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Increased advanced glycation end product and meat consumption is associated with childhood wheeze: analysis of the National Health and Nutrition Examination Survey

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ABSTRACT

We examined 4388 children from the 2003 to 2006 National Health and Nutrition Examination Survey and used survey-design-adjusted multivariable logistic regression to evaluate associations between dietary advanced glycation end product (AGE) and meat consumption frequencies and respiratory symptoms. Higher AGE intake was significantly associated with increased odds of wheezing (adjusted OR 1.18; 95% CI 1.02 to 1.36), wheeze-disrupted sleep (1.26; 95% CI 1.05 to 1.51) and exercise (1.34; 95% CI 1.08 to 1.67) and wheezing requiring prescription medication (1.35; 95% CI 1.13 to 1.63). Higher intake of non-seafood meats was associated with wheeze-disrupted sleep (2.32; 95% CI 1.11 to 4.82) and wheezing requiring prescription medication (2.23; 95% CI 1.10 to 4.54).

INTRODUCTION

Dietary advanced glycation end products (AGEs) are highly oxidant, proinflammatory compounds, with highest levels present in cooked meats.[1] AGEs are ligands for the AGE receptor (RAGE), a pattern recognition and danger signal receptor, with highest expression in the lungs, and an important driving force behind downstream Th-2 inflammation.[2] The relationship between AGEs and inflammatory airways disease is unclear.[1] We investigated the effects of dietary AGE intake and meat consumption on respiratory symptoms within a paediatric cohort from the National Health and Nutrition Examination Survey (NHANES), a cross-sectional survey assessing the health and nutritional status of the US population.[3]

METHODS

We included 4388 children aged 2–17 years from NHANES 2003–2006 (online supplemental figure S1), for whom data on dietary patterns (assessed by the 139-item Food Frequency Questionnaire (FFQ)), and respiratory symptoms were available. Only participants from NHANES 2003–2006 were included, as FFQ data were unavailable in other years. We calculated unitless meat consumption frequency scores using the FFQ, and AGE scores in keeping with prior published methodology (online supplemental appendix 1, online supplemental figure S2).[4] The primary outcome was presence of wheezing over the past year; secondary outcomes included the presence of more specific wheezing symptoms (online supplemental table S1).

The covariates age, sex, race/ethnicity, poverty to income ratio, body mass index (BMI) percentile, asthma status, total Healthy Eating Index (HEI) score and total caloric intake were collected from questionnaires. Survey logistic regression models were used to investigate associations between AGE and meat consumption scores, and dichotomised respiratory outcomes, adjusting for covariates. Interaction effects by age, sex, BMI percentile, asthma status and race/ethnicity were assessed, and total HEI score was removed from the multivariable model as a sensitivity analysis. Further analytical details are described in online supplemental appendix 1.

RESULTS

Survey-weighted demographics of the 4388 paediatric participants are summarised in table 1, with 537 (13%) reporting wheezing in the past year (online supplemental table S2). In the adjusted models, higher AGE scores were significantly associated with increased odds of wheezing (OR 1.18; 95% CI 1.02 to 1.36), at least one sleep disturbance due to wheezing (OR 1.26; 95%CI 1.05 to 1.51), wheezing during exercise (OR 1.34; 95%CI 1.08 to 1.67) and wheezing requiring prescription medication (OR 1.35; 95%CI 1.13 to 1.63) in the past year (table 2).

There were no significant interactions by age (p_{int}=0.46), sex (p_{int}=0.31), BMI percentile (p_{int}=0.73) or asthma status (p_{int}=0.26), but a significant interaction effect for race (p_{int}=0.04) (online supplemental appendix 2). Removing the total HEI score covariate from the main model did not significantly alter the associations between AGE intake and respiratory symptoms (data not shown).

We found a moderate but statistically significant positive correlation between AGE score and any non-seafood meat consumption (Pearson's correlation coefficient of r=0.69, p<0.0001) (online supplemental figure S3), consistent with meats containing high amounts of AGEs.[1] Higher consumption of non-seafood meat was associated with increased odds of wheezing requiring prescription medication (OR 2.23; 95%CI 1.10 to 4.54), and wheezing disrupting sleep (OR 2.32; 95%CI 1.11 to 4.82) (figure 1, online supplemental table S3).

DISCUSSION

To our knowledge, this is the first study in a national paediatric population demonstrating an association between dietary AGE intake and wheezing symptoms,

Table 1 Analytical population characteristics in relation to AGE intake

Participant characteristics (categorical variables)	Weighted % (crude frequency)*	Weighted AGE score median (25–75th percentiles)**
Sex		
Male	51.29 (2148)	5.32 (3.19–8.73)
Female	48.71 (2240)	5.99 (3.67–9.09)
Race/ethnicity		
Non-Hispanic whites	63.28 (1320)	5.43 (3.42–8.45)
Non-Hispanic blacks	14.74 (1404)	7.71 (4.52–12.8)
Hispanics	15.85 (1425)	5.43 (2.9–8.57)
Others	6.14 (239)	5.31 (3.5–8.73)
Current asthma		
No	89.02 (3907)	5.74 (3.48–9)
Yes	10.98 (481)	5.05 (2.94–8.32)
Participant characteristics (continuous variables)	**Weighted median (25–75th percentiles)***	
Age (years)	9.3 (5.3–13.2)	
Poverty to income ratio	2.3 (1.1–4.0)	
Body mass index percentile	68.7 (39.1–89.7)	
Total Healthy Eating Index (HEI) score†	46.1 (37.8–54.8)	
AGE score‡	5.70 (3.4–9.0)	
Red meat consumption score§	18.6 (15.8–21.5)	
Poultry consumption score§	8.6 (6.9–10.2)	
Processed meat consumption score§	16.1 (13.5–19.5)	
Non-seafood meat consumption score§	44.4 (38.7–50.5)	
Seafood consumption score§	8.5 (7.0–10.6)	
All meat consumption score§	53.6 (46.9–60.3)	

*Survey procedures were used to account for the NHANES survey design and to obtain population weighted estimates for proportions, medians, and 25–75th percentiles. All continuous variables were non-normally distributed. AGE and meat consumption scores were natural log transformed.
†Total HEI scores range from 0 to 100, with higher scores representing better compliance with the 2010 Dietary Guidelines for Americans.
‡The AGE score represents the daily average AGE intake, standardised for total caloric intake per participant. The score was calculated based on the FFQ and prior published food AGE content data.
§Meat consumption scores were derived using the FFQ in a similar fashion to the AGE score.
AGE, advanced glycation end-product; FFQ, Food Frequency Questionnaire.

including wheezing disruptive enough to interfere with exercise and sleep, and require prescription medication. Similarly, higher non-seafood meat consumption was associated with wheezing interfering with sleep and requiring prescription medication. Importantly, as non-seafood meats are a major dietary source of AGEs,[1] our results suggest that a AGE-rich diet is adversely associated with childhood wheeze independent of overall diet quality.

Our results expand on current literature supporting a link between a proinflammatory dietary pattern and wheezing.[4 5] The Western dietary pattern, characterised by increased consumption of AGE-rich foods, namely meats and saturated fats, may promote activation of the toll-like receptor 4 pathway and NF-κB inflammatory cascade, thereby contributing to airway inflammation and asthma pathogenesis.[6] Additionally, a recent cohort study demonstrated that a pro-inflammatory diet measured by the Dietary Inflammatory Index, was associated with increased wheezing in atopic children.[5] Conversely, the Mediterranean dietary pattern, characterised by lower consumption of saturated fatty acids and red meats, may have important anti-inflammatory effects, with one systematic review and meta-analysis demonstrating that adoption of a Mediterranean diet in children was associated with lower risk of wheezing symptoms and having ever had asthma.[7] Finally, as several cohort studies have suggested an adverse effect of meat consumption on paediatric airways health,[8 9] confirmation of a positive correlation between AGE intake and non-seafood meat consumption in our cohort strengthens our a priori hypothesis that dietary AGEs may have an important role in airway inflammation in children.

Biologic plausibility for our findings draws from emerging literature demonstrating that AGEs interact with RAGE, a critical modulator of type 2 cytokine signal transduction with a prominent role in allergic asthma pathogenesis.[2 10] However, whether increased dietary AGE intake leads to overactivation and upregulation of RAGE-mediated airways inflammation remains unclear and warrants further exploration.

There are several limitations. First, causality cannot be implied due to the cross-sectional design, although these novel, hypothesis-generating findings provide a foundation for subsequent longitudinal studies to assess AGE intake as a modifiable dietary risk factor in the development of airways disease. Second, while there is no validated method of quantifying AGE consumption, AGE scores presented reasonably represent relative intake within our cohort, and associations between meat intake and wheezing were

Table 2 Associations between age scores and respiratory symptoms

Outcomes	Weighted % (crude frequency: 'yes'/total)*	OR	95% CI	P value
Primary outcome				
Wheezing in chest	13.04 (537/4388)	1.18	1.02 to 1.36	0.03
Secondary outcomes				
At least one sleep disturbance due to wheezing	7.04 (304/4388)	1.26	1.05 to 1.51	0.01
Wheezing during exercise	6.91 (292/4385)	1.34	1.08 to 1.67	0.007
At least one doctor, hospital or emergency room visit for wheezing	8.17 (325/4388)	1.05	0.87 to 1.27	0.64
Wheezing requiring prescription medication use	9.45 (399/4388)	1.35	1.13 to 1.63	0.001
Limitation of usual activities due to wheezing	5.39 (229/4388)	1.20	0.98 to 1.47	0.08
At least 1 day of school or work missed due to wheezing	4.31 (169/4245)	1.12	0.90 to 1.40	0.34
Dry nocturnal cough	2.63 (124/4386)	1.15	0.83 to 1.61	0.41

*Survey procedures were used to account for the NHANES survey design and to obtain population weighted estimates for prevalence and OR estimates. OR estimates were based on logistic models adjusted for age, sex, race/ethnicity, poverty to family income ratio, body mass index percentile, current asthma and total Healthy Eating Index score.
AGE, advanced glycation end product; NHANES, National Health and Nutrition Examination Survey.

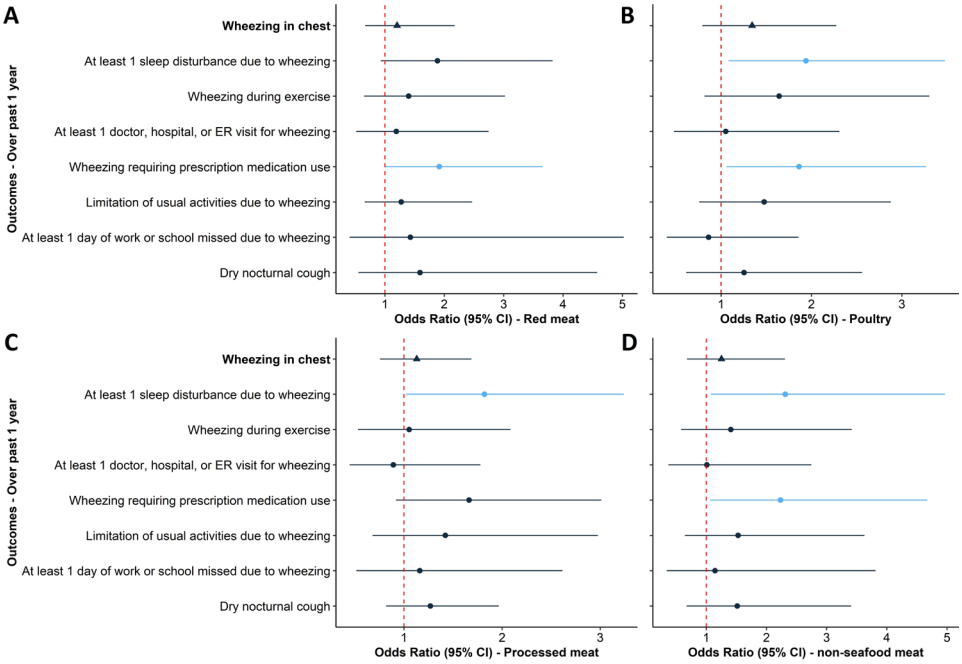

Figure 1 Associations between consumption scores for (A) red meat, (B) poultry, (C) processed meat and (D) any non-seafood meat (combination intake frequencies of red meat, poultry and processed meat) and respiratory symptoms, adjusted for age, sex, race/ethnicity, poverty to family income ratio, body mass index percentile, current asthma, total health eating index score and total caloric intake. ER, emergency room.

consistent with these findings. Finally, we recognise that the aetiology of wheezing may be highly variable in the wide age range of our cohort; however, we demonstrated no significant interaction effects by age.

In conclusion, higher dietary intake of AGEs, in part from non-seafood meat consumption, was associated with increased risk of clinically impactful wheezing in children. Further longitudinal studies examining the impact of dietary AGEs on airways disease in children are warranted.

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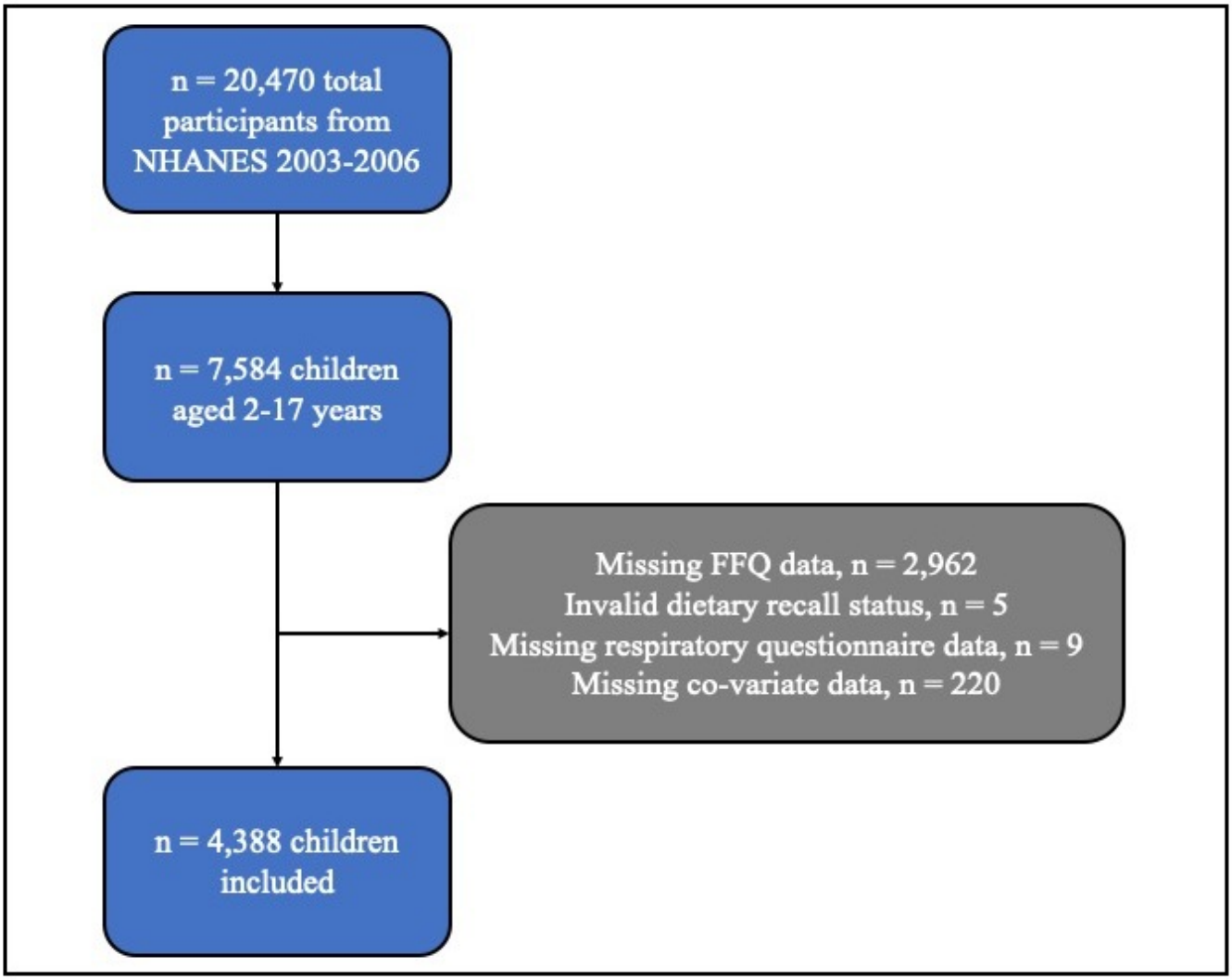
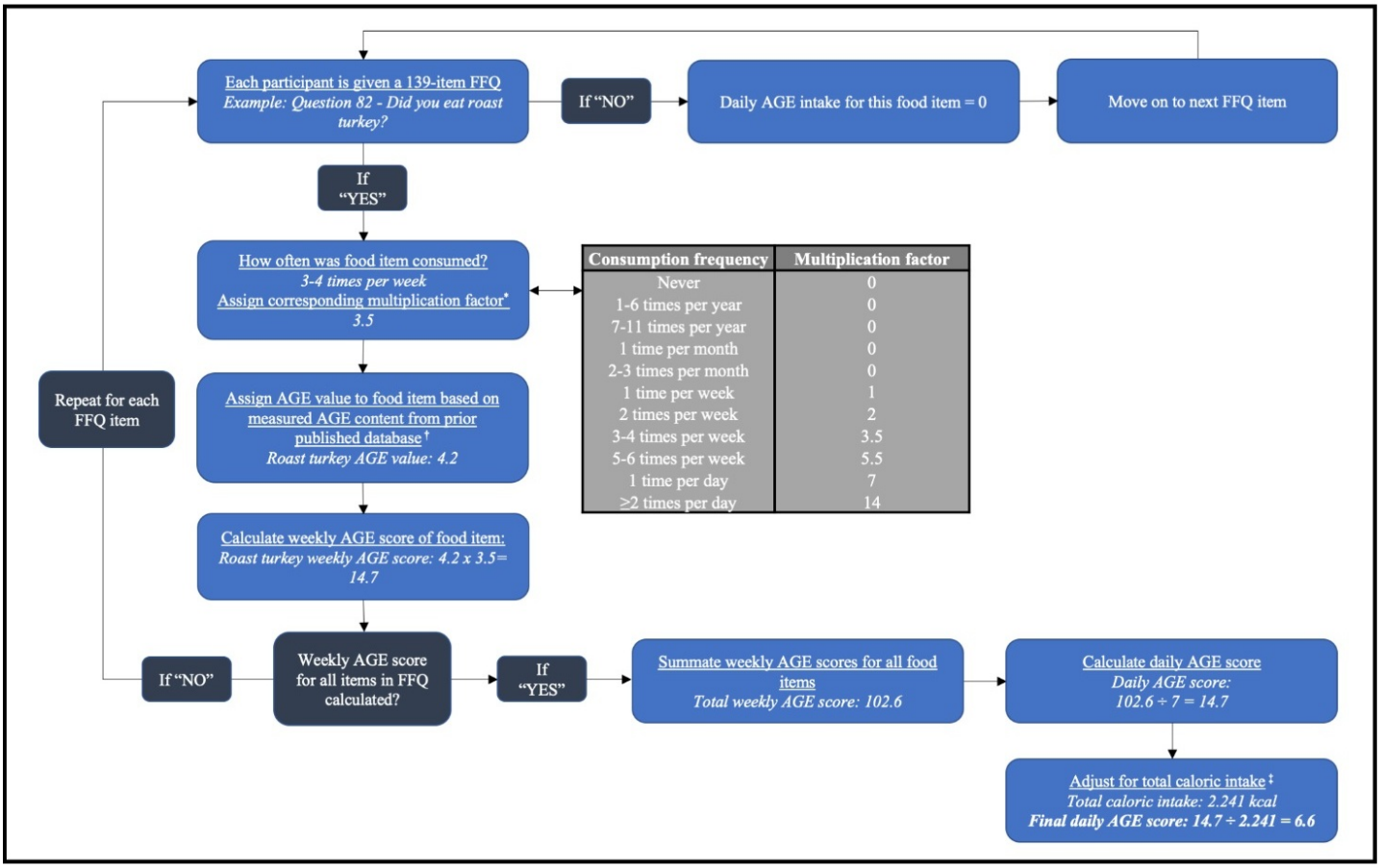
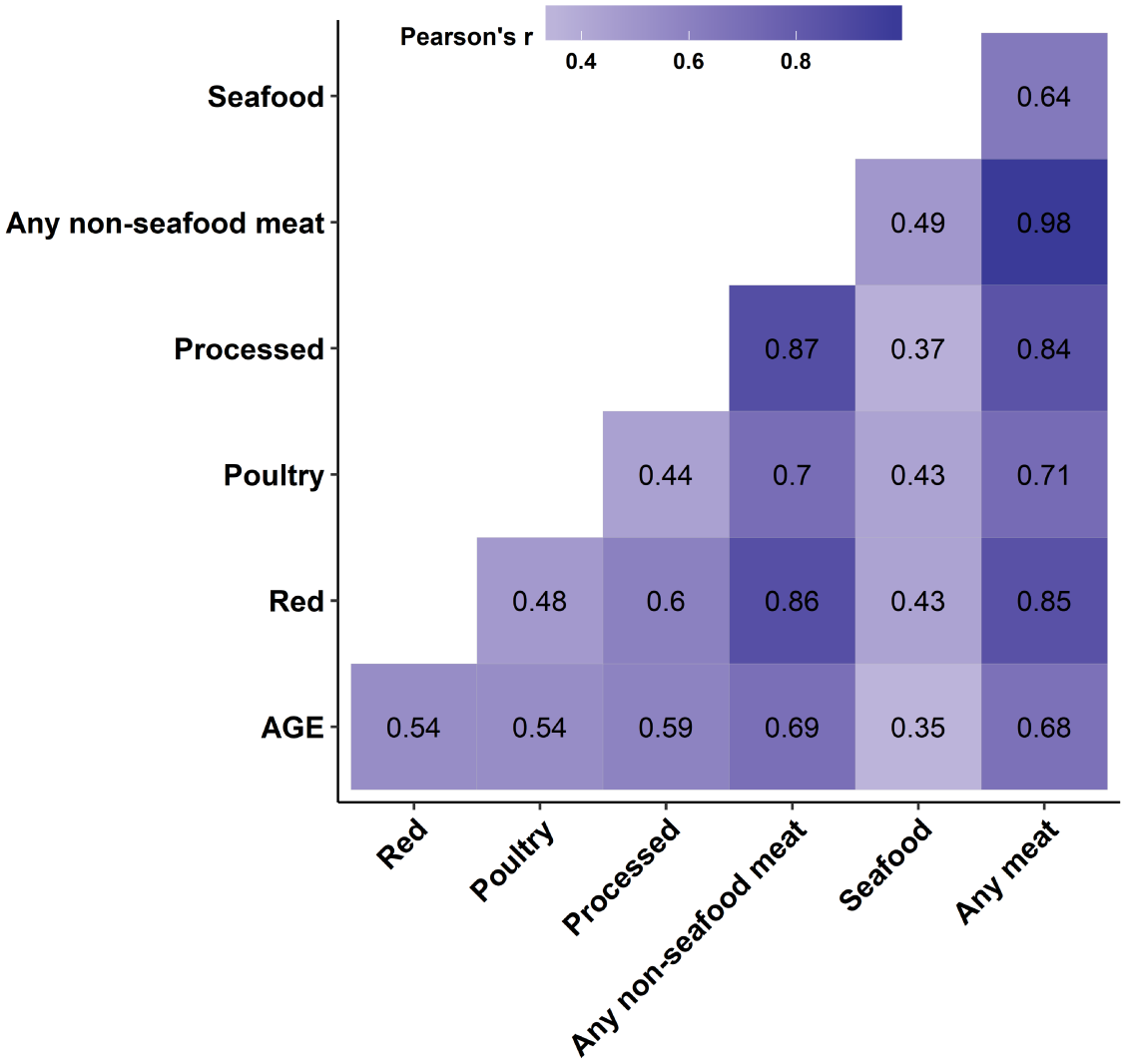
REFERENCES

1 Uribarri J, Woodruff S, Goodman S, *et al*. Advanced glycation end products in foods and a practical guide to their reduction in the diet. *J Am Diet Assoc* 2010;110:911–6.
2 Oczypok EA, Perkins TN, Oury TD. All the "RAGE" in lung disease: The receptor for advanced glycation endproducts (RAGE) is a major mediator of pulmonary inflammatory responses. *Paediatr Respir Rev* 2017;23:40–9.
3 Zipf G, Chiappa M, Porter KS, *et al*. National health and nutrition examination survey: plan and operations, 1999-2010. *Vital Health Stat 1* 2013;1:1–37.
4 Guilleminault L, Williams EJ, Scott HA, *et al*. Diet and asthma: is it time to adapt our message? *Nutrients* 2017;9. doi:10.3390/nu9111227. [Epub ahead of print: 08 Nov 2017].
5 Han Y-Y, Forno E, Shivappa N, *et al*. The dietary inflammatory index and current wheeze among children and adults in the United States. *J Allergy Clin Immunol Pract* 2018;6:834–41.
6 Wood LG, Gibson PG. Dietary factors lead to innate immune activation in asthma. *Pharmacol Ther* 2009;123:37–53.
7 Garcia-Marcos L, Castro-Rodriguez JA, Weinmayr G, *et al*. Influence of Mediterranean diet on asthma in children: a systematic review and meta-analysis. *Pediatr Allergy Immunol* 2013;24:330–8.
8 Nagel G, Weinmayr G, Kleiner A, *et al*. Effect of diet on asthma and allergic sensitisation in the International study on allergies and asthma in childhood (Isaac) phase two. *Thorax* 2010;65:516–22.
9 Hallit S, Raherison C, Abou Abdallah R, *et al*. Correlation of types of food and asthma diagnosis in childhood: a case-control study. *J Asthma* 2018;55:966–74.
10 Oczypok EA, Milutinovic PS, Alcorn JF, *et al*. Pulmonary receptor for advanced glycation end-products promotes asthma pathogenesis through IL-33 and accumulation of group 2 innate lymphoid cells. *J Allergy Clin Immunol* 2015;136:747–56.







Online supplementary

Appendix 1 – METHODS

AGE score calculation

There currently lacks a standardized and validated method of estimating AGE consumption. As a result, we adapted a previously published strategy of approximating AGE intake,[1] albeit in an adolescent population. This strategy utilizes the NHANES FFQ and assumes consumption of standard portion sizes, as the FFQ does not report portion sizes. Further, as the NHANES FFQ does not capture detailed cooking methodology, which may affect AGE content particularly for meats,[2] we assumed conventional methods of cooking, specifically with the use of high, dry heat (roasting, grilling, broiling, frying and searing). Despite these assumptions, our AGE score may still be used as a reasonable comparison of relative AGE intake across this cohort.

Consistent with published methodology, each food item was assigned a multiplication factor based on consumption frequency (0 if consumption was at most 2-3 times per month, 1 if at most once per week, 2 if twice per week, 3.5 if 3-4 times per week, 5.5 if 5-6 times per week, 7 for daily and 14 for twice per day) (Figure S2).[1] We then calculated estimated AGE intake per week for each food item as the product of the multiplication factor and the approximate AGE content of the food item from an existing database.[2] A daily average AGE intake was calculated from the weekly estimated AGE intake for all food. Finally, this value was standardized by reported total kilocalorie intake from the 24-hour dietary recall to arrive at an AGE score representing an estimate of the daily average AGE consumption per participant (Figure S2).[2]

Explanation of covariates

Poverty to income ratio (PIR) was calculated according to the annual Department of Health and Human Services' poverty guidelines by dividing family income by a family size-specific poverty threshold with scores ranging from 0 (no income) to 5 (≥ 5 times the federal poverty level).[3,4] BMI percentile was used to classify BMI in accordance with expert committee recommendations from the American Medical Association,[5] and calculated using the SAS code based on the 2000 Centers for Disease Control and Prevention growth chart.[6] A self-reported diagnosis of current asthma was defined as an affirmative response to the following questions: "Has a doctor or other health professional ever told you that you have asthma?" and "Do you still have asthma?" The total HEI score is a validated measure of overall diet quality and adherence to recommendations from the 2010 Dietary Guidelines for Americans (DGA).[7] Scores range from 0 to 100 with higher scores representing better adherence. For each participant, the total HEI score was calculated using the 24-hour dietary recall interview and the recommended HEI scoring algorithms from the National Institutes of Health.[8]

Statistical analyses

For the multivariable models, respiratory questions that elicited a numerical or ordinal response were dichotomized, indicating either presence or absence of the symptom. AGE and meat consumption scores were natural-log-transformed. Pearson's correlation was conducted between non-survey-weighted AGE and meat consumption scores. Bonferroni correction for multiple testing was applied

for the seven secondary outcomes using $\alpha<0.007$ for statistical significance. All analyses were conducted using Statistical Analysis Software (SAS) (version 9.4) and figures were made using R (version 3.5).

Appendix 2 - RESULTS

Bonferroni corrections

After Bonferroni correction for the 7 secondary outcomes, higher AGE scores were significantly associated with wheezing requiring prescription medication use at $\alpha = 0.007$, while there were no significantly increased odds of having wheezing or cough symptoms with higher consumption of any meat at $\alpha = 0.007$.

Interaction by race/ethnicity

When race/ethnicity was defined as non-Hispanic whites, non-Hispanic blacks, Hispanics and others, there was no significant overall interaction by race/ethnicity ($p_{int} = 0.05$). However, there was a significant interaction effect ($p_{int} = 0.04$) when non-Hispanic blacks were compared with non-Hispanic whites. Stratified analysis showed that only non-Hispanic whites had increased odds of wheezing with higher AGE intake (OR 1.29; 95% CI 1.02 to 1.63).

Appendix 3 – FIGURES AND TABLES

Figure S1. Selection flowchart for study participant inclusion. Participants were included if they had valid responses to the FFQ and 24-hour dietary recall interview, and had completed the interview questionnaire on respiratory symptoms. Participants were also required to provide responses to relevant covariates including age, sex, race/ethnicity, the ratio of family income to poverty, body mass index percentiles, and asthma status. Children younger than 2 years of age were excluded as the FFQ was conducted amongst participants at least 2 years of age. Participants with missing responses or the responses "Don't know" or "Refused" were excluded. NHANES: National Health and Nutrition Examination Survey; FFQ: Food frequency questionnaire.

Table S1. Interview questions included from the NHANES Respiratory Health Questionnaire on respiratory symptoms

Questions	Target population	Range of responses
Primary outcome		
In the past 12 months, have you had wheezing or whistling in your chest?	Both males and females 1-150 years old	Yes No Refused Don't know Missing
Secondary outcomes		
In the past 12 months, how often, on average, has your sleep been disturbed because of wheezing? Would you say this happens…	Both males and females 1-150 years old	Never ≥1 nights per week <1 night/week Refused Don't know Missing
In the past 12 months, has your chest sounded wheezy during or after exercise or physical activity?	Both males and females 1-150 years old	Yes No Refused Don't know Missing
In the past 12 months, how many times have you gone to the doctor's office or the hospital emergency room for one or more of these attacks of wheezing or whistling?	Both males and females 1-150 years old	0-15 ≥20 Refused Don't know Missing
In the past 12 months, have you taken medication, prescribe by a doctor, for wheezing or whistling?	Both males and females 1-150 years old	Yes No Refused Don't know Missing
During the past 12 months, how much did you limit your usual activities due to wheezing or whistling? Would you say…	Both males and females 1-150 years old	Not at all A little

		A fair amount A moderate amount A lot Refused Don't know Missing
During the past 12 months, how many days of work or school did you miss due to wheezing or whistling?	Both males and females 6-69 years old	None 1-7 8-30 ≥31 Refused Don't know Missing
In the past 12 months, have you had a dry cough at night not counting a cough associated with a cold or chest infection lasting 14 days or more?	Both males and females 1-150 years old	Yes No Refused Don't know Missing

Figure S2. Flow diagram depicting step-wise approach used for each participant in determining a daily AGE score representative of approximate AGE intake (adapted from Saha et al.)[1]

*Multiplication factors representative of food consumption frequency, adapted from Saha et al.[1]

†As the NHANES FFQ does not describe detailed cooking methodology or portion size consumed, exact AGE intake could not be obtained. Consequently, a unitless AGE value was derived to approximate AGE intake, utilizing a published database of measured AGE content for various foods.[2] This derived AGE value represents approximate AGE intake per one-time consumption of food item, assuming standard portion sizes.

‡Total caloric intake for each participant taken from NHANES 24-hour dietary recall interview.

FFQ: Food frequency questionnaire

AGE: Advanced glycation end-product

NHANES: National Health and Nutrition Examination Survey

Table S2. Frequency of respiratory symptoms

Respiratory symptoms (over past 1 year)	**Weighted % (Crude Frequency: "Yes"/Total)** *
Wheezing in chest	13.04 (537/4388)
At least 1 sleep disturbance due to wheezing	7.04 (304/4388)
Wheezing during exercise	6.91 (292/4385)
At least 1 doctor, hospital, or emergency room visit for wheezing	8.17 (325/4388)
Wheezing requiring prescription medication use	9.45 (399/4388)
Limitation of usual activities due to wheezing	5.39 (229/4388)
At least 1 day of school or work missed due to wheezing	4.31 (169/4245)
Dry nocturnal cough	2.63 (124/4386)

*Survey procedures were used to take into account the NHANES survey design and to obtain population weighted estimates for prevalence.

Figure S3. Heat map showing Pearson's correlation co-efficients between advanced glycation end-product (AGE) and meat consumption scores. Any non-seafood meat is defined as the total consumption frequencies of red meat, processed meat and poultry. Any meat includes the consumption frequencies of red meat, processed meat, poultry and seafood. All correlations were statistically significant at $p < 0.0001$.

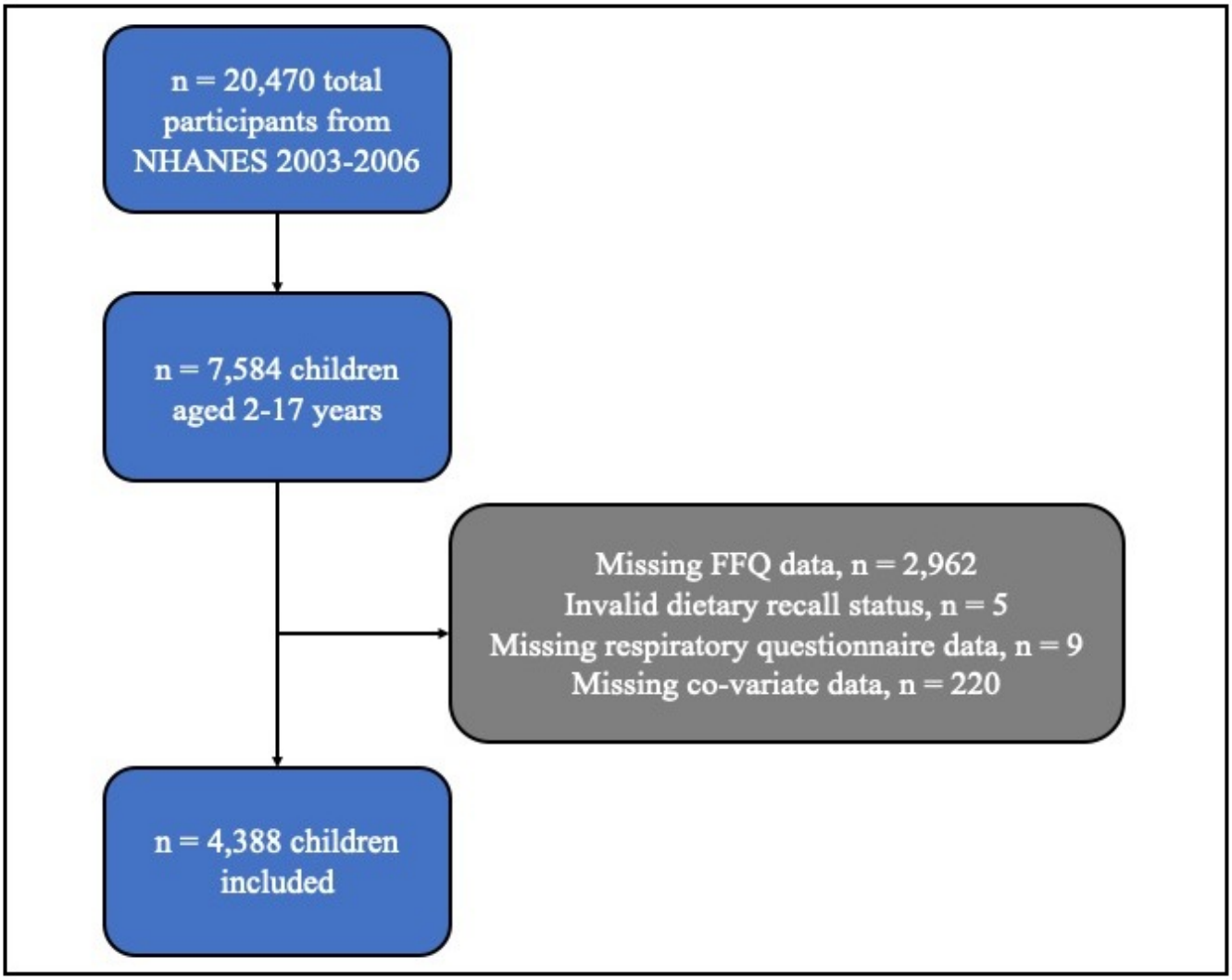
Table S3. Survey-design-adjusted multivariable logistic regression of associations between meat consumption frequencies and respiratory symptoms

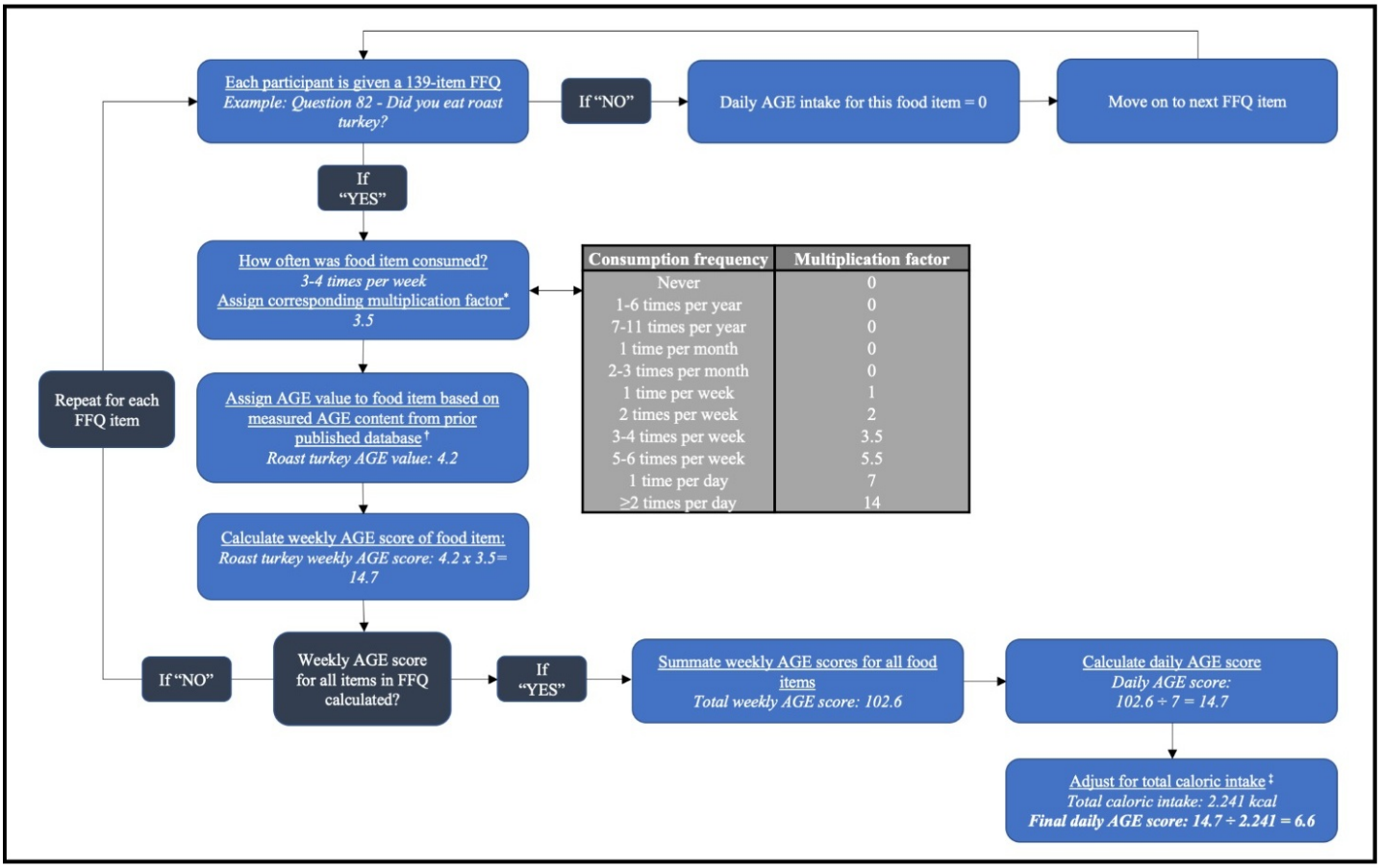
Respiratory outcomes (over past 1 year)	**Odds Ratio**	**95% Confidence Interval**	**p value**
Red meat			
Wheezing in chest	1.20	0.68-2.12	0.52
At least 1 sleep disturbance due to wheezing	1.89	0.96-3.72	0.07
Wheezing during exercise	1.40	0.67-2.93	0.37
At least 1 doctor, hospital, or emergency room visit for wheezing	1.19	0.53-2.66	0.67
Wheezing requiring prescription medication use	**1.92**	**1.03-3.57**	**0.04**
Limitation of usual activities due to wheezing	1.27	0.68-2.41	0.45
At least 1 day of school or work missed due to wheezing	1.43	0.43-4.78	0.56
Dry nocturnal cough	1.59	0.58-4.39	0.37
Poultry			
Wheezing in chest	1.34	0.81-2.23	0.25
At least 1 sleep disturbance due to wheezing	**1.94**	**1.11-3.40**	**0.02**
Wheezing during exercise	1.64	0.84-3.21	0.15
At least 1 doctor, hospital, or emergency room visit for wheezing	1.05	0.50-2.24	0.89
Wheezing requiring prescription medication use	**1.86**	**1.09-3.19**	**0.02**
Limitation of usual activities due to wheezing	1.48	0.78-2.80	0.23
At least 1 day of school or work missed due to wheezing	0.86	0.41-1.80	0.69
Dry nocturnal cough	1.26	0.63-2.49	0.51
Processed meat			
Wheezing in chest	1.13	0.77-1.66	0.53
At least 1 sleep disturbance due to wheezing	**1.82**	**1.05-3.17**	**0.03**
Wheezing during exercise	1.05	0.55-2.03	0.88
At least 1 doctor, hospital, or emergency room visit for wheezing	0.89	0.50-1.73	0.73
Wheezing requiring prescription medication use	1.66	0.94-2.94	0.08
Limitation of usual activities due to wheezing	1.42	0.70-2.89	0.33
At least 1 day of school or work missed due to wheezing	1.16	0.53-2.53	0.71
Dry nocturnal cough	1.27	0.83-1.93	0.27
Any non-seafood meat			
Wheezing in chest	1.25	0.70-2.25	0.45
At least 1 sleep disturbance due to wheezing	**2.32**	**1.11-4.82**	**0.02**
Wheezing during exercise	1.41	0.60-3.30	0.43
At least 1 doctor, hospital, or emergency room visit for wheezing	1.01	0.39-2.64	0.99

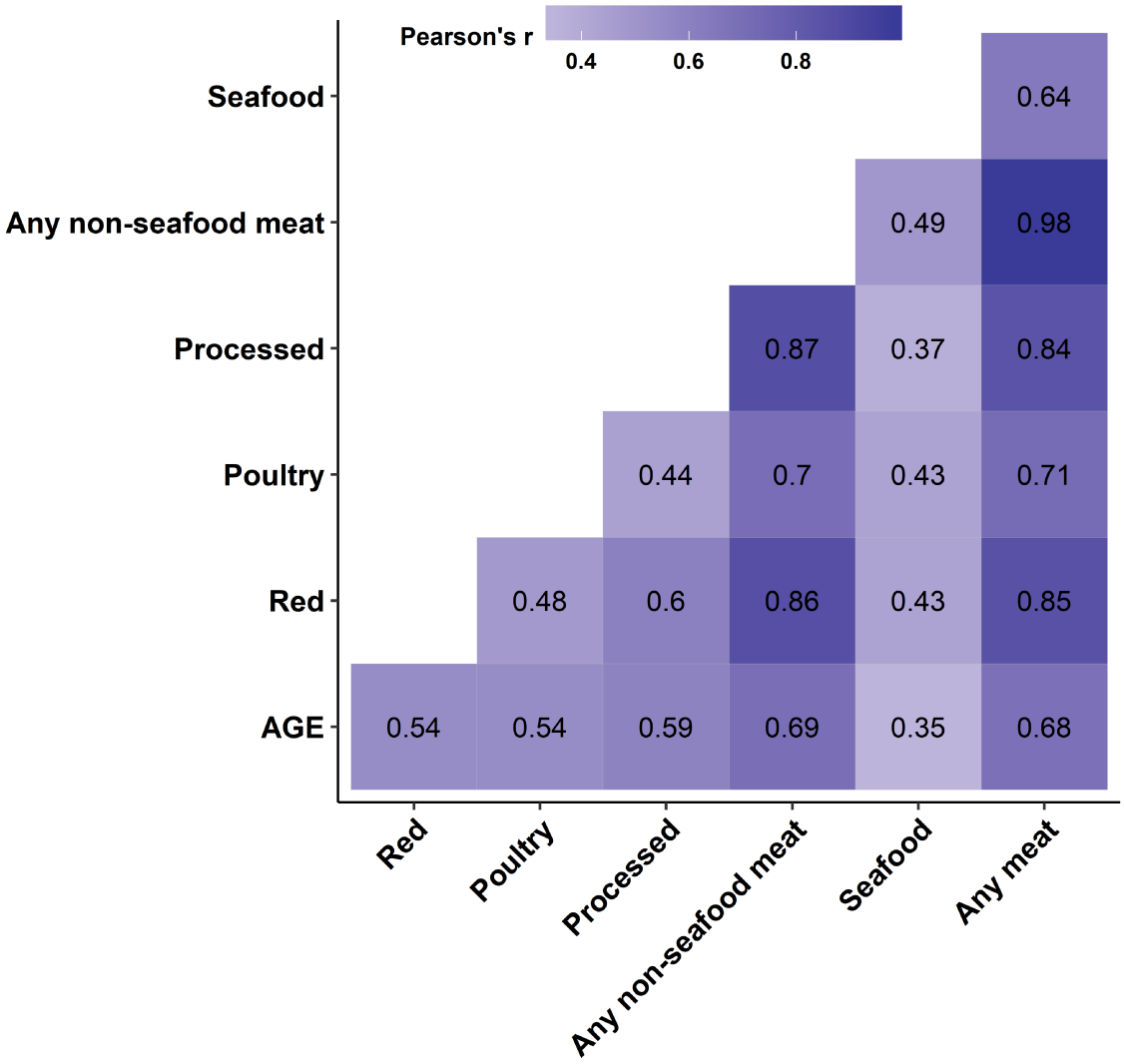
Wheezing requiring prescription medication use	**2.23**	**1.10-4.54**	**0.03**
Limitation of usual activities due to wheezing	1.53	0.67-3.51	0.32
At least 1 day of school or work missed due to wheezing	1.15	0.36-3.63	0.82
Dry nocturnal cough	1.52	0.70-3.30	0.29
Seafood			
Wheezing in chest	1.17	0.72-1.89	0.53
At least 1 sleep disturbance due to wheezing	1.82	0.83-3.99	0.13
Wheezing during exercise	1.09	0.65-1.84	0.75
At least 1 doctor, hospital, or emergency room visit for wheezing	1.13	0.51-2.55	0.76
Wheezing requiring prescription medication use	1.46	0.71-3.02	0.30
Limitation of usual activities due to wheezing	1.11	0.60-2.06	0.75
At least 1 day of school or work missed due to wheezing	1.54	0.77-3.06	0.22
Dry nocturnal cough	1.08	0.63-1.85	0.78

Appendix - References

1. Saha A, Poojary P, Chan L, et al. Increased odds of metabolic syndrome with consumption of high dietary advanced glycation end products in adolescents. *Diabetes Metab* 2017;43(5):469-71. doi: 10.1016/j.diabet.2017.01.001 [published Online First: 2017/02/06]
2. Uribarri J, Woodruff S, Goodman S, et al. Advanced glycation end products in foods and a practical guide to their reduction in the diet. *J Am Diet Assoc* 2010;110(6):911-16 e12. doi: 10.1016/j.jada.2010.03.018 [published Online First: 2010/05/26]
3. Zipf G, Chiappa M, Porter KS, et al. National health and nutrition examination survey: plan and operations, 1999-2010. *Vital Health Stat 1* 2013(56):1-37. [published Online First: 2014/08/01]
4. Fryar CD, Wright JD, Eberhardt MS, et al. Trends in nutrient intakes and chronic health conditions among Mexican-American adults, a 25-year profile: United States, 1982-2006. *Natl Health Stat Report* 2012(50):1-20. [published Online First: 2012/07/19]
5. Barlow SE. Expert committee recommendations regarding the prevention, assessment, and treatment of child and adolescent overweight and obesity: summary report. *Pediatrics* 2007;120 Suppl 4:S164-92. doi: 10.1542/peds.2007-2329C [published Online First: 2007/12/18]
6. Kuczmarski RJ, Ogden CL, Guo SS, et al. 2000 CDC Growth Charts for the United States: methods and development. *Vital Health Stat 11* 2002(246):1-190. [published Online First: 2002/06/05]
7. Guenther PM, Casavale KO, Reedy J, et al. Update of the Healthy Eating Index: HEI-2010. *J Acad Nutr Diet* 2013;113(4):569-80. doi: 10.1016/j.jand.2012.12.016 [published Online First: 2013/02/19]
8. NIH - National Cancer Institute. Division of Cancer Control & Population Sciences. Healthy Eating Index SAS Code 2019 [Available from: https://epi.grants.cancer.gov/hei/sas-code.html accessed October 18, 2019.







Online supplementary

Appendix 1 – METHODS

AGE score calculation

There currently lacks a standardized and validated method of estimating AGE consumption. As a result, we adapted a previously published strategy of approximating AGE intake,[1] albeit in an adolescent population. This strategy utilizes the NHANES FFQ and assumes consumption of standard portion sizes, as the FFQ does not report portion sizes. Further, as the NHANES FFQ does not capture detailed cooking methodology, which may affect AGE content particularly for meats,[2] we assumed conventional methods of cooking, specifically with the use of high, dry heat (roasting, grilling, broiling, frying and searing). Despite these assumptions, our AGE score may still be used as a reasonable comparison of relative AGE intake across this cohort.

Consistent with published methodology, each food item was assigned a multiplication factor based on consumption frequency (0 if consumption was at most 2-3 times per month, 1 if at most once per week, 2 if twice per week, 3.5 if 3-4 times per week, 5.5 if 5-6 times per week, 7 for daily and 14 for twice per day) (Figure S2).[1] We then calculated estimated AGE intake per week for each food item as the product of the multiplication factor and the approximate AGE content of the food item from an existing database.[2] A daily average AGE intake was calculated from the weekly estimated AGE intake for all food. Finally, this value was standardized by reported total kilocalorie intake from the 24-hour dietary recall to arrive at an AGE score representing an estimate of the daily average AGE consumption per participant (Figure S2).[2]

Explanation of covariates

Poverty to income ratio (PIR) was calculated according to the annual Department of Health and Human Services' poverty guidelines by dividing family income by a family size-specific poverty threshold with scores ranging from 0 (no income) to 5 (≥ 5 times the federal poverty level).[3,4] BMI percentile was used to classify BMI in accordance with expert committee recommendations from the American Medical Association,[5] and calculated using the SAS code based on the 2000 Centers for Disease Control and Prevention growth chart.[6] A self-reported diagnosis of current asthma was defined as an affirmative response to the following questions: "Has a doctor or other health professional ever told you that you have asthma?" and "Do you still have asthma?" The total HEI score is a validated measure of overall diet quality and adherence to recommendations from the 2010 Dietary Guidelines for Americans (DGA).[7] Scores range from 0 to 100 with higher scores representing better adherence. For each participant, the total HEI score was calculated using the 24-hour dietary recall interview and the recommended HEI scoring algorithms from the National Institutes of Health.[8]

Statistical analyses

For the multivariable models, respiratory questions that elicited a numerical or ordinal response were dichotomized, indicating either presence or absence of the symptom. AGE and meat consumption scores were natural-log-transformed. Pearson's correlation was conducted between non-survey-weighted AGE and meat consumption scores. Bonferroni correction for multiple testing was applied

for the seven secondary outcomes using $\alpha<0.007$ for statistical significance. All analyses were conducted using Statistical Analysis Software (SAS) (version 9.4) and figures were made using R (version 3.5).

Appendix 2 - RESULTS

Bonferroni corrections

After Bonferroni correction for the 7 secondary outcomes, higher AGE scores were significantly associated with wheezing requiring prescription medication use at $\alpha = 0.007$, while there were no significantly increased odds of having wheezing or cough symptoms with higher consumption of any meat at $\alpha = 0.007$.

Interaction by race/ethnicity

When race/ethnicity was defined as non-Hispanic whites, non-Hispanic blacks, Hispanics and others, there was no significant overall interaction by race/ethnicity ($p_{int} = 0.05$). However, there was a significant interaction effect ($p_{int} = 0.04$) when non-Hispanic blacks were compared with non-Hispanic whites. Stratified analysis showed that only non-Hispanic whites had increased odds of wheezing with higher AGE intake (OR 1.29; 95% CI 1.02 to 1.63).

Appendix 3 – FIGURES AND TABLES

Figure S1. Selection flowchart for study participant inclusion. Participants were included if they had valid responses to the FFQ and 24-hour dietary recall interview, and had completed the interview questionnaire on respiratory symptoms. Participants were also required to provide responses to relevant covariates including age, sex, race/ethnicity, the ratio of family income to poverty, body mass index percentiles, and asthma status. Children younger than 2 years of age were excluded as the FFQ was conducted amongst participants at least 2 years of age. Participants with missing responses or the responses "Don't know" or "Refused" were excluded. NHANES: National Health and Nutrition Examination Survey; FFQ: Food frequency questionnaire.

Table S1. Interview questions included from the NHANES Respiratory Health Questionnaire on respiratory symptoms

Questions	Target population	Range of responses
Primary outcome		
In the past 12 months, have you had wheezing or whistling in your chest?	Both males and females 1-150 years old	Yes No Refused Don't know Missing
Secondary outcomes		
In the past 12 months, how often, on average, has your sleep been disturbed because of wheezing? Would you say this happens…	Both males and females 1-150 years old	Never ≥1 nights per week <1 night/week Refused Don't know Missing
In the past 12 months, has your chest sounded wheezy during or after exercise or physical activity?	Both males and females 1-150 years old	Yes No Refused Don't know Missing
In the past 12 months, how many times have you gone to the doctor's office or the hospital emergency room for one or more of these attacks of wheezing or whistling?	Both males and females 1-150 years old	0-15 ≥20 Refused Don't know Missing
In the past 12 months, have you taken medication, prescribe by a doctor, for wheezing or whistling?	Both males and females 1-150 years old	Yes No Refused Don't know Missing
During the past 12 months, how much did you limit your usual activities due to wheezing or whistling? Would you say…	Both males and females 1-150 years old	Not at all A little

		A fair amount A moderate amount A lot Refused Don't know Missing
During the past 12 months, how many days of work or school did you miss due to wheezing or whistling?	Both males and females 6-69 years old	None 1-7 8-30 ≥31 Refused Don't know Missing
In the past 12 months, have you had a dry cough at night not counting a cough associated with a cold or chest infection lasting 14 days or more?	Both males and females 1-150 years old	Yes No Refused Don't know Missing

Figure S2. Flow diagram depicting step-wise approach used for each participant in determining a daily AGE score representative of approximate AGE intake (adapted from Saha et al.)[1]

*Multiplication factors representative of food consumption frequency, adapted from Saha et al.[1]

†As the NHANES FFQ does not describe detailed cooking methodology or portion size consumed, exact AGE intake could not be obtained. Consequently, a unitless AGE value was derived to approximate AGE intake, utilizing a published database of measured AGE content for various foods.[2] This derived AGE value represents approximate AGE intake per one-time consumption of food item, assuming standard portion sizes.

‡Total caloric intake for each participant taken from NHANES 24-hour dietary recall interview.

FFQ: Food frequency questionnaire

AGE: Advanced glycation end-product

NHANES: National Health and Nutrition Examination Survey

Table S2. Frequency of respiratory symptoms

Respiratory symptoms (over past 1 year)	**Weighted % (Crude Frequency: "Yes"/Total)** *
Wheezing in chest	13.04 (537/4388)
At least 1 sleep disturbance due to wheezing	7.04 (304/4388)
Wheezing during exercise	6.91 (292/4385)
At least 1 doctor, hospital, or emergency room visit for wheezing	8.17 (325/4388)
Wheezing requiring prescription medication use	9.45 (399/4388)
Limitation of usual activities due to wheezing	5.39 (229/4388)
At least 1 day of school or work missed due to wheezing	4.31 (169/4245)
Dry nocturnal cough	2.63 (124/4386)

*Survey procedures were used to take into account the NHANES survey design and to obtain population weighted estimates for prevalence.

Figure S3. Heat map showing Pearson's correlation co-efficients between advanced glycation end-product (AGE) and meat consumption scores. Any non-seafood meat is defined as the total consumption frequencies of red meat, processed meat and poultry. Any meat includes the consumption frequencies of red meat, processed meat, poultry and seafood. All correlations were statistically significant at $p < 0.0001$.

Table S3. Survey-design-adjusted multivariable logistic regression of associations between meat consumption frequencies and respiratory symptoms

Respiratory outcomes (over past 1 year)	**Odds Ratio**	**95% Confidence Interval**	**p value**
Red meat			
Wheezing in chest	1.20	0.68-2.12	0.52
At least 1 sleep disturbance due to wheezing	1.89	0.96-3.72	0.07
Wheezing during exercise	1.40	0.67-2.93	0.37
At least 1 doctor, hospital, or emergency room visit for wheezing	1.19	0.53-2.66	0.67
Wheezing requiring prescription medication use	**1.92**	**1.03-3.57**	**0.04**
Limitation of usual activities due to wheezing	1.27	0.68-2.41	0.45
At least 1 day of school or work missed due to wheezing	1.43	0.43-4.78	0.56
Dry nocturnal cough	1.59	0.58-4.39	0.37
Poultry			
Wheezing in chest	1.34	0.81-2.23	0.25
At least 1 sleep disturbance due to wheezing	**1.94**	**1.11-3.40**	**0.02**
Wheezing during exercise	1.64	0.84-3.21	0.15
At least 1 doctor, hospital, or emergency room visit for wheezing	1.05	0.50-2.24	0.89
Wheezing requiring prescription medication use	**1.86**	**1.09-3.19**	**0.02**
Limitation of usual activities due to wheezing	1.48	0.78-2.80	0.23
At least 1 day of school or work missed due to wheezing	0.86	0.41-1.80	0.69
Dry nocturnal cough	1.26	0.63-2.49	0.51
Processed meat			
Wheezing in chest	1.13	0.77-1.66	0.53
At least 1 sleep disturbance due to wheezing	**1.82**	**1.05-3.17**	**0.03**
Wheezing during exercise	1.05	0.55-2.03	0.88
At least 1 doctor, hospital, or emergency room visit for wheezing	0.89	0.50-1.73	0.73
Wheezing requiring prescription medication use	1.66	0.94-2.94	0.08
Limitation of usual activities due to wheezing	1.42	0.70-2.89	0.33
At least 1 day of school or work missed due to wheezing	1.16	0.53-2.53	0.71
Dry nocturnal cough	1.27	0.83-1.93	0.27
Any non-seafood meat			
Wheezing in chest	1.25	0.70-2.25	0.45
At least 1 sleep disturbance due to wheezing	**2.32**	**1.11-4.82**	**0.02**
Wheezing during exercise	1.41	0.60-3.30	0.43
At least 1 doctor, hospital, or emergency room visit for wheezing	1.01	0.39-2.64	0.99

Wheezing requiring prescription medication use	**2.23**	**1.10-4.54**	**0.03**
Limitation of usual activities due to wheezing	1.53	0.67-3.51	0.32
At least 1 day of school or work missed due to wheezing	1.15	0.36-3.63	0.82
Dry nocturnal cough	1.52	0.70-3.30	0.29
Seafood			
Wheezing in chest	1.17	0.72-1.89	0.53
At least 1 sleep disturbance due to wheezing	1.82	0.83-3.99	0.13
Wheezing during exercise	1.09	0.65-1.84	0.75
At least 1 doctor, hospital, or emergency room visit for wheezing	1.13	0.51-2.55	0.76
Wheezing requiring prescription medication use	1.46	0.71-3.02	0.30
Limitation of usual activities due to wheezing	1.11	0.60-2.06	0.75
At least 1 day of school or work missed due to wheezing	1.54	0.77-3.06	0.22
Dry nocturnal cough	1.08	0.63-1.85	0.78

Appendix - References

1. Saha A, Poojary P, Chan L, et al. Increased odds of metabolic syndrome with consumption of high dietary advanced glycation end products in adolescents. *Diabetes Metab* 2017;43(5):469-71. doi: 10.1016/j.diabet.2017.01.001 [published Online First: 2017/02/06]
2. Uribarri J, Woodruff S, Goodman S, et al. Advanced glycation end products in foods and a practical guide to their reduction in the diet. *J Am Diet Assoc* 2010;110(6):911-16 e12. doi: 10.1016/j.jada.2010.03.018 [published Online First: 2010/05/26]
3. Zipf G, Chiappa M, Porter KS, et al. National health and nutrition examination survey: plan and operations, 1999-2010. *Vital Health Stat 1* 2013(56):1-37. [published Online First: 2014/08/01]
4. Fryar CD, Wright JD, Eberhardt MS, et al. Trends in nutrient intakes and chronic health conditions among Mexican-American adults, a 25-year profile: United States, 1982-2006. *Natl Health Stat Report* 2012(50):1-20. [published Online First: 2012/07/19]
5. Barlow SE. Expert committee recommendations regarding the prevention, assessment, and treatment of child and adolescent overweight and obesity: summary report. *Pediatrics* 2007;120 Suppl 4:S164-92. doi: 10.1542/peds.2007-2329C [published Online First: 2007/12/18]
6. Kuczmarski RJ, Ogden CL, Guo SS, et al. 2000 CDC Growth Charts for the United States: methods and development. *Vital Health Stat 11* 2002(246):1-190. [published Online First: 2002/06/05]
7. Guenther PM, Casavale KO, Reedy J, et al. Update of the Healthy Eating Index: HEI-2010. *J Acad Nutr Diet* 2013;113(4):569-80. doi: 10.1016/j.jand.2012.12.016 [published Online First: 2013/02/19]
8. NIH - National Cancer Institute. Division of Cancer Control & Population Sciences. Healthy Eating Index SAS Code 2019 [Available from: https://epi.grants.cancer.gov/hei/sas-code.html accessed October 18, 2019.