Pediatric RESEARCH

POPULATION STUDY ARTICLE

Association of physical fitness with skin autofluorescence-derived advanced glycation end products in children

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BACKGROUND: Advanced glycation end products (AGEs) accumulate with age and development of cardiovascular disease. Higher AGEs have been shown in children with diabetes but little is known about their association with lifestyle conditions in childhood. We hypothesized that BMI, blood pressure and cardiorespiratory fitness (CRF) are associated with subcutaneous AGEs formation in children.

METHODS: In this cross-sectional study, 1075 children (aged 7.2 ± 0.4 years) were screened for subcutaneous AGEs (skin autofluorescence; SAF), body mass index (BMI), blood pressure (BP), and CRF using standardized procedures. Group comparisons were performed in clinical BP and BMI categories and tertiles of CRF.

RESULTS: Children with higher physical fitness showed lower SAF (0.99(1.03;1.10)au) compared to children with low CRF (1.09 (1.03;1.05)au, $p < 0.001$). An increase of one shuttle run stage was associated with a mean reduction in SAF of −0.033(CI: −0.042; −0.024)au, independent of BMI and BP ($p < 0.001$). BMI and BP were not independently associated with SAF-derived AGEs in this large cohort of primary school children.

CONCLUSIONS: Low physical fitness but not BMI and BP were associated with higher levels of AGEs. Primary prevention programs in young children may need to focus on improving physical fitness in game settings in order to reduce the growing prevalence of metabolic disorders during childhood.

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INTRODUCTION

Advanced glycation end products (AGEs) form when proteins or lipids interact with reduced sugars for an extended period of time, subsequently undergoing progressive irreversible molecular transformation. Growing evidence suggested that AGEs interact with cell surface receptors for AGEs (RAGE) under hyperglycemic conditions, leading to increased oxidative stress and inflammation.[1,2] Interactions of AGEs with RAGE impart distinct and maladaptive remodeling of cross-linked collagen in the vascular wall.[3,4]

Concentrations of serum AGEs correlate with AGEs accumulation in the skin.[5] Subcutaneous AGEs seem to be related to long-term diabetic risk factors and glycemic control.[5] Data on the relationship between obesity and AGEs formation are scarce and inconsistent. den Engelsen et al.[6] found no association of central obesity and subcutaneous AGEs. In contrast, a more recent study demonstrated an association of skin AGEs with incidence metabolic syndrome, higher waist circumference and elevated blood pressure (BP) in adults.[7] However, no association of subcutaneous AGEs with obesity was found in participants without the metabolic syndrome.[7] In a recent meta-analysis of patients with high CV risk, skin AGEs has been shown to be predictive of CV and all-cause mortality.[8] Sedentary lifestyle and unbalanced diet have been associated with an accumulation of AGEs.[9] The combination of exercise and diet seems to be an effective means to reduce AGEs accumulation.[10] A recent study has shown that life-long exercise can counteract the age-related accumulation of AGEs.[11] In older adults it was recently shown that higher physical activity was associated with lower AGEs levels.[12] Hypertension and arterial stiffness have both been associated with increased plasma concentrations of AGEs in adults.[13]

There are very few studies on AGEs in children. A previous review on the role of dietary AGEs in childhood suggested that AGEs are involved in the pathogenesis of adiposity and β-cell failure.[14] Jaisson et al. found that serum AGEs were elevated on first diagnosis of diabetes mellitus and may play a role in developing long-term complications.[15] Children with 5 years exposure to diabetes have been reported to have higher accumulation of skin autofluroescence (SAF)-derived AGEs, comparable to levels of healthy adults and equivalent to about 25 years of chronologic aging.[16] Our study, for the first time, aimed to examine the association of BMI, body fat (BF), BP and cardiorespiratory fitness (CRF) with subcutaneous accumulation of AGEs in an unselected population of young children. We hypothesized that cardiovascular risk factors such as obesity, high blood pressure and low physical fitness were associated with higher concentration of SAF-derived AGEs early in life and that high fitness levels on the other hand would be associated with lower SAF-derived AGEs.

METHODS

Study design and participants

This cross-sectional study was embedded in the large-scale, cross-sectional EXAMIN YOUTH study in Switzerland. The study protocol was approved by the Ethics Committee of the University of Basel (Switzerland, EKBB: 258/12). The study was performed in

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accordance with the Helsinki Declaration of Guideline For Good Clinical Practice[17] and the Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) guidelines.[18] In a predominantly Caucasian population, children aged 6−8 years were screened for BMI, BF, BP, CRF and subcutaneous AGEs accumulation. An informed parental written informed consent was obtained from all participants. The study was registered a priori in a clinical trials registry (ClinicalTrials.gov: NCT02853747).

Measurements

Advanced glycation end products. AGEs were assessed by subcutaneous SAF. Measurements of SAF were performed using the validated AGE Reader® device (DiagnOptics Technologies BV, Groningen, Netherlands).[5,19] The AGE Reader® involves an integrated spectrometer to analyze reflected excitation light. The ratio between the emission light and reflected light multiplied by 100 was used to calculate SAF, expressed in arbitrary units (au). The emission light ranges between 420 and 600 nm, whereas the reflected excitation light ranges between 300 and 420 nm. SAF allows the noninvasive, validated method to analyze AGEs in connective tissue and it is strongly correlated to AGEs accumulation in the blood serum.[5] For further analysis, the arithmetic mean of three measurements at different areas at the right ventral side of the forearm was used.

Anthropometrics. Body height was measured with a wall-mounted stadiometer (Seca 206, Basel, Switzerland), weight and percentage BF were measured with the InBody device (InBody 170 Biospace device; InBody Co., Seoul, Korea). According to cutoff points for BMI (kg/m^2) incorporating age and sex, children were classified in BMI categories by use of percentiles.[20] Children with a BMI over the 85th percentile in their sex and age group were categorized as being overweight and over the 95th percentile as children with obesity. BP parameters were assessed using an automated oszillograph (Oscillomate, CAS Medical Systems, Branford, CT). All children were measured in a sitting position after a 5-min rest based on the recommendations of the American Heart Association.[21] BP was measured five times in series and the mean of the three measurements with the smallest variation were taken for the further analysis. According to the population-based German KiGGS study, children over the 90th percentile were categorized as having high-normal BP and over the 95th percentile as children with hypertension.[22]

Physical fitness. The CRF assessment took place during the physical education lessons with the same equipment used for every school. After a 5-min warm-up, a 20-m shuttle run was performed. This is a well-established and validated test to measure physical fitness.[23,24] A previous meta-analysis has demonstrated the feasibility and validity of the 20-m shuttle run as a surrogate marker for CRF in children.[25] It is concluded that, although spiroergometry remains to be the gold standard, the shuttle run is an established alternative if a laboratory-based test is not feasible. In this progressive endurance test, the children had to run back and forth between two lines of 20 m with an initial running speed of 8.0 km/h and an increase of 0.5 km/h every minute, paced by beeps from an audio device programmed for the timing of the shuttle run test. The individual maximum was reached if the child did not cross the line for two consecutive 20-m trials within the given time, defined by the audio beeps. A 2-m range for crossing the line was allowed. The score was assessed by the numbers of stages (1 stage = 1 min) reached with a precision of 0.5 stages.

Statistical analysis

Mean SAF was analyzed across the clinical categories of BMI, BP and tertiles of CRF using univariate analysis of covariance (ANCOVA). Bivariate analysis was performed to compare clinically relevant BMI categories, physical fitness and SAF. Pearson's correlation was used to assess the association of CRF with AGEs. Multiple linear regression models were used to compare changes in SAF with changes in BMI, BF, BP and CRF. Four different models were fitted to adjust for age and sex as well as BMI, BP and CRF. The predictor variables were preselected and, therefore, we did not perform a stepwise regression. We have selected these predefined predictors and confounders as these are classic parameters known to have an impact on the primary outcome. We found no evidence for interactions effects between the variables.

Variance homogeneity of residuals was assessed using the Tukey−Anscombe Plots. To assess normality, we used normal QQ plots of the residuals. Ninety-five percent confidence intervals were presented for measures of effect to indicate the amount of uncertainty and a two-sided level of significance of 0.05 denotes statistical significance. For analyses and graphics, an up-to-date version of Stata 15 (StataCorp LP, College Station, TX) was used. The sample size of the cross-sectional study was given by the number of children and parents giving their consent.

RESULTS

Participants

From the 3068 children that were invited in the study, 1690 (55%) had a written consent from their parents to participate. A total of 221 children dropped out because of illness or were otherwise absent. Due to skin pigmentation or a temporary technical default of the device (AGE Reader®), 394 children had to be excluded from the data analysis, leaving 1075 children with complete measurements (Fig. 1). Age, body weight and height, BF, BMI and shuttle run data of the 615 excluded children are presented in Supplement Table S1. Excluded children had slightly higher fitness levels compared to children included in the study. Population characteristics are shown in Table 1. Based on a modified questionnaire survey,[26] 95% of children were Caucasian. In our cohort, 87% of children presented with normal weight ($n = 934$), 10% with overweight ($n = 103$) and 3% ($n = 38$) with obesity. Based on systolic BP, 77% were categorized as children with normal BP ($n = 827$), 9% as having high-normal BP ($n = 99$) and 14% as children with hypertension ($n = 149$). Boys were fitter (CRF: 4.0 ± 1.6 stages) but showed higher subcutaneous AGEs (SAF: 1.07 ± 0.2 au)

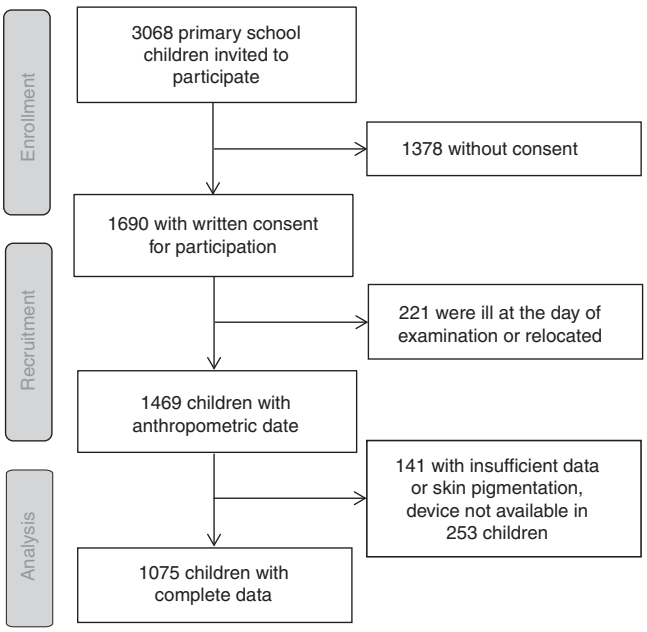

Fig. 1 Flow diagram.

Table 1. Population characteristics of the study.

Parameter	Total Mean ± SD (range)	*n*	Boys Mean ± SD (range)	*n*	Girls Mean ± SD (range)	*n*	*p*
Age (years)	7.2 ± 0.4 (6.2;8.3)	1075	7.2 ± 0.4 (6.4;8.3)	509	7.2 ± 0.4 (6.2;8.2)	566	0.073
Height (cm)	124.4 ± 5.5 (109.0;144.0)	1075	124.8 ± 5.4 (109.0;144.0)	509	124.0 ± 5.5 (109.2;141.0)	566	0.025
Weight (kg)	24.7 ± 4.7 (16.2;48.7)	1075	25.0 ± 4.7 (16.5;45.8)	509	24.4 ± 4.6 (16.2;48.7)	566	0.068
BMI (kg/m^2)	15.9 ± 2.2 (10.0;26.3)	1075	15.9 ± 2.2 (10.0;26.1)	509	15.8 ± 2.2 (12.1;26.3)	566	0.295
Percentage body fat (%)	15.6 ± 7.7 (3.0;44.8)	1075	14.1 ± 7.3 (3.0;44.3)	509	16.9 ± 7.8 (3.0;44.8)	566	<0.001
Heart rate (bpm)	85.8 ± 10.3 (47.7;120.7)	1075	85.5 ± 10.2 (57.0;120.7)	509	86.2 ± 10.5 (47.7;116.7)	566	0.278
Systolic BP (mmHg)	103.7 ± 7.7 (82.0;134.0)	1075	103.6 ± 7.6 (85.7;134.0)	509	103.9 ± 7.8 (82.0;131.3)	566	0.547
Diastolic BP (mmHg)	64.2 ± 6.8 (42.0;85.0)	1075	64.1 ± 6.8¨ (46.0;85.0)	509	64.3 ± 6.8 (42.0;84.0)	566	0.569
SAF (au)	1.05 ± 0.20 (0.27;2.79)	1075	1.07 ± 0.20 (0.27;2.04)	509	1.03 ± 0.20 (0.50;2.79)	566	<0.001
20-m shuttle run (stages)	3.7 ± 1.5 (0.5;8.5)	1075	4.0 ± 1.6 (0.5;8.5)	509	3.4 ± 1.3 (0.5;7.5)	566	<0.001

BMI body mass index, *BP* blood pressure, *SAF* skin autofluorescence, *au* arbitrary units, *SD* standard deviation, *n* number

Table 2. Advanced glycation end products in relation to clinical categories of body mass index, blood pressure and tertiles of shuttle run.

Parameter	*n*	SAF (au) Mean (95% CI)	*p*
BMI[a]			0.685
Normal weight	934	1.05 (1.04;1.06)	
Overweight	103	1.03 (0.99;1.07)	
Obese	38	1.04 (0.98;1.11)	
BF[a]			0.975
First (lowest)	347	1.04 (1.02;1.07)	
Second	354	1.05 (1.02;1.07)	
Third	339	1.05 (1.03;1.07)	
Systolic BP[b]			0.799
Normotensive	827	1.05 (1.03;1.06)	
High-normal BP	99	1.04 (1.01;1.08)	
Hypertensive	149	1.04 (1.00;1.07)	
Diastolic BP[b]			0.181
Normotensive	840	1.04 (1.03;1.05)	
High-normal BP	84	1.07 (1.03;1.11)	
Hypertensive	151	1.06 (1.03;1.10)	
Shuttle run[c]			<0.001
First	476	1.09 (1.03;1.05)	
Second	275	1.03 (1.03;1.11)	
Third (fittest)	324	0.99 (1.03;1.10)	

Data adjusted for age and sex
P value across lowest and highest category (univariate analysis of covariance)
BMI body mass index, *BF* body fat, *BP* blood pressure, *SAF* skin autofluorescence, *au* arbitrary units, *CI* confidence interval
[a]Additionally adjusted for shuttle run, systolic and diastolic blood pressure
[b]Additionally adjusted for shuttle run and BMI
[c]Additionally adjusted for BMI, systolic and diastolic blood pressure

compared to girls (CRF: 3.4 ± 1.3 stages; SAF: 1.03 ± 0.2 au, $p < 0.001$).

Group differences

The results for between group differences are shown in Table 2. Clinical BMI, systolic and diastolic BP categories were not associated with SAF in our cohort of children. Children with higher physical fitness showed lower SAF (0.99 (1.03;1.05) au) compared to children with low CRF (1.09 (1.03;1.05) au, $p < 0.001$). The bivariate analysis illustrates the interrelation between fitness, BMI and SAF levels. Children with low fitness and overweight or obesity had the highest subcutaneous AGEs levels (Fig. 2).

Regression analysis

In the regression analysis, BMI was associated with SAF (0.006 (0.2E-3;0.011) au, $p = 0.042$, $R^2 = 0.004$) with little evidence for an association after adjustment for age and sex (Table 3). Percentage BF was associated with increased SAF (0.003 (0.001;0.004) au, $p = 0.001$, $R^2 = 0.026$). After adjustment for BP and CRF, no statistically significant association was found. Likewise, there was little evidence for an association of systolic and diastolic BP with SAF. One stage increase in shuttle run was significantly associated with decreased SAF, independent of BMI and BP (Fig. 3a, b). CRF alone explained 5% of the SAF variance. As expected, children with higher CRF had lower BMI ($p < 0.001$) and lower systolic ($p < 0.001$) and diastolic BP ($p = 0.035$).

DISCUSSION

This is the first study to examine the association of BMI, BF, BP, and CRF with SAF-derived AGEs accumulation in children. Our findings demonstrate that low physical fitness is associated with increased accumulation of subcutaneous AGEs in young children. In contrast to our hypothesis, we found no independent association of BMI, BF and BP with SAF-derived AGEs.

Physical fitness and AGEs

Higher CRF was associated with reduced subcutaneous AGEs formation, indicating a favorable glucose metabolism and a reduction of associated CV risk in these children. The confidence intervals show evidence for and association of CRF with SAF-derived accumulation of AGEs in young children. Data in a Slovak population suggested that regular self-reported physical activity is associated with lower SAF during lifespan.[27] Two studies have previously measured subcutaneous AGEs in a small number of healthy young children.[27,28] Our study is the first to assess SAF-derived AGEs in a large population-based unselected cohort of 6−8-year-old children, offering reliable normal values for young Caucasian children (mean SAF 1.05 ± 0.20 au) and demonstrating the inverse association with objectively measured CRF.

CRF affects glycation processes in children, as at least 5% of the variance of SAF was explained by CRF in our cohort. One unit increase in CRF was associated with a 0.03 au decrease in SAF and the difference in SAF between the lowest and the fittest tertile was 0.10 au. In comparison, in adolescents with type 1 diabetes one

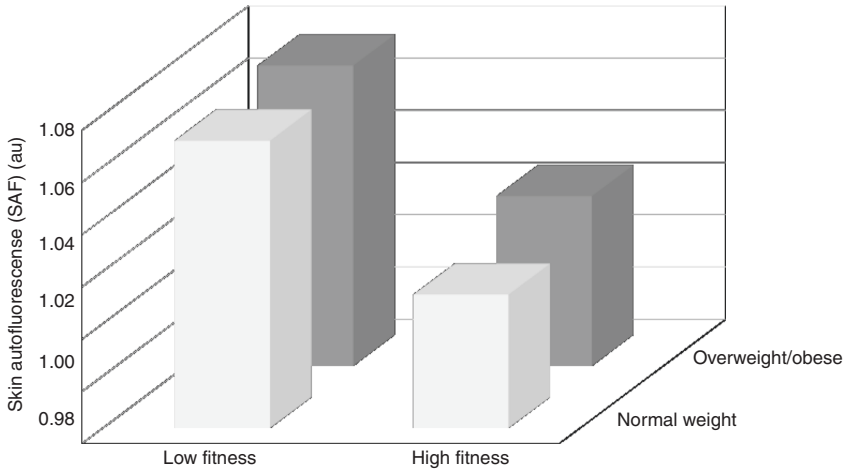

Fig. 2 Skin autofluorescence in relation to body mass index categories and median of physical fitness.

Table 3. Regression analysis for the association of body composition, peripheral blood pressure, physical fitness and activity with advanced glycation end products.

Parameter	Model	SAF (au change per unit)		
		B (95% CI)	*p*	R^2
BMI (kg/m^2)	1	0.005 (−0.001;0.010)	0.085	0.019
	2	−0.002 (−0.008;0.004)	0.439	0.069
Percentage body fat (%)	1	0.003 (0.001;0.004)	0.001	0.026
	2	0.2E-2 (−0.002;0.002)	0.861	0.066
Systolic BP (mmHg)	1	0.001 (−0.001;0.002)	0.575	0.017
	3	−0.6E-4 (−0.001;0.002)	0.936	0.069
Diastolic BP (mmHg)	1	0.001 (−0.001;0.002)	0.542	0.017
	3	0.2E-3 (−0.002;0.002)	0.815	0.069
20-m shuttle run (stages)	1	−0.032 (−0.040;−0.024)	<0.001	0.068
	4	−0.033 (−0.042;−0.024)	<0.001	0.069

Model 1 = adjusted for age and sex
Model 2 = model 1 plus adjusted for systolic, diastolic blood pressure and shuttle run (stages)
Model 3 = model 1 plus adjusted for BMI and shuttle run (stages)
Model 4 = model 1 plus adjusted for BMI, systolic and diastolic blood pressure
BMI body mass index, *BP* blood pressure, *SAF* skin autofluorescence, *au* arbitrary units, *CI* confidence interval

unit increase in hemoglobin A1c (HbA1c) has been associated with a 0.06 au increase in SAF.[29] In their cohort, the difference in SAF between type 1 diabetes and healthy controls was 0.26 au. In light of this evidence, it may be concluded that our findings are potentially of clinical relevance. Future studies will have to determine the long-term clinical and predictive value of subcutaneous AGEs for the development of cardiometabolic disease and the potential of physical fitness to counteract accumulation of AGEs during childhood and later in life.

Body mass index, body fat and AGEs

No independent association between BMI and SAF was found in our cohort of children. The association of BF with SAF was not independent of BP and CRF.

A previous study in adults also showed no association of subcutaneous AGEs with obesity in the absence of the metabolic syndrome.[7] A prior study in children suggested that BMI, BF and fat mass were associated with soluble RAGE in older children aged 12−14 years.[30] It is therefore possible that obesity only affects SAF-derived AGEs formation after a longer-term exposure time to an increased BMI. In addition, sex seems to be a nonmodifiable factor for the SAF-derived accumulation of AGEs in children. In our population of young children, boys showed higher subcutaneous AGE accumulation compared to girls independent of BMI and BP. In contrast to the gender differences in our children, it has been shown that plasma accumulation of AGEs is higher in women compared to men aged around 20 years.[31] Our children were examined in pre-puberty, whereas the aforementioned study investigated young adults. Sex-related differences in childhood development and puberty seems to be the most likely explanation for this conundrum.

Blood pressure and AGEs

In the regression analysis, no association of systolic and diastolic BP with SAF was found. However, there was a weak but significant association of diastolic BP and SAF in children categorized as children with high-normal BP and hypertension. In adults, hypertension has been associated with increased accumulation of AGEs in plasma.[13] A recent study found an association of systolic and diastolic BP with subcutaneous AGEs in a general adult population.[32] In patients with the metabolic syndrome, high-normal BP was also associated with subcutaneous AGEs.[7]

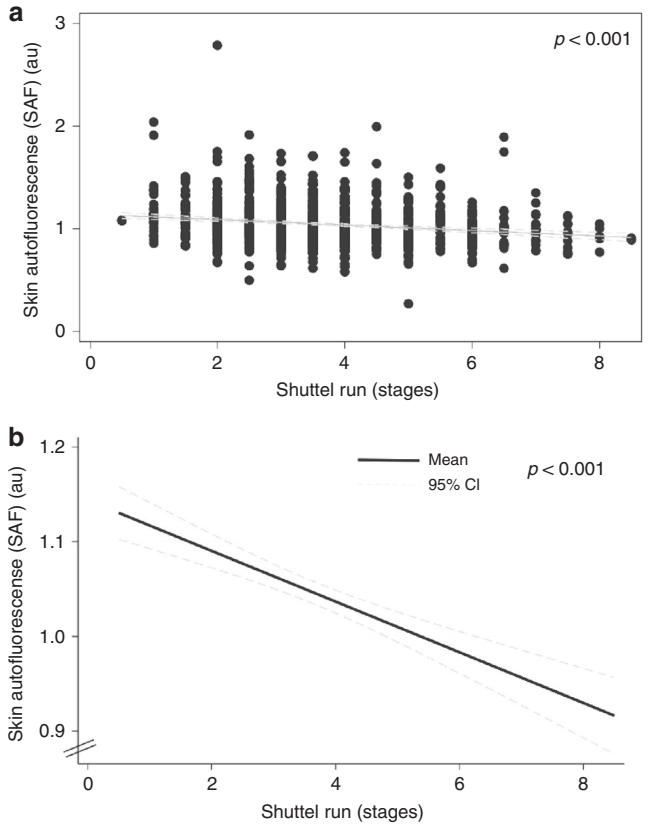

Fig. 3 Skin autofluorescence and cardiorespiratory fitness. a, **b** The association of cardiorespiratory fitness with skin autofluorescence. CI confidence interval.

Childhood BP has been shown to predict development of CV disease in adulthood.[33] In children, higher BP does not seem to directly and independently affect SAF-derived AGEs accumulation and metabolic health. As argued before, exposure time to high BP may not be long enough to affect SAF-derived AGEs accumulation in young children. Based on our findings the clinical application of subcutaneous AGE's in young children to differentiate cardiometabolic risk would appear premature. It remains to be determined if and to what extent BP, and indeed BMI, affect SAF-derived AGEs accumulation in older children and adolescents.

Potential mechanisms

Endothelial dysfunction and obesity-related inflammation are mediated through oxidative stress conditions.[34] It is well known that oxidative stress is a main determinant for increased formation of AGEs.[35,36] Sedentary behavior is characterized by reduced mitochondrial capacity and increased oxidative stress and exercise has the potential to reverse oxidative conditions.[37] Proteins are glycated to form AGEs through the so-called Maillard reaction. Early non-enzymatic glycation and formation of Schiff bases and Amadori products represent reversible cross-links between proteins and sugars.[38] We hypothesize that exercise can reverse the formation of early glycation products preventing irreversible cross-links and tissue accumulation of AGEs forming fluorescent derivatives. In addition, exercise-induced formation of soluble RAGE may play an important role in reducing AGEs accumulation and associated oxidative stress. The circulating soluble RAGE binds to AGE and acts as a competitive inhibitor of ligands that activate RAGE. Long-term physical activity and exercise lead to an increase in soluble RAGE, which blocks RAGE activation.[39]

Improvement of AGEs metabolism may be achieved by physical fitness interventions rather than measures focusing on classical risk factors such as BMI and BP reduction. From a pathophysiological point of view, it is possible that other sensitive metabolites of the AGEs metabolism, such as RAGE or protein-bound AGEs and markers of dicarbonyl stress, may be associated with BMI and BP in young children. SAF-derived AGEs accumulation may occur at later stages compared to increases in circulating serum and urine biomarkers of AGEs metabolism. Future studies will have to analyze blood or urine samples of young children to clarify the clinical relevance and differences of circulating AGEs metabolites as compared to subcutaneous AGEs accumulation.

Strengths and limitations

This is a cross-sectional design and does not investigate temporal development of the associations. However, an inverse relationship between CRF and SAF-derived AGEs was found and a long-term follow-up is warranted to proof causal associations between lifestyle-related risk factors and metabolic health in children. Furthermore, only 3% were children with obesity in our cohort. Studies in populations with a higher prevalence of children with obesity may help to further differentiate the association of obesity and AGEs in children. CRF was assessed by a 20-m shuttle run and not by spiroergometry as the gold standard for objective assessment of physical fitness. Twenty-meter shuttle run, however, is an appropriate surrogate if spiroergometry is not available. It is a valid tool to assess CRF in all children including children with obesity.[25,40] With respect to measuring SAF-derived AGEs, the device cannot be applied in children with dark skin and, therefore, a selection bias is given for technical reasons. Of note, the measurement of SAF-derived AGEs only detects complex mixtures of AGE and non-AGE fluorophores in a qualitative manner. This cross-sectional study is limited to explaining PA-mediated mechanisms of action on SAF-derived AGEs. Interventional studies are warranted, which ideally include the full spectrum of AGEs metabolites in serum. This was beyond the scope of our population-based approach. The R^2 values show that there is a high variability in our cohort and a large sample size is needed to confirm our findings. However, the CIs indicate that there is evidence that CRF is associated with SAF-derived accumulation of AGEs. This result was independent of BMI and blood pressure. Only about 7% of the variation in SAF can be explained by a single factor such as CRF. The mechanisms involved are likely to be multifactorial as is often the case in complex physiological systems. Future longer-term studies have to investigate if this association is predictive for the development of cardiometabolic disease later in life. One strength of our study includes the large sample size and the use of standardized procedures to measure BMI, BF, BP and CRF in children.

CONCLUSIONS

In conclusion, our results demonstrate that physical fitness was associated with SAF-derived AGEs in young children without evidence for statistically significant associations with body mass and BP. Analysis of skin AGEs is a feasible tool to differentiate the effects of physical fitness on tissue glycation and metabolism. We postulate that exposure times to a higher BMI and BP are too short to affect AGEs deposition in tissue of young otherwise healthy children. Associations may still become apparent at later stages and during adolescents. From a clinical perspective, higher AGEs have been linked with development of diabetes mellitus in children[15,16] but, as our findings demonstrate, this is not the case for childhood obesity and hypertension. In children with increased AGEs and referenced to our normal values, treatment strategies should still focus on reducing AGEs accumulation to counteract the growing prevalence of metabolic disease in childhood and later in life. Primary prevention programs may need to focus on improving physical activity and fitness as treatment options to achieve this ambitious long-term goal.

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AUTHOR CONTRIBUTIONS

S.K. planned and conducted the study, collected data, performed the statistical analysis, prepared and revised the manuscript. K.E. designed the study and revised the manuscript. M.T. and M.M. collected data and revised the manuscript. L.Z. designed the study and revised the manuscript. H.H. conceptualized and designed the study, discussed the statistical analysis, prepared and critically revised the manuscript. All authors approved the final manuscript as submitted and agreed to be accountable for all aspects of the work.

ADDITIONAL INFORMATION

The online version of this article (https://doi.org/10.1038/s41390-019-0694-z) contains supplementary material, which is available to authorized users.

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REFERENCES

1. Yamagishi, S. Potential clinical utility of advanced glycation end product cross-link breakers in age- and diabetes-associated disorders. *Rejuvenation Res.* **15**, 564–572 (2012).
2. Tahara, N. et al. Positive association between serum level of glyceraldehyde-derived advanced glycation end products and vascular inflammation evaluated by [18F]Fluorodeoxyglucose positron emission tomography. *Diabetes Care* **35**, 2618–2625 (2012).
3. Vasan, S. et al. An agent cleaving glucose-derived protein crosslinks in vitro and in vivo. *Nature* **382**, 275–278 (1996).
4. Brownlee, M. et al. Aminoguanidine prevents diabetes-induced arterial wall protein cross-linking. *Science* **232**, 1629–1632 (1986).
5. Meerwaldt, R. et al. Simple non-invasive assessment of advanced glycation endproduct accumulation. *Diabetologia* **47**, 1324–1330 (2004).
6. den Engelsen, C. et al. Advanced glycation end products measured by skin autofluorescence in a population with central obesity. *Dermatoendocrinol* **4**, 33–38 (2012).
7. van Waateringe, R. P. et al. Skin autofluorescence, a non-invasive biomarker for advanced glycation end products, is associated with the metabolic syndrome and its individual components. *Diabetol. Metab. Syndr.* **9**, 42 (2017).
8. Cavero-Redondo, I. et al. Skin autofluorescence-indicated advanced glycation end products as predictors of cardiovascular and all-cause mortality in high-risk subjects: a systematic review and meta-analysis. *J. Am. Heart Assoc.* **7**, e009833 (2018).
9. Kim, C.-S., Park, S. & Kim, J. The role of glycation in the pathogenesis of aging and its prevention through herbal products and physical exercise. *J. Exerc. Nutr. Biochem.* **21**, 55–61 (2017).
10. Macías-Cervantes, M. H. et al. Effect of an advanced glycation end product-restricted diet and exercise on metabolic parameters in adult overweight men. *Nutr. Burbank Los Angel. Cty. Calif. März* **31**, 446–451 (2015).
11. Couppé, C. et al. Life-long endurance running is associated with reduced glycation and mechanical stress in connective tissue. *Age Dordr. Neth.* **36**, 9665 (2014).
12. Drenth, H. et al. Advanced glycation end-products are associated with physical activity and physical functioning in the older population. *J. Gerontol. A Biol. Sci. Med. Sci.* **73**, 1545–1551 (2018).
13. McNulty, M., Mahmud, A. & Feely, J. Advanced glycation end-products and arterial stiffness in hypertension. *Am. J. Hypertens. März* **20**, 242–247 (2007).
14. Gupta, A. & Uribarri, J. Dietary advanced glycation end products and their potential role in cardiometabolic disease in children. *Horm. Res. Paediatr.* **85**, 291–300 (2016).
15. Jaisson, S. et al. Early formation of serum advanced glycation end-products in children with type 1 diabetes mellitus: relationship with glycemic control. *J. Pediatr.* **172**, 56–62 (2016).
16. Shah, S. et al. Advanced glycation endproducts in children with diabetes. *J. Pediatr.* **163**, 1427–1431 (2013).
17. World Medical Association. World Medical Association Declaration of Helsinki: ethical principles for medical research involving human subjects. *JAMA* **310**, 2191–2194 (2013).
18. von Elm, E. et al. The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement: guidelines for reporting observational studies. *Lancet Lond. Engl.* **370**, 1453–1457 (2007).
19. Meerwaldt, R. et al. The clinical relevance of assessing advanced glycation end-products accumulation in diabetes. *Cardiovasc. Diabetol.* **7**, 29 (2008).
20. Cole, T. J. et al. Establishing a standard definition for child overweight and obesity worldwide: international survey. *BMJ* **320**, 1240–1243 (2000).
21. Pickering, T. G. et al. Recommendations for blood pressure measurement in humans: an AHA Scientific Statement from the Council on High Blood Pressure Research Professional and Public Education Subcommittee. *J. Clin. Hypertens.* **7**, 102–109 (2005).
22. Neuhauser, H. K. et al. Blood pressure percentiles by age and height from non-overweight children and adolescents in Germany. *Pediatrics* **127**, e978–e988 (2011).
23. Bös, K. & Wohlmann, R. Allgemeiner Sportmotorischer Test <AST 6-11> zur Diagnose der konditionellen und koordinativen Leistungsfaehigket. *Sportunterricht* **36**, S. 145–156 (1987).
24. van Mechelen, W., Hlobil, H. & Kemper, H. C. Validation of two running tests as estimates of maximal aerobic power in children. *Eur. J. Appl. Physiol.* **55**, 503–506 (1986).
25. Mayorga-Vega, D., Aguilar-Soto, P. & Viciana, J. Criterion-related validity of the 20-M shuttle run test for estimating cardiorespiratory fitness: a meta-analysis. *J. Sports Sci. Med.* **14**, 536–547 (2015).
26. Zahner, L. et al. A school-based physical activity program to improve health and fitness in children aged 6-13 years („Kinder-Sportstudie KISS"): study design of a randomized controlled trial [ISRCTN15360785]. *BMC Public Health* **6**, 147 (2006).
27. Simon Klenovics, K. et al. Reference values of skin autofluorescence as an estimation of tissue accumulation of advanced glycation end products in a general Slovak population. *Diabet. Med. J. Br. Diabet. Assoc.* **31**, 581–585 (2014).
28. Koetsier, M. et al. Reference values of skin autofluorescence. *Diabetes Technol. Ther.* **12**, 399–403 (2010).
29. van der Heyden, J. C. et al. Increased skin autofluorescence of children and adolescents with type 1 diabetes despite a well-controlled HbA1c: results from a cohort study. *BMC Endocr. Disord.* [Internet]. **16** (2016) [zitiert 11. Oktober 2018]. Verfügbar unter: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5017065/
30. Accacha, S. et al. Plasma advanced glycation end products (AGEs), receptors for AGEs and their correlation with inflammatory markers in middle school-age children. *Horm. Res. Paediatr.* **80**, 318–327 (2013).
31. Tóthová, L. et al. Sex differences of oxidative stress markers in young healthy subjects are marker-specific in plasma but not in saliva. *Ann. Hum. Biol.* **40**, 175–180 (2013).
32. Botros, N. et al. Advanced glycation end-products (AGEs) and associations with cardio-metabolic, lifestyle, and dietary factors in a general population: the NQplus study. *Diabetes Metab. Res. Rev.* **33** (2017).
33. Lurbe, E. & Ingelfinger, J. R. Blood pressure in children and adolescents: current insights. *J. Hypertens.* **34**, 176–183 (2016).
34. Petrie, J. R., Guzik, T. J. & Touyz, R. M. Diabetes, hypertension, and cardiovascular disease: clinical insights and vascular mechanisms. *Can. J. Cardiol.* **34**, 575–584 (2018).
35. El-Saeed, G. S. M. et al. Advanced glycation end products and soluble receptor as markers of oxidative stress in children on hemodialysis. *Ren. Fail.* **37**, 1452–1456 (2015).
36. Mulder, D. J. et al. Skin autofluorescence, a novel marker for glycemic and oxidative stress-derived advanced glycation end products: an overview of current clinical studies, evidence, and limitations. *Diabetes Technol. Ther.* **8**, 523–535 (2006).
37. Accattato F., et al. Effects of acute physical exercise on oxidative stress and inflammatory status in young, sedentary obese subjects. *PLoS ONE* [Internet], **12** (2017) [zitiert 21. März 2018]. Verfügbar unter: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5459463/
38. Ahmed, N. Advanced glycation endproducts—role in pathology of diabetic complications. *Diabetes Res. Clin. Pract.* **67**, 3–21 (2005).
39. Lindsey, J. B. et al. Receptor for advanced glycation end-products (RAGE) and soluble RAGE (sRAGE): cardiovascular implications. *Diab. Vasc. Dis. Res.* **6**, 7–14 (2009).
40. Quinart, S. et al. Evaluation of cardiorespiratory fitness using three field tests in obese adolescents: validity, sensitivity and prediction of peak V ˙ O 2. *J. Sci. Med. Sport.* **17**, 521–525 (2014).