DIETARY INTAKE AND OBESITY IN ASTHMA CONTROL AND PREVENTION

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ACADEMIC DISSERTATION

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"Someone once asked me what I regarded as the three most important requirements for happiness. My answer was: A feeling that you have been honest with yourself and those around you; a feeling that you have done the best you could both in your personal life and in your work; and the ability to love others. But there is another basic requirement: that is the feeling that you are, in some way, useful. Usefulness, whatever form it may take, is the price we should pay for the air we breathe and the food we eat and the privilege of being alive. And it is its own reward, as well, for it is the beginning of happiness. "

Eleanor Roosevelt, in "You Learn by Living: Eleven Keys for a More Fulfilling Life"

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CONTENTS

The present thesis is based on the following publications:

STUDY I

Barros R, Moreira A, Fonseca J, Delgado L, Graça Castel-Branco M, Haahtela T, Lopes C, Moreira P. Dietary intake of a-linolenic acid and low ratio of n-6:n-3 PUFA are associated with decreased exhaled NO and improved asthma control. *Br J Nutr*. 2011; 29:1-10.

STUDY II

Barros R, Moreira A, Padrão P, Teixeira VH, Carvalho P, Delgado L, Lopes C, Severo M, Moreira P. Dietary patterns and asthma prevalence: evidence fom a National Health Survey. *Allergy 2013;* Vol.68 (Suppl.97): 542-542.
Submitted manuscript

STUDY III

Barros R, Moreira A, Padrão P, Teixeira VH, Carvalho P, Delgado L, Moreira P. Obesity increases prevalent and incident asthma and worsens asthma severity: evidence from the Portuguese National Health Survey. *Obesity Reviews 2010*; Vol.11, (Suppl.1): 55-56.
Submitted manuscript

STUDY IV

Barros R, Moreira A, Padrão P, Teixeira VH, Carvalho P, Delgado L, Moreira P. Height: are asthmatic different? *Allergy 2012;* Vol.67 (Suppl.96): 76-77.
Submitted manuscript

AUTHOR CONTRIBUTION:

STUDY I

Author planned the study together with Professor Pedro Moreira and Professor André Moreira; worked on the clinical patient's enrolment, and data assessment and collection, at the Immunoallergology Department, Hospital São João, EPE; worked on statistical analyses, interpretation and critical discussion of results; and wrote the first draft and critical review the final publication, in cooperation with research team (co-authors and supervisors).

STUDIES II, III AND IV

Author planned the studies together with Professor Pedro Moreira and Professor André Moreira, based on the Portuguese National Health Survey database; worked on statistical analyses, interpretation and critical discussion of results; and wrote the first drafts and critical review the final manuscripts, in cooperation with research team (co-authors and supervisors).

ABSTRACT

Asthma is one of the most common chronic pathological conditions throughout the world and represents a serious economic and social impact in the health care systems and patient´s quality of life. The increase of asthma prevalence in westernized societies has been suggested to be related to environment exposures and lifestyle changes in the last decades.

Several hypotheses have been proposed, including changes in dietary patterns and nutritional intake (namely in fatty acid profile and antioxidant micronutrients), and parallel increase in prevalence of obesity and physical inactivity. We previously reported that adherence to Mediterranean diet, a recognized healthy dietary pattern, have a protective role in asthma control in adult patients, defined by symptoms, lung function and airway inflammation. Traditional Mediterranean diet foods are food sources of antioxidants and n-3 polyunsaturated fatty acids, namely fruit and nuts, and emerged as positively associated with and asthma control and lung function, respectively. However, the study of the association between diet and asthma, by dietary patterns approach is still poorly addressed and results have been controversial.

The parallel increase in the prevalence of obesity and asthma lead to the interest in potential mechanisms linking these two epidemics. We have previous reported a negative association between obesity and airway inflammation, measured by exhaled NO, in asthmatic patients, providing support for the mechanical hypothesis linking obesity and asthma. However, obesity has been associated with asthma diagnosis, respiratory symptoms, poor lung function, increased airway hyperreactivity and worst treatment response, suggesting that obesity increases the risk of asthma and changes prevalent asthma toward a more difficult-to-control phenotype.

Additionally, chronic diseases (e.g. type 1 diabetes, food allergy and asthma) commonly affect growth, leading to an impaired final adult height. Moreover, socio-economic and education factors might also influence growth in asthma, and therefore height could be an indicator of health conditions and population´s lifestyle. Although inhaled corticosteroids are the first line treatment for persistent asthma, the long-term effect on growth and adult height is controversial and a clinical concern.

The general aim of this thesis was to investigate the association between lifestyle factors, such as nutritional intake, dietary patterns, body weight and height with asthma control and prevention.

We have proposed to primarily investigate: (1) the association between dietary intake of several types of fatty acids, antioxidant micronutrients, and asthma control measured by symptoms, lung function and airway inflammation, in asthmatic adult patients (study I); (2) the association between dietary patterns and asthma prevalence in a nationally representative population (study II); (3) the association between obesity and asthma prevalence in a nationally representative population (study III); (4) to compare height between asthmatics and non-asthmatics adults in a nationally representative population (study IV).

For the study I, a cross-sectional study was developed in adult asthmatics attending an outpatient Asthma and Allergy clinic at University Hospital. Dietary intake was obtained by a Food Frequency Questionnaire, and nutritional content was calculated using Food Processor Plus software (ESHA Research, Inc., Salem, OR, USA). Good asthma control was defined by the combination of forced expiratory volume during the first second, exhaled NO (eNO) and Asthma Control Questionnaire (ACQ) score. Gender, education, age, energy intake, body mass index, physical activity, smoking, atopy, rhinitis and inhaled corticosteroid were assessed and analysed as potential confounders. Multiple linear and unconditional logistic regression models were performed to analyse the associations between nutrients and asthma outcomes, adjusting for confounders.

For the studies II, III and IV, we analysed data from the 4th Portuguese National Health Survey. A representative sample of 41193 participants (32644 adults) was selected from households, using a multi-stage random probability design, according to the Portuguese territorial units. Following asthma definitions were used: ever asthma (ever medical doctor asthma diagnosis), current asthma (asthma within 12 months), current persistent asthma (asthma drugs within 12 months), current severe asthma (emergency because of asthma within 12 months), and incident asthma (diagnosis within 12 months). Dietary patterns were assessed by Latent Trait Model analysis, based on previous food-items at meals and snacks. Body mass index (BMI) was calculated based on self-reported weight and height and categorized according to WHO classification. Age, gender, education, family income, proxy reporting information, smoking, physical activity, and dietary patterns were analysed as confounders. Logistic regression were performed to analyse whether dietary patterns and BMI were associated with asthma prevalence, incidence and control in Portuguese adult population, adjusted for confounders; and generalized linear models by gender were performed to compare height between asthmatic and non-asthmatic participants.

The main results and conclusions of this thesis were:

(1) Higher intakes of n-3 polyunsaturated fatty acids and alpha-linolenic acid (ALA) were associated with good asthma control, while the risk for uncontrolled asthma increased with a higher n-6 to n-3 PUFA ratio. The present results introduce a protective effect of ALA in asthma control, independent of marine n-3 fatty acids, and provide a rationale to dietary intervention studies in asthma.

(2) A "*Fish, vegetables and fruit"* dietary pattern was inversely associated with current and persistent asthma prevalence. Conversely *"high fat, sugar and salt"* dietary pattern, based on high energy density/low micronutrient density foods, was associated with severe asthma prevalence, independent of other socio-economic and lifestyle factors. These data supports the rational for diet and lifestyle intervention studies in asthma based on whole dietary patterns and physical activity.

(3) Obesity is associated with increase prevalent and incident asthma, and seems to increase the odds of a more persistent and severe asthma phenotype independently of socio-demographic determinants, physical activity and dietary patterns.
(4) Adult height in asthmatics was significant lower than non-asthmatics, independent of socio-demographic determinants. These differences in height were consistent between genders, and seem to increase with persistency and severity of asthma. Monitoring health conditions that may impact stature in asthmatics should be recommended to optimize height attainment.

Taken together our studies provide further support to the lifestyle hypothesis in asthma, including dietary intake and obesity, with clinical impact on asthma control and prevention. As future perspectives we believe that the results of this thesis will provide rationale for future lifestyle intervention studies, based on whole dietary patterns and physical activity, and for the development of dietary guidelines in asthma, as a complementary public health nutrition strategy to asthma primary prevention, and a clinical complementary approach to asthma treatment.

A asma é uma das doenças crónicas mais prevalentes em todo o mundo e representa um impacto económico e social grave nos sistemas de saúde, bem como na qualidade de vida dos doentes. O aumento da prevalência de asma nas sociedades ocidentais, observado nas últimas décadas, tem sido relacionado com alterações na exposição do ambiente e no estilo de vida.

Várias hipóteses têm sido propostas, incluindo modificações nos padrões alimentares e na ingestão nutricional (nomeadamente ao nível do perfil lipídico e na ingestão de micronutrimentos antioxidantes), o aumento paralelo da prevalência de obesidade e do sedentarismo. Anteriormente, tínhamos mostrado que a adesão à Alimentação Mediterrânica, um modelo alimentar saudável cientificamente reconhecido, tem um papel protetor no controlo da asma em doentes adultos, quando definido pelos sintomas, função pulmonar e inflamação das vias aéreas. Alguns dos alimentos tradicionais da Alimentação Mediterrânica, nomeadamente a fruta e os frutos gordos e oleaginosos (fontes de antioxidantes e de ácidos gordos polinsaturados n-3), evidenciaram uma associação positiva com o controlo da asma e a função pulmonar, respetivamente. No entanto, o estudo da associação entre a alimentação e asma, através de uma abordagem de padrões alimentares, está ainda pouco explorado e os resultados têm sido controversos.

O aumento concomitante da prevalência da obesidade e da asma têm aumentado o interesse sobre os potenciais mecanismos que relacionem estas duas epidemias. Anteriormente, tínhamos mostrado uma associação negativa entre a obesidade e inflamação das vias aéreas, medida através do NO exalado, em doentes asmáticos obesos, o que suportou a hipótese mecânica de ligação entre obesidade e asma. No entanto, a obesidade tem sido associada ao diagnóstico de asma, sintomas respiratórios, diminuição da função pulmonar, aumento da hiperreatividade das vias aéreas e menor resposta ao tratamento, sugerindo que a obesidade aumenta o risco de asma e altera a prevalência da asma no sentido de um fenótipo de asma de difícil controlo.

Adicionalmente, a presença de doenças crónicas (como por exemplo, diabetes tipo 1, alergia alimentar e asma) parecem afetar frequentemente o crescimento, levando a uma diminuição na altura final em idade adulta. Além disso, fatores socioeconómicas e educacionais também podem influenciar o crescimento nos doentes asmáticos, contribuindo nesse sentido para que a altura possa também constituir um indicador das condições de saúde e de estilo de vida da população. Embora os corticosteroides inalados constituam a primeira linha de tratamento na asma persistente, o efeito a longo prazo no crescimento e na altura dos adultos é controversa e constitui uma preocupação clínica.

O objetivo geral deste trabalho foi investigar a associação de fatores relacionados com o estilo de vida, como a ingestão nutricional, padrões alimentares, o peso, e a altura, com o controlo e prevenção da asma.

Especificamente, nesta tese propusemo-nos principalmente a investigar: (1) a associação entre a ingestão de ácidos gordos, micronutrimentos antioxidantes e o controlo da asma, avaliado por sintomas, função pulmonar e inflamação das vias aéreas, em doentes adultos com asma (estudo I); (2) a associação entre padrões alimentares e a prevalência e incidência de asma, numa amostra representativa da população adulta a nível nacional (estudo II); (3) a associação entre obesidade e a prevalência e incidência de asma, numa amostra representativa da população adulta a nível nacional (estudo III); (4) a comparação da altura em idade adulta, entre adultos asmáticos e não-asmáticos, numa amostra representativa da população adulta a nível nacional (estudo IV).

No estudo I, foi desenvolvido um estudo transversal em doentes asmáticos adultos seguidos na consulta externa de Imunoalergologia do Hospital de S. João. A ingestão alimentar foi avaliada através de um questionário semi-quantitativo de frequência alimentar e a ingestão nutricional foi calculada utilizando o programa informático *Food Processor Plus (ESHA Research, Inc., Salem, OR, EUA)*. Um bom controlo da asma foi definido pela combinação de sintomas (volume expiratório forçado no primeiro segundo), de marcadores de inflamação das vias aéreas (NO no ar exalado) e pela pontuação final no Questionário de Controlo da Asma (ACQ). O género, a escolaridade, a idade, a ingestão energética total, o índice de massa corporal, a atividade física, tabagismo, atopia, rinite e tratamento com corticosteroides inalados foram analisadas e consideradas como potenciais variáveis confundidora. A análise estatística incluiu principalmente modelos de regressão linear múltipla e de regressão logística para estudar as associações entre a ingestão de nutrimentos e os *outcomes* da asma, ajustados para as variáveis confundidoras.

Nos estudos II, III e IV, foram analisados os dados do 4.º Inquérito Nacional de Saúde Português. A amostra incluiu uma amostra representativa de 41.193 participantes (32.644 adultos), e foi selecionada a partir de agregados familiares, através de uma metodologia probabilística aleatória multi-etapas, de acordo com as unidades territoriais portuguesas. Foram utilizadas as seguintes definições de asma: diagnóstico de asma (diagnóstico médico de asma), asma atual (asma nos últimos 12 meses), asma persistente (utilização de medicação para a asma nos últimos 12 meses), asma grave (recurso ao serviço de urgência por asma nos últimos 12 meses), e incidência de asma (diagnóstico médico de asma nos últimos 12 meses). Os padrões alimentares foram avaliados por através de *Latent Trait Analysis*, com base nos itens alimentares reportados nas refeições principais e merendas intercalares no dia anterior. O Índice de Massa Corporal (IMC) foi calculado com base no peso e altura referidos, e classificado de acordo com a classificação da Organização Mundial de Saúde. Foram analisadas como

potenciais variáveis confundidoras, a idade, o género, a escolaridade, o rendimento familiar, informação reportado por familiares, tabagismo, atividade física e padrões alimentares. A análise estatística incluiu principalmente modelos de regressão logística para análise da associação entre os padrões alimentares e o IMC, com a prevalência, incidência e controlo da asma na população adulta portuguesa, ajustada para variáveis confundidoras; e modelos de regressão linear, por género, para comparação da altura entre os participantes asmáticos e não-asmáticos.

Os principais resultados e conclusões da presente tese foram:

(1) Ingestões mais elevadas de ácidos gordos polinsaturados (AGP) n-3 e de ácido alfa-linolénico (ALA) foram associadas a um bom controlo da asma, enquanto um maior rácio de AGP n-6: n-3 foi associado a pior controlo da asma. Estes resultados introduziram um efeito protetor do ALA no controlo da asma, independente de outros AGP n-3, suportando a necessidade do desenvolvimento de estudos de intervenção alimentar na asma.

(2) O padrão alimentar "futa, produtos hortícolas e peixe" foi inversamente associado à prevalência de asma e de asma persistente. Por outro lado, o padrão alimentar "gordura, açúcar e sal", baseado por alimentos de elevada densidade energética e baixo valor nutricional, foi associado à prevalência de asma grave, independentemente de outros fatores socioeconómicos e fatores relacionado com o estilo de vida. Estes dados suportam a pertinência da realização de estudos de intervenção alimentar e de estilo de vida em doentes asmáticos, com base nos padrões alimentares e atividade física.

(3) A obesidade foi associada ao aumento da prevalência e incidência de asma, sugerindo um fenótipo mais grave e persistente de asma nos doentes asmáticos com obesidade, independentemente de determinantes sociodemográficos, dos padrões alimentares e da atividade física.

(4) A altura dos adultos asmáticos foi significativamente inferior a dos participantes não-asmáticos, independentemente de determinantes sociodemográficos. Estas diferenças na altura foram consistentes entre os dois géneros e parecem aumentar com a persistência e gravidade da asma, recomendando a monitorização de condições de saúde que podem afetar a estatura em doentes asmáticos, de modo a otimizar o crescimento e a altura na idade adulta.

Tendo em consideração os resultados obtidos, o presente estudo fornece dados adicionais que suportam a hipótese de estilo de vida na asma, incluindo a alimentação e a obesidade, com

impacto clínico na prevenção e controlo da asma. No futuro, os resultados desta tese suportam a pertinência da realização de estudos de intervenção com base nos fatores de estilo de vida, nomeadamente com base nos padrões alimentares e atividade física, e o desenvolvimento de recomendações alimentares na asma, como estratégias complementares de saúde pública na prevenção primária de asma, e como abordagem clínica ao tratamento da asma.

ACQ	Asthma control questionnaire
ALQ	Asthma life quality test
ATS	American Thoracic Society
BMI	Body mass index
EPA	Eicosapentaenoic acid
DHA	Docosahexanoic acid
CI	Confidence interval
DP	Dietary patterns
FEV1	Forced expiratory volume during the first second
FFQ	Food Frequency Questionnaire
ICS	Inhaled corticosteroid
IPAQ	International Physical Activity Questionnaire
MET	Standard metabolic equivalent
MLR	Multiple linear regression
MUFA	Monounsaturated fatty acids
LR	Linear regression
LTM	Latent Trait Model
NO	Nitric oxide
NHS	National Health Survey
NIS	National Institute of Statistics
PA	Physical activity
Ppb	Parts per billion
OR	Odds ratio
SD	Standard deviation
SFA	Saturated fatty acids
WHO	World Health Organization

ASTHMA OVERVIEW

Asthma is a complex and heterogeneous disease, usually characterized by chronic airway inflammation, and defined by history of respiratory symptoms such as wheeze, short of breath, chest tightness and cough, that vary over the time and intensity, together with variable airflow limitation (1, 2). Asthma is a multifactorial disease, wherein environmental factors play a determinant role in development and expression of asthma, even though gene-environment interactions could be complex. Additionally, recognizable clusters of demographic, clinical and pathophysiological characteristics are now often called asthma phenotypes (3, 4). Asthma is an inflammatory disorder of the airways, which involves multiple inflammatory cells and mediators that contribute to characteristic clinical and pathophysiological changes (1, 5). This inflammation is strongly associated with early life exposures (6), airway hyper-responsiveness and asthma symptoms. The pattern of inflammation in allergic asthma is predominantly characterized by T helper 2 lymphocytes (Th2) inflammatory phenotype, with a predominance of th2 cytokines, such as TNF-α and interleukins (IL-4, IL-5, IL-9 and IL-13), increased IgE concentrations, activated mast cells, and eosinophil-mediated inflammation (1, 7). In some cases, especially severe asthma, neutrophils may also contribute to innate response (8, 9).

Being one of the most common chronic pathological conditions throughout the world, asthma represents a serious economic and social impact in the health care systems and patient´s quality of life. Despite advances in knowledge, management and treatment of disease, asthma control is still difficult to attend (10). Asthma is a problem worldwide affecting 300 million individuals and causing 346,000 deaths worldwide every year (11, 12). The lack of a precisely and consensual definition of asthma difficult reliable comparison of reported prevalence's from the different parts of the world, however global prevalence seems to range between 1-16% (12-14), with a decrease in western Europe and increase in low income regions were prevalence were previous low (15).

The increase of asthma prevalence in westernized societies has been suggested to be related to environment exposures and lifestyle changes in the last decades (16, 17). Several hypotheses have been proposed including changes in indoor environment and exposure to the house dust mite; hygiene and cleanliness, associated with reduced exposure to microorganisms and widespread antibiotic therapy, as highlighted by the "hygiene hypothesis" (18-20); parallel increase in the prevalence of obesity (21); inactivity (22, 23); and changes in dietary patterns and nutritional intake, such as changes in fatty acids profile (24), decrease antioxidant micronutrients (25), and vitamin D intake and exposure (26, 27).

The prevalence of asthma and allergy has markedly increased in westernised countries since about 1960. Some western dietary pattern characteristics have been associated with asthma prevalence, namely by an increase consumption of processed foods (high energy density/ low micronutrient foods), a decrease in vegetable and fruits (antioxidant micronutrients), and by a decrease in milk and butter (saturated fatty acids) and fish (n-3 polyunsaturated fatty acids), follow by an increase in vegetable oils and margarines (n-6 fatty acids), increasing the n-6 to n-3 PUFA ratio (16, 27).

These antioxidant and lipid hypotheses have been supported by several epidemiological studies reporting beneficial associations between asthma risk and dietary intake of n-3 fatty acids, fish and nuts (28-30), antioxidant vitamins (A, C, D, E) and antioxidant rich foods such as fruit and vegetables (27), and more recently between also methyl donors through epigenetic mechanisms (folate, choline and vitamin B12) (31, 32). However RCTs and intervention studies with single nutrient supplements have been provided conflicting evidence (33-36), reflecting the complexity of dietary intake, and suggesting that looking at the whole foods and dietary patterns is an important shift in approaching the synergistic effects between nutrients, foods, dietary patterns and lifestyle.

We previous reported that Mediterranean diet, a recognized healthy dietary pattern characterized by antioxidant and anti-inflammatory properties (37-40), have a protective role in asthma control in adult patients, defined by symptoms, lung function and airway inflammation, measured by eNO (41). In addition, Mediterranean diet protection has been also observed in several epidemiological studies during pregnancy (42) and childhood (43). In the opposite, fast food intake was also associated with asthma symptoms and exercise-induced bronchial hyper responsiveness (BHR) in childhood (44), and more recently, data from the International Study of Asthma and Allergies in childhood (ISAAC) phase 3 reported that fast food intake (≥ 3 times per week) was associated with an increased risk of severe asthma in children and adolescents (45). Moreover, high-total fat meals, namely high-trans fatty acids meal, was associated with increase airway inflammation in asthmatic patients, resulted by an increase in sputum neutrophils and an increased TLR4mRNA expression in sputum cells, supporting that high fat intake activates innate immune response; this fat induced inflammation also impairs the airway response to bronchodilator, worsening clinical outcomes, namely in non-atopic patients (46).

The assessment of dietary intake in nutritional epidemiology is complex could be confound by lifestyle and other factors (27, 47). Considering the importance of the whole picture of diet, several authors have proposed assessing dietary patterns, and exploratory approaches have been widely used to derive dietary patterns, namely exploratory factor analysis, principal component analysis and cluster analysis (48). More recently, it has been proposed to derive dietary patterns by using other exploratory methods, however Latent Models analysis is still little used in this field (49, 50). Studies addressing the association between diet and asthma by dietary patterns are still scarce and have been providing conflicting results, mainly during pregnancy (51-53). In children “western dietary pattern” was associated with asthma symptoms (54), while in Japanese university

students (55) “Fast food and soft drinks dietary pattern” was associated wheeze, and “Fish, fruit and milk” was associated with educe the asthma risk. In the E3N female study, “western dietary pattern” was also associated with increasing risk of reporting asthma attacks (56). The results from the three countries in the European Community Respiratory Health Survey-II, in five study centres (Germany, UK and Norway) were inconclusive, probably due to the heterogeneity and the uncontrolled confounding of a multi-centre observational study. However, study concludes and report a potential association between diet and asthma, with beneficial effects of fish, fruits and vegetable consumption on current asthma and asthma symptoms (57), strengthening the relevance of interpret results with caution when studies are cross-sectional and confounders were unmeasured and the relevance of dietary patterns studies.

OBESITY AND ASTHMA

Asthma is a multifactorial disease, wherein environmental factors play a determinant role in development and expression of asthma, even though gene-environment interactions and immune function could be also important. Despite advances in knowledge, management and treatment of this disease, asthma control stills difficult to attend (10). The concomitant increase in the prevalence of obesity and asthma lead to the interest in potential mechanisms linking these two epidemics (21). Increases in body mass index (BMI) have been associated with increased prevalence of asthma, however the mechanisms behind this association are not totally clear (58, 59).

Obesity has been suggested as a pro-inflammatory state (60), but the links with airway inflammation are still scarce. Plausible links between asthma and obesity include mechanical factors, inflammatory conditions and stress models. The increased abdominal and chest wall mass causes decreased functional residual capacity and reduced lung and tidal volumes (61, 62). Obesity is also a state of chronic and low-grade systemic inflammation with increased levels of the pro-inflammatory leptin and plasminogen activator inhibitor and decreased serum levels of the protective anti-inflammatory adiponectin (63) . The relation between BMI and exhaled NO has provided conflicting evidence. In children, BMI had no association with exhaled NO (64-66) while in non-asthmatic and non-obese adults a positive association has been reported (67). Recently, a case-control study described a positive association in normal weight and obese healthy adults, but no significant association was observed in asthma group. Although BMI was correlated with serum leptin levels in both groups, exhaled NO was not related also with serum leptin levels in asthmatic patients (68). We have previous reported a negative association between body mass index and exhaled nitric oxide in overweight and obese asthmatics, providing additional support to the mechanical hypothesis linking obesity and asthma (69).

Several cross-sectional and case-control studies have found obesity to be associated with asthma diagnosis, respiratory symptoms, poor lung function and increased airway hyperreactivity (AHR) (70). In addition, weight reduction in obese patients with asthma has been demonstrated

to improve lung function, symptoms, morbidity, and health status (71), and better asthma control. Moreover, weight loss has been associated with better asthma control and reduced asthma exacerbations (72), and data from meta-analysis showed that being overweight or obese increased the odds of incident asthma in a dose-dependent way (73). A randomized trial to compare effects of dietary restriction and exercise in obese asthmatic patients, shows that a 5-10% weight loss resulted in clinically improvements to asthma control (58%) and quality of life in 83% of patients, with reduced in neutrophilic airway inflammation, suggesting that reducing of weight is recommended for clinical management of obese-asthma phenotype (74). A recent meta-analysis from the European Academy of Allergy and Clinical Immunology showed that weight gain, more than be obese, almost double the odds of incident asthma, suggesting weight loss interventions in overweight population to prevent asthma incidence and improve asthma control in asthmatic patients with obesity (75).

Severe asthma is considered a heterogeneous disease in which a variety of clinical, physiological and inflammatory markers determine the disease severity, with distinct clinical phenotypes classification (76). Evidence suggests that obesity increases the risk of asthma and changes prevalent asthma toward a more difficult-to-control phenotype (1, 75, 77). Moreover, obesity have impact on the asthma treatment response with increased severity of illness (78). Inhaled corticosteroids (ICS) are the first line treatment for persistent asthma (79), however the long-term effect of ICS on growth and adult height is a clinical concern (80, 81). Additionally, children with chronic diseases (e.g. type 1 diabetes, food allergy and asthma) are commonly affected by a variable degree of growth failure, leading to an impaired final adult height (82). However, the long term effects of asthma in adult height are still not clear.

We have previous reported that obesity was not related with asthma prevalence in the previous data from the National Health Survey (1998-99) (83), suggesting that further research should considerer also other clinical outcomes, socio-economic determinants (84), and lifestyle factors such as dietary intake and physical activity, underlying the recent changes in obesity and asthma phenotype.

The general aim of the present thesis was to investigate the association between lifestyle factors, as dietary intake and obesity, in asthma prevalence, incidence and control in adults.

Therefore, we have proposed to primarily investigate:

(1) The association between dietary intake of several types of fatty acids, antioxidant micronutrients, and asthma control measured by symptoms, lung function and airway inflammation, in asthmatic adult patients (study I);

(2) The association between dietary patterns and asthma in a nationally representative population (study II);

(3) The association between obesity and asthma prevalence in a nationally representative population (study III).

(4) To compare height between asthmatics and non-asthmatics adults in a nationally representative population (study IV).

This thesis was based on data from a clinical sample attending Immunoallergology Department, at Hospital de São João, EPE (study I), and data from the 4th Portuguese National Health Survey (studies II, III and IV).

STUDY I

Participants and study design

Two hundred and nineteen consecutive patients, older than 16 years old, attending an outpatient Asthma and Allergy clinic at a University Hospital, with medical diagnosis of asthma, were invited to participate in a cross-sectional study. Exclusion criteria were: food allergy, changing of dietary patterns in the last 12 months; pregnancy; presence of diseases which involved specific nutritional therapy and dietary planning; acute illness in the last four weeks; or inability to comply with the measurement instruments. This study was conducted according to the guidelines laid down in the Declaration of Helsinki and all procedures involving human patients were approved by the Institutional ethics committee. Written informed consent was obtained from all patients previous to inclusion.

Nutritional intake

Dietary intake was obtained by a self-administered, semi-quantitative food frequency questionnaire (FFQ), validated for Portuguese adults (85). The FFQ is an 86-item questionnaire that assess usual dietary intake over the previous 12 months, including usual foods groups and beverages. Food intake was estimated by multiplying frequency of consumption (about 9 possibilities from “never or less than 1 time per month”, to “6 or more times a day”), by the weight of the standard portion size of the food-item. A seasonal variation factor was considered for foods which production and consumption is not regular over the year (mean of 3 months). Nutritional intake was calculated using an adapted Portuguese version of the software Food Processor Plus® (ESHA Research Inc, Salem, Oregon, USA), a nutritional analysis software that converts food intake into total energy and nutrients, based on food composition tables available from the USDA (United States Department of Agriculture) and National data from typically Portuguese foods. Dietary intake of different types of fatty acids (n-3 and n-6 PUFA, SFA and MUFA) and micronutrients involved in antioxidant status and potentially relevant for asthma (vitamins E, C, carotene, retinol, magnesium and zinc) (16, 86), were selected as primary independent variables of interest. Although selenium as a role as cofactor of glutathione peroxidise and potential suppressor of asthma inflammation, it was not considered in the nutritional analysis. Considering the wide variation in the content of the major selenium food sources (depending of geographic origin and soil levels), food composition data for selenium measure by FFQ are considered unreliable (87).

Anthropometry and physical activity assessment

Body mass index (BMI) was calculated after body weight and height measurements with the subject lightly clothed and bare-footed, using a mechanical balance with stadiometer, Seca model 700® (Seca Headquarter, Hamburg, Germany). Weight and height were determined to the nearest 0.1 kilogram and 0.5 centimetres, respectively. BMI was calculated as weight (kilograms) divided by the square of height (metres).

Physical activity (PA) was measured using the International Physical Activity Questionnaire (IPAQ) – short version (88). The short 7 days self-administered version is a 7-item questionnaire that provides information about frequency and duration of 4 domains: sedentary activity, time spent walking and moderate- and vigorous-intensity PA. Physical activity within domains was estimated by weighting the reported frequency (events per week) by duration (minutes per event) and by a MET level assigned to each activity (Walking = 3.3; Moderate-intensity PA = 4.0; and Vigorous-intensity PA = 8.0). A combined total physical activity was computed as the sum of the activity domains scores (Total PA = Walking + Moderate-intensity PA + Vigorous-intensity PA) and reported as a continuous measure (Total PA score = total MET-min/ week).

Asthma control and quality of life: definitions and assessment

Asthma Control was defined combining the results of lung function, exhaled NO, and the Asthma Control Questionnaire (ACQ) score (89). Subjects were classified as having “controlled” asthma if simultaneously had FEV1 ≥ 80% predicted (90, 91), exhaled NO ≤ 35ppb (92), and ACQ score below 1.00 (93). If any of these features was not present, subject were classified as “non-controlled”. Lung function was measured by determination of forced expiratory volume 1 second (FEV1) using the PIKO-1® (Ferraris Respiratory Europe Ltd, Hertford, UK) (94). Patients were asked to perform a set of three technically acceptable manoeuvres and the highest FEV1 measurement was registered and expressed as percent predicted, as recommended by the American Thoracic Society.

Exhaled nitric oxide (NO) was measured with the NIOX® system (Aerocrine, Stockholm, Sweden), using the online technique recommended by the American Thoracic Society (ATS) (95), at a flow rate of 50 ml per second.

The 7-item ACQ was designed to assess the clinical asthma control during the previous week. A 7-point scale (0=no impairment, 6= maximum impairment) was used and score was calculated as the mean of the 7 items, ranging from 0 (totally controlled) to 6 (severely uncontrolled) (96).

Asthma quality of life was measured by the Asthma Life Quality Test (ALQ), developed by the American College of Allergy, Asthma and Immunology, and validated in Portuguese (97). The self-administered ALQ, includes 20 questions of dichotomous answer (yes/no) assessing 6 domains: activity and sleep; symptoms; triggers; unscheduled health care use; medication; and

psychological. Total score was calculated as the sum of affirmative responses, ranging from 0 to 20 (lower values indicate better asthma quality of life).

Statistical analysis

Mean (SD) and proportions (%) were used as descriptive statistics, except for physical activity and several nutrients data that were presented as median (range) given the non-normal distribution. Exhaled NO was logarithmical transformed to attain normal distribution and presented as geometric mean and 95% confidence intervals. Atopic status, defined by positive skin prick tests, medical diagnosis of allergic rhinitis, current use of inhaled corticosteroid (ICS), education (≤ 4, 5 to 9, and ≥ 10 years) and smoking status (non-smoker, past smoker and current smoker) were also recorded.

Nutritional variables were adjusted for total energy intake using the nutrient residual model (98). In this model, energy-adjusted nutrient intakes are computed as the residuals from the regression analysis, with total energy intake as independent variable and absolute intakes as dependent variables. The associations between nutritional intake and asthma outcomes were performed using linear regression (LR), multiple linear regression (MLR) and unconditional logistic regression models. LR was initially fitted to analyse the associations between nutrient intake (independent variables) and asthma outcomes (dependent variables). MLR models adjusted for confounders were performed separately for exhaled NO, FEV1, ALQ and ACQ scores (categorical confounder variables were transformed into dummy variables). Logistic regression models were also performed to analyse the associations between nutritional intake and asthma control level. Nutrients energy-adjusted were categorised into tertiles. Odds ratios (OR) were calculated by reference with the lowest tertile.

Gender, education, age, energy intake, BMI, PA score, smoking, atopy, rhinitis and ICS were analysed as potential confounders. Only the variables significantly associated with each of asthma outcomes in univariated analysis were considered in the final regression and logistic models. Considering that smoking status and physical activity were not significant associated with exhaled NO, FEV1, ALQ or ACQ scores, and that their inclusion as confounders did not influenced the effects, these variables were therefore not included in final models. Considering the biological plausibility related with dietary intake, gender, age and total energy intake, these were considered in all models. A 0.05 level of significance and 95% confidence intervals (95% CI) were considered. The data analysis was performed using the statistical package SPSS ®, 17.0 version (SPSS Inc; Chicago, IL).

Participants and study design

Data from the 4th Portuguese National Health Survey (NHS), carried out by the National Institute of Health and National Institute of Statistics (NIS), between 2005 and 2006, was analysed. The methodology of the NHS has been previously described (99, 100). In summary, the sampling frame was made based on census data and participants were selected from households during that period, using a multi-stage random probability design (hospitals, prisons, military houses and community care institutions were excluded). A representative sample of 41193 participants was included, according to the 7 main Portuguese territorial units (NUTS II), namely *North, Center, Lisbon region, Alentejo, and Algarve, and Madeira and Azores archipelagos*. The primary sampling unit (PSU) was the housing unit and sampling was based on the population and housing census. Two levels were defined within each NUTSII: the parish (corresponding to townships); and geographically defined units of 240 lodgings (within parish). The PSUs were then randomly selected within each territorial unit, and subjects living in the sampling unit were surveyed. Interviews were conducted in the households by trained staff interviewers, and the questionnaire included information on social and demographic characteristics, health, and chronic diseases including asthma. The survey response rate was 76%, as reported by NIS (99, 100). For the purposes of this study, a representative subsample of all NHS surveyed adults (≥20 years-old) was analysed (32644 participants).

Asthma

The following asthma definitions were used: ever asthma (ever medical doctor asthma diagnosis), current asthma (asthma within previous 12 months), current persistent asthma (asthma drugs within previous 12 months), current severe asthma (emergency because of asthma within 12 months), and incident asthma (diagnosis within previous 12 months).

Dietary intake

Dietary patterns were identified by Latent Trait Models (LTM), based on a list of 20 dichotomous questions (“Did you consumed any of the following foods yesterday?”), addressing the previous day dietary intake of usual foods and beverages at meals (milk, cheese and yogurt; vegetable soup; bread and cereal products; meat; fish; potatoes, rice and pasta; pulses; vegetables; fruit; and pastry, chocolates or sweet desserts), and snacks (fruit; bread or sandwiches; milk, cheese and yogurt; juices; soft drinks; pastry, chocolates or sweet desserts; other candies; salty snacks; chips; and alcoholic beverages).

Body mass index

Body mass index (weight/height^2) was calculated based on self-reported weight (kilograms) and height (meters), and categorized according to the World Health Organization BMI Classification (101), in underweight (<18.5 kg/m^2), normal weight (18.5-24.9 kg/m^2), overweight (25.0-29.9 kg/m^2), obesity class I (30.0-34.9 kg/m^2), class II (35.0-39.9 kg/m^2) and class III (≥ 40.0 kg/m^2). Considering the low prevalence of underweight and the clinical higher risk of morbid obesity, for descriptive and logistic regression analyses, BMI classes were also categorized into fourth classes: underweight and normal weight; overweight; obesity class I and class III; and obesity class III.

Socio-demographic and lifestyle

Age, gender, education, family income, proxy reporting information, smoking, physical activity level were also addressed by NHS, and analysed as potential confounders.

Statistical analysis

Study II

Descriptive analysis, including mean (standard deviation) and proportions, was performed to characterize the sample, and conducted considering the sampling weights.

Dietary patterns were identified by Latent Variable Models (49, 50), based on addressed food items intake at meals and snacks. Latent models can be divided into two frameworks: latent trait models (LTM), which assume that the latent variables are metrical (factors), and latent class models (LCM) that consider latent variables as categorical (clusters). Latent trait model is a Binary Data Factor Analysis that considers one or more factors. LTM assume that the dietary intake of each individual is explained by one or more factors, usually called 'latent variables'. Interpretation of the model is done considering the standardized factor loadings, witch expresses the correlation coefficient between the latent variable and an underlying continuous variable obtained from each food item. Coefficient correlations (r) > 0.350 and < -0.350 were considered for selecting food items. Geominrotation was applied to simplify the standardized factor loadings matrix, and the following criteria to determine the number of latent variables in the model was considered: the model with lowest value in the Bayesian information criteria, the global goodness of fit of the considered latent models assessed through the likelihood ratio test, via parametric boostrapping (100 samples) and the residuals inspection.

Unconditional logistic regression models were performed to analyse whether dietary patterns were associated with asthma prevalence, adjusted for confounders (age, gender, BMI, education, family income, proxy reporting information, smoking and physical activity). A 0.05 level of

significance and 95% CI were considered. Data analysis was performed using the statistical package SPSS®, version 18.0 (SPSS Inc., Chicago, IL, USA), and the MPlus (v5.2; Muthen & Muthrn, Los Angeles, California, USA).

Study III

Descriptive analysis, including mean (standard deviation) and proportions, was performed to characterize the sample, and were conducting with sampling weights. Student's t-tests, ANOVA, and chi-squared tests were performed to determine the association between obesity, asthma, and confounder variables. Unconditional logistic regression models were performed to analyse whether classes of BMI were associated with asthma prevalence and incidence. Age, gender, education, family income, proxy reporting information, smoking, physical activity and dietary patterns were analysed as confounders. A 0.05 level of significance and 95% CI were considered. Data analysis was performed using the statistical package SPSS®, version 18.0 (SPSS Inc., Chicago, IL, USA).

Study IV

Descriptive analysis, including mean (standard deviation) and proportions, was performed to characterize the sample, and were conducting with sampling weights. Hypothesis test for comparing height means between groups. Self-reported height (m) was analysed by gender and generalized linear models (GLM) were performed to compare height between asthmatic and non-asthmatic participants, adjusted for confounders. Age, education, family income, proxy reporting information, and weight were analysed as confounders. A 0.05 level of significance and 95% CI were considered. Data analysis was performed using the statistical package SPSS®, version 18.0 (SPSS Inc., Chicago, IL, USA).

STUDY I

Barros R, Moreira A, Fonseca J, Delgado L, Graça Castel-Branco M, Haahtela T, Lopes C, Moreira P. Dietary intake of a-linolenic acid and low ratio of n-6:n-3 PUFA are associated with decreased exhaled NO and improved asthma control. *Br J Nutr*. 2011; 29:1-10.

STUDY II

Barros R, Moreira A, Padrão P, Teixeira VH, Carvalho P, Delgado L, Lopes C, Severo M, Moreira P. Dietary patterns and asthma prevalence: evidence from the Portuguese National Health Survey.

Manuscript submitted

STUDY III

Barros R, Moreira A, Padrão P, Teixeira VH, Carvalho P, Delgado L, Moreira P. Obesity increases prevalent and incident asthma and worsens asthma severity: evidence from the Portuguese National Health Survey.

Manuscript submitted

STUDY IV

Barros R, Moreira A, Padrão P, Teixeira VH, Carvalho P, Delgado L, Moreira P. Height: are asthmatic different?

Manuscript submitted

Barros R, Moreira A, Fonseca J, Delgado L, Graça Castel-Branco M, Haahtela T, Lopes C, Moreira P. Dietary intake of a-linolenic acid and low ratio of n-6:n-3 PUFA are associated with decreased exhaled NO and improved asthma control. *Br J Nutr*. 2011; 29:1-10.

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doi:10.1017/S0007114511000328

Dietary intake of α-linolenic acid and low ratio of *n*-6:*n*-3 PUFA are associated with decreased exhaled NO and improved asthma control

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(Received 16 August 2010 – Revised 13 January 2011 – Accepted 17 January 2011)

Abstract

As recently described, adherence to the Mediterranean diet is associated with improved asthma control. However, evidence of how specific nutrients such as fatty acids and antioxidants may affect this relationship remains largely unknown. We aimed to examine the association between dietary intake of fatty acids and antioxidants and asthma control. A cross-sectional study was developed in 174 asthmatics, mean age of 40 (SD 15) years. Dietary intake was obtained by a FFQ, and nutritional content was calculated using Food Processor Plus™ software (ESHA Research, Inc., Salem, OR, USA). Good asthma control was defined by the combination of forced expiratory volume during the first second, exhaled NO (eNO) and Asthma Control Questionnaire (ACQ) score (control: forced expiratory volume in the first second ≥80 %; eNO ≤35 ppb; ACQ <1·0, scale 0–6 score). Multiple linear and logistic regression models were performed to analyse the associations between nutrients and asthma outcomes, adjusting for confounders. A high *n*-6:*n*-3 PUFA ratio predicted high eNO, whereas high intakes of *n*-3 PUFA, α-linolenic acid (ALA) and SFA were associated with low eNO. Odds for controlled asthma improved along with an increased intake of *n*-3 PUFA (OR 0·14, 95 % CI 0·04, 0·45; *P* for trend=0·001), SFA (OR 0·36, 95 % CI 0·13, 0·97; *P* for trend=0·047) and ALA (OR 0·18, 95 % CI 0·06, 0·58; *P* for trend=0·005). A high *n*-6:*n*-3 PUFA ratio increased the odds for uncontrolled asthma (OR 3·69, 95 % CI 1·37, 9·94; *P* for trend=0·009), after adjusting for energy intake, sex, age, education and use of inhaled corticosteroids. Higher intakes of *n*-3 PUFA, ALA and SFA were associated with good asthma control, while the risk for uncontrolled asthma increased with a higher *n*-6:*n*-3 PUFA ratio. The present results introduce a protective effect of ALA in asthma control, independent of marine *n*-3 fatty acids, and provide a rationale to dietary intervention studies in asthma.

Key words: Asthma: Airway inflammation: Diet: Fatty acids: α-Linolenic acid: Antioxidants

The increase in asthma prevalence in the last decades has been suggested to be related to environment and lifestyle changes, from which diet and physical activity (PA) appear as obvious[1]. The following two dietary hypotheses have been proposed: (1) an increase in the consumption of vegetable oils and margarines and a decline in fat of animal origin and fish consumption shifting the *n*-6:*n*-3 ratio of dietary PUFA from 1:1 to 15–17:1[2]; (2) a decrease in the intake of fresh fruit, vegetables and whole cereals leading to a reduction in dietary antioxidants[3]. Several non-experimental studies have provided evidence supporting the lipid[2] and the antioxidant[3] hypotheses; however, the same has not been reported in intervention trials. Potential benefits of marine *n*-3 PUFA, namely EPA (20 : 5*n*-3) and DHA (22 : 6*n*-3), in inflammatory modulation and asthma have been proposed, whereas the link with the precursor α-linolenic acid (ALA; 18 : 3*n*-3) is still scarce.

We have recently reported that high adherence to a Mediterranean dietary pattern was associated with improved asthma control[4]. This was particularly relevant as asthma control definition incorporated symptoms, lung function

Abbreviations: ACQ, Asthma Control Questionnaire; ALA, α-linolenic acid; eNO, exhaled NO; FEV1, forced expiratory volume in the first second; PA, physical activity.

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and airway inflammation[5]. Among Mediterranean diet food items, nuts (high in ALA) and fresh fruit emerged as positively associated with lung function and asthma control, respectively. However, few data exist on the associations of individual fatty acids, micronutrients and asthma control.

In the present study, we aimed to investigate the association between several types of fatty acids, antioxidant micronutrients and asthma control, measured by symptoms, lung function and airway inflammation, and we hypothesised that *n*-3 PUFA and antioxidant micronutrients provided from the diet could be associated with improved asthma control in asthmatic patients.

Materials and methods

Participants and study design

A total of 219 consecutive patients, older than 16 years old, attending an outpatient Asthma and Allergy clinic at a University Hospital, with a medical diagnosis of asthma, were invited to participate in a cross-sectional study. Exclusion criteria were food allergy, changing of dietary patterns in the last 12 months, pregnancy, presence of diseases which involved specific nutritional therapy and dietary planning, acute illness in the last 4 weeks or inability to comply with the measurement instruments. The present study was conducted according to the guidelines laid down in the Declaration of Helsinki, and all procedures involving human patients were approved by the Institutional ethics committee. Written informed consent was obtained from all patients before inclusion.

Nutritional intake

Dietary intake was obtained by a self-administered, semi-quantitative FFQ, validated for Portuguese adults[6]. The FFQ is an 86-item questionnaire that assessed usual dietary intake over the previous 12 months, including usual food groups and beverages. Food intake was estimated by multiplying the frequency of consumption (about nine possibilities from 'never or less than 1 time/month', to '6 or more times/d') by the weight of the standard portion size of the food item. A seasonal variation factor was considered for foods in which production and consumption are not regular over the year (mean of 3 months). Nutritional intake was calculated using an adapted Portuguese version of the software Food Processor Plus® (ESHA Research, Inc., Salem, OR, USA), nutritional analysis software that converts food intake into total energy and nutrients, based on food composition tables available from the US Department of Agriculture and national data from typical Portuguese foods. Dietary intake of different types of fatty acids (*n*-3 and *n*-6 PUFA, SFA and MUFA) and micronutrients involved in antioxidant status and potentially relevant for asthma (vitamins E, C, carotene, retinol, Mg and Zn)[1] were selected as primary independent variables of interest. Although Se plays a role as a cofactor of glutathione peroxidase and as the potential suppressor of asthma inflammation, it was not considered in the nutritional analysis. Considering the wide variation in the content of the major Se food sources (depending on the geographic origin and soil levels), food composition data for Se measure by FFQ are considered unreliable[7].

Anthropometry and physical activity assessment

BMI was calculated after body weight and height measurements with the subject lightly clothed and barefooted, using a mechanical balance with a stadiometer (Seca model 700®; Seca Headquarter, Hamburg, Germany). Weight and height were determined to the nearest 0·1 kg and 0·5 cm, respectively. BMI was calculated as the weight (kg) divided by the square of the height (m^2).

PA was measured using the International Physical Activity Questionnaire – short version[8]. The short 7 d self-administered version is a seven-item questionnaire that provides information about the frequency and duration of four domains: sedentary activity, time spent walking, and moderate- and vigorous-intensity PA. PA within domains was estimated by weighting the reported frequency (events/week) by duration (min/event) and by a metabolic equivalent level assigned to each activity (walking = 3·3; moderate-intensity PA = 4·0 and vigorous-intensity PA = 8·0). A combined total PA was computed as the sum of the activity domain scores (total PA = walking + moderate-intensity PA + vigorous-intensity PA) and reported as a continuous measure (total PA score = total metabolic equivalent-min/week).

Asthma control and quality of life: definitions and assessment

Asthma control was defined by combining the results of lung function, exhaled NO (eNO) and the Asthma Control Questionnaire (ACQ) score[5]. Subjects were classified as having 'controlled' asthma if simultaneously they had forced expiratory volume in the first second (FEV1) ≥80 % predicted[9], eNO ≤35 ppb[10] and ACQ score below 1·00[11]. If any of these features were not present, subjects were classified as 'non-controlled'. Lung function was measured by the determination of FEV1 using PIKO-1® (Ferraris Respiratory Europe Limited, Hertford, Herts, UK)[12]. Patients were asked to perform a set of three technically acceptable manoeuvres, and the highest FEV1 measurement was registered and expressed as percentage predicted, as recommended by the American Thoracic Society.

eNO was measured with the NIOX® system (Aerocrine, Stockholm, Sweden), using the online technique recommended by the American Thoracic Society[13], at a flow rate of 50 ml/s.

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Table 1. Characteristics of participants according to asthma control*

(Mean values, standard deviations, number of subjects and percentages)

	Controlled asthma (*n* 40)		Non-controlled asthma (*n* 134)		
	Mean	SD	Mean	SD	*P*
Demographic					
Age (years)	42·9	13·4	39·7	15·6	0·206†
Sex (*n*)					0·445‡
Female	31		111		
Male	9		23		
Education					0·723‡
≤4 years					
n	14		55		
%	35		41		
5–9 years					
n	10		34		
%	25		25		
≥ 10 years					
n	16		45		
%	40		34		
BMI (kg/m^2)	26·8	4·5	27·3	5·3	0·620†
Physical activity (MET-min/week)					0·328‖
Median	1405		1844		
Range	0–8739		0–9492		
Present smoker					0·119‡
n	6		7		
%	15		5		
Clinical					
Atopic					0·138‡
n	24		97		
%	65		77		
Allergic rhinitis					0·753‡
n	26		90		
%	65		68		
Present ICS					0·007‡**
n	22		103		
%	55		77		
Exhaled NO (ppb)					<0·001‖**
Mean	19·5		33·0		
95 % CI	16·8–22·6		28·8–37·9		
FEV1	103·8	22·3	82·7	22·3	<0·001†**
ALQ score	10·1	3·8	11·7	4·2	0·027†**
ACQ score	0·4	0·3	1·5	1·0	<0·001†**
Nutritional intake					
Total energy intake (kJ/d)	14 121	4091·95	13 459·92	5133·76	0·457†
Protein (% TEV)	16·3	2·9	15·8	3·7	0·433†
Total carbohydrates (% TEV)	35·6	6·7	35·0	7·6	0·667†
Sugars§	15·0	5·8	14·5	5·2	0·638†
Total fat (% TEV)	46·2	8·7	43·7	9·6	0·148†
PUFA	6·2	1·4	6·3	1·6	0·752†
n-6	4·9	1·4	5·1	1·5	0·614†
n-3	0·7	0·1	0·6	0·2	0·090†
ALA (g)	2·09	0·73	1·80	0·77	0·032†
EPA (g)					0·016‖
Median	0·12		0·09		
Range	0·00–0·40		0·00–1·70		
DHA (g)					0·021‖
Median	0·25		0·21		
Range	0·10–1·00		3·90–0·00		
EPA + DHA (g)					0·020‖
Median	0·36		0·31		
Range	1·43–0·13		5·58–0·01		
MUFA	16·7	3·9	15·8	3·7	0·183†
SFA					0·081‖
Median	20·3		16·7		
Range	9·0–32·0		8·0–36·0		
Trans-FA	0·6	0·3	0·6	0·3	0·832†
n-6:*n*-3 ratio					0·017‖**
Median	7·2		8·2		
Range	3·5–15·2		3·5–24·2		

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Table 1. *Continued*

	Controlled asthma (*n* 40)		Non-controlled asthma (*n* 134)		
	Mean	SD	Mean	SD	*P*
MUFA:SFA ratio					0·325‖
Median	0·86		0·90		
Range	0·6–1·6		0·5–1·8		
Ethanol (% TEV)					0·015‖**
Median	1·4		3·7		
Range	0·0–24·8		0·0–37·0		
Cholesterol (mg)	644·0	209·0	590·0	245·0	0·211†
Total dietary fibre (g)	28·5	9·2	26·0	12·5	0·254†
Carotene (μg RAE)					0·360‖
Median	478·3		430·4		
Range	113·5–1871·6		52·7–2501·5		
Retinol (μg RAE)	1483·5	4384·1	1402·0	3884·0	0·212‖
Vitamin E (mg α-TE)	10·3	14·4	8·5	37·0	0·074‖
Median	10·3		8·5		
Range	5·2–19·5		2·4–39·4		
Vitamin C (mg)	143·7	207·8	131·0	136·1	0·338‖
Mg (mg)	416·9	103·4	386·3	151·5	0·233†
Zn (mg)	20·4	7·8	18·7	8·3	0·259†

MET, metabolic equivalent; ICS, inhaled corticosteroid; exhaled NO, fraction of exhaled NO; ppb, parts per billion; FEV1, forced expiratory volume in the first second; ALQ, asthma life quality test; ACQ, Asthma Control Questionnaire; TEV, total energy value; ALA, α-linolenic acid; FA, fatty acid; RAE, retinol A equivalents; TE, tocopherol equivalents.
* Macro- and micronutrients are presented as unadjusted variables.
† *t* test.
‡ χ^2 test.
§ Sugars refer to all monosaccharides and disaccharides added to foods by the manufacturer, cooking or consumer, plus sugars naturally present in honey, syrups and fruit juices.
‖ Mann–Whitney *U* test
** $P<0·05$.

The seven-item ACQ was designed to assess clinical asthma control during the previous week. A seven-point scale (0 = no impairment, 6 = maximum impairment) was used, and the score was calculated as the mean of the seven items, ranging from 0 (totally controlled) to 6 (severely uncontrolled)[14].

Asthma quality of life was measured by the asthma life quality test, developed by the American College of Allergy, Asthma and Immunology, and validated in Portuguese[15]. The self-administered asthma life quality test includes twenty questions of dichotomous answer (yes/no) assessing six domains: activity and sleep; symptoms; triggers; unscheduled health care use; medication; psychological. Total score was calculated as the sum of affirmative responses, ranging from 0 to 20 (lower values indicate better asthma quality of life).

Statistical analysis

Descriptive statistics are expressed as means and standard deviations, and proportions (%), whereas PA and several nutrient data are presented as medians and ranges given the non-normal distribution. eNO was logarithmically transformed to attain normal distribution and is presented as geometric means and 95 % CI. Atopic status, defined by positive skin prick tests, medical diagnosis of allergic rhinitis, present use of inhaled corticosteroid, education (≤4, 5 to 9 and ≥10 years) and smoking status (non-smoker, past smoker and present smoker) were also recorded.

Nutritional variables were adjusted for total energy intake using the nutrient residual model[16]. In this model, energy-adjusted nutrient intakes are computed as the residuals from the regression analysis, with total energy intake as independent variables and absolute intakes as dependent variables. The associations between nutritional intake and asthma outcomes were performed using linear regression, multiple linear regression and unconditional logistic regression models. Linear regression was initially fitted to analyse the associations between nutrient intake (independent variables) and asthma outcomes (dependent variables). Multiple linear regression models adjusted for confounders were performed separately for eNO, FEV1, asthma life quality and ACQ scores (categorical confounder variables were transformed into dummy variables). Logistic regression models were also performed to analyse the associations between nutritional intake and asthma control level. Energy-adjusted nutrients were categorised into tertiles. OR were calculated by reference with the lowest tertile.

Sex, education, age, energy intake, BMI, PA score, smoking, atopy, rhinitis and inhaled corticosteroid were analysed as potential confounders. Only the variables that were significantly associated with each of the asthma outcomes in the univariate analysis were considered in the final regression and logistic models. Considering that

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Table 2. Associations between nutrient intake and airway inflammation, lung function, asthma quality of life and Asthma Control Questionnaire score (ACQ)
(β Coefficients and 95 % confidence intervals)

	Exhaled NO (ppb)		FEV1 (% predicted)		AQL score		ACQ score	
	Confounder-adjusted†		Confounder-adjusted‡		Confounder-adjusted§		Confounder-adjusted‖	
	β¶	95 % CI	β	95 % CI	β	95 % CI	β	95 % CI
Fatty acids								
n-6 PUFA (g)	−0·024	−0·035, 0·083	0·786	−1·164, 2·734	0·035	−0·248, 0·318	−0·037	−0·110, 0·036
n-3 PUFA (g)	−0·502*	−0·928, −0·075	9·690	−4·373, 23·753	−1·163	−3·222, 0·896	−0·260	−0·792, 0·272
ALA (g)	−0·357*	−0·608, −0·105	3·180	−5·086, 11·446	−0·998	−0·202, 0·207	−0·283	−0·593, 0·027
EPA (g)	−0·003	−0·753, 0·747	16·056	−8·850, 40·962	−0·422	−4·082, 3·239	−0·232	−1·176, 0·711
DHA (g)	−0·005	−0·343, 0·332	6·477	−4·752, 17·706	−0·182	−1·831, 1·467	−0·100	−0·525, 0·325
EPA + DHA (g)	−0·003	−0·236, 0·230	4·632	−3·111, 12·376	−0·127	−1·265, 1·010	−0·070	−0·363, 0·223
n-6:*n*-3 ratio	0·053*	0·017, 0·089	−0·072	−1·278, 1·134	0·072	−0·104, 0·248	−0·002	−0·047, 0·044
MUFA	−0·022	−0·050, −0·007	−0·222	−0·663, 1·108	0·028	−0·158, 0·103	−0·007	−0·041, 0·026
SFA	−0·021*	−0·038, −0·004	−0·032	−0·580, 0·515	−0·051	−0·131, 0·029	−0·009	−0·030, 0·011
MUFA:SFA ratio	0·637*	0·075, 1·199	3·417	−14·765, 21·600	1·902	−0·724, 4·529	0·355	−0·321, 1·030
Antioxidant micronutrients								
Carotene (μg RAE)	0·001	−0·000, 0·001	0·017	−0·009, 0·042	0·001	−0·005 0·003	−0·001	−0·002, 0·000
Retinol (μg RAE)	−0·001	−0·001, 0·000	0·007	−0·006, 0·021	−0·002	−0·004, 0·000	0·000	−0·001, 0·000
Vitamin E (mg α-TE)	−0·074	−0·166, 0·019	2·055	−0·841, 4·950	0·086	−0·340, 0·519	−0·051	−0·161, 0·058
Vitamin C (mg)	0·002	−0·002, 0·005	0·061	−0·056, 0·177	0·008	−0·009, 0·025	−0·001	−0·006, 0·003
Mg (mg)	−0·001	−0·004, 0·003	0·106	−0·009, 0·222	0·010	−0·007, 0·027	−0·003	−0·007, 0·002
Zn (mg)	0·006	−0·068, 0·080	0·159	−2·224, 2·543	−0·095	−0·444, 0·255	−0·008	−0·098, 0·082

ppb, Parts per billion; FEV1, forced expiratory volume in the first second; AQL, asthma quality of life; ALA, α-linolenic acid.
* $P<0·05$.
† Linear regression; multiple linear regression adjusted for: energy intake, sex, age, BMI, education, rhinitis and atopic status.
‡ Linear regression; multiple linear regression adjusted for: energy intake, sex, age, rhinitis and education.
§ Linear regression; multiple linear regression adjusted for: energy intake, sex, age, BMI, education and inhaled corticosteroid (ICS).
‖ Linear regression; multiple linear regression adjusted for: energy intake, sex, age, education and ICS.
¶ Representing the adjusted ratio of geometric means.

Table 3. Association between nutrient intake and asthma control
(Odds ratios and 95 % confidence intervals)

	Energy-adjusted†			Confounder-adjusted‡		
	OR	95 % CI	*P* for trend	OR	95 % CI	*P* for trend
Fatty acids						
n-6 PUFA (g/d)						
< 5·19	1·00	Reference		1·00	Reference	
5·19–7·12	0·58	0·25, 1·35		0·53	0·22, 1·30	
> 7·12	1·25	0·49, 3·18	0·659	1·23	0·45, 3·35	0·707
n-3 PUFA (g/d)						
< 0·73	1·00	Reference		1·00	Reference	
0·73–0·94	0·21*	0·07, 0·67		0·18*	0·05, 0·62	
> 0·94	0·14*	0·04, 0·44	0·001*	0·14*	0·04, 0·45	0·001*
ALA (g/d)						
< 1·54	1·00	Reference		1·00	Reference	
1·54–1·96	0·21*	0·07, 0·61		0·19*	0·06, 0·59	
> 1·96	0·23*	0·08, 0·67	0·010*	0·18*	0·06, 0·58	0·006*
EPA (g/d)						
< 0·07	1·00	Reference		1·00	Reference	
0·07–0·14	0·85	0·32, 2·24		0·80	0·29, 2·21	
> 0·14	0·34	0·14, 0·83	0·496	0·37	0·15, 0·99	0·689
DHA (g/d)						
< 0·16	1·00	Reference		1·00	Reference	
0·16–0·30	0·82	0·32, 2·07		0·82	0·31, 2·19	
> 0·30	0·48	0·20, 1·17	0·086	0·55	0·22, 1·40	0·189
EPA + DHA (g/d)						
< 0·23	1·00	Reference		1·00	Reference	
0·23–0·43	0·78	0·30, 2·02		0·82	0·30, 2·21	
> 0·43	0·21	0·18, 1·03	0·045	0·49	0·19, 1·23	0·107
n-6:*n*-3 ratio						
< 6·45	1·00	Reference		1·00	Reference	
6·45–8·11	2·18	0·95, 4·99		2·24	0·93, 5·36	
> 8·11	4·14*	1·59, 10·74	0·004*	3·69*	1·37, 9·94	0·009*
MUFA (g/d)						
< 16·12	1·00	Reference		1·00	Reference	
16·12–20·06	0·58	0·23, 1·47		0·50	0·19, 1·35	
> 20·06	0·44	0·18, 1·10	0·083	0·47	0·17, 1·27	0·154
SFA (g/d)						
< 16·84	1·00	Reference		1·00	Reference	
16·84–23·90	0·46	0·18, 1·19		0·51	0·19, 1·34	
> 23·90	0·39*	0·15, 0·98	0·056	0·36*	0·13, 0·97	0·047*
MUFA:SFA ratio						
< 0·81	1·00	Reference		1·00	Reference	
0·81–1·0	1·45	0·62, 2·39		1·44	0·59, 3·52	
> 1·0	1·69	0·71, 4·01	0·203	1·89	0·75, 4·78	0·072
Antioxidant micronutrients						
Carotene (μg RAE/d)						
< 152	1·00	Reference		1·00	Reference	
152–238	1·00	0·41, 2·46		1·10	0·43, 2·77	
> 238	0·69	0·29, 1·61	0·310	0·76	0·31, 1·87	0·412
Retinol (μg RAE/d)						
< 95	1·00	Reference		1·00	Reference	
95–849	0·33*	0·13, 0·83		0·32*	0·12, 0·87	
> 849	0·55	0·21, 1·46	0·334	0·54	0·19, 1·50	0·355
Vitamin E (mg α-TE/d)						
< 3·22	1·00	Reference		1·00	Reference	
3·22–4·02	0·89	0·35, 2·29		0·84	0·31, 2·26	
> 4·02	0·43	0·18, 1·03	0·049*	0·43	0·17, 1·10	0·079
Vitamin C (mg/d)						
< 52	1·00	Reference		1·00	Reference	
52–66	0·83	0·35, 1·94		0·84	0·35, 2·04	
> 66	1·11	0·46, 2·69	0·740	1·17	0·46, 3·02	0·667
Mg (mg/d)						
< 149	1·00	Reference		1·00	Reference	
149–175	0·66	0·26, 1·63		0·72	0·28, 1·85	
> 175	0·55	0·22, 1·33	0·184	0·63	0·25, 1·61	0·338
Zn (mg/d)						
< 5·73	1·00	Reference		1·00	Reference	
5·73–7·26	0·84	0·35, 2·01		0·97	0·38, 2·45	
> 7·26	0·80	0·33, 1·92	0·606	0·79	0·31, 2·01	0·657

ALA, α-linolenic acid, RAE, retinol A equivalents.
* $P<0·05$.
† Logistic regression adjusted for energy intake.
‡ Logistic regression adjusted for energy intake, sex, age, education and inhaled corticosteroid.

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smoking status and PA were not significantly associated with eNO, FEV1, asthma life quality or ACQ scores, and that their inclusion as confounders did not influence the effects, these variables were therefore not included in the final models. Considering the biological plausibility related to dietary intake, sex, age and total energy intake, these were considered in all models. A 0·05 level of significance and 95 % CI were considered. Data analysis was performed using the statistical package SPSS®, version 17.0 (SPSS Inc., Chicago, IL, USA).

Results

From the 219 patients invited, forty-five were excluded (twenty-one did not fulfil the inclusion criteria, nine had dietary changes in the last 12 months, eight had incomplete data records, four were considered as energy intake outliers and three refused to participate). Energy intake outliers were previously excluded from the study and were defined as having energy intake values above the arithmetic mean (SD 2), and implausibly low intakes (< 2092 kJ (< 500 kcal) for women and < 3347·2 kJ (< 800 kcal) for men). The characteristics of excluded patients, regarding age, education, smoking status and asthma severity, were similar to the 174 patients (81 %) included in the analysis.

According to asthma control definition, 23 and 77 % of the subjects were classified, respectively, as having controlled and non-controlled asthma (Table 1). Considering the energy contribution of macronutrients, no significant differences between these two groups were observed for total carbohydrates, total fat and SFA intake. However, controlled patients had a significantly lower *n*-6:*n*-3 PUFA ratio intake compared with non-controlled patients ($P=0·017$) and had a significantly higher intake of ALA ($P=0·022$), EPA ($P=0·016$), DHA ($P=0·021$) and EPA + DHA ($P=0·020$); dietary intakes of *n*-3 PUFA ($P=0·090$), SFA ($P=0·081$) and vitamin E ($P=0·074$) were higher in controlled asthmatics, but these differences were not statistically significant.

The associations of the dietary intake of fatty acids and antioxidant micronutrients with markers of asthma adjusted for energy intake, sex, age, BMI, education, atopy, rhinitis and inhaled corticosteroids are presented in Table 2. Higher *n*-6:*n*-3 PUFA and MUFA:SFA ratios were associated with higher eNO, whereas higher intakes of *n*-3 PUFA and SFA were associated with lower eNO. Higher ALA intake was associated with lower eNO, even after adjustment for marine *n*-3 PUFA (R −0·356, 95 % CI −0·609, −0·105; $P=0·006$). No significant associations were found for EPA + DHA and asthma outcomes. Energy-adjusted MUFA and Mg were associated with eNO and FEV1, respectively; however, after adjusting for confounders, these associations were no longer significant.

The OR for asthma control accordingly with the dietary intake of fatty acids and antioxidant micronutrients are given in Table 3. Intake of *n*-3 PUFA between 0·73 and 0·94 g/d and above 0·94 g/d reduced the odds of non-controlled asthma (second tertile: OR 0·18, 95 % CI 0·05, 0·62; third tertile: OR 0·14; 95 % CI 0·04, 0·45; P for trend=0·001), while *n*-6:*n*-3 PUFA above 8·45 had the opposite effect (third tertile: OR 3·69, 95 % CI 1·37, 9·94; P for trend=0·009), after adjusting for energy intake, sex, age, education and inhaled corticosteroids. Dietary intake of ALA between 1·54 and 1·96 and above 1·96 g/d reduced the odds of non-controlled asthma (second tertile: OR 0·19, 95 % CI 0·06, 0·59; third tertile: OR 0·18, 95 % CI 0·06, 0·58; P for trend=0·006), after adjusting for confounders. After adjusting also for alternate *n*-3 PUFA, the protective effect of ALA in asthma control still remained significant (second tertile: OR 0·19, 95 % CI 0·06, 0·60; third tertile: OR 0·18, 95 % CI 0·06, 0·58; P for trend=0·005), independent of EPA + DHA. No significant association was observed for EPA + DHA and asthma control.

The higher intake of SFA (> 23·9 g/d) also decreased the probability of having non-controlled asthma (OR 0·36, 95 % CI 0·13, 0·97; P for trend=0·047), after controlling for confounders. Considering micronutrients, dietary intake of retinol between 95 and 849 μg retinol A equivalents/d decreased the odds of having non-controlled asthma; however, the trend according to the retinol intake category was not significant (P for trend=0·355). A protective trend was also observed for energy-adjusted vitamin E and asthma control (OR 0·43, 95 % CI 0·18, 1·03; $P=0·049$); however, this association was no longer significant after the final adjustment.

Discussion

In the present study, higher dietary intakes of *n*-3 PUFA and SFA were associated with a decreased levels of eNO and improved likelihood of asthma being under control, while a high ratio of *n*-6:*n*-3 PUFA had the opposite effect. In addition, higher dietary intake of ALA was associated with lower eNO and reduced the likelihood of non-controlled asthma, independent of marine *n*-3 PUFA. No significant associations between the dietary intake of EPA + DHA and antioxidant vitamins and minerals and asthma outcomes were observed.

The present results are limited by the cross-sectional design of the study which leaves open any possible cause–effect relationship and the role of other factors. Nevertheless, an inverse causal relationship is not probable and we assessed established lifestyle factors that could have an important role in asthma and that influence nutrient intake, such as total energy intake, PA and BMI, and the association between nutrients and asthma outcomes was extensively adjusted for confounders.

To the best of our knowledge, this is the first study exploring the association between different types of dietary fatty acids and antioxidant nutrient intake, and asthma control. Moreover, we assess the dietary intake of vegetable

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(ALA) and marine (EPA + DHA) *n*-3 PUFA, and report for the first time the protective effect of ALA in asthma control. The score we used to assess control, which included different dimensions of the disease such as inflammation, lung function and symptoms, has been shown to explain 77 % of the variability of asthma control[5]. Another important strength of the present study was the FFQ that we used, since it has been validated for Portuguese adults[6], and it has been shown to provide reliable estimates for *n*-3 PUFA and SFA[6].

In the present study, a higher dietary intake of *n*-3 PUFA (>0·94 g/d) and ALA (>1·96 g/d) reduced the odds of non-controlled asthma. Considering that the prevalence of non-controlled asthma in the present study was high, the OR may be biased towards overestimating the risk. Nevertheless, even though we could admit an overestimation of the protective effect, the reverse result should not be expected.

ALA is the major plant-based *n*-3 PUFA and exerts main effects through conversion to EPA and DHA, when dietary intake of marine PUFA is low[17,18]. Long-chain *n*-3 PUFA decreases the production of inflammatory mediators, competitively inhibiting the metabolism of arachidonic acid (generating less active prostenoids and leukotrienes), suppressing IgE production, and thereby potentially acting to reduce airway inflammation and bronchoconstriction in asthma[18,19]. However, results were inconclusive. A systematic review from Cochrane of the clinical effects of *n*-3 PUFA fish oil supplementation in established asthma suggests that the results are not consistent and that there is little evidence to recommend such supplementation in order to improve asthma control[20]. Reconciling the data from experimental and observational studies is difficult, most probably, due to different methods of assessment of dietary intake and different definitions of asthma. Taken the data into account from previous cross-sectional studies, it seems that dietary or serum *n*-3 PUFA levels are directly associated with lung function, at least in asthmatics[21–23] and atopy[24], and are protective for the risk of asthma or atopy. Recently, in a large population-based study, asthma risk was doubled in subjects who had never eaten fish during childhood and a minimum of weekly fish intake in adulthood was protective against asthma symptoms[25]. In a small study, fish oil supplementation failed to provide any benefit in eNO, lung function or asthma control in asthmatic women[26]. In the present study, dietary intake of *n*-3 PUFA and ALA was associated with improved asthma control and lower eNO, independent of EPA + DHA. Higher intake of ALA (and also not EPA or DHA) was previously associated with a decreased risk of allergic sensitisation and allergic rhinitis in adults[27]. However, the link between ALA and asthma is still poorly addressed. In the present study, dietary intake of EPA and DHA is very similar between controlled and non-controlled subjects, and we have found no significant associations between EPA or DHA and asthma outcomes. There is evidence suggesting that ALA, EPA and DHA might have heterogeneous and potentially independent effects on inflammation, gene expression and chronic diseases; therefore, a better understanding of the individual role of *n*-3 PUFA in inflammatory diseases, such as asthma, is needed[17,18]. It has been suggested that higher margarine intake rich in *n*-6 PUFA is associated with an increased risk of asthma[28,29] and hay fever[30] in adulthood, and eczema and allergic sensitisation in children[31]. Dietary intake of *n*-6 PUFA was similar among controlled and non-controlled subjects, and therefore, no significant associations for *n*-6 PUFA and asthma outcomes were observed. Nevertheless, the ratio of *n*-6:*n*-3 PUFA above 8·45, which was more than tripled the odds of non-controlled asthma, was associated with increased levels of eNO.

In the present study, we analysed the total SFA intake, irrespective of the specific types. However, different types of SFA could have different effects. Foods high in SFA, such as butter[32], whole milk[32,33] and non-pasteurised farm milk[34–36], have been consistently associated with a reduced risk of asthma. For milk, it is not clear whether associations should be attributed to SFA, vitamin A or even to microbial agents (in the case of whole non-pasteurised milk or farm-related co-exposures)[34–36]. Therefore, the present results on SFA could also be a proxy of a dietary pattern high in milk and dairy products. Several epidemiological studies have reported beneficial associations for higher intake of nutrients that may be relevant in the redox mechanisms, such as vitamin C[21,37,38], vitamin E[37,39], carotenoids[40,41], Se[7] and Mg[42]. However, these findings are not conclusive[43] and intervention studies with single nutrient supplementation have been disappointing[44–47]. Inverse associations with asthma have also been observed for foods rich in these micronutrients, such as fresh fruit[7,33,48,49] and vegetables[48], and nuts[4,50,51], and additional benefits may arise from the synergistic effects between nutrients in foods and specific dietary patterns. Nuts contain a high proportion of ALA, fibre, vitamins, minerals and many bioactive compounds that may modulate redox status, and inflammatory and immune responses[30,52]. We have previously reported that intake of nuts is positively associated with lung function, and high adherence to an overall healthy dietary pattern, such as the Mediterranean diet, is associated with an improved asthma control in adults, independent of other lifestyle factors[4].

In summary, the present results provide additional support for the benefits of adequate dietary advice and give a rationale to nutritional intervention studies in asthmatics. Healthy eating in asthma, providing foods high in ALA, such as nuts, and an adequate balance between *n*-6 and *n*-3 PUFA, may reduce disease severity and improve asthma control, independent of other lifestyle factors[4].

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Acknowledgements

We thank all patients who participated in the present study; to Sandra Gomes for assistance with patient recruitment and to Milton Severo for support with statistical analyses. R. B., A. M., J. F. and M. G. C.-B. were responsible for patient enrolment and data collection; R. B. and A. M. conducted the data analyses; J. F., C. L. and P. M. advised on the statistical analysis; C. L. and P. M. advised on the nutritional analyses and supervised the study; R. B., A. M., L. D., T. H., C. L. and P. M. interpreted and critically discussed the results; R. B. wrote the first draft and all authors contributed to and critically reviewed the manuscript. The present study received no specific grant from any funding agency in the public, commercial or not-for-profit sectors. None of the authors has any conflicts of interest.

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NS British Journal of Nutrition

Barros R, Moreira A, Padrão P, Teixeira VH, Carvalho P, Delgado L, Lopes C, Severo M, Moreira P. Dietary patterns and asthma prevalence, incidence and control: evidence from the Portuguese National Health Survey.

Manuscript submitted

Dietary patterns and asthma prevalence, incidence and control: evidence from the Portuguese National Health Survey

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Abstract (243/250)

The increased asthma prevalence in westernized societies has been suggested to be related to environment exposures and lifestyle changes, particularly diet. This study aimed to explore the association between dietary patterns and asthma prevalence, incidence and control in a nationally representative population.

Data from 32644 adults, 53% female, from the 4th Portuguese National Health Survey were analysed. Prevalence of asthma was 5.3%, current asthma 3.5%, current persistent asthma 3.0%, current severe asthma 1.4%, and incident asthma 0.2%. Dietary patterns (DP) were identified by Latent Trait Models based on dietary intake. Unconditional logistic regression models were performed to analyse association between DP and asthma. Two of the five identified DP were associated with asthma: *'high fat, sugar and salt"* DP (positively correlated with pastry, chocolate and sweet desserts, candies, salty snacks, chips, fruit juices, soft drinks and alcoholic beverages consumption at snacks) was associated with asthma prevalence (OR=1.13, 95%CI=1.03, 1.24) and current severe asthma (OR=1.23, 95%CI=1.03, 1.48), while *"fish, fruit and vegetables"* DP (positively correlated with fish, vegetables and fruit intake at meals) was negatively associated with current (OR=0.84, 95%CI=0.73, 0.98), and current persistent asthma (OR=0.84, 95%CI=0.72, 0.98), after adjustment for confounders.

Our results suggest a protective association between *"fish, vegetables and fruit"* DP and current and persistent asthma, and a detrimental association between *"high fat, sugar and salt*" DP and severe asthma prevalence, further supporting the rational for diet and lifestyle intervention studies in asthma based on whole dietary patterns and physical activity.

Key-words: asthma, control, diet, dietary patterns, prevention.

Word count: (2437/2500)

References count: (35/35)

Introduction

The increased asthma prevalence in westernized societies has been suggested to be related to environment exposures and lifestyle changes, particularly diet. Actually, some of the western dietary characteristics have been associated with asthma: (1) an increased consumption of processed foods, particularly vegetable oils and margarines, and a decrease in butter, milk and fish intake, increasing the n-6/n-3 polyunsaturated fatty acids (PUFA); and (2) a reduction in vegetables and fruits consumption, decreasing the intake of antioxidant micronutrients (1). These hypotheses have been supported by epidemiological studies reporting beneficial associations between asthma risk and dietary intake of n-3 PUFA, fish and nuts (2, 3), vitamins and antioxidant rich foods such as fruit and vegetables (1). However, intervention studies with single foods or nutrients provide conflicting evidence (4-6), reflecting the complexity of dietary intake, and suggesting that looking at the whole dietary pattern (DP) is an important shift in approaching the synergistic effects between the interconnected systems of nutrients, foods, food groups, and meal structures.

There is scarce evidence on the association between DP and asthma. However, beneficial effects of *Mediterranean diet* on asthma control (7), and symptoms (8) have been reported made. In contrast, a *"westernized"* DP has been associated with asthma symptoms in children (9) and young adults (10), and asthma exacerbations in the E3N female study (11). Additionally, fast food intake has been associated with asthma symptoms, exercise-induced bronchial hyper responsiveness (BHR) and increased risk of severe asthma in children (12, 13). Effects of DP on adult asthma are still not clear and even with conflicting evidence (14, 15). Therefore, we aimed to determine major dietary patterns and their association with measures of asthma prevalence, incidence and control in a nationwide representative adult population.

Methods

Participants and study design

Data from the 4th Portuguese National Health Survey (NHS), carried out by the National Institute of Health and National Institute of Statistics (NIS), between 2005 and 2006, was analysed. The methodology of the NHS has been previously described (16, 17). In summary, the sampling frame was made based on census data and participants were selected from households during that period, using a multi-stage random probability design (hospitals, prisons, military houses and community care institutions were excluded). A representative sample of 41193 participants was included, according to the 7 main Portuguese territorial units (NUTS II), namely *North, Center, Lisbon region, Alentejo, and Algarve, and Madeira and Azores archipelagos*. The primary sampling unit (PSU) was the housing unit and sampling was based on the population and housing census. Two levels were defined within each NUTSII: the parish (corresponding to townships); and geographically defined units of 240 lodgings (within parish). The PSUs were then randomly selected within each territorial unit, and subjects living in the sampling unit were surveyed. Interviews were conducted in the households by trained staff interviewers, and the questionnaire included information on social and demographic characteristics, health, and chronic diseases including asthma. The survey response rate was 76%, as reported by NIS (16, 17). For the purposes of this study, a representative subsample of all NHS surveyed adults (≥20 years-old) was analysed (32644 participants).

Asthma

The following asthma definitions were used: ever asthma (ever medical doctor asthma diagnosis), current asthma (asthma within previous 12 months), current persistent asthma (asthma drugs within previous 12 months), current severe asthma (emergency because of asthma within 12 months), and incident asthma (diagnosis within previous 12 months).

Dietary intake

Dietary patterns were identified by Latent Trait Models (LTM), based on a list of 20 dichotomous questions (“Did you consumed any of the following foods yesterday?”), addressing the previous day dietary intake of usual foods and beverages at meals (milk, cheese and yogurt; vegetable soup; bread and cereal products; meat; fish; potatoes, rice and pasta; pulses; vegetables; fruit; and pastry, chocolates or sweet desserts), and snacks (fruit; bread or sandwiches; milk, cheese and yogurt; juices; soft drinks; pastry, chocolates or sweet desserts; other candies; salty snacks; chips; and alcoholic beverages).

Socio-demographic, lifestyle and anthropometry

Age, gender, education, family income, smoking, physical activity level (PAL), weight, and height were also addressed by NHS, and analysed as potential confounders. Body mass index (BMI) was calculated based on self-reported weight and height and categorized according to the WHO classification.

Statistical analysis

Descriptive analysis, including mean (standard deviation) and proportions, was performed to characterize the sample, and conducted considering the sampling weights.

Dietary patterns were identified by Latent Variable Models (18, 19), based on addressed food items intake at meals and snacks. Latent models can be divided into two frameworks: latent trait models (LTM), which assume that the latent variables are metrical (factors), and latent class models (LCM) that consider latent variables as categorical (clusters). Latent trait model is a Binary Data Factor Analysis that considers one or more factors. LTM assume that the dietary intake of each individual is explained by one or more factors, usually called 'latent variables'. Interpretation of the model is done considering the standardized factor loadings, witch expresses the correlation coefficient between the latent variable and an underlying continuous variable obtained from each food item. Coefficient correlations (r) > 0.350 and < -0.350 were considered for selecting food items. Geominrotation was applied to simplify the standardized factor loadings matrix, and the following criteria to determine the number of latent variables in the model was considered: the model with lowest value in the Bayesian information criteria, the global goodness of fit of the considered latent models assessed through the likelihood ratio test, via parametric boostrapping (100 samples) and the residuals inspection. Unconditional logistic regression models were performed to analyse whether dietary patterns were associated with asthma prevalence, adjusted for confounders (age, gender, BMI, education, family income, proxy reporting information, smoking and physical activity). A 0.05 level of significance and 95% CI were considered. Data analysis was performed using the statistical package SPSS®, version 18.0 (SPSS Inc., Chicago, IL, USA), and the MPlus (v5.2; Muthen & Muthrn, Los Angeles, California, USA).

Results

The final analysis included 32644 adults, 52.6% female. **Table 1** summarizes socio-demographic characteristics of participants. Prevalence of ever asthma was 5.3%, current asthma 3.5%, current persistent asthma 3.0%, current severe asthma 1.4%, and incident asthma 0.2%.

Considering dietary patterns analysis, five factors (F) were identified. The goodness-of-fit test suggests that the 5-factor LTM was the best solution, and it was improved after elimination of 2 food items from the 20 initially available and included (meat at meals and bread/sandwiches at snacks). The standardized loadings for the 5-factor latent trait model (LTM) are presented in **Table 2**. F1 *("dairy and fruit"* DP*)* was positively correlated with milk and dairy products (r=0.575) at meals and snacks, fruit (r=0.484) at snacks, and negatively correlated with alcoholic beverages (r=-0.405) at snacks. F2 ("*soup and starchy foods" DP*) was positively correlated with vegetable soup (r=0.526), bread (r=0.443) and pulses (r=0.432) at meals. F3 ("*high fat, sugar and salt"* DP) was positively correlated with pastry, chocolate and sweet desserts (r=0.370), other candies (r=0.630), salty snacks (r=0.711), chips (r=0.918), fruit juices (r=0.568), soft drinks (r=0.648), and alcoholic beverages (r=0.392) at snacks. F4 ("*fish, fruit and vegetables"* DP) was positively correlated with fish (r=0.443), vegetables (r=0.538) and fruit (r=0.448) at meals. F5 ("*sugary and fatty foods"* DP) was positively correlated with, pastry, chocolate and sweet desserts at meals (r=0.464) and snacks (r=0.395).

After adjustment for the confounders (age, gender, BMI, education, family income, proxy reporting information, smoking), F3 *("high fat, sugar and salt"* DP*)* was associated with ever asthma prevalence (OR=1.13, 95% CI =1.03, 1.24) and current severe asthma (OR=1.23, 95% CI=1.03, 1.48). F4 *("fish, fruit and vegetables"* DP) was negatively associated with current asthma (OR=0.84, 95% CI=0.73, 0.98), and current persistent asthma (OR=0.84, 95% CI=0.72, 0.98). The three other identified factors of DP had no significant associations with asthma outcomes (**Table 3** and **Figure 1**).

In the NHS, physical activity was only addressed in 14% of participants. Nevertheless we also performed logistic regression models adjusted for PAL in this subsample. F4 *("fish, fruit and vegetables"* DP) still remains negatively associated with current asthma (OR=0.61, 95% CI=0.42, 0.89), but not with current persistent asthma (OR=0.77, 95% CI=0.50, 1.165); while F3 *("high fat, sugar and salt"* DP) was no longer significantly associated with ever asthma prevalence (OR= 1.09, 95% CI=0.85, 1.38) and current severe asthma (OR= 1.14, 95% CI=0.73, 1.79).

Discussion

This study shows a protective association between essential components of a *Mediterranean diet*, such as eating *"fish, vegetables and fruit",* and asthma prevalence and persistency. In contrast, *"high fat, sugar and salt'* dietary patterns, based on high-energy density/low micronutrient foods are associated with increased prevalence and severity of asthma, independent of other socio-economic and lifestyle factors.

Our study has some limitations. First, it is limited by the cross sectional nature of data not allowing to establish causal relationship. Secondly, dietary intake assessment was based on a checklist of usual foods intake at meals and snacks in the previous 24h, which could be considered less informative than other dietary assessment methods on frequency, portion weight, variety (other foods outside the list could have been excluded), and specificity (e.g. milk, cheese and yogurt are aggregated in the same question). Nonetheless, the European Food Safety Authority, recommend 24h diet recall for the collection of national food consumption data in adults (20). Thirdly, asthma prevalence, incidence and control were addressed by self-report. However, this has been the usual approach in large-scale studies assessing asthma and allergic diseases prevalence. Our findings are also supported by important strengths. It is a nationwide representative observational cross-sectional study, with a large sample of adults from both genders addressing DP and asthma. Additionally, this is the first study on asthma that derives DP by LTM analysis (18, 19), which allows an increase in the discrimination of dietary intake and will better support public health dietary recommendations. In addition, the NHS includes important socio-economic and lifestyle confounders regarding diet and asthma, such as age, gender, education, family income, BMI, smoking, proxy reporting information, and physical activity level. Although data of physical activity was available for only 14% participants, logistic regression analysis was performed for these showing that *"fish, fruit and vegetables"* DP still remained negatively associated with current asthma, while *"high fat, sugar and salt"* DP was no longer associated with ever asthma prevalence and current severe asthma. Despite these results cannot be extrapolated for the whole sample, they suggests that a *"fish, fruit and vegetables"* DP may have a protective role on asthma independently of physical activity, while physical activity could modulate the detrimental association between a *"westernized"* DP and asthma prevalence and severity (21, 22).

There are few studies addressing diet and asthma by dietary patterns approach, and results have been controversial (15, 23, 24). However, *"fish, fruit and milk"* has been previously associated with reduced asthma risk in young adults (10). Comparison of dietary patterns between studies is complex, due to differences in the socio-geographic characteristics of the studied populations, and the methodological approaches to assess dietary intake and food patterns analysis (25), particularly in cross-sectional studies and with unmeasured confounders. Nevertheless, our findings are consistent with the reported literature, even when different dietary intake assessment methods were applied. In the European Community Respiratory

Health Survey II, authors described a potential beneficial effect of fish, fruits and vegetable on current asthma and asthma symptoms (14). Fish, vegetables and fruit are essential components of *Mediterranean Diet* (26). We have previously reported that dietary intake of fruit (≥300g) was associated with better asthma control in adults (7), and data from ISAAC phase 3 reported also a protective effect of fruit (≥ 3 times/week) on severe asthma in children and adolescents (13). Dietary intake of fish, fruit, nuts and vegetables has been beneficially associated with asthma outcomes in several epidemiological studies during lifecycle (1-3, 27). Actually, these foods are rich in n-3 PUFA (EPA, DHA, alpha-linolenic acid), vitamins, minerals and methyl donors. The interaction between all these nutrients, foods and meals in a whole dietary pattern could explain the antioxidant, anti-inflammatory and immunomodulator mechanisms in asthma. In addition, fruit, vegetables, nuts and pulses (28) are also fiber food sources that seems to increase the proportion of *Bacterioidacea* in the microbiota. Dietary fiber have effects on the gut microbiota balance, in the short-chain fatty acids (SCFAs) production, including propionate, which potentially reduce AHR and the severity of allergic airway inflammation (29, 30). These Mediterranean healthful food components (26) can have a favourable impact on several chronic inflammatory diseases (31-33), including reduced asthma risk in prospective studies in children (8), and better asthma control in adults (7). Nevertheless, intervention studies based on whole healthy dietary patterns are still lacking.

Conversely, we also observed a detrimental association between *"high fat, sugar and salt"* DP and severe asthma prevalence. A *"Western"* DP has been previous associated with asthma symptoms in children (9), young adults (10), and asthma attacks in the E3N female study (11). In addition, fast food intake has been associated with asthma symptoms, exercise-induced BHR and increased risk of severe asthma in children (12, 13). Looking to the potential mechanisms, high-total fat meals and high-trans fatty acids meal have been associated with increase airway inflammation in asthmatic patients, resulted by an increase in sputum neutrophils and TLR4mRNA expression in sputum cells, activating the innate immune response. This fat induced inflammation impairs the airway response to bronchodilator and worse clinical outcomes, namely in non-atopic patients (34), suggesting that nutritional intervention strategies aimed at modifying dietary fat may be useful in reducing airway neutrophilia, with clinical impact in asthma control and severity.

Our study supports the rational for lifestyle intervention studies in asthma, based on whole dietary patterns that includes fruit, vegetables and fish, such as *Mediterranean* or *prudent healthy* dietary patterns (35), and reduced intake of high energy density/low micronutrient foods, such as *"high fat, sugar and salt"* DP. Moreover, DP approach allows to better understand the interactions between nutrients, foods and meals on asthma, being conceptually easier for dietary recommendations. Incident asthma and poor asthma control are associated with higher medical costs, increased productivity loss, and substantial reduction in quality of life. Therefore, nutritional intervention based on a healthy dietary pattern, adequate body weight maintenance,

and regular physical activity could contribute as a low cost public health lifestyle strategy to reduce asthma prevalence and incidence, and improve asthma control management.

In summary, our results suggest a protective association between *"fish, vegetables and fruit"* DP and current and persistent asthma and, conversely, a detrimental association between *"high fat, sugar and salt*" DP and severe asthma prevalence, independent of other socio-economic and lifestyle factors. These data supports the rational for diet and lifestyle intervention studies in asthma based on whole dietary patterns and physical activity.

Tables and figures

Table 1. Characteristics of participants

	Total (n = 32644)
Gender, female (%)	52.4
Age (%)	
20 – 39 y	37.8
40 – 64 y	40.5
65 – 84 y	19.8
≥ 85 y	1.9
Education (%)	
None	12.7
1st cycle	34.1
2nd and 3rd cycles	27.8
High school	12.2
College	13.1
Family income (%)	
≤ 500€/month	23.7
501 – 1200€/month	44.1
1201€ - 2000€/month	20.0
> 2000€/month	12.2
Proxy reporting information (%)	29.8
Asthma prevalence (%)	
Ever asthma	5.3
Incident asthma	0.2
Current asthma	3.5
Current persistent asthma	3.0
Current severe asthma	1.4
BMI Classes (%)	
Underweight and normal weight	46.4
Overweight	37.6
Obesity classes I and II	15.2
Obesity class III	0.8
Level of Physical activity* (%)	
Low	39.0
Moderate	46.1
Hight	14.9
Smoking (%)	
Current smoker	21.4
Past smoker	16.8

Data presented as proportions (%), and sample weighted pond 1; *Level of physical activity (pond 4)

Table 2. Standardized loadings for the 5-factor latent trait model (LTM) with the food items intake

		LTM				
Food-items		**5-factors Model**				
	Total n=32644	Z1 (r)	Z2 (r)	Z3 (r)	Z4 (r)	Z5 (r)
Meals						
Milk, cheese and yogurt	85.7%	**0.58**	0.05	-0.04	0.0.	0.28
Vegetable soup	69.0%	**0.26**	**0.53**	-0.02	0.03	-0.04
Bread and cereal products	94.2%	0.02	**0.44**	0.00	-0.01	0.35
Meat*	76.6%	-	-	-	-	-
Fish	49.8%	-0.02	-0.06	0.00	**0.44**	-0.02
Potatoes, rice and pasta	87.5%	-0.08	0.12	0.09	0.21	0.23
Pulses	22.6%	-0.04	**0.43**	0.23	0.01	0.01
Vegetables	69.9%	0.16	-0.01	0.01	**0.54**	0.00
Fruit	81.9%	0.22	0.07	-0.05	**0.45**	0.14
Pastry, chocolates or sweet dessert	23.7%	-0.06	0.00	**0.37**	0.06	**0.46**
Snacks						
Fruit	41.2%	**0.48**	0.05	0.15	0.17	-0.20
Bread or sandwiches*	43.8%	-	-	-	-	-
Milk, cheese and yogurt	36.8%	**0.70**	-0.11	0.23	-0.01	-0.04
Juices	10.1%	0.16	-0.02	**0.57**	0.00	0.06
Soft drinks	4.4%	0.02	0.09	**0.65**	-0.11	0.02
Pastry, chocolates or sweet desserts	10.0%	0.03	-0.16	**0.56**	-0.02	**0.40**
Other candies	3.1%	0.04	-0.17	**0.63**	-0.04	0.15
Salty snacks	3.0%	-0.19	0.00	**0.71**	0.22	-0.04
Chips	1.3%	0.02	0.05	**0.92**	0.00	-0.02
Alcoholic beverages	10.6%	**-0.41**	0.04	**0.39**	0.05	-0.13

LTM: Latent trait model. *Food-items removed from the LTM analysis. Five factors (Z) were identified by latent trait model (r > 0.35): factor 1 (positive correlated with milk and dairy products, fruit and negative with alcoholic beverages in snacks); factor 2 (positive correlated with vegetable soup, bread, pulses); factor 3 (positive correlated with pastry, chocolate and sweet desserts, other candies, salty snacks, chips, fruit juices, soft drinks and alcoholic beverages at snack); factor 4 (positive correlated with fish, vegetables and fruit at meals); and factor 5 (positive correlated with pastry, chocolate and sweet desserts).

Table 3. Association between dietary patterns and asthma prevalence, incidence and control

	Ever Asthma		Current Asthma		Current Persistent Asthma		Current Severe Asthma		Incident Asthma	
Dietary Patterns	**aOR (95%CI)**	**p**	**aOR (95%CI)**	**p**	**aOR (95%CI)**	**p**	**aOR (95%CI)**	**p**	**aOR (95%CI)**	**p**
'"Dairy and fruit" DP	0.94(0.86; 1.02)	0.147	0.96 (0.86; 1.06)	0.412	0.99 (0.89; 1.11)	0.901	0.85 (0.71; 1.01)	0.061	1.21 (0.70; 2.09)	0.501
"Soup and starchy foods" DP	0.95 (0.83; 1.08)	0.404	1. 01 (0.87; 1.18)	0.882	1.02 (0.87; 1.20)	0.773	0.87 (0.68; 1.12)	0.282	1.81 (0.79; 4,17)	0.163
"High fat, sugar and salt" DP	**1.13 (1.03; 1.24)**	0.010	1.10 (0.98; 1.23)	0.094	1.05 (0.93; 1,19)	0.405	**1.23 (1.03; 1.48)**	0.024	1.14 (0.63; 2.06)	0.659
"*Fish, fruit and vegetables*" DP	0.92 (0.82; 1.05)	0.218	**0.84 (0.73; 0.98**)	<0.001	**0.84 (0.72; 0.98)**	0.030	0.88 (0.68; 1.12)	0.308	0.79 (0.36; 1.73)	0.552
"Sugary and fatty foods" DP	1.01 (0.90; 1.13)	0.889	1.01 (0.89; 1.16)	0.844	1.06 (0.92; 1.22)	0.444	0.96 (0.77; 1.19)	0.695	0.82 (0.40; 1,72)	0.607

Unconditional logistic regression models adjusted for gender, age, BMI, education, family income, proxy reporting information and smoking. DP: Dietary patterns; aOR: adjusted Odds Ratio; p-value ≤0.05

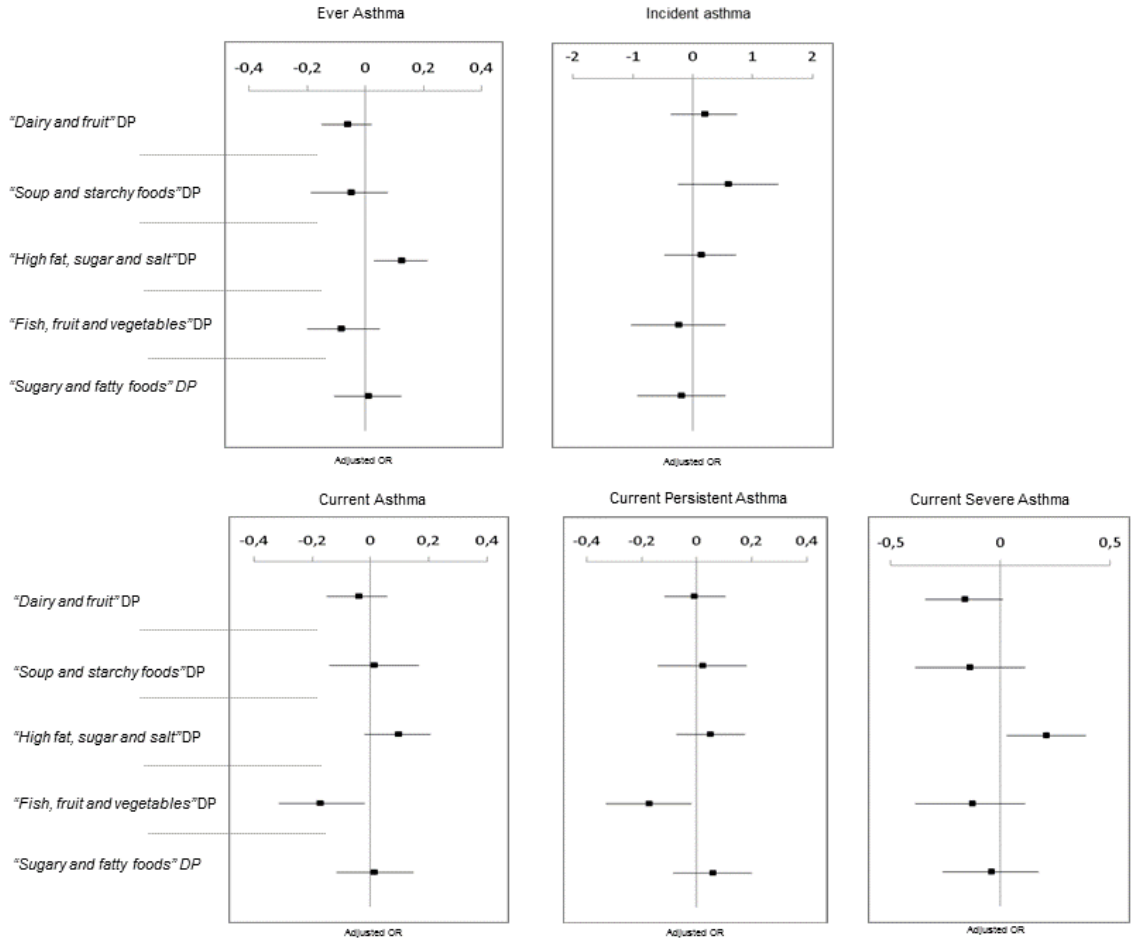

Figure 1. Association between dietary patterns and asthma prevalence, incidence and control

Unconditional logistic regression models adjusted for gender, age, bmi, education, family income, proxy reporting information and smoking. DP: Dietary patterns; aOR: adjusted Odds Ratio; p-value ≤0.05

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Barros R, Moreira A, Padrão P, Teixeira VH, Carvalho P, Delgado L, Moreira P. Obesity increases prevalent and incident asthma and worsens asthma severity: evidence from the Portuguese National Health Survey.

Manuscript submitted

Obesity increases prevalent and incident asthma and worsens asthma severity: evidence from the Portuguese National Health Survey

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Sources of support: None declared

Competing interests: None declared

Abstract (296/300)

Background: We aimed to explore the association between obesity and asthma prevalence and incidence in a representative sample of Portuguese adults.

Methods: A representative sample from the 4th Portuguese National Health Survey was analysed. Following asthma definitions were used: ever asthma (ever medical doctor asthma diagnosis), current asthma (asthma within 12 months), current persistent asthma (asthma drugs within 12 months), current severe asthma (emergency because of asthma within 12 months), and incident asthma (diagnosis within 12 months). Body mass index (BMI) was calculated based on self-reported weight and height and categorized according to WHO classification. Logistic regression models were performed to analyse whether BMI was associated with asthma, adjusted for confounders (age, gender, education, family income, proxy reporting information, smoking, physical activity and dietary patterns, assessed by Latent Trait Models analysis).

Results: Final analysis included 32644 adults, ≥20 years-old, 52.6% female. Prevalence of ever asthma was 5.3%, current asthma 3.5%, current persistent asthma 3.0%, current severe asthma 1.4%, and incident asthma 0.2%. Prevalence of obesity was 16%, overweight 37.6%, normal weight 44.6% and underweight 0.2%.

Overweigh, obesity class I and II, and obesity class III were associated with ever asthma (respectively OR=1.22, 95%CI 1.21, 1.24; OR=1.39, 95%CI 1.36, 1.41; OR=3.24, 95%CI 3.08, 3.40), current asthma (OR=1.16, 95%CI 1.14, 1.18; OR=1.86, 95%CI 1.82, 1.90; OR=4.73, 95%CI 4.49, 4.98), current persistent asthma (OR=1.08, 95%CI 1.06, 1.10; OR=2.06, 95%CI 2.01, 2.10; OR=5.24, 95%CI 4.96, 5.53), and current severe asthma (OR=1.36, 95%CI 1.32, 1.40; OR=1.50, 95%CI 1.45, 1.55; OR=3.70, 95%CI 3.46, 3.95), adjusted for confounders. Considering incident asthma, being obese more than quadrupled the odds of incident asthma (OR=4.46, 95%CI 4.30, 4.62).

Conclusion: Obesity is associated with increase prevalent and incident asthma, and seems to increase the odds of a more persistent and severe asthma phenotype independently of socio-demographic determinants, physical activity and dietary patterns.

Key-words: asthma, prevalence, obesity, body mass index, phenotypes, severe.

Word count: 2379/4000

References: 39/60

Introduction

Asthma and obesity are both complex and multifactorial chronic health conditions, wherein host and environmental factors play a determinant role, representing a serious economic and social burden in health care systems and patient's quality of life (1, 2). Although the advances in knowledge, management and treatment, prevalence of asthma is still globally increasing and asthma control stills difficult to attend (3). The lack of a precisely and consensual definition of asthma difficult reliable comparison of reported prevalence's from the different countries, however global prevalence seems to range between 1-16%, with a decrease in western Europe and a recent increase in low income regions were prevalence were previous low (2, 4, 5). Considering time trends in obesity adults from USA and NHANES data, the prevalence of obesity was higher in participants with asthma than without asthma, almost one in three subjects with asthma were obese, and prevalence of obesity between subjects with current asthma increased from 21.3% (NHANES I) to 32.8% (NHANES III) (6). Adittionaly, a recent meta-analysis from EAACI showed that weight gain, more than be obese, almost double the odds of incident asthma (7).

The concomitant increase in the prevalence of obesity and asthma lead to the interest in potential mechanisms linking these two epidemics (1). Increases in body mass index (BMI) have been associated with increased prevalence of asthma, however the mechanisms behind this association are not totally clear (8). Plausible links between asthma and obesity include mechanical factors, inflammatory conditions and stress models (9, 10). Obesity is a state of chronic and low-grade systemic inflammation with increased levels of the pro-inflammatory leptin and plasminogen activator inhibitor and decreased serum levels of the protective anti-inflammatory adiponectin (11). Nonetheless, we have previous reported a negative association between BMI and exhaled nitric oxide in overweight and obese asthmatics, providing additional support to the mechanical hypothesis linking obesity and asthma (12).

Several cross-sectional and case-control studies have found obesity to be associated with asthma diagnosis, respiratory symptoms, poor lung function and increased airway hyperreactivity (AHR) (13). Additionally, evidence suggests that obesity increases the risk of asthma and changes prevalent asthma toward a more difficult-to-control phenotype (2, 14). Severe asthma is considered a heterogeneous disease in which a variety of clinical, physiological and inflammatory markers determine the disease severity, with distinct clinical phenotypes classification (15). A meta-analysis showed that being overweight or obese increased the odds of incident asthma in a dose-dependent way (16). Additionally, obesity have impact on the asthma treatment response with increased severity of illness (17). Weight reduction in obese patients with asthma has been demonstrated to improve lung function, symptoms, morbidity, and health status (18), and better asthma control, better quality of life and reduced asthma exacerbations (7, 19-21), suggesting that reducing of weight is recommended for clinical management of obese-asthma phenotype. Previous data from National Health Survey (1998-99) reported that obesity was not related with asthma prevalence (22), suggesting that further research should considerer also other clinical outcomes, socio-economic

determinants (23), and lifestyle factors such as dietary intake and physical activity that could clarify the mechanisms underlying the epidemiology of obesity and asthma phenotype.

Therefore, in this study we aim to explore the association between obesity and asthma in a large representative adult population, adjusting for socio-economic and lifestyle confounders. Our hypothesis was that obesity will be associated with asthma, and that this association will be higher according to the severity of both diseases.

Materials and Methods

Participants and study design

Data from the 4th Portuguese National Health Survey, carried out by the National Institute of Health and National Institute of Statistics (NIS), between 2005 and 2006, was analysed. The methodology of the NHS has been previously detailed and described (24, 25). In summary, the sampling frame was made based on census data and participants were selected from households during that period, using a multi-stage random probability design (hospitals, prisons, military houses and community care institutions were excluded). A representative sample of 41193 participants was included, according to the 7 main Portuguese territorial units (NUTS II), namely North, Center*, Lisbon, Alentejo, and Algarve, and Madeira and Azores* archipelagos. The primary sampling unit (PSU) was the housing unit and sampling was based on the population and housing census. Two levels were defined within each NUTSII: the parish (corresponding to townships); and geographically defined units of 240 lodgings (within the parish). The PSUs were then randomly selected within each territorial unit, and subjects living in the sampling unit were surveyed. Interviews were conducted in the households by trained staff interviewers, and the questionnaire included information on social and demographic characteristics, health and chronic diseases including asthma. The survey response rate reported by NIS, defined as the percentage of households who responded, was 76%. (24, 25). For the purposes of this study, a representative subsample of all adults age NHS surveyed people was analysed (32644 participants).

Asthma

Following asthma definitions were used: ever asthma (ever medical doctor asthma diagnosis), current asthma (asthma within previous 12 months), current persistent asthma (asthma drugs within previous 12 months), current severe asthma (emergency because of asthma within previous 12 months), and incident asthma (diagnosis within previous 12 months).

Body mass index

Body mass index (weight/height^2) was calculated based on self-reported weight (kilograms) and height (meters), and categorized according to the World Health Organization BMI Classification (26), in underweight (<18.5 kg/m^2), normal weight (18.5-24.9 kg/m^2), overweight (25.0-29.9 kg/m^2), obesity class I (30.0-34.9 kg/m^2), class II (35.0-39.9 kg/m^2) and class III (≥ 40.0 kg/m^2).

Considering the low prevalence of underweight and the clinical higher risk of morbid obesity, for descriptive and logistic regression analyses, BMI classes were also categorized into fourth classes: underweight and normal weight; overweight; obesity class I and class III; and obesity class III.

Socio-demographic, lifestyle and dietary patterns

Age, gender, education, family income, smoking physical activity level, and dietary patterns were also addressed by NHS, and analysed as potential confounders. Dietary patterns were identified by Latent Variable Models (27, 28), based on dietary intake list of 20 usual foods and beverages consumed at meals and snacks. Dietary patterns assessment data was previously described (29) and original manuscript will be soon published elsewhere. In summary, considering that the goodness-of-fit test suggests that the 5-factor LTM was the best solution, five dietary patters were identified and analysed as confounders: factor 1 *('dairy and fruit"* DP*);* factor 2 ("*soup and starchy foods"* DP); factor 3 *("high fat, sugar and salt"* DP); factor 4 ("*fish, fruit and vegetables"* DP); and factor 5 ("*sugary and fatty foods"* DP).

Statistical analysis

Descriptive analysis, including mean (standard deviation) and proportions, was performed to characterize the sample, and were conducting with sampling weights. Student's t-tests, ANOVA, and chi-squared tests were performed to determine the association between obesity, asthma, and confounder variables. Unconditional logistic regression models were performed to analyse whether classes of BMI were associated with asthma prevalence and incidence. Age, gender, education, family income, proxy reporting information, smoking, physical activity and dietary patterns were analysed as confounders.

A 0.05 level of significance and 95% CI were considered. Data analysis was performed using the statistical package SPSS®, version 18.0 (SPSS Inc., Chicago, IL, USA).

Results

Final analysis included 32644 adults (≥20 years-old), and 52.6% female. **Table 1** summarizes the socio-demographic characteristics of participants according to asthma prevalence. Prevalence of ever asthma was 5.3%, current asthma 3.5%, current persistent asthma 3.0%, current severe asthma 1.4%, and incident asthma 0.2%. Prevalence of underweight was 2.0%, normal weight 44.6%, overweight 37.6% and obesity was 16.0% (obesity class I 12.7%, class II 2.5% and class III 0.8%).

Considering association between BMI classes and asthma prevalence adjusted for confounders (**table 2**), overweight, obesity class I and II, and obesity class III were associated with ever asthma (respectively OR=1.22, 95%CI 1.21, 1.24; OR=1.39, 95%CI 1.36, 1.41; OR=3.24, 95%CI 3.08, 3.40), current asthma (OR=1.16, 95%CI 1.14, 1.18; OR=1.86, 95%CI 1.82, 1.90; OR=4.73, 95%CI 4.49, 4.98), current persistent asthma (OR=1.08, 95%CI 1.06, 1.10; OR=2.06, 95%CI 2.01, 2.10; OR=5.24, 95%CI 4.96, 5.53), and current severe asthma (OR=1.36, 95%CI 1.32, 1.40; OR=1.50, 95%CI 1.45, 1.55; OR=3.70, 95%CI 3.46, 3.95). Being obese class III more than tripled the odds of having asthma and severe asthma, and more than quintuples the odds of having current persistent asthma. Additionally, considering incident asthma, being obese more than quadrupled the odds of incident asthma (OR=4.46, 95%CI 4.30, 4.62).

Discussion

In this study we observed that overweight and obesity in adult population, were significantly associated with ever asthma, current asthma, current severe asthma and current persistent asthma, independent of other socio-economic and lifestyle confounders, such as dietary patterns and physical activity. Additionally, the odds increases according to a gradient of BMI and asthma persistency and severity, with higher odds for obesity class III, and being obese more than quadrupled the odds of incident asthma.

Our study has some limitations. First, it is limited by the cross sectional nature of data not allowing to establish causal relationship. Secondly, asthma prevalence, incidence and control were addressed by self-report. However, this has been the usual approach in large-scale studies assessing asthma and allergic diseases prevalence. Thirdly, weight and height were also addressed by self-reported. Nevertheless, self-reported data have more viability in the large scale surveys, and significant correlates with weight and height measurements in cohort surveys. Although BMI is not the gold standard to assess body composition, it correlates with total body fat content and has been the most widely used measure to assess overweight or obesity and to monitor changes in body weight. Future research should be considered more complex anthropometric measures (e.g., waist-circumference, skinfold thickness, or bioelectrical impedance analysis) that assess total body mass distribution, including body fat mass measure, although methodologically it is difficult to measure those parameters in national surveys with so large sample.

Our findings are also supported by important strengths. It is a nationwide representative observational cross-sectional study, with a large sample of adults from both genders addressing body mass index and asthma, considering dietary patterns and physical activity in the analysis. Actually, the NHS includes important socio-economic and lifestyle confounders regarding obesity and asthma, such as age, gender, education, family income, BMI, smoking, proxy reporting information, physical activity level and dietary intake. Additionally, this is the first study on obesity and asthma adjusted for diet based on dietary patterns by LTM analyses (27, 28).

Severe asthma is considered a heterogeneous disease in which a variety of clinical, physiological and inflammatory markers determine the disease severity, with distinct clinical phenotypes classification (15). Evidence suggests that obesity increases the risk of asthma and changes prevalent asthma toward a more difficult-to-control phenotype (2, 14). Our results show that obesity increases prevalent asthma and seems to change asthma phenotype towards a more persistent and severe disease independently of socio-demographic and lifestyle determinants, namely physical activity and dietary patterns. In addition, being obese more than quadrupled the odds of incident asthma.

A recent meta-analysis showed that being overweight or obese increased the odds of incident asthma in a dose-dependent way (16). Moreover, obesity have impact on the asthma treatment response with increased severity of illness (17).

Clarifying the mechanisms underlying the epidemiology of obesity and asthma phenotype, will allows to better design weight loss interventions to overweight population to reduce asthma incidence; and for improve asthma control and quality of life asthmatic patients with obesity (7).

Increases in BMI have been associated with increased prevalence of asthma, however the mechanisms behind this association are still not totally clear (8, 30). Plausible links between asthma and obesity include mechanical factors, inflammatory conditions and stress models. Obesity has been suggested as a pro-inflammatory state (11), but the links with airway inflammation are still scarce. The increased abdominal and chest wall mass causes a decrease in functional residual capacity and a reduction in lung and tidal volumes (31, 32). Additionally, obesity is a state of chronic and low-grade systemic inflammation with increased levels of the pro-inflammatory leptin and plasminogen activator inhibitor and decreased serum levels of the protective anti-inflammatory adiponectin (11). Nevertheless, the relation between BMI and airway inflammation has provided conflicting evidence. In children, BMI had no association with exhaled NO (33-35), and we have previously reported a negative association between BMI and exhaled nitric oxide in overweight and obese asthmatics, providing additional support to the mechanical hypothesis linking obesity and asthma (12). Additionally, a case-control study described that although BMI was correlated with serum leptin levels in both groups (asthmatics vs. non-asthmatics), exhaled NO was not related with serum leptin levels in asthmatic patients (36).

Several cross-sectional and case-control studies have found obesity to be associated with asthma diagnosis, respiratory symptoms, poor lung function and increased airway hyperreactivity (AHR) (13). In addition, weight reduction in obese patients with asthma has been demonstrated to improve lung function, symptoms, morbidity, and health status (18), and better asthma control. Moreover, weight loss has been associated with better asthma control and reduced asthma exacerbations (19). A randomized trial to compare effects of dietary restriction and exercise in obese asthmatic patients, shows that a 5-10% weight loss resulted in clinical improvements to asthma control (58%) and quality of life in 83% of patients, with a reduction in neutrophilic airway inflammation, suggesting that reducing of weight is recommended for clinical management of obese-asthma phenotype (20). These results taken together suggest that nutritional intervention and dietary strategies aimed at modifying dietary fat intake may be useful in reducing airway neutrophilia, with clinical impact in asthma and control and severity. Additionally, physical activity have also an important role (37-39) in energy balance and adequate body weight maintenance, and therefore in immunomodulation linking obesity and asthma. Therefore, our results gives rational for future lifestyle intervention studies in asthma, based on dietary patterns and physical activity, for weight reduction in overweight and obese asthmatic patients.

In summary, our results show that obesity increases both prevalent and incident asthma and seems to change asthma phenotype towards a more persistent and severe disease

independently of socio-demographic and lifestyle determinants Furthermore, our study suggest that adequate body weight maintenance should be recommended as a low cost public health strategy for asthma primary prevention and minimize the burden of asthma in global health care systems and patients quality of life, and that weight management should be considered in asthma treatment, for better asthma control and reduce severity of obese-asthma phenotype.

Table 1. Characteristics of participants according to asthma prevalence

	Total n = 32644	Ever Asthma			Current Asthma			Current Persistent Asthma			Current Severe Asthma			Incident Asthma		
		Yes = 5.3%	No = 95.7%	p	Yes = 3.5%	No = 96.5%	p	Yes = 3.0%	No = 97%	p	Yes = 1.4%	No = 98.6%	p	Yes = 0.2%	No = 99.8%	p
Gender (%)																
Female	52.4	60.1	51.9	<0.01	61.3	52.1	<0.01	62.0	52.1	<0.01	65.4	52.2	<0.01	46.0	52.4	<0.01
Male	47.6	39.9	48.1		38.7	47.9		38.0	47.9		34.6	47.8		54.0	47.6	
Age (%)																
20 – 39 y	37.8	33.2	38.0	<0.01	25.7	38.2	<0.01	25.3	38.2	<0.01	28.7	37.9	<0.01	30.8	37.8	<0.01
40 – 64 y	40.5	37.9	40.7		42.1	40.5		39.4	40.6		34.9	40.6		49.4	40.5	
65 – 84 y	19.8	26.5	19.4		29.0	19.4		31.7	19.4		31.0	19.6		19.8	19.8	
≥ 85 y	1.9	2.4	1.9		3.2	1.9		3.6	1.9		5.4	1.9		0.0	1.9	
Education (%)																
None	12.7	17.0	12.4	<0.01	21.1	12,4	<0.01	20.8	12.4	<0.01	23.2	12.5	<0.01	17.5	12.7	<0.01
1st cycle	34.1	34.2	34.1		39.3	33.9		38.8	33.9		39.5	34.0		43.3	34.1	
2nd and 3rd cycles	27.8	21.8	28.1		16.1	28.2		16.3	28.1		19.2	27.9		12.6	27.8	
High school	12.2	10.3	12.3		8.9	12.4		7.7.	12.4		9.6	12.3		8.9	12.2	
College	13.1	16.5	12.9		14.6	13.1		16.4	13.0		8.5	13.2		17.8	12.7	
Family income (%)																
≤ 500€/month	23.7	29.0	23.4	<0.01	32.8	23.4	<0.01	31.5	23.5	<0.01	33.9	23.6	<0.01	28.9	23.7	<0.01
501 – 1200€/month	44.1	40.1	44.3		39.6	44.3		41.4	44.2		40.8	44.1		47.3	44.1	
1201€ - 2000€/month	20.0	17.1	20.1		14.2	20.2		16.0	20.1		15.5	20.0		0.0	20.0	
> 2000€/month	12.2	13.9	12.1		13.4	12.1		11.1	12.2		9.8	12.2		23.9	12.2	
Proxy reporting information (%)	29.8	24.5	30.0	<0.01	22.0	30.0	<0.01	22.7	30.0	<0.01	21.5	29.9	<0.01	6.9	29.8	<0.01

Data presented as proportions (%), and mean (SD). Sample weighted for all variables (pond 1); DP: dietary patterns. Qui-square test; p-value ≤0.05

Table 1. Characteristics of participants according to asthma prevalence (cont.)

	Total n = 32644	Ever Asthma			Current Asthma			Current Persistent Asthma			Current Severe Asthma			Incident Asthma		
		Yes = 5.3%	No = 95.7%	p	Yes = 3.5%	No = 96.5%	p	Yes = 3.0%	No = 97%	p	Yes = 1.4%	No = 98.6%	p	Yes = 0.2%	No = 99.8%	p
Smoking (%)																
No smoker	61.7	63.2	61.6	<0.01	63.4	61.7	<0.01	65.3	61.6	<0.01	63.2	61.7	<0.01	67.8	61.7	<0.01
Current smoker	21.4	17.7	21.6		14.8	21.6		12.8	21.7		18.8	21.4		11.7	21.4	
Past smoker	16.8	19.1	16.7		21.7	16.7		21.9	16.7		18.0	16.8		20.5	16.8	
Level Physical activity (%)																
Low	39.0	52.5	38.2	<0.01	56.2	38.3	<0.01	55.9	38.4	<0.01	64.4	38.5	<0.01	28.3	39.0	<0.01
Moderate	46.1	37.4	46.6		31.2	46.7		30.7	46.6		27.3	46.5		0.0	46.2	
Hight	14.9	10.0	15.2		12.6	15.0		13.4	15.0		8.3	15.0		71.7	14.9	
Dietary patterns, mean (SD)																
'Dairy and fruit" DP	-0.05 (0.66)	-0.30 (0.66)	-0.47 (0.66)	<0.01	-0.06 (0.65)	-0.04 (0.66)	<0.01	-0.03 (0.63)	-0.05 (0.66)	<0.01	-0.11 (0.64)	-0.04 (0.66)	<0.01	-0.7 (0.57)	-0.05 (0.66)	<0.01
"*Soup and starchy foods"* DP	-0.03 (0.56)	-0.09 (0.57)	-0.02 (0.56)	<0.01	-0.05 (0.58)	-0.03 (0.56)	<0.01	-0.06 (0.55)	-0.03 (0.56)	<0.01	-0.11 (0.59)	-0.03 (0.56)	<0.01	0.18 (0.52)	-0.03 (0.56)	<0.01
"High fat, sugar and salt" DP	0.17 (0.63)	0.20 (0.68)	0.17 (0.63)	<0.01	0.16 (0.68)	0.17 (0.63)	<0.01	0.14 (0.66)	0.17 (0.63)	<0.01	0.21 (0.71)	0.17 (0.63)	<0.01	0.06 (0.43)	0.17 (0.63)	<0.01
"Fish, fruit and vegetables" DP	-0.01 (0.58)	-0.04 (0.60)	0.01 (0.58)	<0.01	-0.03 (0.61)	0,01 (0.58)	<0.01	-0.03 (0.58)	0.01 (0.58)	<0.01	-0.10 (0.64)	0.01 (0.58)	<0.01	0.17 (0.44)	0.01 (0.58)	<0.01
"Sugary and fatty foods" DP	0.02 (0.50)	0. 00 (0.50)	0.02 (0.50)	<0.01	0.00 (0.50)	0.02 (0.50)	<0.01	0.01 (0.50)	0.20 (0.50)	<0.01	0.01 (0.48)	0.02 (0.50)	<0.01	-0-08 (0.37)	0.01 (0.50)	<0.01
BMI Classes (%)																
Underweight/normal weight	46.4	44.3	46.6	<0.01	40.1	46.7	<0.01	42.1	46.6	<0.01	45.1	46.5	<0.01	34.2	46.5	<0.01
Overweight	37.6	33.6	37.8		33.7	37.7		32.4	37.7		30.2	37.7		19.8	37.6	
Obesity classes I and II	15.2	20.2	15.0		22.9	15.0		23.0	15.0		22.6	15.1		34.4	15.2	
Obesity class III	0.8	1.9	0.7		3.2	0.7		2.5	0.7		2.1	0.7		11.7	0.7	

Data presented as proportions (%), and mean (SD). Sample weighted for all variables (pond 1); DP: dietary patterns. Qui-square test; p-value ≤0.05

Table 2. Association between BMI classes and asthma prevalence

	Ever Asthma		Current Asthma		Current Persistent Asthma		Current Severe Asthma		Incident Asthma	
	aOR (95%CI)*	**p-trend**	**aOR(95%CI)***	**p-trend**	**aOR 95%CI)***	**p-trend**	**aOR (95%CI)***	**p-trend**	**aOR (95%CI) ^**	**p-trend**
Overweight (25.0 – 29.9 kg/m^2)	**1.22 (1.21; 1.24)**	<0.001	**1.16 (1.14; 1.18)**	<0.001	**1.08 (1.06; 1.10)**	<0.001	**1.36 (1.32; 1.40)**	<0.001		
Obesity Class I and II (30.0 – 39.9 kg/m^2)	**1.39 (1.36; 1.41)**		**1.86 (1.82; 1.90)**		**2.06 (2.01; 2.10)**		**1.50 (1.45; 1.55)**			
Obesity Class III (≥ 40.0 kg/m^2)	**3.24 (3.08; 3.40)**		**4.73 (4.49; 4.98)**		**5.24 (4.96; 5.53)**		**3.70 (3.46; 3.95)**		**4.46 (4.30; 4.62)**	<0.001

aOR: adjusted Odds Ratio; p-value ≤0.05

*Unconditional logistic regression models with sampling weight, adjusted for gender, age, education, family income, proxy reporting information, smoking, physical activity level, and dietary patterns; p< 0.05

^ Unconditional logistic regression between obesity (≥ 30.0 kg/m^2) and incident asthma with sampling weight, adjusted for gender, age, education, family income, proxy reporting information, smoking, and dietary patterns; p<0.05

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Barros R, Moreira A, Padrão P, Teixeira VH, Carvalho P, Delgado L, Moreira P. Height: are asthmatic different?

Manuscript submitted

Adult height: are asthmatics different?

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Sources of support: None declared

Competing interests: None declared

Key-words: height, growth, corticosteroids, persistent, asthma, prevalence.

Words count: 913/1000

References: 9/9

Capsule summary: 35/35

Height in adult asthmatics is significant lower than non-asthmatics, and these differences increase with persistency and severity of asthma. Monitoring health conditions that may impact stature in asthmatics should be recommended to optimize height attainment.

To the Editor:

Asthma may exhibit a deleterious effect on growth attainment and adult height. Inhaled corticosteroids (ICS) are the first line treatment for persistent asthma (1), however the long-term effect of ICS on growth and adult height is controversial and a clinical concern (2, 3). Therefore, we aimed to compare height between asthmatics and non-asthmatics adults.

A representative sample from the 4th Portuguese National Health Survey (NHS) was analyzed, with a survey response rate of 76%. The final analysis included a representative sample of 32644 adults (≥20 years-old, 52.6% female). The NHS methodology has been previously detailed and described (4). Briefly, the sampling frame was made based on census data. Participants were selected from households, using a multi-stage random probability design, according to 7 territorial units (NUTSII). The primary sampling unit (PSU) was the housing unit that was randomly selected and subjects were surveyed. Interviews were conducted by trained interviewers, and included information on socio-demographic characteristics and chronic diseases, including asthma.

The following asthma definitions were used: ever asthma (EA, ever medical doctor asthma diagnosis), current asthma (CA, asthma within 12 months), current persistent asthma (CPA, asthma drugs within 12 months), current severe asthma (CSA, emergency because of asthma within 12 months), and incident asthma (IA, diagnosis within 12 months). Self-reported height (m) was analyzed by gender. Age, weight, education, family income and proxy reporting information were analyzed as confounders. Statistical analysis included hypothesis test and generalized linear models (GLM). A p-value ≤0.05 and 95%CI were considered, and SPSS® 18.0 (SPSS Inc., Chicago IL, USA) was used.

Asthma prevalence and characteristics of participants are summarized on **table 1**. Prevalence of asthma was significantly higher in women than men: EA (6.1%; 4.4%), CA (4.1%; 2.8%), CPA (3.6%; 2.4%), and CSA (1.8%; 1.0%). The mean (SD) height (m) for women and men was 1.59 (0.07) and 1.71 (0.76), respectively. Considering GLM (**table 2)**, comparing height between asthmatics and non-asthmatics, the mean difference in height (cm) for women and men was, respectively: EA (-1.0; -0.8), CA (-0.8; -1.3), CPA (-1.3; -1.5), CSA (-1.3; -1.7), and IA (-5.8; -2.7).

Our study identified significant differences in adult height between asthmatic and non-asthmatic patients that were consistent between genders, and seem to increase with persistency and severity of the disease, independent of socio-demographic determinants.

These findings are supported by important strengths. Data are based on a nationwide representative observational health survey, with the large sample of adults from both genders. Additionally, we included relevant socio-economic and lifestyle confounders regarding height and asthma. Nevertheless, several limitations should be considered: the cross-sectional nature of data, not allowing

establishing causal relationship; asthma definitions addressed by self-reporting, although being a widely used approach to assess asthma prevalence in large-scale studies; and NHS did not provide data on oral or inhaled corticosteroids (ICS).

ICS are the first line treatment for persistent asthma in children, and have well-established benefits in asthma control (1). However, the potential negative effects of ICS on bone mineral density and growth are still not clear (3, 5). The use of ICS has been shown to reduce growth velocity in prepubertal children, resulting in a linear growth reduction of approximately 1cm in the first years of therapy. Although growth velocity seems to returns to normal, the long-term effect on adult height is still inconclusive (2, 3, 6, 7). Retrospective cross-sectional studies have reported no differences between children with or without asthma, and treated or not-treated with corticosteroids (6). Nevertheless, results from CAM clinical trial showed that the initial decrease in attained height in prepubertal children persisted in adult height (3), and Cochrane recently reports a significant lower in growth velocity in children from higher ICS group (2). Additionally, there is now evidence that a longer duration of asthma was an independent risk factor for decreased height, that uncontrolled asthma can result in growth reduction, and that systemic effects of ICS are dose-dependent (3). Despite NHS have no data on corticosteroids use our results suggest that patients with more persistent, severe and difficult to treat asthma had also more impact on growth and adult height, independent of age and weight. Nevertheless, pre-school children with less weight (<15kg) have reduced linear growth, potentially because of higher corticosteroid exposure, suggesting that variations in ICS response are possible in potentially unrecognized subgroups (7).

Children with chronic diseases (e.g. type 1 diabetes, food allergy and asthma) are commonly affected by a variable degree of growth failure, leading to an impaired final adult height (8). Behind corticosteroids, several other mechanism related to asthma has been proposed, namely early disease onset, duration and severity of disease, chest deformity, chronic hypoxia, reduced lung function, chronic infection, sleep disturbance, long-term stress, under nutrition, and enhancing metabolic demands by increased work of breathing (6, 8). Additionally, growth retardation is frequently observed in chronic inflammatory states (9). The development and bone growth is variably affected by proinflammatory cytoquines (IL-1β, TNF-α, and IL-6), and other factors (e.g. vitamin D metabolites and sex steroids) (8, 9). Moreover, socio-economic and education factors might also influence growth in asthma, and then height could be an indicator of health conditions and population´s lifestyle (8). Therefore, an adequate nutritional status and low doses of ICS should be considered whenever initiating asthma treatment (1, 3, 5).

Our results strengthen the evidence that adult height in asthmatics is significant lower than non-asthmatics in both genders, and that these differences increases with persistency and severity of disease. Monitoring health conditions that may impact stature in asthmatics should be recommended to optimize height attainment.

Tables

Table 1. Characteristics of participants

	Total (n = 32644)
Gender, female (%)	52.4
Age (%)	
20 – 39 y	37.8
40 – 64 y	40.5
65 – 84 y	19.8
≥ 85 y	1.9
Education (%)	
None	12.7
1st cycle	34.1
2nd and 3rd cycles	27.8
High school	12.2
College	13.1
Family income (%)	
≤ 500€/month	23.7
501 – 1200€/month	44.1
1201€ - 2000€/month	20.0
> 2000€/month	12.2
Proxy reporting information (%)	29.8
Asthma prevalence (%)	
Ever asthma	5.3
Incident asthma	0.2
Current asthma	3.5
Current persistent asthma	3.0
Current severe asthma	1.4
BMI Classes (%)	
Underweight and normal weight	46.4
Overweight	37.6
Obesity classes I and II	15.2
Obesity class III	0.8
Level of Physical activity (%)	
Low	39.0
Moderate	46.1
Hight	14.9
Smoking (%)	
Current smoker	21.4
Past smoker	16.8

Data presented as proportions (%), and sample weighted pond 1.

Table 2. Height according to asthma prevalence by gender

Height (m)	Ever Asthma				Current Asthma				Current Persistent Asthma				Current Severe Asthma				Incident asthma			
	Yes	No	Mean Difference	Adjusted p-value	Yes	No	Mean Difference	Adjusted p-value	Yes	No	Mean Difference	Adjusted p-value	Yes	No	Mean Difference	Adjusted p-value	Yes	No	Mean Difference	Adjusted p-value
Women n = 17181	1.584 (0.070)	1.594 (0.069)	**-1.0 cm**	<0.001	1.585 (0.069)	1.593 (0.069)	**-0.8 cm**	<0.001	1.581 (0.068)	1.594 (0.069)	**-1.3 cm**	<0.001	1.580 (0.069)	1.593 (0.069)	**-1.3 cm**	<0.001	1.536 (0.065)	1.594 (0.069)	**-5.8 cm**	<0.001
Men n = 15463	1.699 (0.082)	1.707 (0.076)	**- 0.8 cm**	<0.001	1.694 (0.078)	1.707 (0.076)	**- 1.3 cm**	<0.001	1.692 (0.080)	1.707 (0.076)	**- 1.5 cm**	<0.001	1.690 (0.090)	1.707 (0.075)	**- 1.7 cm**	<0.001	1.706 (0.076)	1.733 (0.086)	**- 2.7 cm**	<0.001

Height Mean (SD); Height (m): height in meters; Mean difference (cm): centimetres; General linear models with sampling weight, adjusted for age, weight, education, family income, and proxy information. Adjusted p-value < 0.05

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5.1. DIETARY INTAKE IN ASTHMA CONTROL AND PREVENTION

5.1.1. DIETARY INTAKE OF FATTY ACIDS AND AIRWAY INFLAMMATION AND ASTHMA CONTROL (STUDY I)

In our study, higher dietary intake of n-3 PUFA and SFA were associated with decreased levels of exhaled nitric oxide and improved likelihood of asthma being under control, while high ratio of n-6:n-3 PUFA had the opposite effect. In addition, higher dietary intake of ALA was associated with lower eNO and reduced the likelihood of non-controlled asthma, independent from marine n-3 PUFA. No significant associations between dietary intake of EPA plus DHA and antioxidant vitamins and minerals and asthma outcomes were observed.

In our study higher dietary intake of n-3 PUFA (> 0.94g/ day) and ALA (> 1.96g/day) reduced the odds of non-controlled asthma. Considering that prevalence of non-controlled asthma in our study was high, the odds ratio may be biased towards overestimating the risk. Nevertheless, even though we could admit an overestimation of the protective effect, the reverse result should not be expected. ALA is the major plant-based n-3 PUFA and exerts mainly effects through conversion to EPA and DHA, when dietary intake of marine PUFA is low (102, 103). Long chain n-3 PUFA decreases the production of inflammatory mediators, competitively inhibiting the metabolism of arachidonic acid (generating less active prostenoids and leukotrienes), suppressing IgE production and, thereby potentially acting to reduce airway inflammation and bronchoconstrition in asthma (103, 104). However, results have been inconclusive. A systematic review from Cochrane of the clinical effects of n-3 PUFA fish oil supplementation in established asthma, suggests that results are not consistent and that there is little evidence to recommend such supplementation in order to improve asthma control (105). Reconciling the data from experimental and observational studies is difficult most probably due to different methods of assessment of dietary intake and different definitions of asthma. Taken the data from previous cross sectional studies, it seems dietary or serum n-3 PUFA levels are directly associated with lung function, at least in asthmatics (106-108) and atopy (109) and are protective for the risk of asthma or atopy.

Recently in a large population-based study, asthma risk was doubled in subjects who had never eaten fish during childhood and a minimum of weekly fish intake in adulthood was protective against asthma symptoms (110). In a small study, fish oil supplementation failed to provide any benefit in exhaled NO, lung function or asthma control in asthmatic women evidence (36). In our study dietary intake of n-3 PUFA and ALA were associated with improved asthma control and lower eNO, independent of EPA plus DHA. Higher intake of ALA (and also not EPA or DHA) was previous associated with decreased risk of allergic sensitization and allergic rhinitis in adults (111). However the link between ALA and asthma is still poorly addressed. In our study dietary

intake of EPA and DHA is very similar between controlled and non-controlled subjects, and we have found no significant associations between EPA or DHA and asthma outcomes. There is evidence suggesting that ALA, EPA and DHA might have heterogeneous and potentially independent effects on inflammation, gene expression and chronic diseases, therefore a better understanding of the individual role of n-3 PUFA in inflammatory diseases, such as asthma, is needed low (102, 103). It has been suggested that higher margarine intake rich in n-6 PUFA is associated with increased risk of asthma (112, 113) and hay fever (114) in adulthood, and eczema and allergic sensitization in children (115). Dietary intake of n-6 PUFA was similar among controlled and non-controlled subjects and therefore no significant associations for n-6 PUFA and asthma outcomes were observed. Nevertheless, a ratio of n-6/n-3 PUFA above 8.45 more than tripled the odds of non-controlled asthma and was associated with increased levels of exhaled nitric oxide.

In our study we analyse total SFA intake, irrespective to the specific types. However different type of saturated fatty acids could have different effects. Foods high in SFA, such as butter (116), whole milk (116, 117), and non-pasteurized farm milk (118-120) have been consistently associated with reduced risk of asthma. For milk it is not clear whether associations should be attributed to the SFA, vitamin A, or even microbial agents (in the case of whole non-pasteurized milk or farm-related co-exposures arming environments) (118-120). Therefore, our results on SFA could also be a proxy of a dietary pattern high in milk and dairy products. Several epidemiological studies have reported beneficial associations for higher intake of nutrients that may be relevant in the redox mechanisms, such as vitamin C (106, 121, 122), vitamin E (121, 123), carotenoids (124, 125), selenium (87) and magnesium (126). However these findings are not conclusive (127) and intervention studies with single nutrient supplementation have been disappointed (128, 129) (130, 131). Inverse associations with asthma have been also observed for foods rich in these micronutrients, such as fresh fruit (87, 117, 122, 132), vegetables (132), and nuts (41, 133, 134), and additional benefits may arise from the synergistic effects between nutrients in foods and specific dietary patterns. Nuts contain a high proportion of ALA, fibre, vitamins, minerals and many bioactive compounds that may modulate redox status, and inflammatory and immune response (114, 135). We have previous reported that intake of nuts was positively associated with lung function and high adherence to an overall healthy dietary pattern such as the Mediterranean diet is associated with improved asthma control in adults, independent of other lifestyle factors.

In summary, our results provide additional support for the benefits of adequate dietary advice and give rational to nutritional intervention studies in asthmatics. Healthy eating in asthma, providing foods high in ALA, such as nuts, and an adequate balance between n-6 and n-3 FUFA, may reduce disease severity and improve asthma control, independent of other lifestyle factors.

5.1.2. DIETARY PATTERNS AND ASTHMA PREVALENCE, INCIDENCE AND CONTROL (STUDY II)

Our study shows a protective association between essential components of a *Mediterranean diet*, such as eating *"fish, vegetables and fruit",* and asthma prevalence and persistency. In contrast, *"high fat, sugar and salt'* dietary patterns, based on high-energy density/low micronutrient foods are associated with increased prevalence and severity of asthma, independent of other socio-economic and lifestyle factors.

There are few studies addressing diet and asthma by dietary patterns approach, and results have been controversial (51-53). However, *"fish, fruit and milk"* has been previously associated with reduced asthma risk in young adults (55). Comparison of dietary patterns between studies is complex, due to differences in the socio-geographic characteristics of the studied populations, and the methodological approaches to assess dietary intake and food patterns analysis (136), particularly in cross-sectional studies and with unmeasured confounders. Nevertheless, our findings are consistent with the reported literature, even when different dietary intake assessment methods were applied. In the European Community Respiratory Health Survey II, authors described a potential beneficial effect of fish, fruits and vegetable on current asthma and asthma symptoms (57). Fish, vegetables and fruit are essential components of *Mediterranean Diet* (137). We have previously reported that dietary intake of fruit (≥300g) was associated with better asthma control in adults (41), and data from ISAAC phase 3 reported also a protective effect of fruit (≥ 3 times/week) on severe asthma in children and adolescents (45). Dietary intake of fish, fruit, nuts and vegetables has been beneficially associated with asthma outcomes in several epidemiological studies during lifecycle (27, 29, 30, 138). Actually, these foods are rich in n-3 PUFA (EPA, DHA, alpha-linolenic acid), vitamins, minerals and methyl donors.
The interaction between all these nutrients, foods and meals in a whole dietary pattern could explain the antioxidant, anti-inflammatory and immunomodulator mechanisms in asthma. In addition, fruit, vegetables, nuts and pulses (139) are also fiber food sources that seems to increase the proportion of *Bacterioidacea* in the microbiota. Dietary fiber has effects on the gut microbiota balance, in the short-chain fatty acids (SCFAs) production, including propionate, which potentially reduce AHR and the severity of allergic airway inflammation (140, 141). These Mediterranean healthful food components (137) can have a favourable impact on several chronic inflammatory diseases (39, 40, 142), including reduced asthma risk in prospective studies in children (43), and better asthma control in adults (41). Nevertheless, intervention studies based on whole healthy dietary patterns are still lacking.

Conversely, we also observed a detrimental association between *"high fat, sugar and salt"* DP and severe asthma prevalence. A *"Western"* DP has been previous associated with asthma symptoms in children (54), young adults (55), and asthma attacks in the E3N female study (56). In addition, fast food intake has been associated with asthma symptoms, exercise-induced BHR and increased risk of severe asthma in children (44, 45). Looking to the potential mechanisms, high-total fat meals and high-trans fatty acids meal have been associated with increase airway

inflammation in asthmatic patients, resulted by an increase in sputum neutrophils and TLR4mRNA expression in sputum cells, activating the innate immune response. This fat induced inflammation impairs the airway response to bronchodilator and worse clinical outcomes, namely in non-atopic patients (46), suggesting that nutritional intervention strategies aimed at modifying dietary fat may be useful in reducing airway neutrophilia, with clinical impact in asthma control and severity.

Our study supports the rational for lifestyle intervention studies in asthma, based on whole dietary patterns that includes fruit, vegetables and fish, such as *Mediterranean* or *prudent healthy* dietary patterns (143), and reduced intake of high energy density/low micronutrient foods, such as *"high fat, sugar and salt"* DP. Moreover, DP approach allows to better understand the interactions between nutrients, foods and meals on asthma, being conceptually easier for dietary recommendations. Incident asthma and poor asthma control are associated with higher medical costs, increased productivity loss, and substantial reduction in quality of life. Therefore, nutritional intervention based on a healthy dietary pattern, adequate body weight maintenance, and regular physical activity could contribute as a low cost public health lifestyle strategy to reduce asthma prevalence and incidence, and improve asthma control management.

In summary, our results suggest a protective association between *"fish, vegetables and fruit"* DP and current and persistent asthma and, conversely, a detrimental association between *"high fat, sugar and salt*" DP and severe asthma prevalence, independent of other socio-economic and lifestyle factors. These data supports the rational for diet and lifestyle intervention studies in asthma based on whole dietary patterns and physical activity.

5.2. Obesity and height in asthma control and prevention

5.2.1. Obesity and asthma prevalence, incidence and severity (study III)

In this study we observed that overweight and obesity in adult population, were significantly associated with ever asthma, current asthma, current severe asthma and current persistent asthma, independent of other socio-economic and lifestyle confounders, such as dietary patterns and physical activity. Additionally, the odds increases according to a gradient of BMI and asthma persistency and severity, with higher odds for obesity class III, and being obese more than quadrupled the odds of incident asthma.

Severe asthma is considered a heterogeneous disease in which a variety of clinical, physiological and inflammatory markers determine the disease severity, with distinct clinical phenotypes classification (76). Evidence suggests that obesity increases the risk of asthma and changes prevalent asthma toward a more difficult-to-control phenotype (1, 77). Our results show

that obesity increases prevalent asthma and seems to change asthma phenotype towards a more persistent and severe disease independently of socio-demographic and lifestyle determinants, namely physical activity and dietary patterns. In addition, being obese more than quadrupled the odds of incident asthma. A recent meta-analysis showed that being overweight or obese increased the odds of incident asthma in a dose-dependent way (73). Moreover, obesity have impact on the asthma treatment response with increased severity of illness (78).

Clarifying the mechanisms underlying the epidemiology of obesity and asthma phenotype, will allows to better design weight loss interventions to overweight population to reduce asthma incidence; and for improve asthma control and quality of life asthmatic patients with obesity (75). Increases in BMI have been associated with increased prevalence of asthma, however the mechanisms behind this association are still not totally clear (58, 59). Plausible links between asthma and obesity include mechanical factors, inflammatory conditions and stress models. Obesity has been suggested as a pro-inflammatory state (60), but the links with airway inflammation are still scarce. The increased abdominal and chest wall mass causes a decrease in functional residual capacity and a reduction in lung and tidal volumes (61, 62). Additionally, obesity is a state of chronic and low-grade systemic inflammation with increased levels of the pro-inflammatory leptin and plasminogen activator inhibitor and decreased serum levels of the protective anti-inflammatory adiponectin (60). Nevertheless, the relation between BMI and airway inflammation has provided conflicting evidence. In children, BMI had no association with exhaled NO (64-66), and we have previously reported a negative association between BMI and exhaled nitric oxide in overweight and obese asthmatics, providing additional support to the mechanical hypothesis linking obesity and asthma (69). Additionally, a case-control study described that although BMI was correlated with serum leptin levels in both groups (asthmatics vs. non-asthmatics), exhaled NO was not related with serum leptin levels in asthmatic patients (68).

Several cross-sectional and case-control studies have found obesity to be associated with asthma diagnosis, respiratory symptoms, poor lung function and increased airway hyperreactivity (AHR) (70). In addition, weight reduction in obese patients with asthma has been demonstrated to improve lung function, symptoms, morbidity, and health status (71), and better asthma control. Moreover, weight loss has been associated with better asthma control and reduced asthma exacerbations (72). A randomized trial to compare effects of dietary restriction and exercise in obese asthmatic patients, shows that a 5-10% weight loss resulted in clinical improvements to asthma control (58%) and quality of life in 83% of patients, with a reduction in neutrophilic airway inflammation, suggesting that reducing of weight is recommended for clinical management of obese-asthma phenotype (74).

These results taken together suggest that nutritional intervention and dietary strategies aimed at modifying dietary fat intake may be useful in reducing airway neutrophilia, with clinical impact in asthma and control and severity. Additionally, physical activity have also an important role (23, 144, 145) in energy balance and adequate body weight maintenance, and therefore in immunomodulation linking obesity and asthma. Therefore, our results gives rational for future

lifestyle intervention studies in asthma, based on dietary patterns and physical activity, for weight reduction in overweight and obese asthmatic patients.

In summary, our results show that obesity increases both prevalent and incident asthma and seems to change asthma phenotype towards a more persistent and severe disease independently of socio-demographic and lifestyle determinants Furthermore, our study suggest that adequate body weight maintenance should be recommended as a low cost public health strategy for asthma primary prevention and minimize the burden of asthma in global health care systems and patients quality of life, and that weight management should be considered in asthma treatment, for better asthma control and reduce severity of obese-asthma phenotype.

5.2.2. Asthma impact on adult height (study IV)

Our study identified significant differences in adult height between asthmatic and non-asthmatic patients that were consistent between genders, and seem to increase with persistency and severity of the disease, independent of socio-demographic determinants.

ICS are the first line treatment for persistent asthma in children, and have well-established benefits in asthma control (79). However, the potential negative effects of ICS on bone mineral density and growth are still not clear (81, 146). The use of ICS has been shown to reduce growth velocity (GV) in prepubertal children, resulting in a linear growth reduction of approximately 1cm in the first years of therapy. Although GV seems to returns to normal, the long-term effect on AH is still inconclusive (80, 81, 147, 148). Retrospective cross-sectional studies have reported no differences between children with or without asthma, and treated or not-treated with corticosteroids (147). Nevertheless, results from CAM clinical trial showed that the initial decrease in attained height in prepubertal children persisted in AH (81), and Cochrane recently reports a significant lower in GV in children from higher ICS group (80). Additionally, there is now evidence that a longer duration of asthma was an independent risk factor for decreased height, that uncontrolled asthma can result in growth reduction, and that systemic effects of ICS are dose-dependent (81). Despite NHS have no data on corticosteroids use our results suggest that patients with more persistent, severe and difficult to treat asthma had also more impact on growth and AH, independent of age and weight. Nevertheless, pre-school children with less weight (<15kg) have reduced linear growth, potentially because of higher corticosteroid exposure, suggesting that variations in ICS response are possible in potentially unrecognized subgroups (148).

Children with chronic diseases (e.g. type 1 diabetes, food allergy and asthma) are commonly affected by a variable degree of growth failure, leading to an impaired final AH (82). Behind corticosteroids, several other mechanism related to asthma has been proposed, namely early

disease onset, duration and severity of disease, chest deformity, chronic hypoxia, reduced lung function, chronic infection, sleep disturbance, long-term stress, under nutrition, and enhancing metabolic demands by increased work of breathing (82, 147). Additionally, growth retardation is frequently observed in chronic inflammatory states (149). The development and bone growth is variably affected by proinflammatory cytoquines (IL-1β, TNF-α, and IL-6), and other factors (e.g vitamin D metabolites and sex steroids) (82, 149). Moreover, socio-economic and education factors might also influence growth in asthma, and therefore height could be an indicator of health conditions and population´s lifestyle (82). Therefore, an adequate nutritional status and low doses of ICS should be considered whenever initiating asthma treatment (79, 81, 146).

Our results strengthen the evidence that adult height in asthmatics is significant lower than non-asthmatics in both genders, and that these differences increases with persistency and severity of disease. Monitoring health conditions that may impact stature in asthmatics should be recommended to optimize height attainment.

5.3. Methodological considerations

5.3.1. Dietary intake of fatty acids and antioxidants and asthma control (study I)

Our results are limited by the cross sectional design of the study which leaves open any possible cause-effect relationship and the role of other factors. Nevertheless, an inverse causal relationship is not probable and we assessed established lifestyle factors that could have an important role in asthma and that influence nutrients intake, such as total energy intake, physical activity and body mass index, and the association between nutrients and asthma outcomes were extensively adjusted for confounders. To the best of our knowledge, this is the first study exploring the association between different types of dietary fatty acids and antioxidant nutrients intake, and asthma control. Moreover we assess dietary intake of vegetable (ALA) and marine (EPA plus DHA) n-3 PUFA, and report for the first time a protective effect of ALA in asthma control. The score we used to assess control, which included different dimensions of the disease such as inflammation, lung function and symptoms, has been shown to explain 77 % of the variability of asthma control (89). Other important strength of our study was the FFQ that we used, since it has been validated for Portuguese adults (85), and it has been shown to provide reliable estimates for n-3 PUFA and SFA (85).

In our study higher dietary intake of n-3 PUFA (> 0.94g/ day) and ALA (> 1.96g/day) reduced the odds of non-controlled asthma. Considering that prevalence of non-controlled asthma in our study was high, the odds ratio may be biased towards overestimating the risk. Nevertheless, even though we could admit an overestimation of the protective effect, the reverse result should not be expected.

5.3.2. Dietary patterns and asthma prevalence, incidence and control (study II)

Our study has some limitations. First, it is limited by the cross sectional nature of data not allowing to establish causal relationship. Secondly, dietary intake assessment was based on a checklist of usual foods intake at meals and snacks in the previous 24h, which could be considered less informative than other dietary assessment methods on frequency, portion weight, variety (other foods outside the list could have been excluded), and specificity (e.g. milk, cheese and yogurt are aggregated in the same question) (150). Nonetheless, the European Food Safety Authority, recommend 24h diet recall for the collection of national food consumption data in adults (151). Thirdly, asthma prevalence, incidence and control were addressed by self-report. However, this has been the usual approach in large-scale studies assessing asthma and allergic diseases prevalence. Our findings are also supported by important strengths. It is a nationwide representative observational cross-sectional study, with a large sample of adults from both genders addressing DP and asthma. Additionally, this is the first study on asthma that derives DP by LTM analysis (49, 50), which allows an increase in the discrimination of dietary intake and will better support public health dietary recommendations. In addition, the NHS includes important socio-economic and lifestyle confounders regarding diet and asthma, such as age, gender, education, family income, BMI, smoking, proxy reporting information, and physical activity level. Although data of physical activity was available for only 14% participants, logistic regression analysis was performed for these showing that *"fish, fruit and vegetables"* DP still remained negatively associated with current asthma, while *"high fat, sugar and salt"* DP was no longer associated with ever asthma prevalence and current severe asthma. Despite these results cannot be extrapolated for the whole sample, they suggests that a *"fish, fruit and vegetables"* DP may have a protective role on asthma independently of physical activity, while physical activity could modulate the detrimental association between a *"westernized"* DP and asthma prevalence and severity (22, 23).

5.3.3. Obesity and asthma prevalence, incidence and control (study III)

Our study has some limitations. First, it is limited by the cross sectional nature of data not allowing to establish causal relationship. Secondly, asthma prevalence, incidence and control were addressed by self-report. However, this has been the usual approach in large-scale studies assessing asthma and allergic diseases prevalence. Thirdly, weight and height were also addressed by self-reported. Nevertheless, self-reported data have more viability in the large scale surveys, and significant correlates with weight and height measurements in cohort surveys. Although BMI is not the gold standard to assess body composition, it correlates with total body fat content and has been the most widely used measure to assess overweight or obesity and to monitor changes in body weight. Future research should be considered more complex anthropometric measures (e.g., waist-circumference, skinfold thickness, or bioelectrical impedance analysis) that assess total body mass distribution, including body fat mass measure,

although methodologically it is difficult to measure those parameters in national surveys with so large sample. Our findings are also supported by important strengths. It is a nationwide representative observational cross-sectional study, with a large sample of adults from both genders addressing body mass index and asthma, considering dietary patterns and physical activity in the analysis. Actually, the NHS includes important socio-economic and lifestyle confounders regarding obesity and asthma, such as age, gender, education, family income, BMI, smoking, proxy reporting information, physical activity level and dietary intake. Additionally, this is the first study on obesity and asthma adjusted for diet based on dietary patterns by LTM analyses (49, 50).

5.3.4. Asthma impact on adult height (study IV)

Our findings are supported by important strengths. Data are based on a nationwide representative observational health survey, with the large sample of adults from both genders. Additionally, we included relevant socio-economic and lifestyle confounders regarding height and asthma. Nevertheless, several limitations should be considered: the cross-sectional nature of data, not allowing establishing causal relationship; asthma definitions addressed by self-reporting, although being a widely used approach to assess asthma prevalence in large-scale studies; and NHS did not provide data on oral or ICS.

5.4. Clinical implications and future research

Asthma is a complex and multifactorial disease, wherein environmental factors play a determinant role in development and expression of asthma, even though gene-environment interactions have to be considered. Dietary patterns, obesity and physical activity seems to modulate prevalence and incidence of asthma, and control and severity of disease in asthmatic patients, therefore our findings support lifestyle hypothesis and have a relevant impact on clinical nutrition and public health nutrition on asthma.

Incident asthma and poor asthma control are associated with higher medical costs, increased productivity loss, and substantial reduction in patient's quality of life. Currently, the Global Initiative for Asthma Guidelines does not include dietary recommendations for asthma. Nevertheless, Global Strategy for Asthma Management and Prevention, reinforces that both pharmacological and non-pharmacological therapies and strategies are important in asthma management, and encourage asthmatic patients to engage in regular physical activity, to consume a diet high in fruits and vegetables for its general health benefits (evidence A), and include weight reduction in the treatment plan for obese patients with asthma (evidence B).

Our findings support the benefits of dietary recommendation as low cost public health nutrition strategy for asthma primary prevention, based on healthy lifestyle, namely dietary patterns that include fresh fruit, vegetables, nuts, pulses and fish, and less high energy density/low micronutrient foods (namely high fat, sugar or salt foods), regular physical activity and adequate weight.

Additionally, body weight and obesity strongly depends on energetic balance between total energy intake from dietary intake, and total energy expenditure from physical activity level. Therefore, nutritional and physical activity intervention for reducing of weight in patients with overweight and obesity is recommended for clinical management of obese-asthma phenotype, with clinical impact in asthma control and severity. Monitoring nutritional status, physical activity level, body weight and health conditions that may impact also stature in asthmatics should be considered as complementary clinical approach on asthma treatment to optimize adult height attainment and global health condition.

In summary, taken together our studies provides further support to the role of lifestyle factors in asthma. Therefore, nutritional intervention based on a healthy dietary pattern, adequate body weight maintenance, and regular physical activity could contribute as a low cost public health lifestyle strategy for primary prevention of asthma, and to improve asthma control management. As future perspectives we believe that the results of this thesis will provide rationale to future dietary guidelines in asthma, and well-designed lifestyle intervention studies, based on whole dietary patterns and physical activity in asthma, and for weight reduction in overweight and obese asthmatic patients.

The main conclusions of the thesis are the following:

(1) Higher intakes of n-3 polyunsaturated fatty acids and alpha-linolenic acid (ALA) were associated with good asthma control, while the risk for uncontrolled asthma increased with a higher n-6 to n-3 PUFA ratio. The present results introduce a protective effect of ALA in asthma control, independent of marine n-3 fatty acids, and provide a rationale to dietary intervention studies in asthma.

(2) A "*Fish, vegetables and fruit"* dietary pattern was inversely associated with current and persistent asthma prevalence. Conversely *"high fat, sugar and salt"* dietary pattern, based on high energy density/low micronutrient density foods, was associated with severe asthma prevalence, independent of other socio-economic and lifestyle factors. These data supports the rational for diet and lifestyle intervention studies in asthma based on whole dietary patterns and physical activity.

(3) Obesity is associated with increase prevalent and incident asthma, and seems to increase the odds of a more persistent and severe asthma phenotype independently of socio-demographic determinants, physical activity and dietary patterns.

(4) Adult height in asthmatics was significant lower than non-asthmatics, independent of socio-demographic determinants. These differences in height were consistent between genders, and seem to increase with persistency and severity of asthma. Monitoring health conditions that may impact stature in asthmatics should be recommended to optimize height attainment.

Taken together our studies provides further support to the role of lifestyle hypothesis in asthma, including dietary intake, obesity and height, with clinical impact on asthma control and prevention. As future perspectives we believe that the results of this thesis will provide rationale for future dietary guidelines in asthma, and dietary intervention studies, based on whole dietary patterns and physical activity, as a complementary public health strategy to asthma primary prevention, and to improve asthma control management and quality of life of asthmatic patients.

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ACKNOWLEDGEMENTS

Agradeço profundamente a todos os que me acompanharam nesta viagem e que contribuíram para a concretização de um sonho!

Felizmente, ao longo desta viagem, tive o privilégio, a honra e o orgulho de ser orientada pelos melhores Professores… E as palavras não chegam para descrever a imensa gratidão e o papel que têm tido na minha vida e no meu crescimento, como estudante, como nutricionista, como investigadora, como assistente, como pessoa… Serão sempre os meus Professores!
Ao Professor Pedro Moreira e ao Professor André Moreira, agradeço profundamente todos estes anos de trabalho conjunto, de profunda inspiração e admiração! Pela beleza, humildade e generosidade com que sempre partilharam o conhecimento científico, clínico, docente e pedagógico… Por serem brilhantes! Por serem eternamente responsáveis pelo meu gosto pela nutrição, pela docência, e em especial pela investigação na área da nutrição humana e das doenças alérgicas. Pela motivação, por todos os desafios e oportunidades que me proporcionaram, e pela colaboração fundamental que têm tido em todo o meu percurso académico. Pelo privilégio de poder trabalhar numa *Dream Team*, pela riqueza das experiências e alegrias partilhadas! Por toda a disponibilidade, dedicação, apoio e paciência, nos momentos mais difíceis. Pela amizade, e por tudo o que são… Muito obrigada por tudo! Esta tese é vossa!...

À Dr.ª M. Graça Castel-Branco e a toda a equipa do Departamento de Imunoalergologia do Hospital de São João, EPE, e ao Professor Luís Delgado e a toda a equipa do Laboratório de Imunologia, da Faculdade de Medicina da Universidade do Porto, um agradecimento muito especial pela colaboração preciosa que tiveram na inclusão dos doentes no primeiro estudo, pela amabilidade e disponibilidade com que me acolheram, pela dedicação, pela motivação, pelo trabalho de equipa, pela aprendizagem constante, pelas discussões científicas, pelas oportunidades proporcionadas e alegrias partilhadas, e por me fazerem sempre sentir em casa.

A todos os coautores e amigos, com quem tenho tido o privilégio de trabalhar nesta área, nomeadamente ao Dr. Ferraz de Oliveira, ao Professor João Fonseca, à Dr.ª Cristina Lopes, à Professora Carla Lopes, ao Professor Milton Severo, ao Professor Vítor Hugo Teixeira, ao Dr. Pedro Carvalho e à Professora Patrícia Padrão pela colaboração fundamental que tiveram nesta jornada, pela disponibilidade, pela inspiração, pela discussão e partilha de conhecimentos, pela aprendizagem constante, pela motivação, pelo apoio, disponibilidade e espírito de entreajuda, pela amizade e pelas alegrias partilhadas.

A special acknowledgement to Professor Tari Haahtela, for his kindly and important collaboration on the first study, for his brilliant knowledge and simplicity in science and life, for his friendship, motivation and inspiration!

A special acknowledgement to Professor Philip Calder, for all the improvements on the first study manuscript and final publication.

Ao Instituto Nacional de Saúde Dr. Ricardo Jorge e ao Instituto Nacional de Estatística, pela autorização da utilização da base de dados do INS 2004-06, para análise, investigação e desenvolvimento dos estudos 2, 3 e 4.

À Professora Maria Daniel Vaz de Almeida, Diretora da FCNAUP, instituição em que tive a honra de me licenciar, de desenvolver o mestrado e o doutoramento em nutrição clínica, e na qual tenho o privilégio de trabalhar, pela possibilidade que me proporcionou de desenvolver o projeto de doutoramento em simultâneo com as funções de docente na FCNAUP, pela aprendizagem constante, pelo apoio e motivação ao longo desta etapa.

À Professora Flora Correia, Coordenada do Doutoramento em Nutrição Clínica, pela disponibilidade e dedicação sempre demonstradas, por todo o apoio e motivação durante esta etapa, pela amizade e pelas oportunidades de colaboração no Mestrado em Nutrição Clínica, na área da alergia alimentar.

A toda a comunidade FCNAUP, pela motivação e apoio diário. A Dr.ª Marta Azevedo, pela disponibilidade e apoio na revisão das referências bibliográficas. Um agradecimento especial ao Professor Pedro Graça, à Professora Patrícia Padrão e à Dr.ª Maria João Gregório, pelo apoio fundamental nesta etapa comum, pelo companheirismo, pelo espírito de entreajuda, pela motivação, pelos trabalhos conjuntos e alegrias partilhadas, pela amizade. Aos nossos estudantes, pela energia, pela inspiração, pelo desafio e responsabilidade de aprender mais e partilhar melhor.

À minha grande família e aos meus amigos, por tudo o que sou, por tudo que são… Pelos valores, pela inspiração, pela motivação, por terem acreditado sempre, pelo apoio incondicional nos momentos mais difíceis, por me fazerem sorrir todos os dias e pela felicidade indescritível que é conseguir chegar convosco até aqui! Muito obrigada por tudo! Esta tese é vossa!...