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Title: Diet-induced weight loss in obese children with asthma: A randomized controlled trial

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Abstract

Background: Obesity is highly prevalent in asthmatic children and associated with worse clinical outcomes. The use of energy restriction to induce weight loss in asthmatic children has not been investigated in a RCT.

Objective: To investigate the effect of diet-induced acute weight loss on respiratory outcomes in obese asthmatic children.

Methods: In a 10-week pilot RCT, obese asthmatic children, aged 8-17yrs, were randomised to a wait-list control (WLC) (n=15) or dietary-intervention group (DIG) (n=13). Lung function, Asthma Control Questionnaire (ACQ) score, and sputum and systemic inflammation were assessed at baseline and post-intervention. (Australian New Zealand Clinical Trials Registry: ACTRN12610000955011).

Results: Body mass index (BMI) *z*-score reduced significantly in the DIG versus the WLC (-0.2[-0.4, -0.1] vs. 0.0[-0.1, 0.0], p=0.014). ERV increased significantly within the DIG, but not compared to the WLC (0.7[0.0, 1.0]L vs. 0.3[0.0, 0.8]L, p=0.355). ACQ improved significantly in the DIG, compared to the WLC (-0.4[-0.7, 0.0] vs. 0.1[0.0, 0.6], p=0.004). Airway and systemic inflammation did not change within the DIG. In comparison, CRP increased significantly in the WLC (-0.4[-0.5, 0.4] vs. 0.7[-0.1, 1.9], p=0.037). ΔBMI *z*-score correlated with ΔCRP (*r*=0.47, p=0.012) and ΔeNO (*r*=0.46, p=0.034), and ΔACQ was associated with ΔCRP(*r*=0.43, p=0.029).

Conclusion: Dietary intervention can induce acute weight loss in obese asthmatic children with subsequent improvements in static lung function and asthma control. Systemic and airway inflammation did not change following weight loss. However, changes in BMI *z*-score were associated with changes in airway and systemic inflammation and this requires further investigation in a larger RCT.

Key Words

asthma; body mass index; body composition; child; diet; inflammation; weight loss; pediatric; respiratory function tests; obesity

Introduction

Obesity is highly prevalent in the asthmatic population, with almost one in two children with asthma carrying excess weight[1], compared to approximately one quarter of the general population[2]. Addressing the high prevalence of obesity in children with asthma is of critical importance. Once a child becomes overweight or obese the risk of obesity tracking into adulthood is dramatically increased[3]. The detrimental effects of obesity upon adult respiratory status has been well documented[4], with significant lung restriction[5], steroid resistance[6], altered airway inflammation[7, 8], and raised systemic inflammation[8] characterising factors. There is also increasing evidence to suggest that excess weight in children with asthma is associated with worse asthma control and increased risk of exacerbations[4, 9], reduced static lung function[10, 11] and reduced steroid efficacy[12], complicating their management.

Despite the heterogeneity in interventions and measurement outcomes, weight loss in asthmatic populations consistently demonstrates a significant improvement in asthma outcomes[13, 14], notably lung function[15-18], asthma control and severity[15-18], airway responsiveness[15, 16, 18], medication use[16, 18], and quality of life[15, 18]. With the exception of one uncontrolled study[17], previous weight loss interventions in the asthmatic population have been conducted in adults. The majority of these studies have investigated surgically-induced weight loss[13], while few have used very low to low calorie diets[14] or combination therapy[15, 17]. Surgical intervention has achieved up to 35-45% weight loss within 1-3yr follow-up in asthmatic adults[13, 16], with associated improvements in dynamic and static lung function, airway reactivity and medication usage[13, 16, 18]. Dietary interventions conducted in asthmatic adults have ranged from 8-26weeks in duration and achieved approximately 8-19% weight loss, with associated improvements in asthma control, lung function, bronchodilator response, quality of life, self-reported dyspnea and rescue medication use[14, 15].

Evidence suggests that interventions including a dietary component are effective in achieving weight loss in non-asthmatic children[19]. Only one weight loss study has been conducted in obese asthmatic adolescents, which used a 12-month combined dietary, physical activity, psychological and medical intervention[17]. This uncontrolled pre-post study reported significant improvements in lung function, asthma severity and symptoms, with approximately 13% weight loss[17]. However, whether an energy-restricted diet can induce acute weight loss in obese children with asthma and achieve significant improvements in asthma outcomes has not been investigated in a randomised controlled trial (RCT).

Therefore, the aim of this study was to investigate whether: a) acute weight loss can be achieved in children with asthma in a 10-week RCT using dietary intervention alone; and b) dietary-induced weight loss is associated

77 with changes in asthma outcomes, including systemic and airway inflammation, lung function and asthma
78 control.
79

Methods

Study design

Participants

Obese children (BMI *z*-score ≥1.64 standard deviation score (SDS)), aged 8-17 years, with a physician diagnosis of asthma were recruited from the John Hunter Children's Hospital (JHCH) outpatients, local medical centres and the general community in Newcastle, Australia. Participants were randomised by a statistician to one of two groups: (1) Dietary intervention (DIG); or (2) Wait-list control (WLC), who received the intervention after the initial 10-week control period **(Fig 1)**. Exclusion criteria included unexplained weight change during the past 3mths, inflammatory or endocrine disorders, and respiratory disorders other than asthma. Participant assent and guardian consent were obtained. The study was registered with the Australian New Zealand Clinical Trials Registry (ACTRN12610000955011) and approved by the Hunter New England and University of Newcastle Human Research Ethics Committees (09/05/20/5.08).

Intervention

Participants randomised to the intervention group underwent a 10 week dietary intervention, which targeted a 500-kcal/day energy reduction from individually calculated age- and gender-appropriate energy requirements (Schofield equation to estimate basal metabolic rate using activity factor of 1.55)[20]. Participants were required to attend counselling sessions with an Accredited Practising Dietitian (MEJ) in weeks 0, 1, 2, 4, 6, 8, and 10 and were contacted via telephone in alternate weeks. Sessions involved theoretical and practical education on selection of foods and appropriate serving sizes to optimise macronutrient and micronutrient intakes within an energy-restricted diet; identification and resolution of barriers to dietary change; and goal-setting. Materials included individually adapted meal plans and a commercial calorie counter. Participants were encouraged to self-monitor energy intake using a food diary throughout the study period.

Clinical visits

Clinical assessment

Participants attended JHCH after an overnight fast (≥12hrs) and withholding antihistamines and asthma medications (≥24hrs). Asthma stability was confirmed, defined as no exacerbation, respiratory tract infection or oral corticosteroid use in the past 4 weeks. Clinical asthma pattern, current asthma status, and quality of life were assessed using Global Initiative for Asthma (GINA) guidelines[21], Juniper Asthma Control Questionnaire (ACQ)[22], and Paediatric Asthma Quality of Life Questionnaire (standardised) (PAQLQ(s))[23], respectively, in their original unmodified form. Atopy was determined by positive skin prick test to common allergen(s)

(Aspergillus fumigatus, Alternaria tenius, Dermatophagoides Pteronyssinus, Cockroach mix, Grass mix). Tobacco exposure was measured by urinary cotinine (NicAlert, Nymox Pharmaceutical Corp, USA NJ). Dynamic and static lung function was measured using spirometry (Windows KoKo PFT System Version 4.9 2005, PDS Inc Louisville USA) and plethysmography (MedGraphics Elite Series Plethysmograph, USA; Breeze Suite 6.4.1.14 Version 510 2008, MedGraphics Corp., USA). FEV_1 and FVC values were expressed as a percentage of the predicted values[24], and obstruction as FEV_1/TLC(%)[25].

Anthropometry

Weight and height were measured using 150 kg max scales (EB8271 NuWeigh, Newcastle Weighing Services NSW, Australia) and 2 m wall-suspended measuring tape with wall stop (Surgical and Medical Supplies Pty Ltd SA, Australia). BMI was calculated (weight (kg) / height $(m)^2$) and converted to BMI *z*-scores[26]. Total body and thoracic fat and lean mass were measured as a percentage (%) of total body weight using dual energy X-ray absorptiometry (GE Lunar Prodigy, Medtel; GE Healthcare encore 2007 software Version 11.40.004, Madison USA).

Airway biomarkers

Participants underwent exhaled Nitric Oxide (eNO) measurement (NiOX chemiluminescent Detector, Aerocrine, Australian Supplier Zynergy Medical) and combined bronchial provocation testing and sputum induction with hypertonic saline (4.5%) (ULTRA-NEB™ ultrasonic nebuliser, DeVilbiss, Model 2000)[27]. Airway hyperresponsiveness (AHR) was defined as a fall in FEV_1 $\geq$15% of their baseline FEV_1. The dose response slope (DRS) and the log-transformed provocation dose ($LogPD_{15}$) were calculated. Opaque mucocellular sputum portions were selected from saliva, processed using dithiothreitol[27, 28], and a total cell count of leukocytes and viability performed[27, 28]. Cytospins were prepared, stained (May-Grunwald Geimsa) and a differential cell count obtained from 400 non-squamous cells.

Systemic biomarkers

Fasting blood samples were centrifuged at 3000 rpm, 4^oC for 10 minutes. All samples underwent duplicate testing. Plasma IL-6 (R&D Systems, Minneapolis MN USA), and serum leptin and adiponectin (Bio-Rad, Hercules CA USA) were measured using commercial ELISAs, with respective sensitivity of 0.039pg/ml, 3.1pg/ml and 32.7pg/ml. Serum high sensitivity C-Reactive Protein (CRP), and plasma cholesterol, high-density lipoprotein cholesterol (HDL-C), triglyceride and glucose were measured using commercial assays (CRP Flex reagent cartridge, CHOL Flex reagent cartridge, HDLC Flex reagent cartridge, TRIG Flex reagent cartridge & GLU Flex reagent cartridge, Dimension Vista System, Siemans Healthcare Diagnostics Inc. 2008, Newark

USA). Plasma insulin was measured using commercial immunoassay (Access Ultrasensitive Insulin assay, Beckman Coulter Inc. 2008, CA USA). Low-density lipoprotein cholesterol (LDL-C) (total cholesterol–(HDL-C)–(0.4545*triglycerides)) and homeostasis model assessment of insulin resistance (HOMA-IR) (glucose (mmol/L)*insulin (mlU/L) / 22.5) were calculated.

Statistical analysis

Data are presented as mean (standard deviation, SD), median [interquartile range, IQR], or proportion (*n*, (%)). Outcome data are reported as change (Δ) from baseline. Continuous data were assessed using a paired mean-comparison *t*-test or Wilcoxon sign-rank test for within-group comparisons, and a two-group mean-comparison *t*-test or Wilcoxon rank-sum test for between-group comparisons. Associations between ΔBMI *z*-score and 5 key pathways were explored using spearman-rank correlation coefficients: mechanical function (ΔERV), airway inflammation (Δ%sputum inflammatory markers, ΔeNO), systemic inflammation (ΔCRP), endocrine function (Δleptin), and metabolic function (ΔHOMA). All tests were of size alpha = 0.05 and unadjusted for multiple comparisons. Statistical analysis was performed using Intercooled Stata Version 11.0 for Windows (StataCorp, College Station, Texas, USA 1984-2005).

Results

Group characteristics were similar at baseline **(Table I)**, except for a significantly higher ACQ score in the DIG compared to the WLC. Following the intervention period, a significant reduction in all adiposity indicators occurred in the DIG **(Table II)**, with a clinically important reduction in BMI *z*-score **(Fig 2a)** and a reduction in %body fat **(Fig 2b)**. However, metabolic markers remained unchanged in the DIG. No significant change in anthropometric or metabolic markers was observed for the WLC, except for a statistically significant increase in body weight and fasting glucose levels **(Table II)**. The change in BMI *z*-score and %body fat was significantly different between groups **(Fig 2a, Fig 2b)**. The change in weight was also significantly different between groups, while changes in metabolic markers were not significantly different **(Table II)**.

There was no significant change in dynamic lung function, within or between groups **(Table III)**. Static lung function, including ERV **(Fig 3a)**, was significantly different within the DIG compared to baseline. However, the difference between groups was not statistically significant. ACQ score improved significantly within the DIG, compared to the WLC **(Fig 3b)**. PAQLQ symptom (0.6[-0.1, 1.2], $p<0.05$) and emotional (0.4[0.3, 1.7], $p<0.05$) domain scores significantly improved in the DIG, but this was not different compared to the change in the WLC (0.1[-0.4, 0.6] and -0.1[-0.4, 0.8], respectively). A trend towards a clinically significant improvement in PAQLQ total (0.7(1.2), $p>0.05$) and activity domain (0.6[-0.3, 1.7], $p>0.05$) scores was also observed in the DIG, but this did not differ from the change in the WLC (0.1(0.7) and 0.1[-0.2, 0.4], respectively).

Airway & systemic inflammation

There was no change in the number or proportion of eosinophils or neutrophils, within or between groups **(Table IV)**. However, a non-significant trend towards a reduction in %neutrophils in the DIG was observed. A statistically significant difference in both absolute and %lymphocytes was observed between groups. However, the change within groups was non-significant. A significant increase in CRP levels was detected in the WLC compared to the DIG, while no change was observed in IL-6, leptin or adiponectin levels, within or between groups **(Table IV)**.

Correlations

Table V presents correlations between ΔBMI *z*-score and key outcomes. Change in BMI *z*-score **was associated with** ΔCRP and ΔeNO **(Table V)**. Change in CRP was positively associated with Δ%body fat (r=0.64, p=0.001) and negatively associated with Δ%lean mass (r=-0.61, p=0.001) and Δ%thoracic lean mass (r=-0.41, p=0.043). Change in eNO was negatively associated with Δ%thoracic lean mass (r=-0.56, p=0.011) only. In addition, ΔHOMA-IR was positively associated with Δ%body fat (r=0.57, p=0.003) and Δ%thoracic fat (r=0.47,

185 p=0.020), while negatively associated with Δ%lean mass (*r*=-0.61, p=0.001) and Δ%thoracic lean mass (*r*=-
186 0.51, p=0.012). Only ΔCRP was associated with ΔACQ (*r*=0.43, p=0.029).
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Discussion

The presented study was a pilot RCT aimed at investigating the efficacy of dietary energy restriction to induce acute weight loss in obese children with asthma, and the subsequent effect on asthma outcomes. Results demonstrate that dietary intervention can induce clinically important weight loss in asthmatic children within 10-weeks. Importantly, acute dietary-induced weight loss was associated with a significant improvement in static lung function, asthma control, and self-reported quality of life in this group of children.

Our data demonstrate that clinically significant rapid weight loss can be achieved in children with asthma, compared to controls, using simple dietary intervention**.** The presented dietary intervention achieved a median 5.7% reduction in body weight within 10-weeks, comparable with the 5.4% weight loss achieved at 6-months using interdisciplinary therapy in a previous study[17]. Notably, in the presented study, the reduction in BMI *z*-score achieved in the intervention group was of clinical importance. Few weight loss intervention studies conducted in children and adolescents have reported age- and sex-standardised BMI, which limits comparability between studies. Nonetheless, of previous weight loss interventions in non-asthmatic children which included a dietary component and reported standardised BMI, reductions of approximately 0.2-0.4BMI-SDS have been reported over time frames ranging from 4-6mths[29-32], while others have found no significant change[33, 34]. Hence, the weight loss achieved in the current study, a median BMI *z*-score reduction of 0.2SDS, suggests our intervention was very successful. This may be due to the frequency of dietetic contact and/or the delivery of the dietary intervention, or that it was motivated by evaluating the impact on asthma outcomes. Participants were required to focus on one aspect of lifestyle modification which may make adjusting to beneficial changes easier than addressing multiple aspects simultaneously. Participation in a weight loss study designed to evaluate the effect upon asthma outcomes may also have motivated adherence to the dietary prescription. Investigation of whether greater weight loss can be achieved with a longer intervention, and follow-up to evaluate weight loss maintenance, is a consideration for future studies.

In both asthmatic and non-asthmatic adults, obesity is associated with reduced static lung function, namely FRC and ERV, while ventilatory function is only moderately affected[5]. A dose-response relationship has been reported with weight loss in adults, whereby greater improvements in FEV_1, FVC and TLC are seen with greater amounts of weight loss[35]. However, the most notable increases in lung function following weight loss have been reported for ERV, with increases of approximately 20% and 60% reported in asthmatic adults following diet-induced[15] and surgery-induced[16] weight loss, respectively. Recently, Boulet et al reported significant improvements in %predicted dynamic and static lung function variables, with the greatest improvement seen in

%ERV, which more than doubled the baseline value by 12-months post-surgery in asthmatic adults[16]. Recent reports describe RV and FRC, expressed as both %predicted and relative to TLC, to be significantly lower in overweight and obese asthmatic children compared to non-obese counterparts[10, 11]. We recently reported that obesity in children with asthma is also associated with a reduced ERV (Jensen et al, unpublished). Conversely, childhood FEV_1 and FVC appear largely unaffected by the presence of obesity, with reports of increased ventilatory function in overweight and obese children compared to non-obese children with asthma[4, 10, 11]. A recent uncontrolled weight-loss study did not measure static lung function, but did demonstrate significant improvements in %predicted spirometry values at 6-months and 12-months post-intervention in asthmatic adolescents[17]. In contrast to this study, our participants had relatively normal baseline $\%FEV_1$ and %FVC, which may explain why a change in spirometry measurements was not observed following acute weight loss. However, similar to adult studies, a significant improvement in ERV for the intervention group was detected, following weight loss. In addition, there was a significant reduction in RV and %RV/TLC, suggesting a reduction in obstruction.

Importantly, the improvement in asthma control in the intervention group approached clinical significance. There was also a clinically significant improvement in self-reported quality of life for the intervention group. However, only the change in the PAQLQ symptom and emotion domains reached statistical significance. Improved quality of life and asthma control has been demonstrated in asthmatic adults, 6 and 12-months following surgically-induced weight loss[16, 18] and 10-weeks following dietary-induced weight loss[15]. Our results demonstrate that acute weight loss can achieve clinically significant improvements in static lung function, and significant improvements in asthma control and quality of life in obese children. Follow-up studies are needed to investigate the effect of long-term weight loss intervention on lung function and asthma control in this group of children.

Non-eosinophilic asthma, characterised by significant neutrophilia, has been described as a defining feature of adult obese asthma[7], and recently, Scott et al reported a significant association between %weight loss and reduced sputum %neutrophils in asthmatic females following a 10-week dietary and/or exercise intervention[15]. Although a recent cluster analysis in asthmatic children identified a distinct cluster characterised by a greater BMI, elevated peripheral neutrophils and poorer FEV_1[36], the presence of airway neutrophilia in paediatric obese asthma has not been described. The presented study is the first paediatric weight loss trial to report airway inflammation. No significant change in eNO or induced sputum inflammatory cells was detected within groups. However, there was a non-significant trend towards a reduction in %neutrophils in

the intervention group. Likewise, there was no significant difference in the change in airway inflammatory markers between groups, with the exception of absolute and %lymphocytes. Interestingly, in asthmatic adults, an increase in %lymphocytes in broncho-alveolar lavage (BAL) samples 12-months post-bariatric surgery has also been reported, while no change in neutrophils or eosinophils were observed[18].

Elevated systemic inflammation has been described in adult obese asthma[8]. Following weight loss, significant increases in adiponectin levels[18], and significant reductions in CRP[16], leptin[15] and IL-6[15] have been reported. Previous weight loss interventions in non-asthmatic children have produced variable changes in systemic inflammation[14]. Both short (3-6 weeks) and long term (3-12 months) weight-loss studies have observed significant reductions in CRP, IL-6 and leptin, and increases in adiponectin[14], while other studies have reported no difference in CRP, TNF-α or adiponectin levels over a 6-12 month period, despite weight loss of up to 0.4BMI-SDS[14, 29, 31]. These studies also failed to detect a significant change in metabolic markers[29, 31]. The recent weight loss intervention by da Silva et al reported modest but statistically significant improvements in CRP, leptin and adiponectin in obese asthmatic and non-asthmatic adolescents at 12-months, where there was no significant change at 6-months[17]. Furthermore, a 12-month longitudinal study in non-asthmatic children demonstrated that improvements in TNF-α, CRP, leptin and adiponectin levels did not occur in children with weight loss <0.5BMI-SDS compared to children with weight loss ≥0.5 BMI-SDS[37, 38]. Although, the presented study achieved a clinically significant reduction in BMI *z*-score in 10-weeks, the median reduction of 0.2BMI-SDS may not have been sufficient to achieve favourable changes in systemic biomarkers. Indeed, systemic inflammatory and metabolic biomarkers remained stable in this group of children, despite acute weight loss. However, moderate but significant increases in glucose and CRP were observed in the control group, even in this short period of time. This may suggest that although this group of children appeared metabolically healthy at baseline, changes in body weight, specifically body composition, may have adverse effects upon the metabolic profile, which increases risk for other chronic conditions, including cardiovascular disease and diabetes mellitus. This is supported by the positive correlations between ΔCRP and ΔHOMA-IR versus fat mass and negative associations versus lean mass.

In an endeavour to understand the mechanism of weight loss in asthma, we explored the associations between ΔBMI *z*-score and key outcomes. Previous reports have suggested that obesity adversely impacts asthma outcomes via mechanical restriction of the chest wall and diaphragm[39]. It is also suggested that obesity alters the airway inflammatory phenotype in asthma, altering management needs[7, 8]. Our results indicate that weight change after 10 weeks is associated with airway and systemic inflammation, as indicated by ΔCRP and ΔeNO.

This may suggest that weight loss, and more specifically, reductions in adiposity and increases in lean mass, can lead to reductions in both systemic and airway inflammation. However, increased systemic inflammation was the only outcome associated with poorer asthma control, supported by a positive association between ΔCRP and ΔACQ. This may suggest that the adverse effect of obesity may operate via alterations in systemic inflammation, and that body composition is an important consideration, as previously hypothesised[4, 14]. However, further investigation is required.

This was the first RCT, designed as a pilot study, to investigate: a) the feasibility of dietary-induced weight loss in asthmatic children; and b) whether improvements in asthma outcomes are observed with acute weight loss. The presented study is limited by the sample size which may have reduced the likelihood of detecting a change in outcome measures. Although this was a randomised trial, the intervention group tended to have a greater proportion of males and a greater proportion were reportedly using ICS. The intervention group also had a significantly poorer ACQ score at baseline, compared to the WLC, which may have confounded the results. In addition, a potential source of bias is improved patient-directed asthma awareness, such as improved medication adherence, purely due to enrolment in a study in which the primary end-points are asthma outcomes. Although there was no significant change in reported medication use during the course of the study in either group, the reliability of this information is subject to the limitations of self-report. Furthermore, the nature of the intervention precluded blinding of participants and research officers to group allocation. However, the key outcomes of the study were objective measurements, which would have minimised bias. Lastly, our study did not include a follow-up period. Recent studies including a follow-up period have indicated that weight loss is maintained in children[29, 30, 32]. Therefore, further investigation into the efficacy of diet-induced weight loss in asthmatic children is needed, which includes an adequate follow-up period.

The presented study demonstrates that rapid weight loss can be achieved safely in obese asthmatic children with dietary intervention alone, resulting in improvements in static lung function, asthma control and quality of life. On the other hand, changes in airway and systemic inflammation were not detected following acute dietary-induced weight loss in this group of children. However, exploratory analysis suggests that body composition changes, specifically greater adiposity and lesser lean mass, are associated with adverse changes in systemic and airway inflammation. In addition, systemic inflammation was associated with poorer asthma control in children, indicating that the role of inflammation should not be discounted and further investigation in larger trials is needed. Our data indicates that weight loss can produce beneficial changes in childhood asthma outcomes and

supports the need for larger RCTs, with an appropriate intervention and follow-up period, to investigate further the efficacy of weight loss intervention in asthmatic children.

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414 **Tables**

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Table I: Subject characteristics at baseline, randomised to dietary intervention or wait-list control group for ten weeks

Subject Characteristics	**Dietary Intervention**	**Wait-list Control**
number; *n*	13	15
Age (years); mean (SD)	11.5(2.1)	12.4(2.4)
Gender (% females); **%**	3(23.1)	8(53.3)
Height (cm); mean (SD)	1.6(0.1)	1.6(0.1)
Weight (kg); median [IQR]	59.9[56.1, 78.6]	71.2[66.5, 82.4]
BMI *z*-score (SDS); median [IQR]	2.1[1.9, 2.3]	2.2[1.8, 2.4]
Total body fat mass (%); mean (SD)	44.7(5.9)	44.8(7.3)
Total body lean mass (%); mean (SD)	53.3(5.6)	53.3(6.8)
Atopic (Y/N); n (%Y)	9(69.2)	10(66.7)
FEV_1 % predicted (%); mean (SD)	90.5(13.6)	96.0(7.6)
FVC %predicted (%); mean (SD)	100.8(10.2)	101.4(6.9)
FEV_1/TLC (%); mean (SD)	59.1(9.0)	64.0(9.0)
ACQ Score; median [IQR]	1.14[0.43, 1.57]*****	0.57[0.29, 0.86]
PAQLQ; median [IQR]	5.52[4.65, 6.26]	6.00[5.65, 6.52]
Airway hyperresponsiveness (%true)†; n (%)	8(72.7)	10(76.9)
$LogPD_{15}$ (ml); mean (SD)	1.4[0.4, 2.0]	1.1[-0.3, 2.4]
Dose response slope (%fall/ml)); median [IQR]	2.6[1.4, 9.2]	1.8[0.5, 14.3]
SABA (true)†; n (%)	11(84.6)	13(86.7)
ICS (true)†; n (%)	7(53.9)	3(20.0)
Beqs; median [IQR]	400[275, 400]	400[233, 400]

BMI, body mass index; SDS, standard deviation score; FEV_1, forced expiratory volume in 1 second; FVC, forced vital capacity; TLC, total lung capacity; ACQ, Asthma Control Questionnaire (Juniper); PAQLQ, Paediatric Asthma Quality of Life Questionnaire (Juniper); $LogPD_{15}$, log-transformed provocation dose; SABA, short acting β-agonist; ICS, inhaled corticosteroid; Beqs, beclomethasone equivalents; *p =0.026 vs control group; †Pearson's Chi-squared test.

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Table II: Change in anthropometric & metabolic variables in obese children with asthma, following randomisation to diet-induced weight loss intervention or no intervention for ten weeks

	Intervention group		Control group		
Anthropometric & metabolic markers	**Baseline**	**Change (Δ) vs. Baseline**	**Baseline**	**Change (Δ) vs. Baseline**	***Change (Δ) between groups: p-value***
Weight (kg); median [IQR]	59.9[56.1, 78.6]	-3.4[-4.8, -2.9]*	71.2[66.5, 82.4]	1.3[0.7, 2.2]*	**0.003**
Total body lean mass (%); mean (SD)	53.3(5.6)	2.1(3.0)*****	53.3(6.8)	0.2(2.5)	0.083
Glucose (mmol/L); median [IQR]	4.3[3.9, 4.4]	0.0[-1.0, 0.3]	4.4[4.0, 4.7]	0.4[-0.2, 0.5]*	0.174
Insulin (mlU/L); median [IQR]	7.3[5.3, 12.0]	-0.7[-6.4, 3.7]	10.1[7.1, 16.2]	0.9[-2.3, 8.1]	0.356
Total cholesterol (mmol/L); median [IQR]	3.9[3.8, 4.1]	-0.1[-0.3, 0.2]	4.1[3.8, 4.7]	0.1[-0.3, 0.3]	0.437
LDL-C (mmol/L); median [IQR]	2.2[2.0, 2.7]	-0.1[-0.3, 0.2]	2.2[1.9, 2.8]	0.1[-0.1, 0.2]	0.423
HDL-C (mmol/L); median [IQR]	1.3(0.2)	-0.0[-0.1, 0.0]	1.1(0.2)	0.1[0.0, 0.1]	0.254
Triglycerides (mmol/L); median [IQR]	1.0[0.7, 1.2]	0.0[-0.4, 0.2]	1.3[0.8, 2.0]	0.0[-0.4, 0.2]	0.923
HOMA-IR; median[IQR]	1.4[0.9, 2.0]	-0.2[-1.0, 1.2]	2.0[1.8, 3.2]	0.3[-0.4, 1.6]	0.317

LDL-C, low density lipoprotein cholesterol; HDL-C, high density lipoprotein cholesterol; HOMA-IR, homeostasis model assessment of insulin resistance. *p <0.05 versus baseline value

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Table III: Change in lung function & clinical asthma outcomes in obese children with asthma, following randomisation to diet-induced weight loss intervention or no intervention for ten weeks

	Intervention group		Control group		
Lung function variables	**Baseline**	**Change (Δ) vs. baseline**	**Baseline**	**Change (Δ) vs. baseline**	***Change (Δ) between groups: p-value***
FEV_1 (L); median [IQR]	2.4[2.0, 2.9]	0.0[-0.2, 0.1]	2.6[2.2, 2.9]	0.0[-0.2, 0.1]	0.489
FVC (L); median [IQR]	3.4[2.7, 3.5]	0.1(0.2)	3.3[2.9, 3.5]	0.0(0.2)	0.191
FEV_1/TLC (%); mean (SD)	59.1(9.0)	2.3[-1.3, 14.1]	64.7(8.9)	-0.5[-4.6, 4.8]	0.217
TLC (L); median [IQR]	4.4[3.4, 4.8]	0.0[-0.5, 0.0]	4.0[3.6, 4.7]	-0.1[-0.2, 0.1]	0.471
FRC (L); median [IQR]	1.9[1.7, 2.1]	0.2(0.5)	1.6[1.5, 2.1]	0.2(0.4)	0.905
RV (L); median [IQR]	0.9[0.8, 1.6]	-0.4(0.5)*	0.9[0.7, 1.1]	-0.1(0.4)	0.237
RV/TLC (%); mean (SD)	25.8(9.3)	-6.9(9.2)*	20.5(9.0)	-1.7(10.2)	0.188
Dose response slope (%fall/ml)); median [IQR]	2.6[1.4, 9.2]	0.4[-0.4, 1.7]	1.8[0.5, 14.3]	0.1[-4.7, 0.6]	0.271
$LogPD_{15}$ (ml); mean (SD)	1.4[0.4, 2.0]	-0.4[-0.8, 0.3]	1.1[-0.3, 2.4]	0.6[-0.2, 1.2]	0.391

FEV_1, forced expiratory volume in 1 second; FVC, forced vital capacity; TLC, total lung capacity; FRC, functional residual capacity; RV, residual volume; $LogPD_{15}$, log-transformed provocation dose. *p <0.05 versus baseline value

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Table IV: Change (Δ) in airway & systemic inflammatory markers in obese children with asthma, following randomisation to diet-induced weight loss intervention or no intervention for ten weeks

	Intervention group		Control group		
Airway & systemic inflammatory markers	**Baseline**	**Change (Δ) vs. baseline**	**Baseline**	**Change (Δ) vs. baseline**	***Change (Δ) between groups: p-value***
Exhaled nitric oxide (ppb); median [IQR]	13.1[8.4, 41.8]	-2.6[-11.3, 0.39]	27.2[10.5, 46.7]	-1.9[-4.0, 0.3]	0.673
Total cell count (x 10^6/ml); median [IQR]	3.2[1.3, 4.6]	0.5[-0.5, 2.4]	2.8[1.9, 5.5]	0.7[0.3, 2.4]	0.596
Neutrophils (%); median [IQR]	10.5[8.0, 18.8]	-4.8[-7.5, -0.6]	10.3[2.8, 27.5]	1.0[-4.5, 14]	0.355
Eosinophils (%); median [IQR]	0.8[0.5, 5.3]	-0.1[-0.5, 5.1]	0.8[0.3, 8.5]	0.0[-0.8, 2.3]	0.938
Macrophages (%); mean (SD)	78.8[71.3, 83.5]	3.4[-3.6, 8.6]	78.0[42.5, 84.8]	3.5[-14.5, 4.0]	0.537
Lymphocytes (%); median [IQR]	2.3[0.3, 7.0]	-2.4[-4.9, -1.0]	1.0[0.0, 1.3]	0.8[0.0, 2.0]	**0.025**
Neutrophils (x 10^6/ml); median [IQR]	0.53[0.15, 1.50]	-0.09[-0.88, 0.48]	0.49[0.14, 1.39]	0.16[-0.24, 0.36]	0.497
Eosinophils (x 10^6/ml); median [IQR]	0.02[0.01, 0.12]	-0.01[-0.02, 0.24]	0.03[0.01, 0.12]	0.01[-0.01, 0.43]	0.396
Macrophages (x 10^6/ml); median [IQR]	2.48[1.25, 4.03]	0.22[-1.69, 3.92]	2.05[1.71, 4.93]	0.41[-0.49, 1.22]	1.00
Lymphocytes (x 106/ml); median [IQR]	0.12[0.05, 0.40]	-0.04[-0.35, 0.00]	0.04[0.02, 0.07]	0.05[0.01, 0.07]	**0.042**
C-Reactive Protein (mg/L); median [IQR]	2.1[1.5, 3.3]	-0.4[-0.5, 0.4]	2.1[0.7, 4.0]	0.7[-0.1, 1.9]*	**0.037**
Interleukin-6 (pg/mL); median [IQR]	1.2[0.7, 2.7]	0.3[-0.3, 0.4]	1.4[0.7, 2.0]	-0.1[-0.5, 0.4]	0.907
Leptin (ng/mL); median [IQR]	6.4[0.5, 27.4]	-0.3[-1.8, 0.2]	3.1[0.7, 18.0]	-0.5[-8.1, 0.1]	0.419
Adiponectin (ug/L); median [IQR]	4.4[3.6, 6.0]	0.7[-0.5, 2.4]	4.8[4.1, 7.2]	1.1[-1.3, 2.3]	0.954

*p <0.05 versus baseline value

Table V: Spearman rank correlation coefficients between change (Δ) in BMI *z*-score, lung function, and airway and systemic biomarkers.

H_0 MOA	**Key variable of interest**	**ΔBMI *z*-score**	
		r [†]	**p-value**
Mechanical	ΔERV	0.12	0.568
Systemic inflammation	ΔCRP	**0.47**	**0.012**
Airway inflammation	Δ%eosinophils	-0.49	0.093
	Δ%neutrophils	0.05	0.873
	Δ%macrophages	0.16	0.591
	Δ%lymphocytes	0.05	0.865
	ΔeNO	**0.46**	**0.034**
Endocrine	Δleptin	0.31	0.136
Metabolic	ΔHOMA-IR	0.33	0.104

H_0 MOA, hypothesised mechanism of action; Δ, change; BMI, body mass index; ERV, expiratory reserve volume; CRP, C-Reactive Protein; eNO, exhaled nitric oxide; HOMA-IR, homeostasis model assessment of insulin resistance. [†]Spearman's rank correlation coefficient

Figure Legends

Figure 1. Participant flow through study

Figure 2. Change from baseline in a) BMI z-score, and b) Total %body fat. ‡p-value<0.05 within group; §p-value<0.05 between groups

Figure 3. Change from baseline in a) Expiratory Reserve Volume (ERV) and b) Juniper's Asthma Control Questionnaire (ACQ) score. ‡p-value<0.05 within group; §p-value<0.05 between groups