Utrecht University

Gene-Environment Interactions and the Effect on Obesity Risk in LMICs: A Systematic Review

Sophia L. Pledger

Assistant Professor Dr Fariba Ahmadizar (Daily Supervisor)

Assistant Professor Dr Patrick Souverein (Second Reviewer)

Department of Epidemiology and Global Health, Julius Centre for Health Sciences and Primary Care, University Medical Center Utrecht, Utrecht University

LAYMAN'S SUMMARY

Obesity is a term used to describe people with high body mass index (BMI) and an excess of body fat that affects their health. It is a disease that increases the chance of developing other diseases, such as heart disease or certain cancers. Obesity is a problem worldwide, however, currently, the proportion of people classified as obese is increasing quicker in low- and middle-income countries (LMICs) compared to high-income countries (HICs). Obesity is also considered an inherited condition, with genetic information playing a big role in how likely people develop it. Since 2008, a type of genetic research known as genome-wide association studies (GWAS) has been used to identify specific genes associated with obesity. These genes, also called single nucleotide polymorphisms (SNPs), vary between individual people and populations. More recent research has shown that different SNPs can interact with a person's environment or lifestyle choices, such as their diet or levels of exercise, to either increase or decrease their risk of obesity. Like GWAS, studies investigating gene-environment interactions (GxE), have mainly been performed in HICs among European populations. However, findings from GxE research in HICs cannot be assumed to be the same in LMICs. This is because people living in LMICs have different ethnic backgrounds with different genetic information, and live in different environments with different lifestyles. At present, research analysing GxE on obesity in LMICs is limited but growing. This review discusses what research is available, and brings to light areas for improvement for future scientific studies. Three different databases, which contain millions of scientific research articles, were searched using combinations of key words such as 'obesity', 'diet' 'LMIC' and 'SNP'. Search results up until November 2022 were checked and irrelevant research was excluded. From these results, eighteen research papers were considered relevant to this review. The risk of bias was assessed for all included studies, which is the risk that a scientific study may have presented misleading results due to how they designed the study or collected scientific data. Overall, 14 different SNPs were found to significantly interact with different lifestyle factors to change the likelihood of an individual being obese. However, significant interactions

were not repeated across different research studies or in different populations. This review also highlighted several problems with the way current scientific studies collected lifestyle information, or how they defined or classified obesity using varied BMI cut-off values. Statistical analyses were also outdated, relied on information found in old European-based GWAS, or were weakened by small numbers of research participants. Future scientific studies in LMICs should therefore focus on improving the quality of their research, by using standardised data-collection methods, updated statistical techniques, and information from GWAS performed in populations of the same ethnicity.

ABSTRACT

BACKGROUND: Obesity represents a major and preventable global health challenge as a complex disease in itself, and a modifiable risk factor for the development of other non-communicable diseases. In recent years, obesity prevalence has risen more rapidly in low- and middle-income countries (LMICs) compared to high-income countries (HICs). Obesity traits are shown to be modulated by an interplay of genetic and environmental factors such as unhealthy diet and physical inactivity in studies from HICs focused on populations of European descent, however genetic heterogeneity and environmental differences prevent the generalisation of study results to LMICs. Primary research investigating gene-environment interactions (GxE) on obesity in LMICs is limited but expanding. Synthesis of current research would provide an overview of the interactions between genetic variants and environmental factors that underlie the obesity epidemic, and identify knowledge gaps for future studies.

METHODS: Three databases were searched systematically using a combination of key words such as "genetic risk", "obesity", "LMIC", "diet" and "physical activity", to find all relevant observational studies published prior to November 2022. Risk of bias was assessed for all included studies.

RESULTS: Eighteen of the 1,373 articles met the inclusion criteria, of which one was a GWAS, thirteen used a candidate gene approach and five were assigned as genetic risk score studies. Six studies were considered to be of high quality, while twelve studies were of moderate quality. Statistically significant findings were true for a total of 14 individual SNPs across 10 different genetic loci, however most studies were of small scale and without replication.

CONCLUSIONS: Although results suggest significant GxE interactions on obesity in LMICs, updated robust statistical techniques with more precise and standardised exposure and outcome measurements are necessary for translatable results. Future research should focus on replication efforts with improved quality, with emphasis on large-scale, long-term longitudinal study designs using multi-ethnic GWAS.

INTRODUCTION

Obesity is a major public health concern worldwide, with 39% of adults over 18 classified as overweight, and 13% obese according to the most recent global estimates by the Word Health Organisation (WHO) (1). Previously considered a disease primarily affecting high-income counties (HICs), obesity is rapidly rising in developing countries with emerging economies. These low- and middle-income countries (LMICs) are now home to 62% of the world's overweight or obese population (2), and make up the top 10 countries with the largest average annual increase in obesity prevalence worldwide (3). In recent decades, LMICs have been faced with an epidemiological transition, characterised by a shift in the main drivers of mortality and morbidity from communicable diseases to non-communicable diseases (NCDs), such as cardiovascular disease, type 2 diabetes and cancer. As both a major metabolic risk factor for NCD development, and a disease by itself, obesity represents a significant epidemiological burden (4). While the prevention and treatment of obesity is a major target within global health systems, it poses significant challenges thanks to its complexity as a disease, and the contribution of a multifaceted interplay of variables which underwrite its development.

Obesity, defined by the WHO as a body mass index (BMI) of ≥ 30 kg/m^2, is determined by a long-term positive imbalance in energy intake versus energy expenditure, driven by an unhealthy diet and reduced physical activity (1,5). Changes in global trade, dietary patterns, and declining physical activity have exposed people living in developing countries to increasingly obesogenic environments (2). The nutrition transition faced by LMICs has been a major contributor to the obesity epidemic, characterised by a shift from traditional dietary habits to increased consumption of energy-dense, nutrient-poor ultra-processed foods and beverages (6). Rapid urbanisation and within-country rural-to-urban migration have also led to a decline in manual labour and active transportation, and an increase in sedentary behaviours (7,8). Genetics has also been shown to play a strong role in an individual's susceptibility to obesity, with obesity heritability estimated to be between 40-70% (9). The advent of genome-wide association studies

(GWAS) accelerated the discovery of obesity-related genetic loci and causative single nucleotide polymorphisms (SNPs), but focussed on adult populations of European ancestry (10,11). The fat mass and obesity-associated (*FTO*) locus was the first obesity-related GWAS-identified locus, and remains the most highly significant and robustly replicated (11,12). The landmark 2007 European study initially found that per risk allele in the *FTO* SNP rs9939609, there was a 1.32-fold increased odds of obesity (13). The effect of *FTO* SNPs on obesity risk, and the prevalence of *FTO* risk alleles has since been shown to vary across different ethnic populations (14), with the risk of obesity per risk allele increasing 1.25-fold in Asians, and 1.15 in Indians (15,16). Since the discovery of *FTO*, a further 1,100 independent genome-wide significant loci have been identified, however, these combined explain only 6% of inter-individual obesity variation (17).

As both a modifiable risk factor and a complex multifactorial condition, obesity results from an interplay of genetic, lifestyle and environmental factors (12). In parallel with GWAS, the number of studies analysing gene-environment interactions (GxE) on obesity risk has increased exponentially over the last decade, however these have focussed primarily on European populations living in resource-rich settings (18). Recent research from HICs has presented evidence that an individual's genetic susceptibility to obesity can be magnified or mitigated in response to environmental factors, such as physical activity (19), alcohol consumption (20), smoking (21), diet (22) and sleep (23). Generalisability of findings from these studies to developing countries is restricted due to the genetic heterogeneity found in different populations and ethnic groups, in addition to differences in the obesogenic environmental exposure (24). So far, observational research focussing on populations from LMIC's is limited but expanding. Synthesis of population-based studies specifically investigating GxE on obesity in LMIC's could provide a more comprehensive understanding of this cause-effect relationship in varied ethnic groups and potentially translate into tailored region-specific lifestyle intervention strategies to combat the global obesity

epidemic. To our knowledge, this will be the first systematic review to examine and discuss population-based studies from LMIC's investigating gene-lifestyle interactions and their effect on obesity.

METHODS

This study was conducted according to the Transparent Reporting of Systematic Reviews and Meta-analyses (PRISMA) guidelines.

ELEGIBILITY CRITERIA

The study inclusion and exclusion criteria for this review were specified using the PECOS elements, as defined in **Table 1**. All observational research articles investigating GxE on obesity risk in adult human populations from LMICs were included. Articles were also excluded if they focussed on specific populations, e.g. only women, or participants with comorbidities such as cancer, cardiovascular or renal disease to reduce concerns for disease labelling bias.

SEARCH STRATEGY

Three electronic databases PubMed, EMBASE and Scopus were systematically searched on October 24th 2022 by one investigator. To identify articles relevant to the research question addressed by this review, the search strategy was subdivided into three main groupings: populations in LMICs, gene-environment interactions, and weight-related outcomes. To define LMICs, articles were searched by title and abstract for key words such as 'low middle income', 'developing country' and 'low resource'. To further increase capture, the names of countries and geographical areas were included in the search syntax according to the World Bank 2022 country classifications (25). To include gene-environment interactions, key words

were used such as 'gene-lifestyle' or 'GxE' or terms relating to genetic susceptibility such as 'polygenic risk score', 'single nucleotide polymorphism' and 'epigenetic' in combination with environmental exposure such as 'diet' or 'physical activity'. Obesity and all other weight-related anthropometric measurements such as 'BMI', 'waist circumference', 'body fat percentage', and 'waist-hip ratio' were included in the final group. Animal, paediatric and intervention studies were excluded. No filters based on language or publication date were applied. Details of the search strategies developed for each specific database can be found in **Supplementary Table 1**.

DATA COLLECTION AND EXTRACTION

Titles and abstracts of all articles identified via database searches were screened by one investigator based on the eligibility criteria previously detailed using Endnote (v20.4.1) (26). Full text articles were assessed using Rayyan, with all reasons for exclusion documented (27). Data extraction was also performed by one investigator using a standardised form with software programme Microsoft Excel 2016. For each included study, the following information was extracted:

- First author, year of study, year of publication, country of coverage, study objectives, and study design.

- Sample size, distribution of study population characteristics (e.g. BMI, age, gender), obesity definition, environmental or lifestyle exposure, genetic exposure (e.g. gene or SNP of interest) and type of genetic analysis.

- Primary and secondary results (e.g. β coefficient or odds ratio where possible) and overall conclusion.

RISK OF BIAS AND QUALITY ASSESSMENT

Risk of bias was assessed by one investigator using the Newcastle-Ottawa-Scale (NOS) for case-control and cohort studies. Studies were judged based on three categories: selection of study participants, comparability of participants, and the assessment of the exposure or outcome for case-control or cohort studies respectively. Studies could be awarded a maximum of four stars for the selection category, two stars for comparability and three stars for exposure. For cross-sectional studies, an adapted version of the NOS was used (28), where a maximum of five stars could be awarded for the selection category, two for comparability and 3 for outcome. A quality threshold score was used to summarise overall study quality, with >7 points indicating high quality, 5-7 fair quality and ≥4 points poor quality.

RESULTS

A total of 1,373 articles were identified, of which 744 were from PubMed, 133 from Embase and 499 from Scopus. After removing 101 duplicate studies, and 3 ineligible studies, titles and abstracts of 1,269 articles were screened and a further 1,233 irrelevant studies were excluded. Full texts were reviewed for 36 articles, of which 18 were excluded. Of these, 11 reported on populations which did not meet the inclusion or exclusion criteria, 4 did not include an environmental interaction, 1 reported an irrelevant outcome, and 1 incorporated an interventional design. Two studies by the same authors reported duplicate populations, studies and outcomes but differed by cross-sectional versus longitudinal analyses. The cross-sectional study was excluded to ensure the strength of evidence was not overestimated, and the most recent longitudinal study was included. In total, 18 studies met the PECOS criteria and were included in this systematic review. **Figure 1** shows the PRISMA flow chart for the selection of studies.

CHARACTERISTICS OF INCLUDED STUDIES

Populations and study designs

A summary of the key features of the 19 included studies is presented in **Table 2**. In short, five studies were conducted in South Asia (Pakistan, Sri Lanka and India), three studies in South-East Asia (Indonesia), four studies in West Asia (Iran and Turkey), five studies in China, and one study in Ghana. Studies from China were assessed to ensure that study populations did not focus on very high-income regions. Across all studies, ages ranged between 18 – 90 years, and sample sizes ranged from 71 – 14,131 participants. Publication time ranged from 2012-2022. The majority of studies were cross-sectional (n = 9) or case-control (n = 8) by design, with only two cohort studies examining longitudinal associations.

Gene-environment exposures

Four articles focussed on a variety of environmental or lifestyle factors and their genetic interactions on the risk of obesity. Five studies assessed dietary interactions only, including dietary components and dietary patterns, three studies investigated the effects of physical activity and sedentary behaviours, three studies looked at smoking and drinking statuses, two studies evaluated sleep patterns and two studies assessed urban-rural differences and effects of within-country migration. With respect to genetic exposures, there was only one GWAS included in this review. Five studies assessed genetic risk through a genetic risk score (GRS), with the number of included SNPs ranging from 2 to 9. Only two studies assigned weights through the use of an external independent study or genome-wide meta-analyses, whilst the other three used an unweighted approach. Thirteen studies used a candidate gene approach to investigate 62 different SNPs, with risk alleles in genetic variants of the *FTO* and melanocortin 4 receptor (*MC4R*) gene most commonly studied. For only two studies, SNPs selection was based on recent GWAS conducted in the same ancestral population as the study participants, while the rest either relied on GWAS of European ancestry or failed to justify.

Obesity outcomes

Anthropometric indices for obesity outcomes included (change in) weight, BMI, waist circumference (WC), hip circumference (HC), waist-hip ratio (WHR), waist-to-height ratio (WHtR), fat mass index (FMI), fat-free mass index (FFMI), and percentage body fat (%BF). Population measures were also examined, including odds of being overweight. For all included studies, obesity outcomes were objectively measured by a healthcare provider or study investigators using validated consistent devices. Definitions of general obesity ranged across the studies, countries and populations from BMI $\geq$ 25 kg/m^2 to BMI $\geq$ 30 kg/m^2.

STUDY QUALITY ASSESSMENT AND RISK OF BIAS

The results from the NOS for observational studies and the adapted NOS for cross-sectional studies risk of bias assessments for the included studies are shown in **Table 3** and **Table 4,** respectively. Scores for case-control and cohort studies ranged from 5 – 8, and scores for cross-sectional studies ranged from 6 – 10. All studies included in the review were assessed as having either a low or fair risk of bias. The most common score-reducing traits were from the selection domain, due to either small or unjustified sample sizes, or due to the potential for bias from the participant selection procedures. Studies generally scored well for comparability, with only one failing to control for age and sex in the statistical analysis. Obesity-related outcome measurements were assessed objectively by trained researchers using standardised equipment for all studies. Lifestyle and environmental exposures were collected either via participant self-report or face-to-face interviews, using a combination of standardised externally validated questionnaires or short self-developed questionnaires.

THEMATIC RESULTS

Diet and food timing

Studies investigating gene-diet interactions on obesity, focus on macronutrient intake including total fat, protein and carbohydrates, and fatty acid intake including saturated (SAFA), monounsaturated (MUFA) and polyunsaturated (PUFA) fatty acids were most prevalent. Within these studies Al-Jawadi et al. (n = 71), Alsulami et al. (n = 302), Daya et al (n = 80). and Wuni et al. (n = 497) all reported significant associations between higher total fat intake and obesity traits, for those carrying risk alleles of obesity-related gene variants, or for those with a high GRSs in Indonesian, Ghanian and Indian populations (29–32). High SFA specifically was also found to interact positively with WC in those with increased genetic

susceptibility to obesity in the cross-sectional studies by Alsulami et al. and Wuni et al. Individuals with high GRS (≥ 2 risk alleles) and high SFA intake (>14 g/day) had a significantly higher WC ($P_{interaction}$ = 0.02) compared to those with low SFA intake after adjustment for age and sex in the Ghanian study, while in an Indian population, those with lower SFA intake (≤23.2 g/day) had a significantly smaller WC (β= -0.01cm, P=0.03) ($P_{interaction}$= 0.006) after adjustment for age, sex and 6 other potential confounders (30,32). However, some inconsistencies were also reported on the modifying effects of dietary fat intake on genetic susceptibility and obesity risk. In the case-control study by Isgin-Atici et al. (n = 400), the same *FTO* SNP rs9939609 variant as the Indonesian case-control study by Daya et al. was investigated both individually and as part of a GRS with one other *FTO* gene variant. Contrastingly, no statistically significant association was found between dietary fat intake on obesity traits in Turkish populations (33). Findings in relation to protein dietary intake and its interaction with *FTO* gene variants on obesity measures were also conflicting in Turkish and Indonesian populations. Carriers of *FTO* risk alleles in the study by Isgin-Atici et al. showed a significant interaction with protein intake on increased WC ($P_{interaction}$ = 0.044) after adjustment for age, sex hypertension and CVD. In contrast, *FTO* risk allele carriers showed no significant interaction for protein intake on obesity in findings by Al-Jawadi et al. after adjustment for age and sex (29,33).

With regards to dietary patterns, two studies examining eating patterns in two separate Iranian populations reported modifying effects in individuals with either high GRSs or those carrying risk alleles of the Cocaine and Amphetamine-Regulated Transcript Prepropeptide (*CARTPT*) gene and their association with obesity-related anthropometric measures. In a cohort study, Hosseini-Esfahani et al. (n = 4292) showed that higher Western dietary pattern scores (namely high intakes of processed foods and drinks, sugar, red meat and high-fat dairy) were associated with increased BMI in subjects with high GRS compared to those with low GRS over time (2.26 ± 0.36, $P_{interaction}$= 0.01) after multivariable adjustment

for age, sex and 5 other confounders (34). In a cross-sectional study, Mahmoudi-Nezhad et al. (n = 287) used the Diet Quality Index—International (DQI-I), an indicator of nutritional variety, moderation and adequacy to show that in individuals with high-scoring quality diets, *CARPT*-DQI-I interactions significantly reduced BMI ($P_{interaction} < 0.001$) following adjustment for age and sex (35). In both studies, however, analyses assessing healthy eating patterns rich in fruits, vegetables, fish and whole grains, quantified by the Healthy Eating Index, showed no significant modifying effects by genotypic groups or GRS for obesity traits.

Significant interactions between food timing and genetic variants on obesity were also demonstrated in both Iranian and Pakistani populations. Rahati et al. (n = 403) reported in a cross-sectional study, that for carriers of Circadian Locomotor Output Cycles Kaput (*CLOCK*) gene polymorphisms, delayed eating times for breakfast and lunch increased the odds of obesity by 2.95 (95% CI = 1.77, 4.90) and 1.53 (95% CI = 1.32, 1.89) respectively (*P*<0.05) after adjustment for age, sex and 6 other confounders (36). Significant interactions between risk alleles in multiple genes including *FTO, MC4R*, and transmembrane protein 18 (*TMEM18)* and random eating patterns were also found to increase BMI (*P*=0.002, *P* = 0.008, *P*=0.001 respectively) in the case-control study by Rana et al. (n = 578) focussing on a Pakistani population after age and sex adjustment (37).

Physical activity and sedentary behaviours

A total of 10 studies using either a candidate gene approach (n = 6) or GRSs (n = 4), reported gene-physical activity (PA) interactions on obesity traits. PA or sedentary behaviour were defined via participant self-reporting in studies by Xue et al., Rana et al., Moore et al., Isgin-Atici et al., and Gong et al., (33,37–40) while articles by Sun et al., Muhammed et al., Alsulami et al., and Ahmad et al., used investigator-administered questionnaires (30,41–43). The standardised international physical activity questionnaire

was most commonly used to assess PA levels across studies (n = 4), which assesses levels of PA relating to work and house-related work, transportation and recreation, calculated and summarised as metabolic equivalent of task units per week (MET-min/wk).

Four of the candidate gene studies showed that interactions between 4 different gene variants, and low levels of PA were significantly associated with obesity-related anthropometrics (33,37,39,42). The study by Moore et al. (n = 1129) used a cross-sectional design to show that in India, for participants with a low PA level of <81 MET-h/wk, the *FTO* s3751812 risk allele was significantly associated with an increased WC (β = 2.86; 95% CI = 1.24, 4.12) after controlling for age, sex, region and religion (39). The same association for variants of the *FTO* candidate gene was also found in 2 other case-control studies, which focused on Turkish (n = 400) and Pakistani populations (n = 578)(33,37). High levels of PA however, defined as >212 MET-h/wk did not produce any significant gene interaction effect on obesity (39). In conflict, there were no significant interactions between putative uncharacterized protein (*FLJ33534*), uncoupling protein 2 (*UCP2*), or olfactory pathway-related candidate genes and PA, or their interaction on obesity-related traits (41–43). The study by Alsulami et al. which used a GRS, comprised partly of *FTO* gene variants, also failed to show any significant interaction between high genetic risk and PA on obesity (30) in a Ghanian population. Two cross-sectional studies specifically investigated sedentary behaviours and their potentially modifying effects in two Chinese populations. Gong et al. (n = 2216) and Xue et al. (n = 3976) consistently showed that increased leisure time sedentary behaviours such as television watching positively accentuated the interaction between high GRS or SNPs and WC and BMI after multivariable adjustment (38,40).

Tobacco smoking and alcohol consumption

Across the studies included in this review, very few investigated GxE assessing the effect of tobacco smoking (n = 3) or alcohol consumption (n = 2) on obesity. In these candidate gene studies, tobacco and alcohol exposures were assessed by a self-administered questionnaire by Wei et al., while Ahmad et al. and Sun et al. collected data using researcher administered validated questionnaires in the form of a structured interview (41,43,44). Both Ahmad et al. (n = 8,193) and Sun et al. (n = 608) using cross-sectional and case-control study designs respectively, showed that for current smokers, the interaction between smoking status and obesity was modified by different gene variants (41,43). In the current smokers from Pakistan, *FLJ33534* risk alleles showed a negative association with BMI ($\beta=-1.51 \pm 0.52$, *P*=0.003) after adjustment for age, sex and genetic ancestry (43). While the Chinese population showed smoking increased the risk of obesity for those with high olfactory receptor family 4 subfamily D member 1 (*OR4D1*) gene scores (OR = 2.67; 95% CI = 1.35, 5.30; *P*=0.005), but decreased the risk of obesity for those with high calmodulin like 3 (*CALML3*) gene scores (OR = 0.25; 95% CI = 0.10, 0.62; *P*=0.003) after adjustment for age, sex, PA and alcohol consumption (41). A separate case-control study in China by Wei et al. (n = 1,836) however showed no significant gene-smoking or gene-alcohol interaction on obesity risk for *MC4R* genotypes (44). Alcohol consumption was also consistently disproved to show any modifying effect on the relationship between gene variants and obesity by Sun et al. (41).

Sleeping patterns

Only two candidate gene studies performed gene-sleep interaction analyses on obesity traits, both of which estimated sleeping patterns and sleep duration using participant self-reporting via study-specific questionnaires. Both studies demonstrated unfavourable outcomes on obesity traits in response to the interaction between genetic variants and reduced sleeping times. In a Pakistani population (n = 578), Rana

et al. applied a case-control study design to demonstrate a significant interaction between *TMEM18,* neuronal growth regulator 1 (*NEGR1*)*, FTO* and *MC4R* gene variants and irregular sleep wake cycle, which was shown to augment BMI, WC, HC, WHR, WHtR and %BF, in carriers of risk alleles after controlling for age and sex (37). In the same study, inadequate sleep, defined as <7 hours/night, was also shown to interact with *FTO, TMEM18* and *NEGR1* gene variants to significantly increase BMI and WC. In a separate cross-sectional study by Rahati et al. the interaction between sleep duration (hours/week) and *CLOCK* rs1801260 genotypes were also assessed in an Iranian population (n = 403), where obese individuals with the CT + CC genotypes had a significantly shorter sleeping time than TT genotype carriers after controlling for age, sex and 6 other variables (36).

Rural-urban differences

Two studies investigating the moderating effects of urban and rural living environments found disparity in their effects on obesity and their interactions with *MC4R* candidate genes in Chinese and Sri Lankan populations. Information pertaining to sociodemographic information and lifestyle factors was collected through face-to-face interviews, or participant self-report via standardised questionnaires by Wang et al. and Illangasekera et al. respectively. The *MCR4* rs17782313 CC + CT genotype was cross-sectionally associated with significantly higher BMI values in Sri Lankans (*P*= 0.03) (n = 528) compared to the TT genotype, a result which was replicated in the Chinese case-control study by Wang et al. (n = 965), which demonstrated significantly higher odds of obesity (OR = 3.01; 95% CI = 1.49, 6.05) for homozygous C allele carriers (45,46). However, on the performance of stratified analysis by urban or rural residence and the interaction with the *MC4R* gene polymorphism on obesity, only the study by Wang et al. found a statistically significant heterogenous association between the two living environments, with an attributable proportion of 0.65 (95% CI = 0.22, 1.17) after controlling for age, sex and 7 other potential

confounders (46). In contrast, Illangasekera et al. showed the *MC4R* non-variant TT carriers of urban residence to record higher mean BMIs (45).

DISCUSSION

To our knowledge, this is the first systematic review to provide an overview of current literature investigating the effects of gene-environment interactions on obesity traits in LMICs. Of the 18 studies (n=26,684) included, approximately two-thirds explored gene-diet or gene-PA interactions. In contrast, more limited numbers explored other emerging obesogenic environmental risk factors, such as urbanicity, irregular or insufficient sleep, and tobacco and alcohol use. Results from this study indicate there may be some consistent associations across developing countries, for interactions between genetic variants and reduced sleeping times, urban living environments, low levels of PA, increased sedentary behaviour and delayed eating patterns on obesity outcomes. However, due to considerable heterogeneity between study outcome definitions, genetic polymorphisms and environmental and lifestyle factors, in combination with the genetic heterogeneity across different ethnic groups in LMICs, genotype-phenotype cross-correlations should be interpreted with caution.

While results from nutrigenetic studies, and studies investigating gene-physical activity interactions on obesity suggest there could be some consistent associations between eating patterns or low levels of physical activity and their genetic interactions on an increased risk of obesity, associations with specific genes variants were not replicated across studies. Across all included studies, statistically significant findings were true for 14 individual SNPs across 10 different genetic loci; *FLJ33544* rs140133294 (43), *FTO* rs1421085 (29,37), rs9939609 (31,45), rs10163409 (33), rs3751812 (39), *CARTPT* rs2239670 (35), *UCP2* rs659366 (42), *CLOCK* rs1801260 (36), *MC4R* rs17782313 (37,45,46), rs12970134 (40,46), *TMEM18* rs7561317 and *NEGR1* rs2815752 (37). However, these associations were either only significant in single trials, or were not replicated in response to the same environmental exposure. Trials which did assess interactions between the same *FTO* genetic variants and macronutrients, including total fat and protein intake on obesity were also conflicting across different LMICs and populations (29,31,33).

Explanations for these inconsistent results could be accounted for in the substantial heterogeneity in exposure and outcome data collection methods, alongside wide-ranging obesity definitions in the included studies. Obesity and overweight were often not differentiated, and using BMI values only as a screening tool for obesity was common, with cut-offs varying from ≥ 25 kg/m^2 to ≥ 30 kg/m^2. Further heterogeneity can be evidenced by the additional incorporation of WC measurements into obesity definitions by some studies, as a measurement of central obesity. Regional and ethnic differences in anthropometry and adiposity prevent the use of standardised global obesity definitions, as evidenced by WHO guidelines, which define obesity as 2.5 kg/m^2 lower in Asian populations compared to the global standard (2,47). However, this review has demonstrated disparity in the use of obesity definitions even amongst populations from the same developing countries, signalling the need for more consistent application of recommended definitions. Assessment and definitions of exposures were also conflicting across studies, and at high risk of recall or reporting bias due to incomplete or ambiguous recording of methods, and use of short self-developed questionnaires and surveys for patient self-report. Confounding was however controlled reasonably well across studies, with age and sex-adjusted for by all researchers as a minimum.

There is a high prevalence of statistically 'significant' GxE on obesity on single genetic variants or environmental exposures without replication. However, even with replication, many of the study designs shown in this review are susceptible to reverse causation, highlighting the need for more well-controlled long term prospective longitudinal research looking at GxE on obesity. Use of *P*-values without effect sizes or confidence intervals to report associations was also prevalent across studies (32–35,42), alongside small sample sizes and erroneous underpowered statistical analyses, resulting in concerns for selective reporting and publication bias.

From what is demonstrated in this review, emerging primary research from LMICs investigating GxE primarily employ a hypothesis-based approach, using pre-specified genes of interest identified in GWAS studies from European populations. The inconsistencies, and lack of replication across study findings could be partly attributed to use of genetic variants identified from GWAS conducted in developed regions with ancestrally homogenous populations. It is therefore likely that the inconsistency in study findings results from a lack of generalisability of the GWAS-identified candidate genes from developed nations, due to varied genetic architecture found in the diverse ethnic populations across LMICs (48). In addition, of the studies using a candidate gene approach, approximately half failed to correct for multiple testing when using multiple regression analysis to investigate several SNPs across different genetic loci (49). Of those which did control for Type I error, sample sizes were very small and consequentially underpowered to reliably detect GxE.

Only five of the studies included in this review used a GRS (30,32–34,38); which while eliminating the loss of statistical power attributable to correction for multiple testing, studies were still underpowered owing to insufficient sample sizes of 400, 497 and 302 for studies by Isgin-Atici et al., Wuni et al. and Alsulami et al. respectively (30,32,33,50). Moreover, only two studies by Xue et al. and Hosseini-Esfahani et al. assigned a weighted method, using external weights from an independent study of the same ancestral population (38), or a multiethnic GWAS meta-analysis respectively (34). Absence of suitable external weights for the studies using unweighted GRSs in Turkish, Indian and Ghanian populations further demonstrates the lack of global diversity in the existing genetic research (30,32,33). As an aggregation of multiple genetic variants, weighted GRS using meta-analysed external weights is considered the gold standard for this genetic approach and is a powerful and bourgeoning tool for identifying GxE (51). New statistical techniques which rely on internal information of effect size distributions could improve the accuracy of research in developing countries using GRS analyses where external genetic information is unavailable (52,53).

Application of more GWAS approaches in LMIC research would eliminate reliance on the strength of a priori evidence, and any previous associations drawn from homogenous populations of European decent, in turn producing quality genetic associations for future gene-interaction studies (54). Only one paper eligible for inclusion in this study used a GWAS approach in a Pakistani population to identify the *FLJ33534* obesogenic locus. This paper demonstrated a strong, high quality significant interaction between the identified genetic variant, smoking and a moderating effect on obesity (43). Transethnic GWAS studies for the estimation of improved GRSs could provide greater predictive power for future GxE studies in developing countries, and their inclusion in future research has been called for in previous reviews (18,55). However, with a recommended genome-wide significance threshold of $P = 5\times10^{-8}$, GWAS require very large sample sizes to reach an adequate statistical power (56). This presents a significant challenge in LMICs, where resources are often limited and participant recruitment can be challenging due to low engagement levels and distrust of the scientific community (48). Expansion of genetic studies in diverse populations is essential, and while an increase in research capacity from LMICs could eliminate the Eurocentric biases surrounding GWAS and GxE interactions, a more equitable and open sharing of technologies, statistical advancements and GWAS summary statistics in diverse populations is needed to improve the quality of future research and eliminate health disparities (57).

STUDY STRENGTHS AND LIMITATIONS

The strength of this systematic review is that it is the first to report on GxE on obesity traits in LMICs through a broad and exhaustive literature search, using rigorous and predetermined inclusion and exclusion criteria. Risk of bias was also assessed for each included study using standardised checklists. Evidence of this topic as a fast developing, and emerging field of research in developing countries can be substantiated by the number of included primary research conducted or published in the last two years. However, several limitations associated with this study should be highlighted. Despite nearly all included

studies reporting significant findings for GxE on obesity, the limited homogeneity among studied SNPs and outcome definitions severely restricted the synthesis and interpretation of results. In addition, no meta-analysis or quantitative data synthesis could be performed, owing to the considerable heterogeneity and sources of error surrounding the included primary research. Imprecise and diverse measurements of exposures and outcomes, alongside small sample sizes and underpowered or improper interaction analyses, which could have yielded false positive or false negative results, mitigate any value in a meta-analytic summary of effect sizes of GxE.

While all decisions regarding data extraction, screening and exclusion have been transparently documented using the Rayyan and Endnote software's, risk of bias cannot be assumed as this review was conducted individually as part of the Graduate School of Life Sciences writing assignment, and thus lacks an independent second reviewer with methodological expertise. It should also be noted that although the search strategy and study eligibility criteria did not exclude studies based on language, only English search terms were used in database searches. In addition, no non-English databases were included in the search strategy due to time and resource constraints. This could explain the lack of any representation of research from developing countries from Central and South America, and the limited representation of studies from developing African countries eligible for inclusion in this review, with only 3 irrelevant non-English studies identified in the initial database search.

CONCLUSION

This review has examined and discussed population-based studies from LMIC's investigating GxE and their effect on obesity, in addition to synthesising the achievements and pitfalls in the currently available primary research. Individual results have shown smoking status to modify the interaction between *FLJ33544* and olfactory pathway genetic loci and obesity traits. At the same time, urban living environments were demonstrated to interact with *MC4R* gene polymorphisms to increase obesity traits. However, the ability to draw concrete conclusions is limited due to concerns over study quality and a high potential for biases. The considerable heterogeneity exhibited across the investigated genetic variants, exposure and outcome measures, statistical analyses and reporting of data has highlighted a need for updated standardised protocols bespoke to LMICS, and advanced statistical techniques and data availability to improve the quality and comparability of future studies.

NONSTANDARD ABBREVIATIONS AND ACRONYMS

BMI; body mass index, CALML3; calmodulin like 3, CARTPT; cocaine and amphetamine-regulated transcript prepropeptide, CI; confidence interval, CLOCK; circadian locomotor output cycles kaput, DQI-I; diet quality index – international, FFMI; fat-free mass index, FLJ33534; putative uncharacterized protein, FMI; fat mass index, FTO; fat mass and obesity-associated, GRS; genetic risk score, GWAS; genome wide association study, GxE; gene-environment interactions, HC; hip circumference, HDL; high density lipoprotein, HIC; high income countries, LDL; low density lipoprotein, LMIC; low- and middle-income countries, MC4R; melanocortin 4 receptor, MET; metabolic equivalent of task, MUFA; monounsaturated fatty acids, NCD; non-communicable disease, NEGR1; neuronal growth regulator 1, NOS; Newcastle-Ottawa-Scale, OR; odds ratio, PA; physical activity, PUFA; polyunsaturated fatty acids, SES; socioeconomic status, SAFA; saturated fatty acid, SNP; single nucleotide polymorphism, TMEM18; transmembrane protein 18, UCP2; uncoupling protein 2, WC; waist circumference, WHR; waist-hip ratio, WHtR; waist-to-height ratio, %BF; percentage body fat.

REFERENCES

1.   Obesity and Overweight Fact Sheet. World Health Organisation. 2021.

2.   Ford ND, Patel SA, Narayan KMV. Obesity in Low- and Middle-Income Countries: Burden, Drivers, and Emerging Challenges. Annu Rev Public Health. 2017 Mar 20;38(1):145–64.

3.   Lobstein T, Brinsden H, Neveux M. World Obesity Atlas 2022. World Obesity Federation. London; 2022. p. 1–289.

4.   FAO, IFAD, UNICEF, WFP, WHO. The State of Food Security and Nutrition in the World 2019 - Safeguarding Against Economic Slowdowns and Downturns. Rome; 2019.

5.   Hill JO, Wyatt HR, Reed GW, Peters JC. Obesity and the Environment: Where Do We Go from Here? Science (80-). 2003 Feb 7;299(5608):853–5.

6.   Popkin BM, Ng SW. The nutrition transition to a stage of high obesity and noncommunicable disease prevalence dominated by ultra-processed foods is not inevitable. Obes Rev. 2022 Jan 10;23(1).

7.   Elshahat S, O'Rorke M, Adlakha D. Built environment correlates of physical activity in low- and middle-income countries: A systematic review. PLoS One. 2020 Mar 17;15(3):e0230454.

8.   Carrillo-Larco RM, Bernabé-Ortiz A, Pillay TD, Gilman RH, Sanchez JF, Poterico JA, et al. Obesity risk in rural, urban and rural-to-urban migrants: prospective results of the PERU MIGRANT study. Int J Obes. 2016 Jan 31;40(1):181–5.

9.   Elks CE, den Hoed M, Zhao JH, Sharp SJ, Wareham NJ, Loos RJF, et al. Variability in the Heritability of Body Mass Index: A Systematic Review and Meta-Regression. Front Endocrinol (Lausanne). 2012;3.

10.   Buniello A, MacArthur JAL, Cerezo M, Harris LW, Hayhurst J, Malangone C, et al. The NHGRI-EBI GWAS Catalog of published genome-wide association studies, targeted arrays and summary statistics 2019. Nucleic Acids Res. 2019 Jan 8;47(D1):D1005–12.

11.   Loos RJF, Yeo GSH. The genetics of obesity: from discovery to biology. Nat Rev Genet. 2022 Feb 23;23(2):120–33.

12.   Qi L, Cho YA. Gene-environment interaction and obesity. Nutr Rev. 2008 Nov 19;66(12):684–94.

13.   Frayling TM, Timpson NJ, Weedon MN, Zeggini E, Freathy RM, Lindgren CM, et al. A Common Variant in the FTO Gene Is Associated with Body Mass Index and Predisposes to Childhood and Adult Obesity. Science (80-). 2007 May 11;316(5826):889–94.

14.   Peng S, Zhu Y, Xu F, Ren X, Li X, Lai M. FTO gene polymorphisms and obesity risk: a meta-analysis. BMC Med. 2011 Dec 8;9(1):71.

15.   Li H, Kilpeläinen TO, Liu C, Zhu J, Liu Y, Hu C, et al. Association of genetic variation in FTO with risk of obesity and type 2 diabetes with data from 96,551 East and South Asians. Diabetologia. 2012 Apr 23;55(4):981–95.

16.   Vasan SK, Karpe F, Gu HF, Brismar K, Fall CH, Ingelsson E, et al. FTO genetic variants and risk of obesity and type 2 diabetes: A meta-analysis of 28,394 Indians. Obesity. 2014 Mar;22(3):964–70.

17.   Yengo L, Sidorenko J, Kemper KE, Zheng Z, Wood AR, Weedon MN, et al. Meta-analysis of genome-wide association studies for height and body mass index in ~700000 individuals of European ancestry. Hum Mol Genet. 2018 Oct 15;27(20):3641–9.

18.   San-Cristobal R, de Toro-Martín J, Vohl M-C. Appraisal of Gene-Environment Interactions in GWAS for Evidence-Based Precision Nutrition Implementation. Curr Nutr Rep. 2022 Aug 11;11(4):563–73.

19.   Ahmad S, Rukh G, Varga T V., Ali A, Kurbasic A, Shungin D, et al. Gene × Physical Activity Interactions in Obesity: Combined Analysis of 111,421 Individuals of European Ancestry. PLoS Genet. 2013 Jul 25;9(7):e1003607.

20.   Rask-Andersen M, Karlsson T, Ek WE, Johansson Å. Gene-environment interaction study for BMI reveals interactions between genetic factors and physical activity, alcohol consumption and

socioeconomic status. PLOS Genet. 2017 Sep 5;13(9):e1006977.

21.   Justice AE, Winkler TW, Feitosa MF, Graff M, Fisher VA, Young K, et al. Genome-wide meta-

analysis of 241,258 adults accounting for smoking behaviour identifies novel loci for obesity

traits. Nat Commun. 2017 Apr 26;8(1):14977.

22.   Wang T, Heianza Y, Sun D, Huang T, Ma W, Rimm EB, et al. Improving adherence to healthy

dietary patterns, genetic risk, and long term weight gain: gene-diet interaction analysis in two

prospective cohort studies. BMJ. 2018 Jan 10;j5644.

23.   Celis-Morales C, Lyall DM, Guo Y, Steell L, Llanas D, Ward J, et al. Sleep characteristics modify the

association of genetic predisposition with obesity and anthropometric measurements in 119,679

UK Biobank participants. Am J Clin Nutr. 2017 Apr;105(4):980–90.

24.   Vimaleswaran KS. A nutrigenetics approach to study the impact of genetic and lifestyle factors on

cardiometabolic traits in various ethnic groups: findings from the GeNuIne Collaboration. Proc

Nutr Soc. 2020 May 31;79(2):194–204.

25.   Hamadeh N, Van Rompaey C, Metreau E, Eapen SG. New World Bank country classifications by

income level: 2022-2023. World Bank Blogs. 2022.

26.   The EndNote Team. EndNote. Philadelphia, PA: Clarivate; 2013.

27.   Ouzzani M, Hammady H, Fedorowicz Z, Elmagarmid A. Rayyan—a web and mobile app for

systematic reviews. Syst Rev. 2016 Dec 5;5(1):210.

28.   Herzog R, Álvarez-Pasquin MJ, Díaz C, Del Barrio JL, Estrada JM, Gil Á. Are healthcare workers'

intentions to vaccinate related to their knowledge, beliefs and attitudes? a systematic review.

BMC Public Health. 2013 Dec 19;13(1):154.

29.   Al-Jawadi AA, Priliani L, Oktavianthi S, Febinia CA, Daya M, Artika IM, et al. Association of FTO

rs1421085 single nucleotide polymorphism with fat and fatty acid intake in Indonesian adults.

BMC Res Notes. 2021 Dec 7;14(1):411.

30. Alsulami S, Nyakotey DA, Dudek K, Bawah A-M, Lovegrove JA, Annan RA, et al. Interaction between Metabolic Genetic Risk Score and Dietary Fatty Acid Intake on Central Obesity in a Ghanaian Population. Nutrients. 2020 Jun 27;12(7):1906.

31. Daya M, Pujianto DA, Witjaksono F, Priliani L, Susanto J, Lukito W, et al. Obesity risk and preference for high dietary fat intake are determined by FTO rs9939609 gene polymorphism in selected Indonesian adults. Asia Pac J Clin Nutr. 2019;28(1):183–91.

32. Wuni R, Adela Nathania E, Ayyappa AK, Lakshmipriya N, Ramya K, Gayathri R, et al. Impact of Lipid Genetic Risk Score and Saturated Fatty Acid Intake on Central Obesity in an Asian Indian Population. Nutrients. 2022 Jun 29;14(13):2713.

33. Isgin-Atici K, Alsulami S, Turan-Demirci B, Surendran S, Sendur SN, Lay I, et al. FTO gene–lifestyle interactions on serum adiponectin concentrations and central obesity in a Turkish population. Int J Food Sci Nutr. 2021 Apr 3;72(3):375–85.

34. Hosseini-Esfahani F, Koochakpoor G, Mirmiran P, Daneshpour MS, Azizi F. Dietary patterns modify the association between fat mass and obesity-associated genetic variants and changes in obesity phenotypes. Br J Nutr. 2019 Jun 1;121(11):1247–54.

35. Mahmoudi-Nezhad M, Farhangi MA, Kahroba H. Cocaine and amphetamine-regulated transcript prepropeptide gene (CARTPT) polymorphism interacts with Diet Quality Index-International (DQI-I) and Healthy Eating Index (HEI) to affect hypothalamic hormones and cardio-metabolic risk factors among obese individuals. J Transl Med. 2020 Dec 9;18(1):16.

36. Rahati S, Qorbani M, Naghavi A, Nia MH, Pishva H. Association between CLOCK 3111 T/C polymorphism with ghrelin, GLP-1, food timing, sleep and chronotype in overweight and obese Iranian adults. BMC Endocr Disord. 2022 Dec 2;22(1):147.

37. Rana S, Sultana A, Bhatti AA. Effect of interaction between obesity-promoting genetic variants and behavioral factors on the risk of obese phenotypes. Mol Genet Genomics. 2021 Jul

9;296(4):919–38.

38. Xue H, Zhang X, Li D, Chen M, Luo J, Gong Y, et al. Relevance of Physical Activities, Sedentary Behaviors, and Genetic Predisposition in Body Fatness: Population-Based Study on Chinese Adults. Obes Facts. 2021;14(4):346–56.

39. Moore SC, Gunter MJ, Daniel CR, Reddy KS, George PS, Yurgalevitch S, et al. Common Genetic Variants and Central Adiposity Among Asian-Indians. Obesity. 2012 Sep;20(9):1902–8.

40. Gong W, Li H, Song C, Yuan F, Ma Y, Chen Z, et al. Effects of Gene-Environment Interaction on Obesity among Chinese Adults Born in the Early 1960s. Genes (Basel). 2021 Feb 13;12(2):270.

41. Sun M, Li S, Ning F, Zhang L, Wang W, Duan H, et al. Association between olfactory pathway gene variants and obesity in Chinese Han population: A case-control study based on genetic score. Gene. 2022 May;825:146442.

42. Muhammad HFL, Sulistyoningrum DC, Huriyati E, Lee YY, Muda WAMW. The interaction between energy intake, physical activity and UCP2-866G/A gene variation on weight gain and changes in adiposity: an Indonesian Nutrigenetic Cohort (INDOGENIC). Br J Nutr. 2021 Mar 28;125(6):611–7.

43. Ahmad S, Zhao W, Renström F, Rasheed A, Zaidi M, Samuel M, et al. A novel interaction between the FLJ33534 locus and smoking in obesity: a genome-wide study of 14 131 Pakistani adults. Int J Obes. 2016 Jan 17;40(1):186–90.

44. Wei B-L, Yin R-X, Liu C-X, Deng G-X, Guan Y-Z, Zheng P-F. The MC4R SNPs, their haplotypes and gene-environment interactions on the risk of obesity. Mol Med. 2020 Dec 8;26(1):77.

45. Illangasekera YA, Kumarasiri RPV, Fernando DJ, Dalton CF. Association of FTO and near MC4R variants with obesity measures in urban and rural dwelling Sri Lankans. Obes Res Clin Pract. 2016 Sep;10:S117–24.

46. Wang Y, Pan L, Wan S, Yihuo W, Yang F, He H, et al. MC4R Gene Polymorphisms Interact With the Urbanized Living Environment on Obesity: Results From the Yi Migrant Study. Front Genet. 2022

Apr 14;13.

47. Nishida C, Barba C, Cavalli-Sforza T, Cutter J, Deurenberg P, Darnton-Hill I, et al. Appropriate body-mass index for Asian populations and its implications for policy and intervention strategies. Lancet. 2004 Jan;363(9403):157–63.

48. Sirugo G, Williams SM, Tishkoff SA. The Missing Diversity in Human Genetic Studies. Cell. 2019 Mar;177(1):26–31.

49. Yi N, Xu S, Lou X-Y, Mallick H. Multiple comparisons in genetic association studies: a hierarchical modeling approach. Stat Appl Genet Mol Biol. 2014 Jan 1;13(1).

50. Hüls A, Krämer U, Carlsten C, Schikowski T, Ickstadt K, Schwender H. Comparison of weighting approaches for genetic risk scores in gene-environment interaction studies. BMC Genet. 2017 Dec 16;18(1):115.

51. Hüls A, Ickstadt K, Schikowski T, Krämer U. Detection of gene-environment interactions in the presence of linkage disequilibrium and noise by using genetic risk scores with internal weights from elastic net regression. BMC Genet. 2017 Dec 12;18(1):55.

52. Song S, Jiang W, Hou L, Zhao H. Leveraging effect size distributions to improve polygenic risk scores derived from summary statistics of genome-wide association studies. PLOS Comput Biol. 2020 Feb 11;16(2):e1007565.

53. Ma Y, Zhou X. Genetic prediction of complex traits with polygenic scores: a statistical review. Trends Genet. 2021 Nov;37(11):995–1011.

54. Ahmad S, Varga T V., Franks PW. Gene x Environment Interactions in Obesity: The State of the Evidence. Hum Hered. 2013;75(2–4):106–15.

55. Aasdahl L, Nilsen TIL, Meisingset I, Nordstoga AL, Evensen KAI, Paulsen J, et al. Genetic variants related to physical activity or sedentary behaviour: a systematic review. Int J Behav Nutr Phys Act. 2021 Dec 22;18(1):15.

56.   Dudbridge F, Gusnanto A. Estimation of significance thresholds for genomewide association

       scans. Genet Epidemiol. 2008 Apr;32(3):227–34.

57.   Martin AR, Kanai M, Kamatani Y, Okada Y, Neale BM, Daly MJ. Clinical use of current polygenic

       risk scores may exacerbate health disparities. Nat Genet. 2019 Apr 29;51(4):584–91.