Fasting indices of glucose-insulin-metabolism across life span and prediction of glycemic deterioration in children with obesity from new diagnostic cut-offs

Maximiliane Chiara Hammel,[a,k] Robert Stein,[a,b,k] Jürgen Kratzsch,[c] Mandy Vogel,[d] Alexander J. Eckert,[e,f] Rima Destya Triatin,[g] Marco Colombo,[a] Christof Meigen,[d] Ronny Baber,[c,d] Juraj Stanik,[a,h] Ulrike Spielau,[a] Anette Stoltze,[a] Kerstin Wirkner,[d,i] Anke Tönjes,[j] Harold Snieder,[g] Reinhard W. Holl,[e,f] Michael Stumvoll,[j] Matthias Blüher,[b,j] Wieland Kiess,[a,d] and Antje Körner[a,b,d,*]

[a]Medical Faculty, University Hospital for Children and Adolescents, Center for Pediatric Research, University of Leipzig, Leipzig, Germany
[b]Helmholtz Institute for Metabolic, Obesity and Vascular Research (HI-MAG) of the Helmholtz Zentrum München at the University of Leipzig and University Hospital Leipzig, Germany
[c]Medical Faculty, Institute of Laboratory Medicine, Clinical Chemistry and Molecular Diagnosis, University of Leipzig, Leipzig, Germany
[d]LIFE Child Leipzig Research Center for Civilization Diseases, University of Leipzig, Leipzig, Germany
[e]University of Ulm, Institute of Epidemiology and Medical Biometry, ZIBMT, Ulm, Germany
[f]German Center for Diabetes Research (DZD), Munich-Neuherberg, Germany
[g]Department of Epidemiology, University of Groningen, University Medical Center Groningen, Groningen, the Netherlands
[h]Medical Faculty, Department of Pediatrics, and DIABGENE Laboratory, Institute of Experimental Endocrinology, Biomedical Research Center, Slovak Academy of Sciences, Comenius University, Bratislava, Slovakia
[i]Institute for Medical Informatics, Statistics and Epidemiology, Medical Faculty, University of Leipzig, Leipzig, Germany
[j]Department of Endocrinology, Nephrology und Rheumatic Diseases, Medical Faculty, University of Leipzig, Leipzig, Germany

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Summary

Background Fasting indices of glucose-insulin-metabolism are an easy and affordable tool to assess insulin resistance. We aimed to establish reference ranges for fasting insulin indices that reflect age-dependent variation over the entire life span and subsequently test their clinical application regarding the prediction of glycemic deterioration in children.

Methods We calculated age- and puberty-dependent reference values for HOMA-IR, HOMA2-IR, HOMA-β, McAuley index, fasting insulin, and fasting glucose from 6994 observations of 5512 non-obese healthy subjects aged 5–80 years. Applying those references, we determined the prevalence of insulin resistance among 2538 subjects with obesity. Furthermore, we investigated the intraindividual stability and the predictive values for future dysglycemia of these fasting indices in 516 children and adolescents with obesity up to 19 years of follow-up. We validated the results in three independent cohorts.

Findings There was a strong age-dependent variation of all indices throughout the life span, including prolonged recovery of pubertal insulin resistance and a subsequent continuous increase throughout adulthood. Already from age 5 years onwards, >40% of children with obesity presented with elevated parameters of insulin resistance. Applying newly developed reference ranges, insulin resistance among children with obesity doubled the risk for future glycemic deterioration (HOMA-IR HR 1.88 (95% CI 1.1–3.21)), fasting insulin HR 1.89 (95% CI 1.11–3.23). In contrast, fasting glucose alone was not predictive for emerging dysglycemia in children with obesity (HR 1.03 (95% CI 0.62–1.71)). The new insulin-based thresholds were superior to fasting glucose and HbA1c in detecting children eventually manifesting with dysglycemia in prospective analyses.

Interpretation The variation of fasting glucose-insulin-metabolism across the life span necessitates age-specific reference ranges. The improved prediction of future glycemic deterioration by indices based on fasting insulin beyond simple glucose measures alone could help to stratify risk characteristics of children with obesity in order to guide patient-tailored prevention and intervention approaches.

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Abbreviations: *ADA*, American diabetes association; *FG*, Fasting glucose; *FI*, Fasting insulin; *HOMA-IR*, Homeostasis model assessment of insulin resistance index; *HR*, Hazard ratio; *IRB*, Institutional review board; *SDS*, Standard deviation score; *TG*, Triglyceride

*Corresponding author. Helmholtz Institute for Metabolic, Obesity and Vascular Research (HI-MAG) of the Helmholtz Zentrum München at the University of Leipzig and University Hospital Leipzig, Germany.
E-mail addresses: Antje.Koerner@medizin.uni-leipzig.de, antje.koerner@helmholtz-munich.de (A. Körner).
[k]Authors contributed equally.

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Keywords: Insulin resistance; Insulin sensitivity; HOMA-IR; McAuley index; Reference values; Childhood obesity; Obesity; Type 2 diabetes; Prediabetes

Research in context

Evidence before this study

We searched for observational studies, reviews and meta-analyses published in any language from inception up until December 2021 in PubMed using the search terms "insulin resistance", "insulin sensitivity", "fasting index", "reference value", "reference interval" and "HOMA-IR". In summary, fasting indices of glucose-insulin-metabolism were used as an easy and affordable tool to assess insulin resistance in a wide range of studies with different cut-offs and ranges proposed. However, no study had considered reference values for fasting indices of glucose-insulin-metabolism over the whole age span including both childhood and adulthood for a broad range of indices. The currently applied reference values have been established decades ago and did not, or only to a narrow extend, consider age dependent variations. Furthermore, we did not find studies testing the clinical implications of the proposed cut-offs regarding the onset of glycemic deterioration among childhood.

Added value of this study

This study establishes new reference values for fasting glucose-insulin-parameters from early childhood to old age. All indices show strong age-dependent dynamics including prolonged recovery of pubertal insulin resistance and a subsequent continuous increase throughout adulthood. Furthermore, we show that as early as from early childhood onwards (age 5 years), there is a high prevalence of disturbed glucose metabolism (>40%) in patients with obesity and that age-related insulin-based indices perform better in prediction of future dysglycemia in children with obesity.

Implications of all the available evidence

The adoption of lifespan-wide age-adjusted reference values for fasting glucose-insulin-indices may allow earlier identification of patients at risk of dysglycemia and thus prompt earlier risk-adjusted intervention.

Introduction

Concomitant with the worldwide increasing prevalence and severity of obesity in childhood,[1] the onset of type 2 diabetes and prediabetes is shifting towards childhood and adolescence.[2] This is especially alarming, as young patients with type 2 diabetes show a three to four fold faster beta-cell deterioration and higher rates of therapeutic failure and early organ damages as compared to subjects with adult-onset type 2 diabetes.[3,4] On the other hand, there is a window of opportunity in childhood for intervention, as comorbidities such as type 2 diabetes are more likely to be reversible, when obesity is treated rigorously.[5] Therefore, it is important to detect and even predict impairment in glucose-insulin metabolism early in life.

Although insulin resistance and beta-cell-dysfunction are major steps preceding the manifestation of type 2 diabetes,[6] diagnostic criteria targeting glucose-insulin metabolism are solely based on fasting glucose (FG), HbA1c and 2 h OGTT glucose according to the American Diabetes Association (ADA) guidelines.[7] However, these glycemia-based parameters are uniformly applied irrespective of age, sex and condition and may not be sufficient. Furthermore, they do not reflect the underlying pathology and risk constellations and recent investigations suggest more complex phenotypes for diabetes and prediabetes.[6,8,9] Additionally, the established age-independent cut-offs were derived from adults and thus do not account for age- and puberty-related dynamics during childhood and adolescence.

Fasting indices based on glucose and insulin levels are useful and feasible criteria to evaluate insulin sensitivity in a clinical and large-scale epidemiological context. Among those, the homeostasis model assessment of insulin resistance index (HOMA-IR) is the most commonly used proxy for insulin resistance as it correlates strongly with the results of euglycemic-hyperinsulinemic clamps.[10] The HOMA-IR has been further improved, resulting in the HOMA2-IR, which shows an even stronger prognostic power for type 2 diabetes in adults, as compared to the HOMA-IR.[10,11] The HOMA-β index, which is widely used but controversially discussed, can be applied to estimate beta-cell function.[12] In addition, it has been suggested that triglyceride (TG) levels in combination with fasting insulin (FI), as in the McAuley index, provide a better prediction of insulin resistance than FI alone.[13]

Nevertheless, reference ranges targeting those indices over the whole life span including the transition

from childhood into adulthood are lacking so far. Furthermore, there are contradictory recommendations in the literature as to which cut-offs should be applied to define insulin resistance.[10,14,15] Moreover, little is known about the intraindividual longitudinal stability of these indices and whether they are useful for the prediction of type 2 diabetes or other obesity related disorders.

Herein, we investigated whether and how fasting glucose-insulin-metabolism changes over the entire life span. Secondly, we derived reference ranges for fasting indices that reflect those changes. Finally, we assessed performance of those indices in predicting glycemic deterioration specifically in children and adolescents, who are the most important group that prevention strategies should be addressed to.

Methods

Design and study population

Leipzig cohorts

Pediatric data were obtained between 1999 and 2023 from the LIFE Child Cohort[16] (recruited from the urban population around the city of Leipzig, registration number at institutional review board (IRB-Nr): 265-10-19042010; NCT02550236) and the obesity-enriched Leipzig Childhood Obesity Cohort (recruited from the local obesity outpatient clinic, IRB-Nr: 007/04, NCT04491344), which have been described elsewhere.[17] Data of adults were obtained from the corresponding LIFE Adult Cohort[18] (IRB-Nr: 263-2009-14122009), which was recruited as a random sample from the urban population around Leipzig between 2011 and 2014. Written informed consent was provided by the legal guardians as well as the subjects themselves from the age of 12 years. All studies meet the ethical standards of the Declaration of Helsinki as revised in 2008 and have been approved by the institutional review board of the Medical Faculty of the University Leipzig, Germany.

Among those cohorts, 12,355 subjects (15,066 observations) aged 5–80 years had a valid assessment of fasting insulin and fasting glucose and a fasting time of at least 8 h. Participants with syndromes or diseases affecting glucose metabolism (Type 1 diabetes, rheumatic diseases, autoimmune diseases, active or currently treated cancer, renal insufficiency, liver cirrhosis, pituitary tumors, congenital adrenal hyperplasia, precocious puberty, delayed puberty, Prader Willi syndrome, Klinefelter syndrome), or medications with potential influence on glucose metabolism (antidiabetics, systemic glucocorticoids, pancreatic hormones, immunosuppressives, betablocker, statins, diuretics, somatostatin, somatotropin, ACTH and systemic sympathomimetics) were excluded. More information on the selection of the study population can be retrieved from Figure S1.

The calculation of reference values was based on a non-obese (BMI SDS <1.88 in children (≤18 years), BMI <30 kg/m^2 in adults) reference cohort (n = 5512 healthy subjects with 6994 observations, Table S2). As in our study cohorts people with overweight were slightly overrepresented, we performed random sampling from the participants with overweight (Figure S1) to achieve a sample reflecting the shares of overweight and normal weight, differentiated by sex and age groups, among the German population.[19,20] To test the clinical implications of those reference values, we used an obesity cohort encompassing 2538 subjects (3551 observations) (Tables S2–S4, Figure S1). Among children and adolescents with obesity, a subset of 516 participants had multiple visits with a follow-up time of more than 6 months, hence serving as follow-up cohort for longitudinal survival analyses (Figure S1).

Pubertal status was determined according to Tanner[21] by trained staff members and categorized into five pubertal stages ranging from 1 (prepubertal) to 5 (complete maturity) (Table S1).

APV-registry

Data were retrieved from the standardized multicenter Adiposity Patient Follow-up (APV) registry (www.a-p-v.de) using the data pool of October 2022. The registry started in 1999 collecting cross-sectional and longitudinal data on children and adolescents, but also adults with obesity from 238 centers in Germany, Austria, and Switzerland. 52 of the 238 centers provided data for this analysis with the Leipzig center excluded. Anthropometric and metabolic anonymized data are transferred and compiled in a cumulative database. Ethical approval for the APV data collection and centralized analysis was obtained by the Ethics Committee of the University of Ulm (IRB-Nr. 133/22), and the consortium of the APV registry warrants the integrity of all data. Data protection and ethical guidelines correspond to local standards and all participating institutions confirm adherence to local data protection regulations.

We included children aged 5–23 years with obesity and at least one complete data set for BMI-SDS, fasting glucose, fasting insulin and triglycerides (Figure S2). Applying exclusion criteria as in the Leipzig cohorts left 10,051 subjects with 14,721 observations (Table S5). For follow-up analyses, patients were selected who were <18 years and did not have dysglycemia at their first visit with a minimum follow-up time of 6 months.

Lifelines cohort

Lifelines is a multi-disciplinary prospective population-based cohort study examining in a three-generation design health and health-related behaviors of 167,729 persons living in the North of the Netherlands, with a special focus on multi-morbidity and complex genetics. Fasting blood was collected from participants aged 8 years and older.[22]

We excluded participants taking medications that may influence glucose metabolism, self-reported of

diseases (i.e., cancer, renal failure, liver cirrhosis and rheumatism), individuals with missing information on body weight or height, or <8 h fasting before blood sampling. Finally, we included a total of 136,475 participants (Figure S3).

Signed informed consent was provided by all participants. Lifelines was conducted according to the principles of the Declaration of Helsinki and following the research code of the University Medical Center Groningen and was approved by the medical ethical committee of the University Medical Center Groningen (2007/152). Characteristics of the study population are provided in Tables S6 and S7.

Sorbian cohort

The Sorbian population in Eastern Germany is an ethnic minority with putative genetic isolation. Between 2005 and 2006 1049 adults have been recruited as described elsewhere.[23] Individuals without a valid measurement of fasting glucose or fasting insulin as well as individuals with obesity were excluded from this analysis, resulting in 639 subjects serving as an independent reference cohort (Figure S4). Details of the study cohort are summarized in Table S8.

For all cohorts, obesity in children was defined as a BMI ≥97th percentile (BMI standard deviation score (SDS) ≥1.88) according to sex- and age-specific national reference values[24,25] and in adults as a BMI ≥30 kg/m^2. A BMI SDS between 1.28 and 1.88 in children and a BMI of 25–30 kg/m^2 in adults was considered as overweight.

Laboratory assessments

All laboratory assessments of the Leipzig and Sorbian cohorts were conducted by the certified local laboratory of the university hospital Leipzig. Blood samples taken after a minimum of 8 h of fasting were either analyzed immediately or centrifuged within 30 min and stored at –80 °C within 2 h. Insulin serum concentrations were determined by the analyzers Cobas 8000 (Roche Diagnostics, Mannheim, Germany) and LIAISON (DiaSorin, Saluggia, Italy). Glucose was either measured in serum by a Cobas 8000 analyzer (Roche Diagnostics, Mannheim, Germany) or in hemolysates by the automated laboratory analyzer Super GL speedy using an enzymatic-amperometric method. We confirmed comparability of respective methods by Bland–Altman plots and Passing-Bablok regression (Figure S5). Triglycerides were assessed by enzymatic color assay in a Cobas analyzer (Roche Diagnostics, Mannheim Germany). For the Sorbian cohort, insulin serum concentrations were determined with the AutoDELFIA insulin assay (PerkinElmer Life and Analytical Sciences, Turku, Finland) and glucose serum concentrations by the Cobas 8000 analyzer (Roche Diagnostics, Mannheim, Germany).

Fasting indices were calculated as follows[13,26]:

$$HOMA-IR=\frac{FG_{(mmol/l)}\times FI_{\left(\frac{mIU}{l}\right)}}{22.5},$$

$$HOMA-\beta=\frac{\left(FI_{\left(\frac{mIU}{l}\right)}\right)\ x\ 20}{FG_{\left(\frac{mmol}{l}\right)}-3.5}\ \%,$$

$$McAuley\ index=$$

$$\exp\left(2.63-0.28\ \ln\left(FI_{\left(\frac{mIU}{l}\right)}\right)-0.31\ \ln\left(TG_{\left(\frac{mmol}{l}\right)}\right)\right).$$

For the calculation of HOMA2-IR, we used the web-based HOMA2-calculator of Oxford University.[27] Laboratory assessments in the APV-registry was conducted by the respective certified local labs. In the Lifelines cohort, fasting glucose was assessed in plasma of fresh blood samples by the local laboratory of the University Medical Center Groningen.

Statistics

The statistical analyses for the Leipzig, Lifelines and Sorbian cohorts were conducted with R Version 3.6.1. To assess the influence of age, sex, and BMI on the respective index, we applied linear mixed effect models with the fasting index as dependent variable and age, sex or BMI (SDS) as independent variables. The subject ID was added as random effect to account for repeated measurements of the same participant.

Reference values were calculated according to the EP28-A3c guideline of the Clinical and Laboratory Standards Institute[28] based on data of healthy participants without obesity (reference cohort). We assigned appropriate subgroups for age and pubertal stage to calculate separate reference intervals by applying the method of Harris and Boyd[29] as well as visual inspection (Tables S2–S4). Within these subgroups, repeated measurements from the same subject were removed by random exclusion. Afterwards, outliers were excluded according to Tukey[30] on Box-Cox transformed data. Percentiles were calculated by two approaches applying the R packages *referenceIntervals* and *gamlss*: i.) non-parametrically and ii.) by fitting a three-parameter Box-Cox Cole and Green distribution to the data set using a non-linear maximization[31] which also enabled the calculation of SDS values.

For longitudinal prediction analyses, the endpoint (dysglycemia) was defined as follows: therapy with antidiabetic drugs or fulfilling at least two of the ADA (pre)diabetes criteria (FG ≥ 5.6 mmol/l, HbA1c ≥ 5.7%, 2 h OGTT glucose ≥7.8 mmol/l).[7] The time until the first onset of dysglycemia was evaluated by event-time-analysis. For this, children were stratified

into a risk group and a non-risk group according to their fasting index value with respect to the previously established reference values at baseline examination. The statistical difference of event-free survival (time free of dysglycemia) between those risk groups was assessed by log-rank test. Additionally, hazard ratios were calculated using Cox proportional hazard regression, hereby also taking the covariates BMI-SDS, sex, age and family history of diabetes into account.

Statistical evaluation of the APV cohort was performed using SAS version 9.4 (Statistical Analysis Software SAS Institute, Cary, NC, USA) in accordance to the workflow described above.

Role of funding source

The study sponsor/funder was not involved in the design of the study; the collection, analysis, and interpretation of data; writing the report; and did not impose any restrictions regarding the publication of the report.

Results

Patterns of fasting indices for glucose-insulin metabolism across the life span

We investigated age-related courses of fasting indices in our cross-sectional Leipzig reference cohort of 5512 healthy subjects without obesity between age 5 and 80 years.

All parameters investigated showed an age-dependent pattern (Fig. 1, Table S9).

FG increased from early childhood to a peak during puberty and subsequently decreased in adolescence and early adulthood. Thereafter, FG constantly rose from the age of 25 years onwards by approximately 0.25 mmol/l per decade reaching a median of 5.5 mmol/l in the 60–80 years age group (Fig. 1, Table 1).

Likewise, FI and HOMA indices more than doubled from early childhood to peak at the age of 13–14 years (Fig. 1). This puberty-related insulin resistance gradually regressed until the age of 25 years without reaching pre-pubertal values again. Subsequently, the levels remained stable until the age of 40 years and increased thereafter. The McAuley index, as a proxy of insulin sensitivity, mirrored the aforementioned distribution of the HOMA-indices. Triglycerides showed a constant increase until 60 years. The course of HbA1c levels resembled FG levels.

To assess the generizability of these age-dependent dynamics in fasting glucose-insulin-parameters, we compared two independent cohorts: Lifelines from Groningen, Netherlands, and the putative genetically isolated population of Sorbians within Germany. Age-dependent patterns were comparable to our results (Figures S6 and S7) as were absolute levels of indices. Glucose levels of the Leipzig cohort tended to be slightly higher than in Lifelines (around 0.2 mmol/l, Figure S9) and insulin levels tended to be slightly higher than in the Sorbian cohort (around 6–21 pmol/l, Figure S10).

In linear model analyses we identified significant associations between the examined fasting indices and BMI, age and sex (Tables S9 and S10). Since a large sample size would achieve statistical significance even with small changes, we applied the method proposed by Harris and Boyd[25] to evaluate to which extent these covariate factors will have a clinically relevant impact and hence should be considered prior to the calculation of reference ranges. Based on this, we calculated reference ranges for six separate age groups (Table 1, Table S13) and three separate pubertal groups (Tables S11 and S12). Sex did not make a relevant difference and thus was not considered for the calculation of reference ranges. The novel reference values are available open-access at the Ped(Z)Pediatric Calculator app[32] and via an R package.[33]

Concerning puberty, a strong age dependency was seen also within the pubertal stages (Figures S13 and S14): Fasting insulin and fasting glucose increased before and during early puberty stages, whereas they were more stable in mid-puberty. Thereafter, insulin and glucose levels regressed significantly with age, even within each pubertal stage. Hence, age conveyed more specific information in addition to the pubertal stage regarding the temporal dynamics of insulin resistance parameters and we emphasized on age-specific references for further analyses.

Parameters of insulin resistance in subjects with obesity

The shape of the age-related pattern of indices for glucose-insulin metabolism in the 2538 subjects with obesity resembled that of the reference cohort (Fig. 1). However, the magnitude of fasting indices of insulin resistance was approximately twice as high in subjects with obesity compared to subjects without obesity across the life span (Fig. 1, Tables S2–S4). Of note, more than 40% of the young children with obesity aged between 5 and 8 years already presented insulin resistance when applying the 97.5th percentile of either HOMA-IR, HOMA2-IR or FI as pathological cut-off (Fig. 2). Likewise, one third of adults with obesity (aged 24 years and older) were classified as insulin resistant (Fig. 2). The HOMA-β as a proxy for beta-cell function was constantly decreasing from the age of 25 years onwards in patients with obesity.

Age-dependent distribution of fasting glucose in the Lifelines cohorts resembled the results of the Leipzig obesity cohort, especially in adults. Of note, glucose levels during childhood tended to be lower in Lifelines in line with a lower mean BMI-SDS in Lifelines (Figures S6 and S9, Tables S3 and S7). When comparing our data to an independent, nation-wide registry of 10,051 children and adolescents with obesity (APV-registry), age- and puberty-related dynamics of glucose-insulin-indices could be confirmed (Figure S8). As such, around one third of children aged 5–8 years had

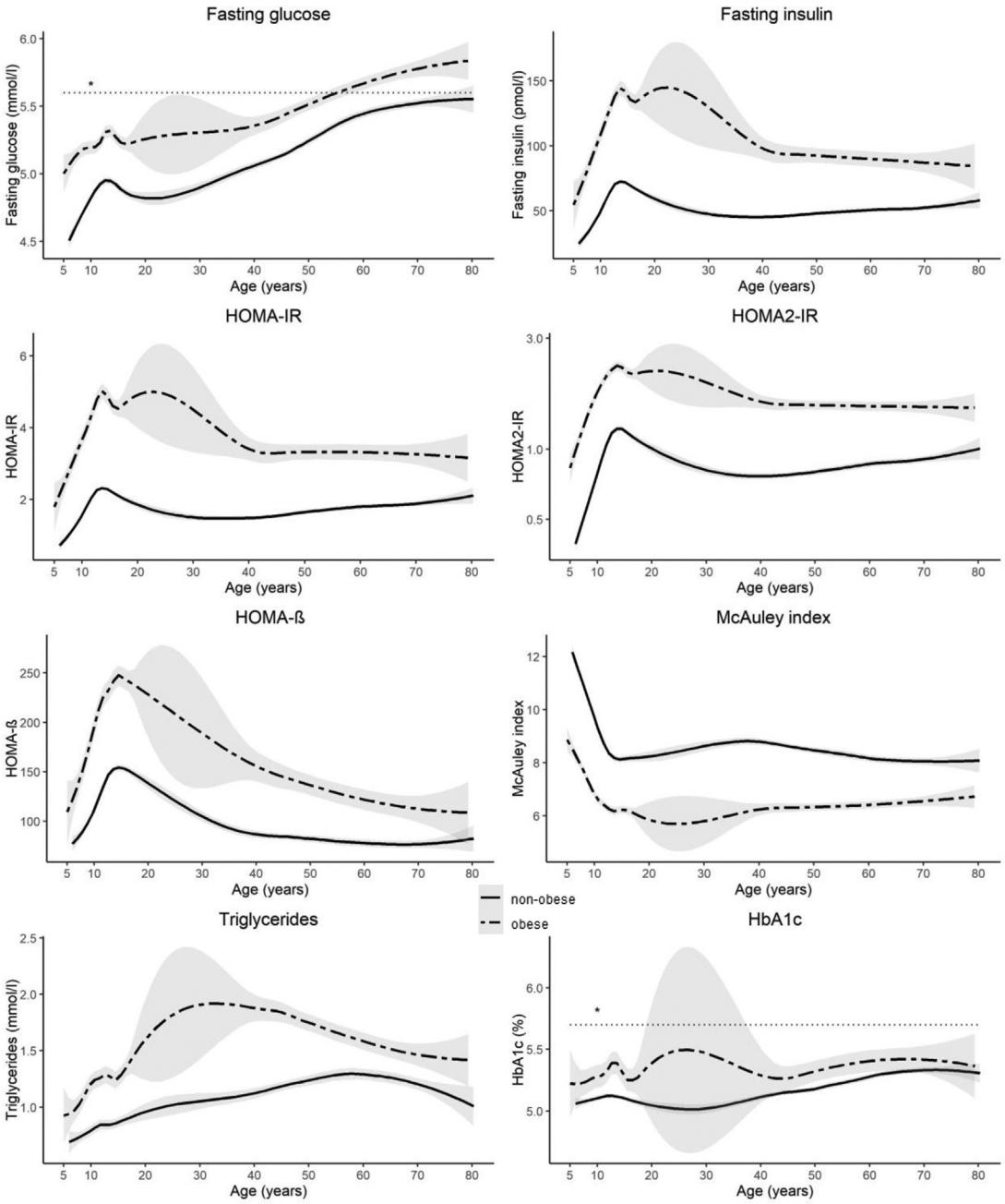

Fig. 1: **Distribution of fasting indices over age (5–80 years).** Observations from the non-obese reference cohort are depicted as solid line, whereas the obesity cohort is depicted as dashed line. Curved lines and ribbons represent local polynomial regression fitting with 95% confidence intervals. *cut-off for the diagnosis of prediabetes according to the American diabetes association; *HOMA-IR,* Homeostasis model assessment of insulin resistance index.

levels above the 97.5th percentile of insulin resistance indices.

As the BMI-based stratification of obesity has some limitations,[34] we furthermore stratified our cohort for waist-to-height ratio. This reproduced comparable rates of disturbed metabolic parameters for children and adolescents, whereas for adults prevalences tended to be higher (Figure S11).

Stability of insulin resistance parameters

We next evaluated the intraindividual stability of insulin resistance in longitudinal analyses in a subgroup of 343

Age groups (years)	N	P2.5 (CI 90%)	P5 (CI 90%)	P10 (CI 90%)	P25 (CI 90%)	P50 (CI 90%)	P75 (CI 90%)	P90 (CI 90%)	P95 (CI 90%)	P97.5 (CI 90%)
FG (mmol/l)										
5–8	461	3.9 (3.8–3.9)	4 (3.9–4.1)	4.1 (4.1–4.2)	4.4 (4.3–4.4)	4.6 (4.6–4.6)	4.9 (4.8–4.9)	5.1 (5.0–5.1)	5.2 (5.2–5.3)	5.4 (5.2–5.4)
9–10	461	4.2 (4.1–4.3)	4.3 (4.2–4.3)	4.4 (4.4–4.5)	4.6 (4.6–4.6)	4.8 (4.8–4.9)	5.0 (5.0–5.1)	5.3 (5.2–5.3)	5.4 (5.4–5.5)	5.6 (5.5–5.7)
11–15	929	4.2 (4.2–4.3)	4.3 (4.3–4.3)	4.4 (4.4–4.5)	4.7 (4.7–4.7)	4.9 (4.9–5.0)	5.1 (5.1–5.2)	5.4 (5.4–5.4)	5.6 (5.5–5.6)	5.7 (5.6–5.8)
16–23	331	4.2 (4.1–4.3)	4.3 (4.2–4.3)	4.4 (4.3–4.4)	4.6 (4.5–4.6)	4.8 (4.8–4.9)	5.0 (5.0–5.1)	5.3 (5.2–5.4)	5.5 (5.4–5.6)	5.6 (5.5–5.7)
24–45	1018	4.3 (4.2–4.3)	4.4 (4.3–4.4)	4.5 (4.5–4.6)	4.8 (4.7–4.8)	5.0 (5.0–5.1)	5.3 (5.3–5.4)	5.7 (5.6–5.7)	5.9 (5.8–5.9)	6.1 (6.0–6.2)
46–59	1650	4.4 (4.4–4.4)	4.5 (4.5–4.5)	4.7 (4.6–4.7)	4.9 (4.9–4.9)	5.2 (5.2–5.3)	5.6 (5.6–5.6)	5.9 (5.9–6.0)	6.2 (6.1–6.2)	6.4 (6.3–6.5)
60–80	1060	4.6 (4.6–4.7)	4.7 (4.7–4.8)	4.9 (4.8–4.9)	5.1 (5.1–5.1)	5.5 (5.4–5.5)	5.8 (5.8–5.9)	6.3 (6.3–6.4)	6.5 (6.4–6.6)	6.6 (6.5–6.7)
FI (pmol//l)										
5–8	461	7.3 (5.6–8.4)	9.5 (7.9–10.4)	12.5 (11.0–13.7)	18.1 (17.1–18.9)	27.8 (25.6–29.5)	40.8 (39.2–43.5)	57.4 (54.1–62.9)	69.1 (61.0–75.8)	78.6 (75.1–81.4)
9–10	461	16.1 (15.2–17.5)	17.5 (15.6–18.2)	22.7 (22.1–24.5)	29.6 (28.6–31.1)	41.1 (38.6–44.0)	61.9 (59.1–64.6)	83.0 (78.1–88.4)	102.1 (85.7–112.6)	124.0 (116.3–135.6)
11–15	929	23.2 (21.7–24.9)	27.0 (25.2–28.4)	33.3 (31.7–34.9)	45.3 (43.8–46.8)	64.3 (62.2–66.6)	84.7 (81.7–86.6)	109.1 (104.0–113.3)	128.0 (122.2–134.9)	144.3 (138.9–150.7)
16–23	331	22.5 (18.7–23.8)	27.2 (24.6–29.3)	32.6 (28.9–35.2)	42.5 (40.2–44.8)	57.0 (54.8–59.9)	71.7 (68.8–74.8)	86.0 (82.4–90.1)	98.6 (88.6–106.9)	114.6 (109.7–125.0)
24–59	2668	17.1 (16.5–17.8)	19.6 (18.9–20.2)	23.2 (22.5–23.8)	30.4 (29.5–31.1)	42.1 (41.3–42.8)	58.2 (57.2–59.5)	78.2 (76.3–80.3)	92.5 (89.1–95.2)	107.7 (103.2–111.8)
60–80	1060	18.9 (17.8–20.1)	22.1 (20.9–22.9)	26.8 (26.0–27.5)	33.8 (32.7–34.5)	47.0 (45.6–48.6)	63.4 (60.4–65.5)	82.6 (79.4–84.7)	97.3 (88.4–103.0)	114.3 (111.7–120.5)
HOMA-IR										
5–8	461	0.2 (0.2–0.2)	0.3 (0.2–0.3)	0.3 (0.3–0.4)	0.5 (0.5–0.5)	0.8 (0.8–0.9)	1.3 (1.2–1.4)	1.7 (1.6–1.8)	2.1 (1.8–2.3)	2.6 (2.3–2.7)
9–10	461	0.5 (0.4–0.5)	0.5 (0.4–0.5)	0.7 (0.6–0.7)	0.9 (0.9–1.0)	1.2 (1.1–1.3)	2.0 (1.9–2.1)	2.6 (2.3–2.7)	3.4 (2.8–3.7)	4.2 (4.0–4.8)
11–15	929	0.6 (0.6–0.7)	0.8 (0.7–0.8)	1.0 (1.0–1.0)	1.4 (1.4–1.5)	2.0 (1.9–2.1)	2.7 (2.7–2.8)	3.6 (3.5–3.7)	4.4 (4.3–4.9)	5.0 (4.8–5.2)
16–23	331	0.7 (0.5–0.7)	0.8 (0.7–0.9)	1.0 (0.9–1.0)	1.3 (1.2–1.3)	1.7 (1.6–1.8)	2.2 (2.1–2.3)	2.8 (2.7–3.0)	3.1 (2.7–3.2)	3.6 (3.2–4.1)
24–59	2668	0.5 (0.5–0.5)	0.6 (0.6–0.6)	0.7 (0.7–0.7)	1.0 (1.0–1.0)	1.4 (1.3–1.4)	2.0 (1.9–2.0)	2.7 (2.6–2.8)	3.3 (3.2–3.4)	3.9 (3.7–4.1)
60–80	1060	0.6 (0.5–0.6)	0.7 (0.7–0.8)	0.9 (0.8–0.9)	1.2 (1.1–1.2)	1.6 (1.6–1.7)	2.3 (2.2–2.4)	3.1 (3.0–3.2)	3.8 (3.7–4.1)	4.2 (3.7–4.3)
HOMA2-IR										
5–8	461	0.1 (0.1–0.2)	0.2 (0.2–0.2)	0.2 (0.2–0.2)	0.3 (0.3–0.3)	0.5 (0.5–0.5)	0.8 (0.7–0.8)	1.1 (1–1.2)	1.3 (1.1–1.4)	1.5 (1.4–1.5)
9–10	461	0.3 (0.3–0.3)	0.3 (0.3–0.3)	0.4 (0.4–0.4)	0.5 (0.5–0.6)	0.8 (0.7–0.8)	1.2 (1.1–1.2)	1.5 (1.5–1.7)	1.9 (1.6–2.1)	2.3 (2.2–2.6)
11–15	929	0.4 (0.4–0.5)	0.5 (0.4–0.5)	0.6 (0.6–0.6)	0.8 (0.8–0.9)	1.2 (1.2–1.2)	1.6 (1.5–1.6)	2.0 (2.0–2.1)	2.4 (2.3–2.5)	2.7 (2.7–2.9)
16–23	331	0.4 (0.3–0.4)	0.5 (0.5–0.6)	0.6 (0.5–0.6)	0.8 (0.8–0.8)	1.1 (1.0–1.1)	1.3 (1.3–1.4)	1.6 (1.5–1.7)	1.8 (1.6–1.9)	2.1 (1.9–2.3)
24–59	2668	0.3 (0.3–0.3)	0.4 (0.4–0.4)	0.4 (0.4–0.4)	0.6 (0.6–0.6)	0.8 (0.8–0.8)	1.1 (1.1–1.1)	1.5 (1.5–1.5)	1.8 (1.7–1.8)	2.1 (2.0–2.1)
60–80	1060	0.4 (0.3–0.4)	0.4 (0.4–0.4)	0.5 (0.5–0.5)	0.7 (0.6–0.7)	0.9 (0.9–0.9)	1.2 (1.2–1.3)	1.6 (1.5–1.6)	1.9 (1.8–2.0)	2.2 (2.1–2.3)
HOMA-β										
5–8	458	24.7 (17.8–25.3)	32.9 (30.5–34.3)	40.2 (37.4–43.7)	54.4 (52.6–57.4)	77.7 (76.3–81.5)	99.1 (93.9–101.5)	136.3 (130.5–148.4)	151.4 (143.8–156.7)	167.4 (150.3–177.8)
9–10	461	37.4 (32.9–40.7)	44.1 (41.7–48.2)	51.9 (49.6–54.4)	65.9 (61.5–68.4)	94.6 (89.9–99.7)	126.7 (118.3–130.9)	166.9 (150.1–174.5)	199.2 (178.7–205.9)	235.1 (226.5–255.6)
11–15	929	51 (47.9–53.9)	59 (53.6–61.6)	71.8 (68.3–74.4)	96.2 (92.8–98.7)	128.4 (122.1–132.7)	177.8 (170.1–183.4)	233.1 (222.1–242.5)	284.8 (263.8–301)	330.6 (307.5–350.9)
16–23	331	53.8 (49.1–60)	63.2 (59.3–71.2)	70.3 (66.3–73.4)	93.2 (89.3–97.9)	123.2 (118.9–129.3)	168.6 (157.5–178.6)	214.8 (196.1–222.6)	250 (233–264.8)	286 (271.5–317.1)
24–59	2668	31.5 (29.9–32.7)	36.2 (35.3–36.9)	42.3 (41.4–43.8)	54.4 (52.5–55.4)	74.7 (73.6–75.9)	101.3 (98.9–103.2)	133.9 (132.3–137.3)	154.8 (148.5–158.6)	181.8 (173.8–187.3)
60–80	1060	30.3 (28.5–31.7)	34.6 (33.1–36.4)	39.9 (38.2–40.8)	51.9 (49.9–53.4)	70.3 (68.4–72.6)	93.7 (91.2–97.2)	121.6 (117.9–126.3)	141.4 (133.5–147)	163.2 (156.4–172)
McAuley index										
5–8	444	7.1 (6.9–7.3)	7.4 (7.2–7.6)	8.2 (7.9–8.5)	9.5 (9.2–9.6)	10.9 (10.7–11.2)	12.9 (12.5–13.2)	15.1 (14.8–15.6)	16.4 (16.0–17.1)	17.3 (16.8–17.9)
9–10	449	5.2 (4.9–5.6)	5.7 (5.1–5.8)	6.7 (6.4–6.9)	8.1 (7.8–8.3)	9.6 (9.4–9.8)	11.3 (11.0–11.6)	12.8 (12.5–13.0)	13.8 (13.5–14.4)	14.4 (13.9–14.8)
11–15	886	5.3 (5.1–5.5)	5.7 (5.6–5.9)	6.2 (6.0–6.3)	7.0 (6.9–7.1)	8.2 (8.1–8.3)	9.4 (9.3–9.4)	10.7 (10.3–10.8)	11.5 (11.3–11.7)	12.1 (11.7–12.4)
16–23	320	5.6 (5.4–5.9)	5.9 (5.7–6.2)	6.4 (6.1–6.6)	7.2 (7.0–7.4)	8.1 (8.0–8.2)	9.4 (9.2–9.6)	10.6 (10.2–10.9)	11.4 (10.9–11.8)	12.1 (11.9–12.3)
24–59	2666	5.0 (5.0–5.2)	5.4 (5.3–5.5)	6.0 (5.9–6.1)	7.1 (7.0–7.2)	8.5 (8.4–8.5)	9.8 (9.8–9.9)	11.2 (11.0–11.3)	11.9 (11.7–12.0)	12.8 (12.6–13.0)
60–80	1060	5.2 (5.1–5.4)	5.5 (5.4–5.7)	6.0 (5.9–6.1)	6.9 (6.8–7.0)	8.0 (7.9–8.1)	9.1 (9.0–9.2)	10.2 (10.1–10.4)	10.9 (10.6–11.1)	11.7 (11.5–12.0)

Reference ranges were determined non-parametrically. *CI*, Confidence interval; *FG*, Fasting glucose; *FI*, Fasting insulin; *HOMA-IR*, Homeostasis model assessment of insulin resistance index; *N*, Number of subjects (multiple measurements of one subject within the same age group were randomly excluded); *P*, Percentile.

Table 1: **Age-specific reference values for fasting indices of glucose-insulin metabolism in children and adults.**

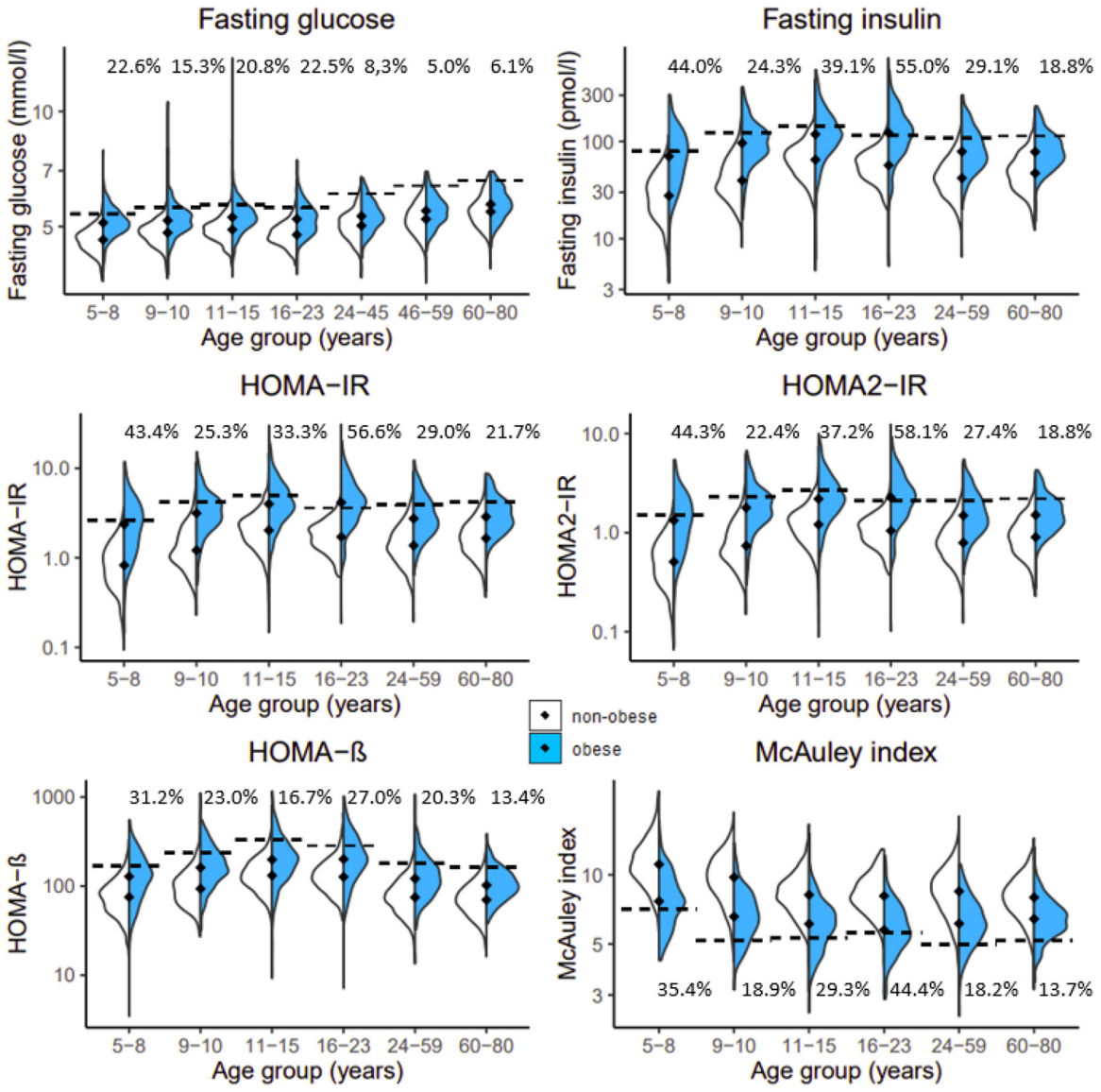

***Fig. 2:* Age-dependent distribution of fasting indices among subjects without and with obesity.** The dashed lines represent the age-specific 97.5th percentile (2.5th percentile respectively for McAuley index) as a cut-off for pathological values. The prevalence of pathological findings among subjects with obesity is depicted next to the violin plots. Black symbols represent the median of subjects without obesity and with obesity.

children, who had a follow-up visit within a maximum of two years. Hereby, two thirds of children with obesity and insulin resistance (defined as fasting index levels >97.5th percentile for e.g., FI, HOMA-IR, HOMA2-IR and McAuley-Index) consistently displayed insulin resistance during the follow-up visit at a maximum of two years later (Table 2). Adolescents with obesity, who were categorized as insulin resistant during puberty (age 11–15 years), had an even higher risk (72%) of persisting insulin resistance (HOMA-IR >97.5th percentile) until adulthood (age 16–23 years, Table 2).

In contrast, when applying the glucose-based ADA prediabetes criteria, only 51% (FG) and 36% (2 h OGTT glucose) of elevated values could be reproduced as above the reference range in the follow-up examination.

Prediction of future dysglycemia

To evaluate the clinical relevance of the indices and to determine the most appropriate cut-off, we performed survival analyses in children and adolescents with obesity targeting dysglycemia as the outcome measure. Dysglycemia was defined as either intake of

Fasting indices	All children (N = 343)			Pubertal → post-pubertal (N = 312)		
	P75	P90	P97.5	P75	P90	P97.5
FG						
Elevated at T0 (%)	59	37	17	59	33	14
Concordantly elevated (%)	79	59	44	67	64	55
FI						
Elevated at T0 (%)	71	54	32	76	57	31
Concordantly elevated (%)	86	78	63	85	77	65
HOMA-IR						
Elevated at T0 (%)	70	55	30	75	55	33
Concordantly elevated (%)	87	74	63	89	83	72
HOMA2-IR						
Elevated at T0 (%)	70	55	32	77	57	34
Concordantly elevated (%)	86	76	64	87	76	63
HOMA-β						
Elevated at T0 (%)	60	38	27	60	38	21
Concordantly elevated (%)	73	62	52	87	88	85
	P25	**P10**	**P2.5**	**P25**	**P10**	**P2.5**
McAuley index						
Diminished at T0 (%)	71	53	29	70	52	29
Concordantly diminished (%)	81	73	60	83	68	58
ADA prediabetes criteria						
FG (≥5.6 mmol/l)						
Elevated at T0 (%)			19			17
Concordantly elevated (%)			51			39
2 h glucose (≥7.8 mmol/l)						
Elevated at T0 (%)			19			20
Concordantly elevated (%)			36			59
HbA1c (≥5.7%/39 mmol/ml)			N = 238			N = 59
Elevated at T0 (%)			16			13
Concordantly elevated (%)			31			21

We included all children with obesity aged 5–18 years at baseline, who had a follow-up visit within two years. In a sub-cohort we assessed concordance of insulin resistance from pubertal age (age 11–15 years) to post-pubertal age (age 16–21 years). Different age-specific percentiles were considered as cut-offs for insulin resistance. *ADA*, American diabetes association; *FG*, Fasting glucose; *FI*, Fasting insulin; *HOMA-IR*, Homeostasis model assessment of insulin resistance index; *N*, Number of subjects; *P*, Percentile; *T0*, Baseline examination.

***Table 2*: Concordance of values above the reference range at baseline and follow-up visits in children with obesity.**

antidiabetics and/or laboratory confirmed prediabetes or diabetes (a combination of at least two pathological values for FG, HbA1c and/or 2 h OGTT glucose). Participants with euglycemia at baseline were stratified into a risk and a non-risk group according to different percentiles (75th, 90th and 97.5th) of the respective fasting index. A subset of 516 participants had multiple visits with a mean follow-up time of 4.3 years (median 2.7 years, range 0.5–19.5 years) and 73 events of dysglycemia (Table S15). Insulin resistance, defined as a fasting index above the reference range, was proven to be a risk factor for future dysglycemia throughout all analyses (Table 3, Fig. 3). For example, children with HOMA-IR levels above the age-specific 90th percentile were twice as likely to develop dysglycemia independent of age, sex, BMI-SDS and family history of type 2 diabetes (Table 3). When comparing different indices and cut-offs, we obtained the highest hazard ratios (HRs) by applying the 90th percentile of HOMA-IR (HR 1.88 (95% CI 1.11–3.21)) and FI (HR 1.89 (95% CI 1.11–3.23)). In contrast, FG percentiles were not suitable for stratifying the baseline cohort into risk groups with significant differences regarding the future onset of prediabetes (HR 1.03 (95% CI 0.62–1.71)).

Out of the 73 subjects who developed dysglycemia at follow-up, two thirds (67.1%) would have been detected already at baseline when applying the newly established 90th percentile of insulin-based indexes, whereas only one out of six showed elevated fasting parameters according to current guidelines at baseline (FG ≥ 5.6 mmol/l: 12.3%; HbA1c ≥ 5.7%: 17.8%; Table S17). We validated our observations in an independent, nation-wide cohort of 1826 children with obesity (APV-registry, mean follow-up time 2.2 years, median 1.7 years, follow-up range 0.5–10 years) and obtained similar results: if insulin resistance is present at baseline (defined by values above the 90th percentile),

	N	P75	P90	P97.5
FG				
Unadjusted HR	516	0.75 (0.47–1.19)	1.01 (0.61–1.66)	0.95 (0.43–2.07)
Adjusted HR	473	0.75 (0.46–1.23)	1.03 (0.62–1.71)	1.11 (0.49–2.52)
FI				
Unadjusted HR	516	1.83 (1.03–3.24)*	1.97 (1.21–3.21)**	1.67 (1.02–2.71)*
Adjusted HR	473	1.60 (0.86–2.99)	1.89 (1.11–3.23)*	1.58 (0.93–2.7)
HOMA-IR				
Unadjusted HR	516	1.90 (1.06–3.43)*	1.95 (1.19–3.2)**	1.49 (0.90–2.44)
Adjusted HR	473	1.72 (0.91–3.25)	1.88 (1.1–3.21)*	1.46 (0.85–2.52)
HOMA2-IR				
Unadjusted HR	516	1.85 (1.05–3.29)*	1.87 (1.14–3.05)*	1.77 (1.10–2.87)*
Adjusted HR	473	1.64 (0.88–3.04)	1.75 (1.03–2.99)*	1.7 (1.0–2.9)*
HOMA-β				
Unadjusted HR	516	1.47 (0.90–2.39)	1.54 (0.97–2.45)	1.52 (0.88–2.63)
Adjusted HR	473	1.34 (0.8–2.27)	1.53 (0.91–2.55)	1.49 (0.81–2.75)
		P25	**P10**	**P2.5**
McAuley index				
Unadjusted HR	505	1.60 (0.92–2.81)	1.84 (1.14–3.0)*	1.86 (1.11–3.11)
Adjusted HR	462	1.45 (0.80–2.63)	1.64 (0.98–2.76)	1.77 (1.03–3.05)*

Participants with obesity but without dysglycemia at baseline (n = 516) were stratified according to their fasting index value. Different age-specific percentiles (P75, P90, P97.5/P25, P10, P2.5) were applied to discriminate between a risk and a non-risk group at baseline. The risk of emerging dysglycemia between those two groups was evaluated by Cox proportional hazards regression both in an univariate model (unadjusted) and in a model adjusted for age, sex, BMI-SDS and family history of type 2 diabetes. The 95% confidence interval is given in brackets; *p < 0.05, **p < 0.01. *FG*, Fasting glucose; *FI*, Fasting insulin; *HR*, Hazard ratio; *HOMA-IR*, Homeostasis model assessment of insulin resistance index; *P*, Percentile.

Table 3: **Prediction of future dysglycemia in children with obesity based on new references.**

children are two-to three times more likely to develop dysglycemia (Figure S12, Tables S14 and S16), whereas fasting glucose was less predictive for future dysglycemia in this cohort.

Discussion

Our observation of the dynamics of fasting glucose-insulin-parameters from early childhood to old age in healthy individuals without obesity prompted us to establish age (group) specific reference ranges for these parameters. Applying these new cut-offs, we identified a high prevalence of disturbed glucose metabolism in patients with obesity as early as from early childhood onwards. Particularly insulin-based indices predicted future dysglycemia within less than 10 years in children with obesity (Fig. 4).

Age-dependent patterns of fasting glucose-insulin indices

In a large population-based cohort, we systematically investigated the pattern of fasting indices of glucose and insulin over the entire range from early childhood to old age in healthy individuals without obesity.

In childhood, the parameters reflected the known puberty-related increase in insulin resistance.[35] In contrast to what has been described so far, however, this increase did not resolve immediately after completion of puberty. Recovery was prolonged beyond adolescence and never reached prepubertal levels again. Mechanisms postulated to underlie the puberty-associated insulin resistance comprise direct or indirect effects of the growth hormone and the insulin-like growth factor, which also rise during puberty, as well as the decrease of glucose oxidation combined with the decreased insulinogenic suppression of free fatty acid oxidation.[36]

Furthermore, after completion of puberty insulin resistance parameters and FG levels steadily increased during adulthood, almost approaching the ADA proposed cut-off for impaired fasting glucose of 5.6 mmol/l in healthy, normal-weight 60–80 years old individuals. We confirmed this age dependent dynamic in FG levels in the independent Lifelines and the Sorbian cohorts.

It should be considered, however, that age per se is likely not to be the only factor affecting metabolic parameters; for example BMI, body fat content and distribution, genetic predisposition, life style factors etc. are strongly related to glucose metabolism.

From early childhood to old age, the obesity group had much increased parameters of glucose-insulin metabolism. Levels of insulin resistance parameters remained on average twice as high among subjects with obesity compared to the reference population. This could be related to lipotoxicity, which is an effect of excess fat accumulation and triggers insulin resistance.[37]

It is also important to note that an increase of FI during puberty is a sign of increased insulin resistance,

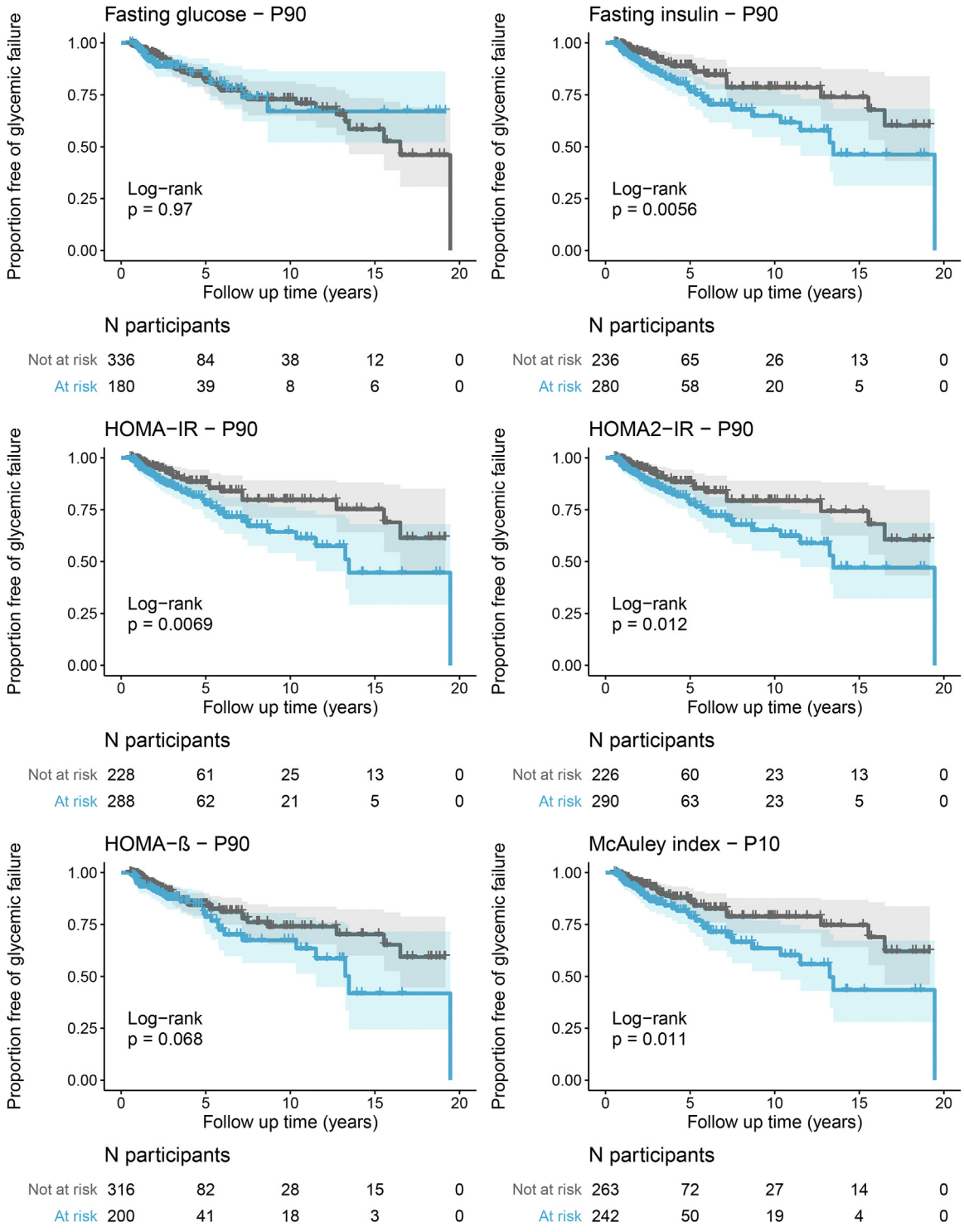

***Fig. 3:* Prediction of future dysglycemia.** Participants with obesity but without dysglycemia at baseline (n = 516) were stratified according to their fasting index value. Hereby, the 90th (P90) percentile (or P10 respectively for McAuley index) serves as the cut-off to separate a risk group (blue) from a non-risk group (gray). Differences of event-free survival (time without occurrence of dysglycemia) were compared between risk groups by log-rank test. Ribbons around the survival curves represent the 95% confidence interval and ticks represent right-censored data.

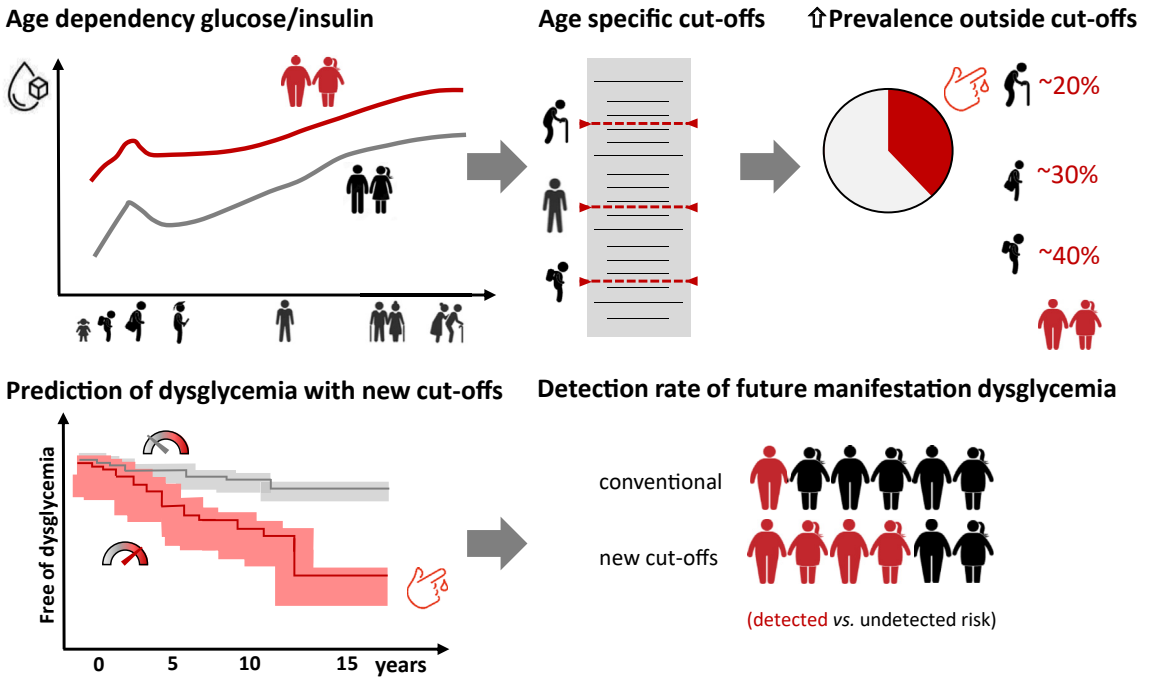

Fig. 4: **Graphical abstract.**

whereas decreasing FI in adults is more likely to reflect beta-cell dysfunction than improvement in insulin sensitivity.[38] This is in line with the age-related rise in glucose levels and the decrease of beta-cell function (HOMA-β) among adults with obesity.

Concerning the question whether to use age- or puberty-dependent reference values in children, the use of age-dependent might be easier in practice as information on the puberty status is not always given and there is still a strong age-dependence even during pre- and postpubertal stages and even within the pubertal stages. However, in children with early or late puberty, puberty-dependent reference ranges might be helpful as a second reference, which we provide in the supplement.

New reference ranges

According to current guidelines, fixed age-stable cut-offs for FG, HbA1c and 2 h glucose are applied for the diagnosis of (pre)diabetes that do not account for the considerable dynamics in patterns of glucose-insulin metabolism parameters across the life span. They have been solely developed for adults and were just subsequently applied for children. We here calculated and provide reference ranges for indices of fasting glucose-insulin metabolism that comprise a wide range of the human life span from 5 to 80 years of age in a population-based cohort.

We found similar reference ranges for FI, FG and HOMA-IR, when comparing our findings to the results from similar but smaller cohorts, that did not cover the whole life span.[39,40] For the HOMA2-IR and the McAuley index, no reference values have been published for children and adolescents yet and published cut-offs among adults do not take age-related dynamics into account.[41] Even though we saw slight differences according to sex in this large sample size, these differences were not considered to be clinically relevant after testing the need for splitting the reference groups by sex as recommended by the CLSI guideline.[42] Hence, we provide the same reference values for male and female individuals.

Of course in clinical practice, the application of age-dependent thresholds as opposed to simple standard cut-offs needs to be finely balanced. However, we advocate that the value in patient risk assessment may go beyond the established practice of simply using identical reference ranges across all age groups. Refinements in reference ranges are of particular importance for children and adolescents at risk, who are in greatest need for early identification and treatment. In fact, our data show that of those children who eventually

developed dysglycemia, 5 out of 6 would have been detected with our new age-related and insulin-based fasting thresholds as opposed to 1 out of 6 with the conventional FG and HbA1c based thresholds.

Prevalence of impaired glucose-insulin-metabolism in children with obesity and prediction of future dysglycemia

Applying these newly established reference ranges, we found insulin resistance as a very early alteration associated with obesity, which manifested already in more than one third of preschool children with obesity. This is especially striking, as insulin resistance is unlikely to spontanously recover over time. Insulin resistance more than doubled the risk for future prediabetes independent of age, sex, BMI-SDS and also of family history of diabetes, even though an important genetic predisposition for is well acknowledged.[43] It will be important to detect those children at high risk, as with rigorous intervention there may be a window of opportunity to relevantly postpone the manifestation of diabetes.[5,44]

It is an ongoing debate which percentile to apply for the definition of insulin resistance. Recommendations in the literature vary from the 66th to the 97.5th percentile.[14,15] In this study, based on follow-up data from children and adolescents with obesity, we determined the 90th percentile as the best cut-off in order to discriminate risk vs. non-risk groups for those indices with the best predictive performance for emerging dysglycemia in children and adolescents (HOMA-IR and FI). It is interesting and important, however, that FI, HOMA-IR, HOMA2-IR and McAuley index all performed better in predicting emerging type 2 diabetes than FG, which is however the only parameter of the ones mentioned above that is recommended in guidelines. However, there is a study from 2010 stating that children with FG-levels above 86 mg/dl (4.8 mmol/l) are 3.40 times more prone to develop pre-diabetes and 2.06 times more prone to develop diabetes in adulthood.[45] This cut-off is equal to the 50th percentile in our reference values, although we did not find significant prediction of future dysglycemia with this threshold in our cohort.

Insulin resistance in childhood and adolescence according to these parameters had a high likelihood to be reconfirmed, while pathological results of conventional glycemia markers (FG, 2 h glucose, HbA1c) were less likely to be reproduced within two years follow-up, which is in line with previous findings.[46] The risk of sustained insulin resistance was especially high among pubertal adolescents with obesity (72%) and therefore pathological values during puberty should not be dismissed as a physiological change among adolescents with obesity in the clinical setting.

Our results and the new reference values may change the diagnostic approach for patients with obesity in that we advocate for age-specific and hence more precise thresholds and particularly the use of insulin based parameters. More than one third of patients are at metabolic risk based on insulin derived parameters outside cut-offs. According to our prediction models, one third is likely to develop dysglycemia within 10 years. Many of these patients (five out of six) would not have been detected with classical parameters FG and HbA1c, whereas two thirds could be detected early with the newly established cut-offs of insulin-based indices.

Strengths and weaknesses of the study

This is the first study to our knowledge to systematically examine the pattern of glucose-insulin metabolism over nearly the entire life span in a large population-based cohort with almost 7000 observations. Furthermore, we tested the clinical relevance of the newly established reference ranges in longitudinal data from children and adolescents with obesity. However, our longitudinal analyses were limited by lower sample size in the young-adult population. Also, we had to apply prediabetes criteria as a proxy for dysglycemia, since overt type 2 diabetes occurs rarely in childhood. Furthermore, it should be noticed that our reference cohort was recruited from the Caucasian, mainly urban, population living around Leipzig. Thus our results might not entirely reflect other populations. We addressed this limitation by reconfirming the age-related dynamics in independent cohorts over the entire age range (Lifelines) and in adults (Sorbian cohort). Nevertheless, considering the limitation in generalizability (e.g., due to ethnic differences, analytical procedures, etc.) we advocate for national references to apply, as for example it is standard for anthropometric measures.

For the obesity cohort, we had an overrepresentation of children with strong obesity by strategy, due to the inclusion of our Leipzig Childhood Obesity—cohort, which might explain the slightly higher glucose as compared to the Lifelines cohort. The performance of the new cutoffs for prediction of future dysglycemia were, however, confirmed in an independent cohort based on the German national childhood obesity registry.

We employed a BMI-based stratification for the normal-weight vs. obesity groups. However, the BMI is limited in assessing body composition and other parameters (e.g., waist-to-hip ratio or waist-to-hight ratio) may predict future impairment of glucose-insulin-metabolism in children better compared to a BMI-based classification[34] with similar diagnostic value correlated to body fat percentage in DXA-scans.[47] However, comparing BMI- and waist-to-height- based stratification did not result in relevant differences in the prevalence of metabolic parameters outside the reference ranges. Therefore, and as in practice the BMI is more widely used and accessible with valid reference ranges, we based our definition of obesity on BMI and BMI-SDS values.

In summary, the variation of fasting indices of glucose-insulin-metabolism across the life span underlines the need for age-specific reference ranges to reliably identify patients at risk for dysglycemia. We herein provide (freely accessible) reference values for subjects from 5 to 80 years. According to these cut-offs, there is a high prevalence of insulin resistance (>40%) in children with obesity, starting as young as preschool age. Insulin-based indices are superior to currently recommended FG in predicting future dysglycemia (Fig. 4). Particularly with respect to the increasing recognition of (pre)diabetes subtypes[6,8] this approach of more refined cut-offs of parameters, extending beyond the classically recommended ones, may help to better identify and classify patient's risk characteristics and stratify them for precision prevention and intervention approaches.

Contributors

CH and RS designed the study, analysed and interpreted the data and drafted the manuscript. CH and RS have directly accessed and verified the underlying data reported in the manuscript. JK contributed to data acquisition, study design and revised the manuscript. MV was involved in conception of the study, data analysis, and revised the manuscript. MC interpreted the data and revised the manuscript. CM contributed to study design and revised the manuscript. RB and KW were involved in study design, data acquisition and revised the manuscript. JS, US and AS contributed to data acquisition and revised the manuscript. MS and MB were involved in study conception and revised the manuscript. WK acquired funding, was involved in study design and revised the manuscript. AK designed the study, acquired funding, interpreted the results and revised the manuscript. For the APV initiative, AJE analysed the data and helped interpreting the data and revised the manuscript, RWH is the scientific coordinator of the APV registry, helped with data interpretation and revised the manuscript. HS was involved in the study design and interpretation of Lifelines cohort results. RDT was involved in data analysis and interpretation of Lifelines cohort results. AT helped with interpreting the data for the Sorbian cohort and revised the manuscript. All authors approved the final version of the manuscript and agreed to be accountable for all aspects of the work.

Data sharing statement

The novel reference values are available open-access at the Ped(Z)Pediatric Calculator app[32] and via an R package.[33] The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request. More information about how to request Lifelines data and the conditions of use can be found on their website (https://www.lifelines.nl/researcher/how-to-apply). For the APV data, remote data access is possible on reasonable request to the APV scientific board.

Declaration of interests

MB received honoraria and consulting fees from Amgen, AstraZeneca, Bayer, Boehringer-Ingelheim, Lilly, Novo Nordisk, Novartis and Sanofi and is part of the advisory board from Boehringer-Ingelheim. All other authors have no conflicts of interest relevant to this article to disclose.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at https://doi.org/10.1016/j.lanepe.2023.100652.

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