruitment and baseline measurements, kindergartens’ municipalities were randomly and automatically (i.e., using a command in Excel) assigned by the project coordinator (Greece) to the intervention or control group (ratio 2:1) to avoid contamination between kindergartens in the same municipality. Kindergartens in the intervention group received the intervention program. Kindergartens in the control group continued with the standard kindergarten curriculum. One year after baseline measurements, follow-up measurements (T1)

took place (i.e., May to June 2013) and consisted of the same measurements as during baseline. Only data from preschoolers in the control group will be used in the analyses.

## Measurements

**Screen time.** Screen time was reflected by two questions assessing television viewing and computer use separately in the Primary Caregiver’s Questionnaire. This questionnaire showed to be a reliable questionnaire [31]. Each of the questions was assessed separately for weekdays and weekend days. For television viewing, the question was formulated as follows: “About how many hours a day does your child usually watch television (including DVDs and videos) in his/her free time?”. Answer possibilities were “never”, “less than 30 min/day”, “30 min to < 1 h/day”, “1–2 h/day”, “3–4 h/day”, “5–6 h/day”, “7–8 h/day”, “8 h per day”, “more than 8 h/day”, and “I don’t know”. For computer use, the question was formulated as follows: “About how many hours a day does your child use the computer for activities like playing games on a computer, game consoles (e.g., PlayStation, Xbox, GameCube) during leisure time?”. Answer possibilities were identical to the television viewing questions. Answer possibilities were recoded into minutes of television viewing and computer playing per day by using the midpoint method [26], and were then added up to reflect the total screen time, separately for weekdays and weekend days. To estimate the percentage of preschoolers adhering with the screen time recommendation of less than one hour of screen time per day, minutes of total screen time were dichotomized into 0 (>60 min of screen time per day) and 1 ( $\leq$ 60 min of screen time per day).

**Sleep duration.** Sleep duration was assessed by one question in the Primary Caregiver’s Questionnaire. This question was formulated separately for weekdays and weekend days as follows: “How many hours of sleep does your child usually have during the night?”. Answer possibilities were “less than 6 h”, “6–7 h”, “8–9 h”, “10–11 h”, “12–13 h”, “14 h”, “more than 14 h”, and “I don’t know”. Answer possibilities stating “10–11 h” and “12–13 h” were recoded into 1, reflecting all preschoolers adhering with the sleep duration guidelines of 10–13 h of sleep per night. Answer possibilities stating sleep duration shorter or longer than 10–13 h of sleep per night were recoded into 0, reflecting all preschoolers not adhering with the sleep duration recommendations. This was conducted separately for weekdays and weekend days.

**Physical activity.** Physical activity was expressed as steps per day using Omron Walking Style Pro pedometers (HJ-720IT-E2) and reflected total physical activity per day. The Omron Walking Style Pro pedometer is validated to measure step counts in preschoolers

[32]. The pedometers were worn on the right hip, secured by an elastic waist band. Preschoolers wore the pedometers during waking hours for six consecutive days (including two weekend days), except for water-based activities. Parents/caregivers received an information letter with all necessary instructions with regards to their child wearing the pedometer. Data from pedometers were downloaded using Omron Health Management Software version E1.012. Data from the first and last day (i.e., fitting day and collection day respectively) were omitted because these days have incomplete data. Following the guidelines by [33], all step counts below 1,000 and above 30,000 steps per day were deleted and treated as missing data [33]. Step count data were only included in data analyses if preschoolers had valid data for a minimum of two weekdays and one weekend day. Mean steps per weekday and mean steps per weekend day were calculated separately. To calculate the proportion of preschool children adhering with the physical activity guideline of being physically active for more than 180 min per day (weekday and weekend day separately), total steps per day were dichotomized into 0 (< 11,500 steps per day) and 1 ( $\geq$  11,500 steps per day) according to the step count guideline of De Craemer et al. [34].

**Anthropometrics.** Body height and weight were measured at kindergarten by trained research assistants according to standardized protocols [35]. Children were measured in light clothing without shoes. Body height was measured with the SECA 225 Leicester Portable stadiometer (accuracy of 0.1 cm). Body weight was measured with a calibrated electronic scale SECA 861 (accuracy of 0.1 kg). Two readings were obtained of each measurement and the mean was used for analyses. An additional measurement was conducted when the two readings differed by more than 1%, and the mean of the two least deferring values was used. Body Mass Index (BMI) was calculated as  $\text{weight}/\text{height}^2$  ( $\text{kg}/\text{m}^2$ ). The International Obesity Task Force thresholds were used to obtain weight status (underweight, normal weight, overweight, obesity) [36]. In addition, BMI z-scores were calculated using the WHO Anthro and AnthroPlus software. Waist circumference was measured with a tape measure (SECA 200 or SECA 201, accuracy of 0.1 cm). Three readings were obtained of each measurement and the mean was used for analyses. Information regarding the procedures and training of research staff and validity and reliability of the measurements can be found elsewhere [31, 35, 37, 38].

**Other variables.** Sex and date of birth were parent-reported by two questions in the Primary Caregiver's Questionnaire.

### Statistical analysis

All statistical analyses were performed using R Statistical Software (v4.3.1). To be included in the analysis, valid data for screen time, sleep duration and physical activity had to be available. If only data for one or two out of three outcomes was available, data were excluded from analysis. Descriptive characteristics are presented as means and standard deviations for continuous variables and as percentages for categorical variables. Descriptive statistics were used to describe the average time spent on total physical activity (steps/day), screen time and sleep (min/day), adherence with the integrated and separate 24-hour movement behavior guidelines (%), for weekdays and weekend days separately. Linear mixed-effects models were used to investigate associations between compliance with 24-hour movement behavior guidelines and changes in adiposity indicators (BMI z-score, waist circumference and waist-to-height ratio) by using the "lme4" package in R. Random intercepts were included at (1) the individual level to account for data clustering within children (i.e., baseline and follow-up) within countries and (2) at country level to consider between country variability. The restricted maximum likelihood method was used to calculate the effect estimates (EE). Estimated marginal means (EMMs) were calculated using the "emmeans" package in R. Statistical significance was set at a  $p$ -value of < 0.05.

### Results

In total, 719 preschoolers (50.9% boys,  $4.76 \pm 0.45$  years old at baseline) provided valid data for screen time, sleep duration and physical activity. Baseline characteristics are provided in Table 1. At T0, 12.5% of preschoolers complied with the 24-hour movement behavior guidelines on weekdays and 6.7% complied with the integrated guidelines on weekend days. No significant differences were observed in preschoolers' sex, weight status and waist-to-height ratio at baseline. In addition, the estimated variance of the random intercept for children within countries suggests low variability in baseline BMI z-score and waist-to-height ratio and moderate variability in baseline waist circumference between children. The random intercept for countries had a variance ranging between 0.00004 and 0.96, suggesting minimal variability between countries in baseline values of the dependent variables.

### Results for weekdays

For BMI z-score as an outcome, a significant interaction effect was found between time (baseline to follow-up) and compliance with the 24-hour movement behavior guidelines at T0 (EE = -0.15,  $p = 0.001$ ). Preschoolers not complying with the 24-hour movement behavior guidelines

**Table 1** Baseline characteristics of the sample with valid data for screen time, sleep duration and physical activity on both weekdays and weekend days

	Total sample (n = 719)
<b>Sex (% boys)</b>	50.9
<b>Age (years)</b>	4.76 ± 0.45
<b>Weight status (%)</b>	
Underweight	7.8
Normal weight	79.4
Overweight	10.8
Obesity	1.9
<b>BMI z-score</b>	0.17 ± 0.94
<b>Waist circumference (cm)</b>	52.2 ± 3.61
<b>Waist-to-height ratio</b>	0.48 ± 0.03
<b>Adherence integrated guidelines (% meeting all guidelines)</b>	
Weekdays	12.5
Weekend days	6.7
<b>Adherence screen time guidelines (% meeting the guideline)</b>	
Weekdays	53.8
Weekend days	24.9
<b>Adherence sleep duration guidelines (% meeting the guideline)</b>	
Weekdays	72.3
Weekend days	82.6
<b>Adherence physical activity guidelines (% meeting the guideline)</b>	
Weekdays	28.2
Weekend days	28.5

Proportions for categorical variables; means and standard deviation for continuous variables

on weekdays at T0 showed an increase in BMI z-score one year later ( $\Delta\text{EEM} = +0.10$ ,  $p < 0.0001$ ) compared with no change in BMI z-score over time in preschoolers complying with the 24-hour movement behavior guidelines on weekdays at T0 ( $\Delta\text{EEM} = 0.05$ ,  $p = 0.191$ , Table 2). There was no significant difference in BMI z-score at T0 between compliers and non-compliers at T0 ( $\Delta\text{EEM} = -0.08$ ,  $p = 0.462$ ) as well as at T1 ( $\Delta\text{EEM} = 0.07$ ,  $p = 0.507$ ).

Looking at waist circumference and waist-to-height ratio as an outcome, no significant interaction effect was found between time (T0 to T1) and compliance with the 24-hour movement behavior guidelines on weekdays at T0 ( $\text{EE} = -0.10$ ,  $p = 0.734$  and  $\text{EE} = -0.00$ ,  $p = 0.72$  respectively; Table 3). This means that the changes in waist circumference and waist-to-height ratio from T0 to T1 are not different according to children's compliance status with the 24-hour movement behavior guidelines on weekdays at T0. No between group effects were found for waist circumference and waist-to-height ratio, both at T0 (waist circumference:  $\Delta\text{EEM} = -0.16$ ,  $p = 0.716$ ; waist-to-height ratio:  $\Delta\text{EEM} = -0.00003$ ,  $p = 0.994$ ) and T1 (waist circumference:  $\Delta\text{EEM} = -0.06$ ,  $p = 0.889$ ; waist-to-height ratio:  $\Delta\text{EEM} = 0.0009$ ,  $p = 0.794$ ). However, significant changes within groups were found with a significant

increase in waist circumference going from T0 to T1 in the group of non-compliers ( $\Delta\text{EEM} = -1.60$ ,  $p < 0.0001$ ) and the group of compliers ( $\Delta\text{EEM} = -1.50$ ,  $p < 0.0001$ ). Waist-to-height ratio significantly decreased over time in both groups (non-compliers:  $\Delta\text{EEM} = 0.014$ ,  $p < 0.0001$ ; compliers:  $\Delta\text{EEM} = 0.015$ ,  $p < 0.0001$ ).

#### Results for weekend days

A significant interaction effect was found for BMI z-score between time and compliance with the 24-hour movement behavior guidelines on weekend days at T0 ( $\text{EE} = -0.12$ ,  $p = 0.046$ ; Table 3). Preschoolers not complying with the 24-hour movement behavior guidelines on weekend days at T0 showed an increase in BMI z-score one year later ( $\Delta\text{EEM} = -0.08$ ,  $p < 0.0001$ ; Table 2) compared with no significant change in BMI z-score in preschoolers complying with the integrated guidelines on weekend days at T0 ( $\Delta\text{EEM} = 0.04$ ,  $p = 0.486$ ). Again, there were no between-group differences in BMI z-score at T0 ( $\Delta\text{EEM} = -0.09$ ,  $p = 0.512$ ) and T1 ( $\Delta\text{EEM} = 0.03$ ,  $p = 0.817$ ).

No significant interaction effect was found for waist circumference ( $\text{EE} = 0.11$ ,  $p = 0.773$ ; Table 3) and waist-to-height ratio ( $\text{EE} = 0.002$ ,  $p = 0.637$ ), and no significant between-group differences were found at T0 (waist



**Table 2** Summary of linear mixed-effects models including the interactions between time (baseline to follow-up) and compliance with the integrated 24-hour movement behavior guidelines at baseline during weekdays and weekend days

BMI z-score					Waist circumference (cm)					Waist-to-height ratio				
	EE	95% CI	p-value	EE	95% CI	p-value	EE	95% CI	p-value	EE	95% CI	p-value		
<b>WEEKDAYS</b>														
Intercept	0.20	0.06–0.35	<b>0.006</b>	52.07	51.21–52.93		0.48	0.47–0.48	<b>&lt;0.001</b>	0.48	0.47–0.48	<b>&lt;0.001</b>		
Compliance at T0 (full)	0.08	-0.13–0.29	0.462	0.16	-0.69–1.01		0.00	-0.01–0.01	0.994	0.00	-0.01–0.01	0.994		
Time (T1)	0.09	0.06–0.12	<b>&lt;0.001</b>	1.60	1.40–1.80		-0.01	-0.02 – -0.01	<b>&lt;0.001</b>	-0.01	-0.02 – -0.01	<b>&lt;0.001</b>		
Compliance at T0 (full) * time (T1)	-0.15	-0.24 – -0.06	<b>0.001</b>	-0.10	-0.66–0.46		-0.00	-0.01–0.00	0.716	-0.00	-0.01–0.00	0.716		
<b>WEEKEND DAYS</b>														
Intercept	0.21	0.06–0.35	<b>0.006</b>	52.08	51.22–52.94		0.48	0.47–0.48	<b>&lt;0.001</b>	0.48	0.47–0.48	<b>&lt;0.001</b>		
Compliance at T0 (full)	0.09	-0.18–0.36	0.513	0.10	-1.01–1.20		-0.00	-0.01–0.01	0.866	-0.00	-0.01–0.01	0.508		
Time (T1)	0.08	0.05–0.11	<b>&lt;0.001</b>	1.59	1.40–1.78		-0.01	-0.02 – -0.01	<b>&lt;0.001</b>	-0.01	-0.02 – -0.01	<b>&lt;0.001</b>		
Compliance at T0 (full) * time (T1)	-0.12	-0.24 – -0.00	<b>0.046</b>	0.11	-0.63–0.84		0.002	-0.01–0.01	0.773	0.002	-0.01–0.01	0.637		

EMM estimated marginal means, CI confidence interval, T0 Baseline measurement, T1 1-year follow-up measurement

EMM using mixed model analysis corrected for the clustered study design

Asterisks (\*) indicate statistically significant within differences over time

Numbers in bold refer to statistically significant interaction effects between time (baseline to follow-up) and compliance with the integrated guidelines at baseline as reported in Table 2

**Table 3** Estimated marginal means and confidence intervals at baseline and follow-up for non-compliers and full compliers with the integrated 24-hour movement behavior guidelines at baseline for the three different adiposity indicators

	BMI z-score EMM (95% CI)		Waist circumference (cm) EMM (95% CI)		Waist-to-height ratio EMM (95% CI)	
	T0	T1	T0	T1	T0	T1
WEEKDAYS						
Non compliance	<b>0.20 (0.05–0.36)*</b>	<b>0.30 (0.14–0.45)*</b>	52.10 (51.20–53.00)*	53.70 (52.80–54.60)*	0.48 (0.47–0.49)*	0.47 (0.46–0.47)*
Full compliance	<b>0.28 (0.08–0.49)</b>	<b>0.23 (0.02–0.43)</b>	52.20 (51.20–53.20)*	53.70 (52.70–54.00)*	0.48 (0.47–0.49)*	0.46 (0.46–0.47)*
WEEKEND DAYS						
Non compliance	<b>0.21 (0.05–0.36)*</b>	<b>0.29 (0.13–0.44)*</b>	52.10 (51.20–53.00)*	53.70 (52.80–54.60)*	0.48 (0.47–0.49)*	0.47 (0.46–0.47)*
Full compliance	<b>0.30 (0.05–0.55)</b>	<b>0.26 (0.00–0.51)</b>	52.20 (51.20–53.20)*	53.70 (52.70–54.70)*	0.48 (0.47–0.49)*	0.46 (0.46–0.47)*

T0 Baseline measurement, T1 1-year follow-up measurement, EE Effect estimates, CI confidence interval, Effect estimates for 'compliance × time' describe the difference between the mean change of the adiposity indicator in full compliers and non-compliers at T0

circumference:  $\Delta EEM = -0.10$ ,  $p = 0.866$ ; waist-to-height ratio:  $\Delta EEM = 0.003$ ,  $p = 0.508$ ) and T1 (waist circumference:  $\Delta EEM = -0.20$ ,  $p = 0.719$ ; waist-to-height ratio:  $\Delta EEM = 0.001$ ,  $p = 0.753$ ). A significant increase was found in waist circumference going from T0 to T1 both in the group of non-compliers ( $\Delta EEM = -1.59$ ,  $p < 0.0001$ ) and the group of compliers ( $\Delta EEM = -1.70$ ,  $p < 0.0001$ ). A significant decrease was found in waist-to-height ratio going from T0 to T1 both in the group of non-compliers ( $\Delta EEM = 0.014$ ,  $p < 0.0001$ ) and the group of compliers ( $\Delta EEM = 0.01$ ,  $p = 0.0002$ ).

Estimated marginal means are presented in Table 2.

## Discussion

The present study aimed to investigate whether compliance with the 24-hour movement behavior guidelines is associated with changes in adiposity indicators (i.e., BMI z-score, waist circumference, and waist-to-height ratio) in preschool children from a European sample. Differences in BMI z-score trajectories were found between preschoolers complying with the integrated 24-hour movement behavior guidelines and preschoolers not complying with the integrated guidelines with less beneficial changes (i.e. a higher increase) to be found in the non-compliant group of children. Interestingly, these results were not confirmed for waist circumference and waist-to-height ratio.

Although no difference in BMI z-score was found between preschoolers complying with the integrated guidelines and those not complying with the integrated guidelines both at baseline and follow-up, the interaction effect showed that the change over time in BMI z-score was different in both groups, for both weekdays and weekend days. Preschoolers not complying with the integrated guidelines on weekdays showed an increase in BMI z-score of 0.10 over time, while there was a small non-significant difference of -0.05 in BMI z-score for those complying with the integrated guidelines. Similar

differences were found for weekend days (i.e., increase of 0.08 in non-complying preschoolers, decrease of 0.04 in complying preschoolers). The US Preventive Services Task Force indicated that a reduction of BMI z-score of 0.20 to 0.25 is the threshold for being a clinically important change in this outcome when it comes to intervention studies focusing on weight loss in this age group [39]. However, this means that an increase of 0.20 to 0.25 in BMI z-score is also a clinically relevant difference when it comes to weight gain. Although there was a significant difference between both groups of preschoolers, results from our study do not show a clinically important increase in BMI z-score ( $< 0.20$ ). In addition, a lower BMI z-score in non-compliant preschoolers compared to compliant preschoolers was found at baseline. Although not statistically different at baseline, regression towards the mean might be a possible explanation for the increase that was found in preschoolers not complying with the integrated guidelines. It is important to take into account that we only have longitudinal data for the duration of one year in a young age group (3- to 5-year-olds). It might be possible that larger differences occur when children are followed up for a longer period of time. Therefore, future (intervention) studies should incorporate longer follow-up periods to see how adiposity indicators change over time and to investigate the possible other factors being the cause(s) of this change.

Height and weight are two simple measures to conduct to have an insight into adiposity in all age groups. However, it does not give insights into abdominal adiposity even though this is associated with cardiometabolic risk factors in children and adolescents [40]. Therefore, our study also looked at waist circumference and waist-to-height ratio with the latter being a promising and useful measure of size-adjusted abdominal adiposity [41], although no specific pediatric reference values are available for these outcome measures. Waist circumference and waist-to-height ratio account for

the weakness in using BMI which is not being able to distinguish between higher adiposity or higher muscle mass. The results of our study found no difference in change in waist circumference and waist-to-height ratio from baseline to follow-up between compliers and non-compliers. Both groups had an increase in waist circumference and a small decrease in waist-to-height ratio, which is related to normal growth. In addition, it might be possible that we do not see a change between compliers and non-compliers because the number of children in the group of compliers is very small compared to the group of non-compliers.

Results were similar for compliance on weekdays and weekend days for all adiposity indicators, showing that complying with the integrated guidelines on weekdays and weekend days have a similar effect on adiposity indicators. Literature in preschool children often show that preschoolers are less active and more sedentary on weekend days compared to weekdays [42–44]. Therefore, efforts should be undertaken to motivate preschoolers and preschoolers' parents to comply with the integrated guidelines on weekend days as well.

Since our study is the first to look at longitudinal changes in BMI z-score, waist circumference and waist-to-height ratio according to compliance with 24-hour movement behavior guidelines in preschoolers, it is not possible to compare our results with other studies. However, a recent study of [25] looked at the longitudinal associations of movement behaviors and body composition in children from four to nine years of age [25]. Body composition was measured with air-displacement plethysmography resulting in fat mass index and fat-free mass index as outcomes. [25] found that none of the movement behaviors at four years of age were associated with fat mass index or fat-free mass index at nine years of age. They also found that a lower fat mass index at four years of age was associated with higher vigorous PA and light PA at nine years of age [25] used a longer follow-up period to look at longitudinal changes in body composition which might be the reason that our results differ from the Swedish study. In addition, results from [25] are based on a small(er) sample in one country, which might be another reason for differences with our results. Lastly, different measures of adiposity were used with [25] using a criterium method to measure body fatness (i.e., air displacement plethysmography) while height, weight and waist circumference were used in the current study. Although criterium methods are preferred to draw firm conclusions, they are not easy to incorporate in large scale studies, such as the ToyBox-study. It is important to consider that the study of [25] used data from a randomized controlled trial in which follow-up data from both intervention and control groups were used in the

data analysis which might have caused some bias in the results.

Strengths of this study include the longitudinal design with a one year follow-up during the same season as the baseline measurement and thus avoiding seasonal bias. In addition, our study only includes data from preschoolers in the control group which means that we included preschoolers having a normal development without the influence of external programs or interventions focusing on lifestyle. Furthermore, data from different European countries were included, enabling us to draw conclusions on a larger basis since countries from different regions in Europe were included (Central Europe, South Europe, Eastern Europe).

One of the most important methodological limitations of the current study is the fact that we were not able to capture the adiposity rebound. This is a period during preschool age where a second rise in BMI occurs, typically between the age of 5 to 7. An early adiposity rebound is associated with an increased risk of overweight [45]. Several anthropometric measurements during growth are necessary to capture adiposity rebound. Future studies should definitely try to incorporate multiple anthropometric measurements (e.g., every three months) to take the adiposity rebound period into account within longitudinal studies concerning weight status in preschool children. Weight gain and a change in weight status or BMI is attributed to the energy balance when energy intake exceeds energy expenditure. Dietary intake plays a considerable role in the energy balance but was not taken into account in the current analyses. Physical activity was measured using pedometers which means that we can only make statements about the total volume/amount of physical activity per day. Although pedometers are easy to use and cost-effective with regards to large-scaled studies, they do not distinguish between different physical activity intensities. However, the WHO guidelines state that preschoolers should engage in at least 180 min of physical activity of which 60 min are spent in energetic play, which corresponds to physical activity at moderate-to-vigorous intensity. Therefore, it was not possible to fully capture adherence with the WHO guidelines since it was not possible to make the distinction in physical activity intensities. Two out of three behaviors were proxy-reported by the parents using a proxy-reported questionnaire. Since preschool children are cognitively not able to fill in a questionnaire, parents had to report on their child's behavior. It might be possible that social desirability occurred when filling in the questionnaire. Finally, the distinction between short and long sleep duration could not be taken into account, although the clinical impact on adiposity indicators of having a short or a long sleep duration might be different.



## Conclusions

The change in BMI z-scores across one year is different for preschoolers who do not comply with the 24-hour movement behavior guidelines compared to preschoolers who do comply with the integrated guidelines. Preschoolers who do not comply with the integrated guidelines have a small increase in BMI z-scores (although not clinically relevant) compared to almost no change in preschoolers who comply with the integrated guidelines. No difference between compliers and non-compliers was found for waist circumference and waist-to-height ratio. Future studies should incorporate longer follow-up periods to see the effect of compliance to the integrated 24-hour movement behavior guidelines on adiposity indicators in preschool children.

## Abbreviations

BMI	Body Mass Index
EEMs	Estimated Marginal Means
SES	socio-economic status
WHO	World Health Organization

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## Authors' contributions

M.D.C. was responsible for conceptualization, methodology, writing the original draft and editing the draft. V.V. was responsible for formal analysis and reviewing and editing the draft. O.A. and Y.M. were responsible for project administration. M.D.C., G.C., M.D., L.M., V.I., B.K., P.S., O.A., Y.M. and V.V. were responsible for reviewing and editing the draft. M.D.C., G.C., M.D., L.M., V.I., B.K., P.S., O.A., Y.M. and V.V. have read and agreed to the published version of the manuscript.

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## Data availability

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

## Declarations

### Ethics approval and consent to participate

Parents/caregivers gave written informed consent to participate in the study. This study was included in the approval of the ToyBox-study by Ethical Committees in all six European countries, in line with national regulations (i.e., the Ethical Committee of Ghent University Hospital (Belgium), Committee for the Ethics of the Scientific Studies (KENI) at the Medical University of Varna (Bulgaria), Ethikkommission der Ludwig-Maximilians-Universität München (Germany), the Ethics Committee of Harokopio University of Athens (Greece), Ethical Committee of Children's Memorial Health Institute (Poland), and CEICA (Comité Ético de Investigación Clínica de Aragón (Spain)).

### Consent for publication

Not applicable.

### Competing interests

The authors declare no competing interests.

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