Received: 16 August 2023 | Revised: 9 March 2024 | Accepted: 18 April 2024

DOI: 10.1002/lim2.103

Lifestyle Medicine Open Access WILEY

ORIGINAL ARTICLE

Long-term preservation of lean mass and sustained loss of fat mass after completion of an intensive lifestyle intervention in older adults with obesity and type 2 diabetes

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Funding information
Danone Nutricia Research; Tromp Medical BV, the Netherlands; Topsector Agri & Food, the Netherlands, Grant/Award Number: AF12174; Nederlandse Organisatie voor Wetenschappelijk Onderzoek, Grant/Award Number: 023.009.065

Abstract

Introduction: Lifestyle interventions combining caloric restriction with resistance exercise have the potential to preserve lean mass during weight loss. Additional protein intake can further improve lean mass. However, it is unclear whether these effects are sustained after completion of the intervention. This study aimed to evaluate the long-term effect of a 3-month lifestyle intervention, with or without supplementation of a protein drink, to preserve lean mass in older adults with obesity and type 2 diabetes at 6 months post-intervention.

Methods: Adults (n = 123) aged ≥55 years with obesity and type 2 diabetes were enrolled in a 3-month intensive lifestyle intervention including a hypocaloric diet, resistance exercise and high-intensity interval training. Participants were randomized to either receive a leucine and vitamin D-enriched protein drink or an isocaloric control drink. The 3-month intervention was followed by a 6-month phase without intervention. At baseline, 3 and 9 months (follow-up) body composition, physical functioning, physical activity and quality of life were assessed. Statistical analyses were performed using linear mixed models.

Results: Body weight loss was largely sustained at follow-up (−2.1 kg compared to baseline, 95% CI [−2.8, −1.5]) and comprised a sustained loss of fat mass (−2.6 kg, 95% CI [−3.2, −2.0]) with a simultaneous gain of lean mass (+0.7 kg, 95% CI [+0.2, +1.2]). Improvements in 400 m walk speed (+0.05 m/s, 95% CI [+0.03, +0.08]) and chair stand test time (−1.5 s, 95% CI [−1.9, −1.1]) were sustained at follow-up. There were no differences in these changes between the protein supplementation group and the control group at follow-up.

Conclusions: Older adults with obesity and type 2 diabetes preserved their lean mass, their loss of fat mass and their improvements in physical functioning at 6 months post-intervention. Protein supplementation during intervention did not affect outcomes at follow-up.

KEYWORDS
combined lifestyle intervention, muscle mass, weight loss

1 | INTRODUCTION

Individuals with type 2 diabetes who are more physically active, less sedentary or consume a healthier diet, have a reduced risk of adverse health outcomes compared to individuals with a less healthy lifestyle. These outcomes include a lower incidence of cardiovascular diseases and extended survival, as shown by a recent systematic review and meta-analysis of prospective cohort studies among individuals with type 2 diabetes.[1] Combined lifestyle interventions are recommended for the treatment of obesity and the prevention and treatment of type 2 diabetes. These interventions consist of advice and guidance on healthy nutrition, eating habits and physical activity[2] and aim to sustainably improve lifestyle behaviour, especially dietary consumption and physical activity.[3] However, implemented lifestyle interventions in health care did not lead to a reduction in cardiovascular or all-cause mortality compared to no advice on lifestyle, standard advice on lifestyle or usual diabetes care, as shown in a recent systematic review of randomized clinical trials in individuals with pre-diabetes and type 2 diabetes.[4] On health markers such as body mass index (BMI) and haemoglobin A1c (HbA1c), combined lifestyle interventions showed mixed results in individuals with type 2 diabetes.[5] In the majority of studies, lifestyle interventions resulted in limited reductions in body weight (< 5%) at the end of intervention, without major effects on metabolic health markers such as HbA1c.

Diet-induced weight loss is accompanied by a decline in lean mass, which is estimated at approximately 25% of the body mass lost.[6] This loss of lean mass during dietary intervention is associated with weight regain,[7] and individuals often experience weight fluctuations or weight loss cycling, which increases the risk for muscle wasting and weakness.[8] Loss of muscle mass during weight loss can also increase mortality risk and risk of disability.[9,10] As skeletal muscle mass is the predominant component of lean mass, preservation of lean mass is important for older adults who engage in weight loss or lifestyle programmes. This is especially relevant for individuals with type 2 diabetes because type 2 diabetes accelerates the age-related loss of muscle mass[11] and skeletal muscle is the major organ of postprandial glucose uptake.[12] However, many lifestyle interventions do not focus on the preservation of lean mass during weight loss,[13] which might have contributed to the limited effects of lifestyle interventions in type 2 diabetes reported so far. Only two intervention studies in older adults with type 2 diabetes showed preservation of muscle mass during weight loss. Dunstan et al. achieved this by the addition of resistance exercise during modest weight loss (2.5 kg in 6 months).[14] However, protein intake was not reported in their study. In addition, we performed a study that combined resistance exercise with protein supplementation during moderate caloric restriction.[15] Both resistance exercise and protein intake play a crucial role in regulating muscle metabolism and muscle mass,[10,16] and this combination has demonstrated efficacy in preserving lean mass during weight loss in older adults with obesity.[17] In our 3-month intensive lifestyle intervention in older adults with type 2 diabetes, lean mass was preserved by ingestion of a leucine and vitamin D-enriched protein drink.[15] The moderate weight loss observed in this study (approximately 2.6 kg) was primarily due to loss of fat mass and was accompanied by a reduction in circulating HbA1c of 5.0 mmol/mol (95% CI [3.5, 6.6], $p < 0.001$). However, the long-term effects of interventions that succeed in preserving lean mass during weight loss are not known, while in general, people tend to relapse into former habits after intervention.[18]

In the present work, we therefore report on the follow-up results of our study. We aim to evaluate whether the preservation of lean mass and changes in fat mass, physical functioning and quality of life are sustained 6 months post-intervention and whether the use of a protein drink in the intervention period modifies these long-term outcomes. In addition, we aim to evaluate to what extent adherence to the intensive lifestyle intervention relates to the changes sustained. We hypothesized that the intensive lifestyle intervention leads to sustained changes 6 months post-intervention and that the effect of the

protein drink observed during the intervention may last 6 months post-intervention.

2 | MATERIALS AND METHODS

2.1 | Study participants and randomization

We recruited older adults (≥55 years) with obesity and type 2 (pre-)diabetes from the Amsterdam area, in the Netherlands. Participants (n = 123) were randomly allocated to a 3-month intensive lifestyle intervention with either a test drink or an isocaloric control drink, as described elsewhere.[15] The 3-month intervention phase was followed by a 6-month follow-up phase. After these 6 months, a follow-up visit was scheduled to re-assess the outcome measurements. Detailed information on the 'Protein and lifestyle intervention to preserve muscle mass in obese older type 2 diabetes patients' (PROBE) study is available in the WHO International Clinical Trials Registry Platform (https://trialsearch.who.int), where the study was prospectively registered under ID NTR4497.

2.2 | Lifestyle intervention and study treatment

The 3-month (13 weeks) intensive lifestyle intervention consisted of bi-weekly individual dietary counselling with a dietitian and alternate bi-weekly educational group sessions on healthy diet and lifestyle under the supervision of a dietitian, in combination with progressive resistance exercise and high-intensity interval training (HIIT). The dietary programme aimed for a hypocaloric diet of 600 kcal below the estimated energy needs according to the Dutch guideline for the treatment of obesity.[19] The exercise programme included 1 h group sessions three times per week, under the supervision of a personal trainer. The lifestyle intervention involved the use of motivational interviewing techniques by the dietitians to promote behaviour change. Following the behaviour change taxonomy of Michie et al.[20] several techniques were employed, including goal setting (1.3), action planning (1.4), self-monitoring of behaviour (2.2), biofeedback (2.6), graded tasks (8.7) and body changes (12.6). More details on the diet and exercise programme can be found in Memelink et al.[15]

During the 3-month intensive lifestyle intervention, participants either received a test drink or a control drink to be consumed during breakfast and after training on training days. Per serving, the test drink contained 21 g of protein, of which 1 g of free leucine, and a mixture of carbohydrates and fat providing 150 kcal per serving, 800 IU vitamin D and a mixture of fibres, minerals and vitamins. The isocaloric control drink contained no protein or micronutrients.[15]

2.3 | Follow-up phase

The 3-month intervention phase was followed by a follow-up phase of 6 months (24 weeks) without active intervention, that is, without dietary counselling, educational group sessions, exercise programme or study drink. Participants returned to their normal lives, receiving standard care (Figure 1).

2.4 | Outcome measurements

Participant characteristics such as age, sex, BMI, duration of type 2 diabetes and diabetes medication were recorded at the baseline visit. At baseline, 3 months, and at the end of the follow-up phase (9 months), participants underwent assessment of body weight, body composition, physical functioning (400 m walk speed, five times chair stand), physical activity level (PAL), quality of life (RAND-36, physical and mental component scores)[21,22] and dietary intake as described previously.[15] Briefly, lean mass, fat mass and visceral adipose tissue were measured using dual-energy x-ray absorptiometry (DXA; Hologic Discovery A, Bedford, USA). PAL was assessed using a 3-day activity diary and by accelerometry (PAM BV, Oosterbeek, the Netherlands), and dietary intake was assessed using a 3-day food record, to be completed for 2 weekdays and 1 weekend day. Total energy and macronutrient intakes were calculated using the Dutch Food Composition Database, version 2013/4.0.[23] Diabetes medication was registered at all study visits. Participants were instructed to bring to the study visits either their diabetes medication or a prescription list from the pharmacy. Two weeks after completion of the intervention, a follow-up phone call was performed to ask for any changes in diabetes medication since the study visit at 3 months.

2.5 | Adherence to the intensive lifestyle intervention

Adherence to the intensive lifestyle intervention during the 3-month intervention phase was calculated for the lifestyle intervention components separately. First, adherence to the exercise programme was expressed in three different ways: (1) exercise programme attendance, expressed as the percentage of training sessions attended; (2) relative strength training volume, expressed as cumulative leg press load adjusted for baseline 10-repetition maximum (10RM); (3) relative HIIT training load, based on maximum power assessed during a steep ramp test on a cycle ergometer. Second, adherence to the hypocaloric diet was assessed by calculating the caloric deficit at the end of the intervention, using energy intake assessed by diet diary, resting energy expenditure by indirect calorimetry and PAL by accelerometry. Third, study product compliance was calculated from a study product calendar that the participants completed.

2.6 | Statistical analyses

Participant characteristics, parameters of adherence to the intensive lifestyle intervention and change in diabetes medication will be presented using descriptive statistics (observed mean ± standard

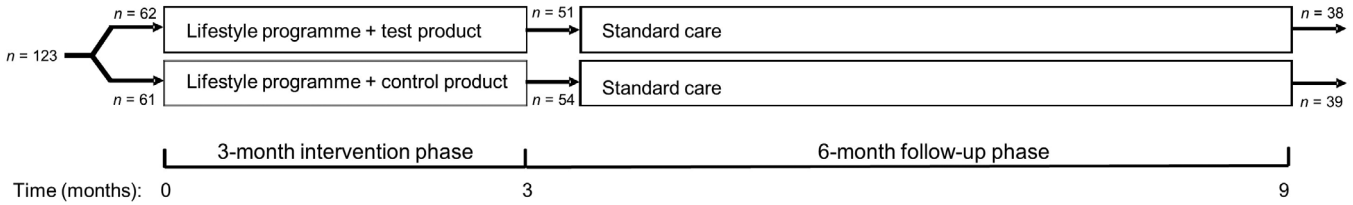

FIGURE 1 Study design of the PROBE study: 3-month intervention phase, followed by a 6-month follow-up phase without intervention. Assessments took place during participant visits to the Amsterdam Nutritional Assessment Center at 0 (baseline), 3 and 9 months.

deviation (*SD*) or percentage). Differences between participants who completed the follow-up visit (follow-up group) and participants who dropped out during the follow-up phase were evaluated using the independent samples *t*-test, Fisher's exact test or Pearson χ^2 where appropriate.

For the main analysis, we first evaluated the outcome measurements at 9 months compared to baseline (0 months) and end of the intervention (3 months), to evaluate the change at the end of the follow-up phase and during the follow-up phase, respectively. A linear mixed model was applied including the baseline value of the outcome in the outcome vector and adjusting for stratification factors (sex and sulfonylurea [SU] derivatives) and time (0, 3, 9 months) as fixed factors. A random intercept for each participant was included, and the 'unstructured' variance–covariance structure was used. Outcome measurements are presented as estimated marginal mean (EMM) ± standard error (SE).

Second, we evaluated the effect of the test drink consumed during the intensive lifestyle intervention on the change in outcome measurements at 9 months, compared to the control drink. Treatment (test, control) and time by treatment were added to the linear mixed model as fixed factors. The significance of the two-way interaction including time and treatment was checked for evaluation of the treatment effect at 9 months, compared to 0 and 3 months.

Third, associations between adherence to the lifestyle intervention components and changes in lean mass, fat mass, visceral fat, physical functioning, physical activity and quality of life at the end of follow-up were evaluated using the Pearson correlation. In the next step, visual outliers were excluded and linear regression analysis was performed for those pairs of independent and dependent variables that had a *p*-value < 0.10 for the correlation coefficient. Regression coefficients β are reported including a 95% confidence interval (CI). Potential confounders sex, age, duration of type 2 diabetes, vitamin D intake, plasma calcidiol and independent adherence parameters were step-by-step included in the regression model and were considered as a confounder when the regression coefficient β changed >10%. β values close to zero were considered irrelevant. The association between adherence to the lifestyle intervention components and changes in diabetes medication was evaluated using the analysis of variance.

Last, sensitivity analyses were performed to evaluate the change in total lean mass using two different assumptions during the follow-up phase for participants that had dropped out during this phase: (1) no change in lean mass (last observation carried forward) and (2) loss of 0.5 kg of lean mass (using a yearly loss of lean mass in older adults of ca. 1%[24] and rounding it to 0.5 kg for a worst-case scenario).

Statistical analyses were performed in IBM SPSS Statistics for Windows v28.0.1.0 (IBM Corp, Armonk, NY, USA). Statistical tests were conducted two-sided with a significance level of 5%. All CIs will be presented with a confidence level of 95%.

3 | RESULTS

3.1 | Study participants

In total, 77 out of 105 participants who had completed the intervention phase completed the 6-month follow-up phase (Figure 2). Two participants withdrew informed consent during the follow-up phase, without specifying a reason. Characteristics of the $n = 77$ participants who completed the follow-up phase (follow-up group) and of the $n = 28$ who dropped out during the follow-up phase are shown in Table 1. Both groups were comparable at baseline. However, the follow-up group had higher adherence to the exercise programme during the 3-month intervention phase, as shown by higher relative training loads for both HIIT and resistance exercise (Table 1). Change in RAND-36 physical component score at 3 months was higher in the follow-up group compared to the drop-outs at follow-up (3.9, 95% CI [1.1, 6.6], $p = 0.007$). None of the other outcome measurements at the end of the 3-month intervention phase differed significantly between the follow-up group and the dropouts at follow-up (Supplementary Table S1). Furthermore, there were no clinically relevant differences in baseline characteristics of the follow-up group between the test and control group (Supplementary Table S2).

3.2 | Outcome measurements

Body weight loss was sustained at 9 months (−2.1 kg, 95% CI [−2.8, −1.5]). The body weight loss was composed of a sustained loss of fat mass (−2.6 kg, 95% CI [−3.2, −2.0]) compared to baseline, with a simultaneous gain in lean mass (+0.7 kg, 95% CI [+0.2, +1.2]) that predominantly occurred during the follow-up phase (Figure 3, Table 2). Loss of visceral adipose tissue was sustained at 9 months (−15.3 cm^2, 95% CI [−22.1, −8.4]) and appendicular lean mass increased (+0.4 kg, 95% CI [+0.2, +0.6]) (Figure 4, Table 2).

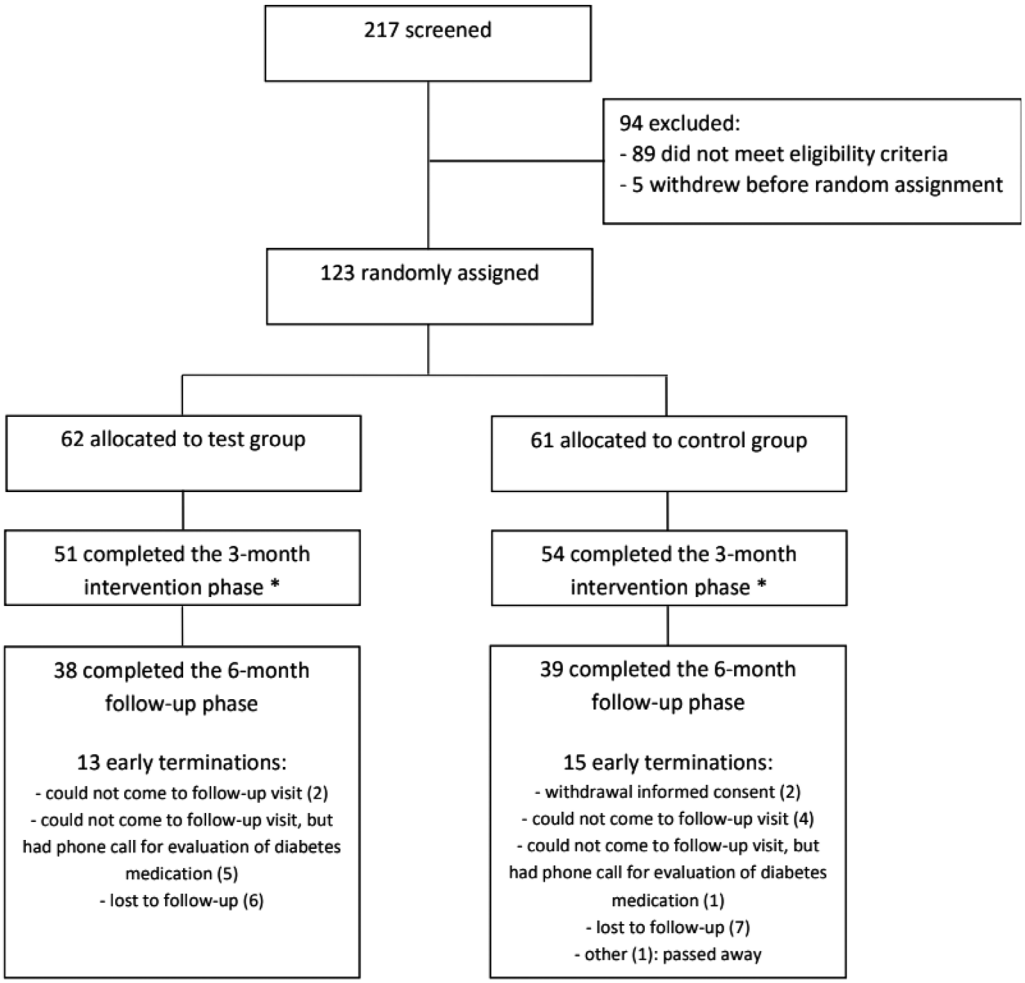

FIGURE 2 Flow chart of the PROBE study participants for the 3-month intervention phase and 6-month follow-up phase. * Reasons for early withdrawal during the 3-month intervention phase can be found in Memelink et al.[15]

Walking speed increased (+0.05 m/s, 95% CI [+0.03, +0.08]) and chair stand test time decreased (−1.5 s, 95% CI [−1.9, −1.1]) at 9 months compared to baseline, and both did not change during the follow-up phase (Figure 4, Table 2). Physical activity level and quality of life at 9 months were similar to baseline level. Energy intake calculated from the diet diary significantly decreased at 9 months compared to baseline and 3 months. Protein intake in g/kg fat-free mass (FFM)/day at 9 months significantly decreased compared to baseline, while relative protein intake (expressed as energy percentage) at 9 months showed no change from baseline (Table 2).

In the follow-up group, 71 out of 77 participants used diabetes medication at the start of the intervention. At 9 months, 28 participants (39.4%) had a reduction in diabetes medication and 4 (5.6%) had an increase.

Evaluation of the effect of the test drink versus the control drink consumed during the 3-month intervention phase showed no statistically significant differences in change from baseline to 9 months in any of the outcome measurements (Supplementary Table S3). During the follow-up phase (from 3 to 9 months), walking speed decreased in the test group compared to the control group and, as was expected per protocol, protein intake decreased in the test group (Supplemental Table S3).

3.3 | Influence of adherence to the intensive lifestyle intervention

A 20% higher exercise programme attendance during the intervention phase was associated with 1.2 s greater improvement in chair stand test time at 9 months ($\beta = -0.06$ s/%, 95% CI [−0.11, −0.01]) and with 0.1 points higher improvement in PAL score evaluated by questionnaire ($\beta = 0.005$ /%, 95% CI [0.000, 0.009]). A 20% higher relative HIIT training load was associated with 0.24 kg lower increase in lean mass (β for appendicular lean mass = −0.012 kg/%, 95% CI [−0.015, −0.003]) (Supplementary Table S4). Change in diabetes medication at 9 months was associated with caloric deficit. More specifically, participants with reduced diabetes medication had a moderate caloric deficit, while participants without a change in diabetes medication seemed to have a smaller caloric deficit, and participants with an increase in diabetes medication were in a caloric surplus (Supplementary Table S5).

TABLE 1 Baseline characteristics and adherence to the lifestyle intervention components for the follow-up group and dropouts at follow-up.

	Follow-up group		Dropouts at follow-up		
	n	Mean ± *SD*	*n*	Mean ± *SD*	*p*-value
Sex (% male)	77	65	28	64	1.00
Age (years)	77	66.3 ± 6.2	28	67.3 ± 5.9	0.454
Body weight (kg)	77	98.9 ± 14.8	28	100.4 ± 16.8	0.665
BMI (kg/m^2)	77	33.2 ± 4.3	28	33.5 ± 5.1	0.744
Waist circumference (cm)	76	115.2 ± 10.1	27	115.4 ± 11.1	0.956
Duration of diabetes (months)	71	85 ± 79	26	95 ± 54	0.531
Use of diabetes medication (%)	83[a]	86	22	91	0.729
- Use of metformin (%)		82		82	1.000
- Use of SU derivatives (%)		29		50	0.077
Fasting glucose (mmol/L)	72	8.08 ± 1.85	27	8.36 ± 1.65	0.490
HbA1c (mmol/mol)	75	50.7 ± 9.0	26	53.5 ± 11.7	0.203
Serum calcidiol (nmol/L)	72	57.3 ± 22.8	27	66.5 ± 23.0	0.078
Handgrip strength (kg)	75	36.6 ± 10.4	27	36.0 ± 10.1	0.791
Usual gait speed (m/s)	77	1.16 ± 0.23	27	1.14 ± 0.19	0.596
Current smoker (%)	77	8	28	11	0.698
Alcohol user (%)	77	68	28	68	1.00
Study product compliance (%)	76	95.0 ± 4.7	27	92.3 ± 10.1	0.069
Caloric deficit (kcal/day)	63	−396 ± 518	24	−409 ± 644	0.925
Exercise programme attendance (%)	76	81.9 ± 10.0	28	76.9 ± 14.6	0.050
Cumulative leg press training load adjusted for baseline 10RM	75	716 ± 341	24	462 ± 194	<0.001
Relative HIIT training load (%)	77	103 ± 30	28	87 ± 34	0.020

Note: Data are presented as mean ± *SD* or as the percentage. *p*-value represents the outcome of Fisher's exact test or Independent samples *t*-test. Abbreviations: BMI, body mass index; HbA1c, haemoglobin A1c; PAL, physical activity level; SU, sulfonylurea.

[a]Including six participants who had a phone call for an evaluation of diabetes medication use instead of a full follow-up visit in the lab.

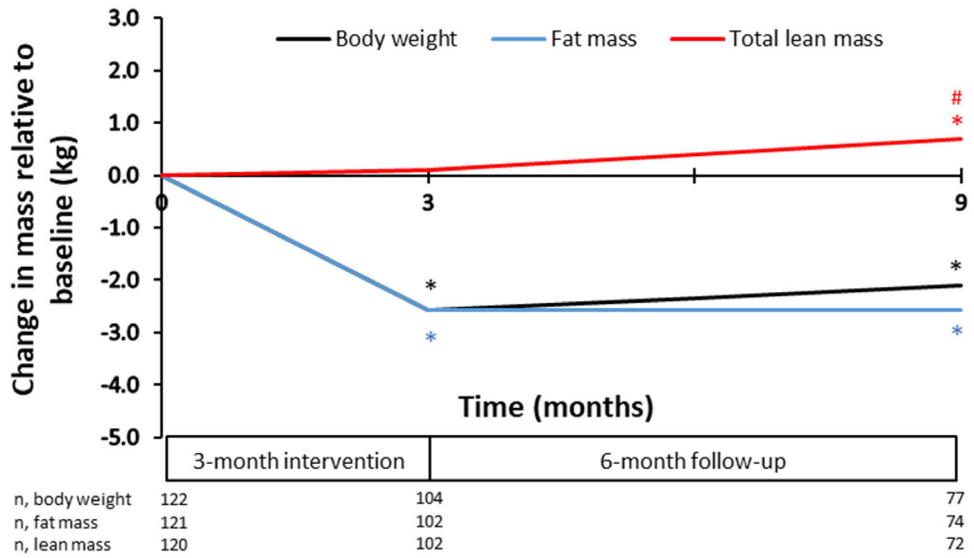

FIGURE 3 Sustained loss of body weight and fat mass and increased lean mass at 9 months (end of follow-up phase) for the whole study population, that is, test and control groups together. Change in mass is presented relative to baseline (0 months). * Significantly different from baseline. # Significantly different from the end of intervention (3 months).

TABLE 2 Outcome measurements at 0 months (baseline), 3 months (end of intervention phase) and 9 months (end of follow-up phase) with estimation of change during the follow-up phase (9 months vs. 3 months) and estimation of change from baseline at end of the follow-up phase (9 months vs. 0 months).

Outcome measurement	Time	EMM ± SE[a]	*n*	Effect size (95% CI)[b] for change during follow-up [9 vs. 3 months]	*p*-value	Effect size (95% CI)[b] for change from baseline [9 vs. 0 months]	*p*-value
Body weight (kg)	0 months	97.6 ± 1.4	122	0.5 [−0.2, 1.1]	0.156	−2.1 [−2.8, −1.5]	<0.001
	3 months	95.0 ± 1.4	104				
	9 months	95.5 ± 1.5	77				
BMI (kg/m^2)	0 months	33.3 ± 0.4	122	0.2 [−0.1, 0.4]	0.138	−0.7 [−0.9, −0.5]	<0.001
	3 months	32.5 ± 0.4	104				
	9 months	32.6 ± 0.4	77				
Leg lean mass (kg)	0 months	19.0 ± 0.3	121	0.2 [0.0, 0.3]	0.042	0.3 [0.1, 0.4]	0.002
	3 months	19.1 ± 0.3	104				
	9 months	19.2 ± 0.3	76				
Appendicular lean mass (kg)	0 months	25.6 ± 0.4	119	0.2 [−0.1, 0.4]	0.133	0.4 [0.2, 0.6]	<0.001
	3 months	25.8 ± 0.4	101				
	9 months	26.0 ± 0.4	73				
Total lean mass (kg)	0 months	61.3 ± 0.8	120	0.6 [0.1, 1.0]	0.012	0.7 [0.2, 1.2]	0.003
	3 months	61.4 ± 0.8	102				
	9 months	62.0 ± 0.8	74				
Skeletal muscle mass index (kg/m^2)	0 months	8.66 ± 0.09	119	0.05 [−0.18, 0.12]	0.142	0.13 [0.06, 0.19]	<0.001
	3 months	8.74 ± 0.09	101				
	9 months	8.79 ± 0.10	73				
Fat mass (kg)	0 months	34.8 ± 0.9	121	0.0 [−0.6, 0.6]	0.976	−2.6 [−3.2, −2.0]	<0.001
	3 months	32.2 ± 0.9	102				
	9 months	32.2 ± 0.9	72				
Fat percentage (%)	0 months	35.5 ± 0.4	121	−0.2 [−0.7, 0.2]	0.355	−1.9 [−2.4, −1.5]	<0.001
	3 months	33.8 ± 0.4	102				
	9 months	33.6 ± 0.5	72				
VAT (cm^2)	0 months	179.2 ± 5.0	122	2.7 [−4.3, 9.6]	0.450	−15.3 [−22.1, −8.4]	<0.001
	3 months	161.3 ± 5.1	104				
	9 months	164.0 ± 5.4	76				
400 m walk speed (m/s)	0 months	1.41 ± 0.03	119	0.00 [−0.03, 0.03]	0.962	0.05 [0.03, 0.08]	<0.001
	3 months	1.47 ± 0.03	99				
	9 months	1.47 ± 0.03	73				
Five times chair stand (s)	0 months	12.0 ± 0.2	120	−0.2 [−0.6, 0.2]	0.314	−1.5 [−1.9, −1.1]	<0.001
	3 months	10.6 ± 0.3	102				
	9 months	10.4 ± 0.3	76				
PAL, by questionnaire (-)	0 months	1.37 ± 0.01	121	−0.02 [−0.06, 0.02]	0.248	0.03 [−0.01, 0.06]	0.186
	3 months	1.41 ± 0.02	98				
	9 months	1.39 ± 0.02	70				
PAL, by accelerometry (-)	0 months	1.19 ± 0.01	109	−0.02 [−0.04, 0.00]	0.074	−0.01 [−0.03, 0.01]	0.260
	3 months	1.19 ± 0.01	88				
	9 months	1.18 ± 0.01	64				
RAND-36 physical component score	0 months	46.6 ± 0.9	123	−0.6 [−2.1, 1.0]	0.458	1.2 [−0.3, 2.7]	0.125
	3 months	48.4 ± 0.9	102				
	9 months	47.8 ± 1.0	76				

(Continues)

TABLE 2 (Continued)

Outcome measurement	Time	EMM ± SE[a]	*n*	Effect size (95% CI)[b] for change during follow-up [9 vs. 3 months]	*p*-value	Effect size (95% CI)[b] for change from baseline [9 vs. 0 months]	*p*-value
RAND-36 mental component score	0 months	50.9 ± 0.9	123	−0.3 [−1.9, 1.3]	0.748	−0.2 [−1.8, 1.4]	0.822
	3 months	51.0 ± 0.9	102				
	9 months	50.7 ± 1.0	76				
Energy intake (kcal/day)	0 months	1807 ± 43	121	−199 [−324, −74]	0.002	−253 [−375, −132]	<0.001
	3 months	1752 ± 47	98				
	9 months	1553 ± 57	61				
Protein intake (g/kg BW/day)	0 months	0.87 ± 0.03	120	N/A		−0.08 [−0.15, 0.00]	0.057
	3 months[c]	0.99 ± 0.03	97				
	9 months	0.79 ± 0.04	61				
Protein intake (g/kg FFM/day)	0 months	1.35 ± 0.04	119	N/A		−0.15 [−0.27, −0.03]	0.017
	3 months[c]	1.51 ± 0.05	95				
	9 months	1.20 ± 0.06	57				
Protein intake (energy %)	0 months	18.6 ± 0.4	121	N/A		1.0 [−0.2, 2.3]	0.109
	3 months[c]	21.2 ± 0.5	98				
	9 months	19.6 ± 0.6	61				

Abbreviations; BMI, body mass index; BW, body weight; CI, confidence interval; EMM, estimated marginal mean; FFM, fat-free mass; PAL, physical activity level; RAND-36, the RAND-36 item health survey; VAT, visceral adipose tissue;.

[a]Data are presented as estimated marginal mean (EMM) with SE.

[b]Estimate of change (effect size) using a linear mixed model including the baseline value in the outcome vector and adjusting for stratification factors (sex and SU-derivate use).

[c]By protocol, protein intake at 3 months differed between treatment groups test and control. Therefore, no effect size for change during the follow-up phase (9 vs. 3 months) is reported. The effect size is specified for the test and control groups separately in Supplementary Table S3.

3.4 | Sensitivity analysis

Sensitivity analysis assuming no change in total lean mass during the follow-up phase for the dropouts did not significantly alter the results for change in total lean mass at 9 months, neither compared to baseline nor compared to 3 months (+0.7 kg, 95% CI [+0.3, +1.1] and +0.6 kg, 95% CI [+0.2, +1.0], respectively). Similarly, sensitivity analysis assuming 0.5 kg loss of total lean mass during the follow-up phase for the dropouts did not significantly alter the results for change in total lean mass at 9 months, neither compared to baseline nor compared to 3 months (+0.7 kg, 95% CI [+0.2, +1.1] and +0.6 kg, 95% CI [+0.1, +1.0]).

4 | DISCUSSION

Our study demonstrated a preservation of fat mass loss, together with increased lean mass in older adults with obesity and type 2 diabetes, 6 months after an intensive exercise and caloric restriction intervention. This was accompanied by sustained improvements in walking speed and chair stand performance. Higher exercise programme attendance during the intervention period was associated with improved chair stand performance and PAL 6 months after completion of the intervention, while higher HIIT training load was associated with a lower increase in lean mass. The addition of a protein drink during the intensive lifestyle intervention did not influence outcomes 6 months after completion of the intervention.

On average, our study participants lost a moderate amount of body weight upon intensive lifestyle intervention, comparable to other interventions involving diet-based approaches using moderate energy restriction in individuals with type 2 diabetes.[5,15] We observed that most of this weight loss was sustained up to 6 months after completion of the intervention, whereas, in general, weight regain is common after lifestyle intervention or weight loss programmes for adults with obesity and type 2 diabetes.[25,26] More specifically, we observed a sustained loss of fat mass, which has, for example, been observed in the DiOGenes trial in adults with overweight or obesity 6 months after an 8-week low-calorie diet. However, the loss of fat-free mass in that study was about 20% of the body weight lost.[27] To our knowledge, sustained fat mass loss with maintained or improved lean mass has not been reported before in older adults with obesity and type 2 diabetes. Some studies reported on successfully sustained weight loss in adults with type 2 diabetes or at high risk of diabetes upon combined lifestyle or weight-management interventions with a duration of up to 2 years in primary care settings, but these studies lack information on changes in body composition.[28–30] In contrast, our intervention was specifically targeted at preserving lean mass during weight loss, using an

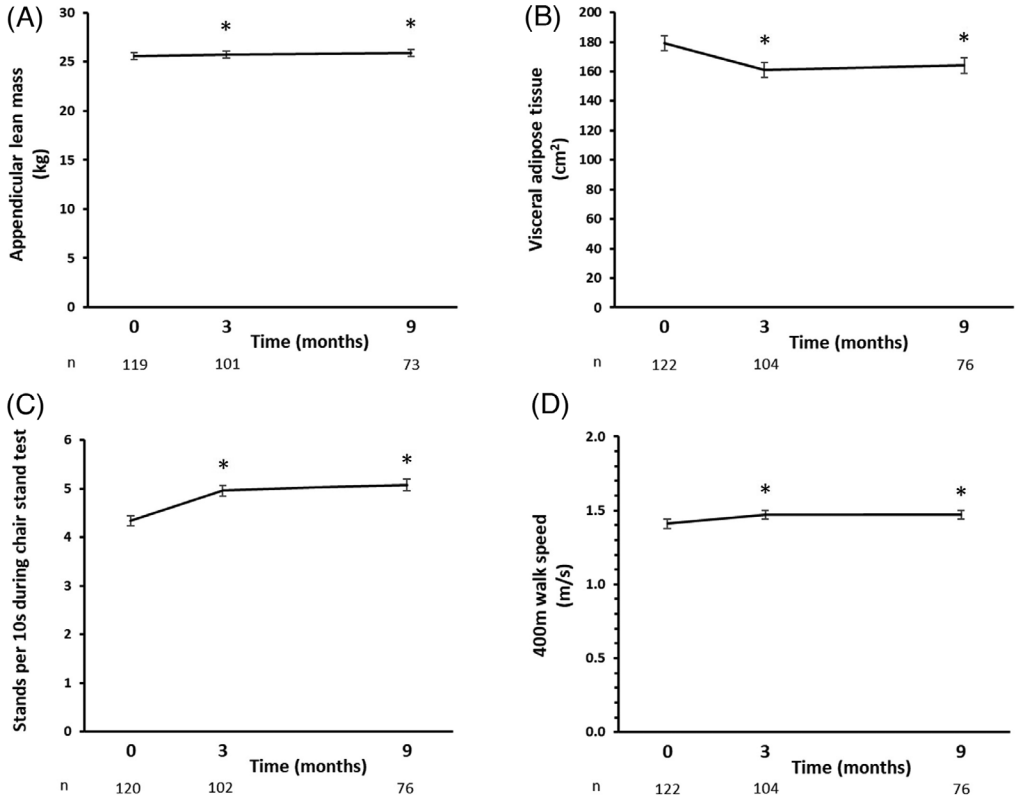

FIGURE 4 Appendicular lean mass (A), visceral adipose tissue (B), chair stand test performance expressed as the number of stands per 10 s (C) and 400 m walk speed (D) at 0 (baseline), 3 (end of intervention phase) and 9 months (end of follow-up phase). Lines represent the whole study population, that is, test and control groups together. * Significantly different from baseline (0 months).

intensive exercise programme.[15] The increase in lean mass that we observed together with a fully sustained fat mass loss 6 months after completion of the intensive lifestyle intervention can be clinically important because loss of lean mass during dietary intervention is associated with weight regain [7] which can be explained by lower basal energy expenditure.[31]

A possible explanation for the observed increase in lean mass after weight loss in our study population may be a reduced anabolic resistance obtained through improved insulin sensitivity,[32] probably due to increased exercise volume, caloric restriction or both. However, a causal relation between improved insulin sensitivity and reduced anabolic resistance is not clear from the literature.[33] Muscle insulin sensitivity was indeed improved during the intensive lifestyle intervention, in participants that could be subtyped as muscle insulin resistant.[34] Individual data from our study revealed that those participants who had an increase in insulin sensitivity or a decrease in HbA1c had similar gains in lean mass during the follow-up period as the entire follow-up population (that is, about 0.6 kg on average). This suggests that not the improved insulin sensitivity but a continuation of exercise in a considerable proportion of the participants is the most plausible explanation for the observed increase in lean mass during follow-up. Exercise, and predominantly resistance exercise, is key to increasing lean mass, muscle strength and physical function in older adults.[35] We did not collect data on participation in programmed exercise activities throughout the follow-up phase, but the follow-up group at least seemed to be more motivated to exercise than the participants who dropped out during the follow-up phase. Participants in the follow-up group had trained harder, and their physical component of the quality of life score had increased compared to the dropouts which may be linked to the higher training intensity. The observed difference in the quality of life is clinically meaningful[36] and may have increased their motivation to sustain healthy lifestyle behaviour and continue training after the intervention.

In general, lifestyle interventions in research settings do not necessarily promote sustainable behaviour change.[37] However, the intensive lifestyle intervention in the present study may have resulted in meaningful lifestyle changes, caused by the following effective components reported for lifestyle interventions: guidance towards a healthy diet, application of behaviour change techniques,[38,39] intense regular exercise, individual and/or group counselling, face-to-face counselling,[3] internal motivation to lose weight, social support and self-efficacy.[40] Sustained dietary behaviour in our study was reflected in a continued self-reported low-energy intake during follow-up, which is important for weight loss maintenance.[41] The observed energy intake suggested a continued caloric restriction; however, body weight did not decrease any further during the follow-up phase. Self-reported energy intake in our study population was most likely underestimated, which is typical for overweight or obesity.[41] Physical activity as assessed by the physical activity diary was not significantly increased compared to baseline. However, walking speed and chair stand test time were both

significantly improved compared to baseline, and the improvement was sustained during follow-up. Such improvements were also observed in the Look AHEAD trial, a long-term lifestyle intervention among middle-aged and older adults with type 2 diabetes.[42] The 1.5-s improvement in chair stand test performance in our study is within the clinically meaningful change range of 0.5–4.7 s for older adults reported by Gonzalez-Bautista et al.[43] The 0.05-m/s improvement in 400 m walk speed is not clinically meaningful according to findings from the LIFE-P study.[44] However, it is when interpreted from the small meaningful change of 20 m reported for the 6-min walk distance test.[45]

Regarding participants' adherence to the intensive lifestyle intervention itself, a higher exercise programme attendance during the intervention was associated with greater improvements in chair stand performance and daily physical activity 6 months after the intervention. This suggests that training frequency or volume matters for physical functioning. Remarkably, a higher relative HIIT training load was associated with a lower increase in lean mass, while HIIT training load was not associated with lean mass at baseline. However, it should be noted that adherence to HIIT training intensity was relatively high. It could be that, in highly compliant participants, the intensive progressive exercise programme may have been sub-optimal for exercise-induced modulation of skeletal tissue in a situation of caloric restriction. Reduction in diabetes medication seemed to be related to an adequate caloric deficit during the intensive lifestyle intervention, which is also seen in the long-term Look AHEAD lifestyle intervention using moderate caloric restriction.[46]

In the 3-month intervention phase, we previously showed that adding a leucine and vitamin D-enriched protein drink resulted in a daily protein intake of about 1.2 g/kg BW that supported the preservation of lean mass during weight loss.[15] Average protein intake at the end of the follow-up phase was 0.79 g/kg BW/day, suggesting that higher protein intakes of about 1.2 g/kg BW/day may not be required for preservation, or even increase, of lean mass during weight maintenance. Similar findings have been shown in the DIOGENES trial where adults with overweight or obesity maintained their weight on a high (23 En%) or low (16 En%) protein diet, without significant difference in the observed increases in fat-free mass.[47] Surprisingly, the reported difference in increase in lean mass between the test and control group at 3 months[15] had disappeared at 9 months, indicating that the persons receiving the control drink during the lifestyle intervention gained on average more lean mass (+0.83 kg) during the follow-up phase compared to persons that received the protein drink (+0.34 kg). Daily protein intake in the test group decreased below 0.8 g/kg BW at follow-up, while intake remained above 0.8 g/kg BW during the whole 9 months in the control group. It seems that participants in the test group had not replaced the protein-containing study drink with other protein-containing foods during the follow-up phase. This could explain why lean mass differences between the test and control group had disappeared at follow-up. Whether continuation of the protein drink during follow-up, with or without voluntary exercise training, would have shown maintenance or further improvement in lean mass gain remains a subject for future research.

4.1 | Clinical implications and recommendations for future research

In the present study, we showed preservation of lean mass in older adults with obesity and type 2 diabetes 6 months after combined lifestyle intervention. Interestingly, this sustained change was obtained after a relatively short intervention duration of 3 months, which is much shorter than the generally advised duration of 1 year.[3] To achieve long-term preservation of lean mass, combined lifestyle interventions likely need to include an intensive exercise programme, on top of nutritional and behavioural support. The cost-effectiveness of such intensive combined lifestyle interventions remains to be evaluated in future research involving measurements of glucose control and cardiovascular disease incidence as well.

4.2 | Strengths and limitations

The main strength of our study is the thorough evaluation of changes in body composition after combined lifestyle intervention in older adults with obesity and type 2 diabetes. Although designed as a research-based lifestyle intervention focusing on the evaluation of lean mass and fat mass after 3 months, lean mass was preserved and the change in fat mass was sustained 6 months post-intervention. This is most likely due to the incorporation of various behaviour change techniques in our lifestyle intervention. Based on qualitative assessments, we did find several indications that behaviour had indeed changed after the 3-month lifestyle intervention. However, we did not measure behaviour objectively and used self-reported measures. Self-reported energy intake in individuals with obesity is most likely underestimated by the diet diary.[41] The 3-day physical activity diary using 30-min intervals possibly limited the sensitivity to detect changes.[48] The combined lifestyle intervention programme in our study seems fairly generalizable to practical settings, which is another strength of the study. The absence of measurements of glucose control at the end of the follow-up phase can be seen as a limitation of our study. Availability of these measurements would have enabled the evaluation of the sustainability of HbA1c reduction 6 months post-intervention. Finally, the number of dropouts at follow-up (28 out of 105) is considerable, which makes our results prone to selection bias. However, baseline characteristics did not differ between the follow-up population and the dropouts at follow-up and sensitivity analysis did not alter the statistical significance of the results on lean mass.

5 | CONCLUSION

Our study showed that older adults with obesity and type 2 diabetes preserved their lean mass, their loss of fat mass and their improvements in physical functioning, 6 months after completing a 3-month intensive lifestyle intervention. The addition of the protein drink during the intervention did not affect the outcomes at follow-up.

ACKNOWLEDGEMENTS

We gratefully acknowledge Mariëtte van Rijmenam, Twan ten Haaf, Minse de Bos Kuil, Anita Tump, Renske Benedictus and Aldo Scafoglieri for their expert assistance during the study; all our study participants for their willingness to participate; our graduate students involved in the study for their dedication; Topsector Agri & Food, the Netherlands (AF12174), for providing financial support for the study; Danone Nutricia Research for providing study products and financial support for conducting the study; Tromp Medical BV, the Netherlands, for providing material support for the DXA scans; and the Dutch Research Council (NWO) for providing a Ph.D. grant for RGM.

CONFLICT OF INTEREST STATEMENT

JdV-vdB is an employee of Danone Nutricia Research. WP and SW are employees of the Netherlands Organisation for Applied Scientific Research (TNO), which is a not-for-profit reserach organization collaborating in several public-private partnerships or business-to-business research projects that receive funding from companies. All other authors declare no conflicts of interest.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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SUPPORTING INFORMATION

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How to cite this article: Memelink RG, Hijlkema A, Valentin B, et al. Long-term preservation of lean mass and sustained loss of fat mass after completion of an intensive lifestyle intervention in older adults with obesity and type 2 diabetes. *Lifestyle Medicine*. 2024;5:e103. https://doi.org/10.1002/lim2.103

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