Full title

Bone turnover and metabolite responses to exercise in people with and without long-duration type 1 diabetes: a case-control study

Short running title

Exercise & bone turnover in type 1 diabetes

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Abstract

Introduction: Exercise acutely alters markers of bone resorption and formation. As fracture risk is increased in patients with type 1 diabetes, understanding if exercise-induced bone turnover is affected within this population is prudent. We assessed bone turnover responses to acute exercise in individuals with long-duration type 1 diabetes and matched controls.

Research Design and Methods: Type 1 diabetes participants (n=15; age: 38.7±13.3, HbA1c: 60.5±6.7mmol/mol; diabetes duration: 19.3±11.4years) and age-, fitness-, BMI-matched controls (n=15) completed 45 minutes of incline walking (60%VO_{2peak}). Blood samples were collected at baseline and immediately, 30, and 60 minutes post-exercise. Markers of bone resorption (type-1 cross-linked β-C-telopeptide [β-CTx]) and formation (procollagen type-1 N-terminal propeptide [P1NP]), parathyroid hormone (PTH), phosphate, and calcium (albumin-adjusted and ionised) were measured. Data (mean±SD) were analysed by a mixed-model ANOVA.

Results: Baseline concentrations of P1NP and β-CTx were comparable between type 1 diabetes participants and controls. P1NP did not change with exercise ($p=0.20$) but β-CTx decreased ($p<0.001$) in both groups, but less so in type 1 diabetes participants compared to controls (-9.2±3.7%; $p=0.02$). PTH and phosphate increased immediately post-exercise in both groups; PTH only, was raised at 30 minutes post-exercise, ($p<0.001$) with no between-group differences ($p>0.39$). Type 1 diabetes participants had reduced albumin and ionised calcium at all sample points ($p<0.01$).

Conclusions: Following exercise, type 1 diabetes participants displayed similar time course changes in markers of bone formation, associated metabolites, but an attenuated suppression in bone resorption. The reduced albumin and ionised calcium may have implications for future

bone health. Further investigation of the interactions between type 1 diabetes, differing modalities and intensities of exercise, and bone health is warranted.

What is already known about this subject?

- Individuals with type 1 diabetes are at increased risk of skeletal fracture and have reduced bone density and turnover, while physical activity has been demonstrated to improve bone health in healthy populations.

What are the new findings?

- This is the first study to investigate the acute effects of exercise upon biochemical markers of bone resorption and formation in individuals with type 1 diabetes.
- Our study demonstrates that exercise has a similar acute time-course effects on bone turnover in individuals with type 1 diabetes compared to age-, sex- and fitness matched non-diabetes controls.
- Individuals with type 1 diabetes may have an attenuated reduction in bone resorption after exercise compared to the controls.

How might these results change the focus of research or clinical practice?

- As the acute bone turnover response exercise is largely normal, exercise may be a viable strategy to reducing the increased incidence of future fractures and osteoporosis in a growing elderly population of patients with type 1 diabetes.
- Research is needed to explore the optimal type, duration, and intensity of exercise to maximise bone turnover in adults with long-term type 1 diabetes, and whether this can translate into reduced incidence of fractures and osteoporosis.

Introduction

Osteoporosis and fractures are common complications of type 1 diabetes with a one- to two-fold increased fracture risk at any skeletal site.(1, 2) Long-term type 1 diabetes is associated with deficits in bone density, structure, microarchitecture, and turnover.(3, 4) Exposure to hyperglycemia and oxidative stress,(5, 6) elevated sclerostin,(4) insulinopenia and decreased gastrointestinal hormones, and chronic inflammation are all potential drivers of this.(7) At the time of clinical manifestation of type 1 diabetes around 80% of β-cell mass is already lost.(8, 9) In parallel, lower levels of bone formation and resorption markers including osteocalcin, procollagen type 1 propeptides amino-terminal (PINP), and crosslinking telopeptides of type 1 collagen β-C-terminal (β-CTx) are observed.(10) In addition to bone remodelling being impaired in recently diagnosed individuals, a diagnosis before or during puberty and poor glycemic control are further associated with reduced bone turnover and bone mineral density.(3, 10, 11)

Human bone is continuously undergoing resorption, its breakdown by osteoclasts releasing calcium and phosphate into the circulation, and formation, a process by which osteoblasts lay down new bone material.(12) Osteocytes, the most abundant bone cell phenotype that regulates bone formation and initiate bone resorption, controls bone remodelling by responding to mechanical strain.(13) After a certain age, varying between individuals, the rate of bone resorption starts to exceed the rate of bone formation, resulting in net bone loss.(12) As the type 1 diabetes population ages, so does the rate of diabetes-induced osteoporotic fractures.(1) In combination with the increased life expectancy seen during the past century due to improvements in management and survival in type 1 diabetes,(14, 15) interventions are needed to reduce the occurrence of osteoporotic fractures in this growing population of older type 1 diabetes individuals.

Regular exercise should be an integral part of modern diabetes management.(16) Structured training and physical activity have the potential to improve glycemic control, reduce inflammation, lower the demand for exogenous insulin, and improve quality of life.(16-18) Exercise training has been shown to increase bone formation and decrease bone resorption in healthy and disease states,(19) thus exercise interventions may be able to improve bone health in those with type 1 diabetes. Whilst endurance exercise programs have not appeared to benefit bone health in diabetic rats,(20, 21) there is limited evidence of the effects of exercise on bone turnover in humans with type 1 diabetes. Previous studies have demonstrated that a 9 month weight bearing and 3 month aerobic exercise interventions were successful at increasing bone mineral density and altering circulating levels of biochemical bone turnover markers in children and adolescents with type 1 diabetes, respectively.(22, 23) However, little is known on the acute effects of physical activity on the markers of bone turnover in individuals with type 1 diabetes.

As over 90% of the organic matrix of bone is type 1 collagen, many of the commonly used markers of bone turnover relate to its synthesis or degradation.(24) These include P1NP, cleaved during the synthesis of type 1 collagen and thus a marker of bone formation, and β-CTx, a product of the degradation of type 1 collagen and thus a marker of bone resorption.(25) As bone resorption and formation are tightly coupled, high levels of either bone resorption markers or bone formation markers signify a high bone turnover rate.(25) Single exercise bouts have been shown to alter circulating concentrations of P1NP and β-CTx, parathyroid hormone (PTH), a regulator of bone remodelling, and related metabolites (calcium and phosphate) in young and old people free-from disease.(26-28) Given the deficits observed in bone turnover within type 1 diabetes patients, it is important to understand if the bone turnover response to acute exercise is comparable to those without diabetes, and what clinical factors are driving any abnormalities.

Studies investigating the bone response to exercise in healthy individuals have mainly been carried out in fasted conditions, often involving intense exercise.(19, 25, 28-30) As moderate-intensity and duration aerobic exercise is the most commonly advocated physical activity by the American Diabetes Association (ADA),(16) and a carbohydrate snack prior to exercise is often required to meet the international consensus advice on pre-exercise blood glucose concentration to reduce the risk of exercise-induced hypoglycemia,(17) it is unclear how applicable this research is to the type 1 diabetes population. Additionally, the challenges of glycemic management, and various comorbidities can make performing intense exercise unrealistic for individuals with type 1 diabetes.

The aim of the present study was to investigate the bone turnover response to moderate-intensity, continuous physical exercise in people with type 1 diabetes compared to healthy controls, replicating real world exercise practices of this population.

Methods

Ethical considerations

This single-centre, case-control trial was performed in line with Good Clinical Practice and the Declaration of Helsinki. Following approval from the NHS HRA North East Tyne & Wear South Research Ethics Committee, fully informed participants gave written consent before any trial related activities. The trial was registered at the ISRCTN registry (http://www.isrctn.com/ISRCTN10346879).

Participants

Eligibility criteria for the type 1 diabetes group comprised age between 18 and 65 years (inclusive), clinical diagnosis of type 1 diabetes (weight loss, ketotic, hyperglycemic and insulin initiation at diagnosis) at least three years before enrolment, glycosylated hemoglobin (HbA1c) < 10.0% (86 mmol/mol), and absence of clinically diagnosed diabetes-related micro-

/ macro-vascular complications (apart from background retinopathy), recent fractures, or abnormal estimated glomerular filtration rate. A minimum duration of diabetes of three years was used to allow a clear gap from the approximate 2 year point often referred to as the 'honey moon period'.(31) Participants had to have stable Multiple Daily Injection (MDI) or Continuous Subcutaneous Insulin Infusion (CSII) regimen without changes over the preceding 6 months. The healthy control group was matched for gender, age, cardio-respiratory fitness (peak oxygen uptake [VO_{2peak}]) and anthropometry. Participants with type 1 diabetes were recruited from the Newcastle Diabetes Centre by posters and clinicians passing the details of interested patients to the study team. The healthy control group were recruited from Newcastle University, using posters and emailing lists.

Screening visit

Participants attended the Newcastle NIHR Clinical Research Facility on two separate occasions. Firstly, participants attended for a screening visit to determine eligibility, medical assessment and resting electrocardiogram. Eligible participants then completed a maximal graded walking to running exercise treadmill test based on the Bruce protocol,(32) as previously described by our group.(33) Participants with type 1 diabetes had their capillary blood glucose concentration measured prior to the maximal test. If blood glucose was below 7 mmol/L (126 mg/dL) then 10-30 grams of carbohydrate was orally administered via a glucose drink.(17, 34) If blood glucose was above 15 mmol/L (270 mg/dl) the test was rescheduled. Breath-by-breath respiratory parameters (MetaLyzer 3B; Cortex, Leipzig, Germany) and heart rate (H10; Polar, Kempele, Finland) were continuously recorded throughout the maximal test. VO_{2peak} was determined by the average oxygen consumption measured over the 30 seconds prior to test termination.

Trial visits

Participants returned to the laboratory at least one week after their screening visit and following an overnight fast (from 10 PM). Participants were instructed to maintain their normal basal insulin regimen, while if they had a hypoglycemic event overnight prior to the study visit, the visit was rearranged. On arrival at ~8.30am, the non-dominant arm of each participant was cannulated. One 10 ml EDTA (Becton, Dickinson and Company, New Jersey, USA) and two serum separation tubes (SSTTM II Advance, Becton, Dickinson and Company, New Jersey, USA) were collected at all-time points (baseline, immediately post-exercise [0 min post], 30 minutes post and 60 minutes post). An additional 4 ml EDTA vacutainer® was drawn at baseline for analysis of HbA1c. The EDTA and one SSTTM vacutainer were centrifuged for 10 minutes at 1500g at 4˚C; serum and plasma were aliquoted and stored at -80˚C in the Faculty of Medical Sciences Biobank facility.

Participants ate a carbohydrate snack (Belvita Soft Bakes Chocolate Chip, Mondelēz International, USA), providing 204 kcal including 31g carbohydrate, immediately after baseline blood tests and remained rested for 30 minutes before starting the trial protocol. After assessment for a safe blood glucose level 7 mmol/L [126 mg/dL] to 12 mmol/L [216 mg/dL] (17) participants completed 45 minutes of steady state incline walking on a treadmill at an exercise intensity set at 60% of VO_{2peak}. If blood glucose dropped below 7 mmol/L during the exercise, the type 1 diabetes participants were given an additional 10g of carbohydrate. Individuals' breath-by-breath respiratory parameters (MetaLyzer 3B; Cortex, Leipzig, Germany) and heart rates (H10; Polar, Kempele, Finland) were continuously recorded throughout the exercise session. Immediately after cessation of exercise, a further blood sample was collected, before participants were seated. Further venous samples were collected at 30 minutes and 60 minutes post-exercise, after which the cannula was removed and the participant discharged from the laboratory if glucose concentration >3.9 mmol/L (70 mg/dL).

Blood sample analysis

Ionised calcium concentrations and HbA1c were analysed by routine hospital clinical biochemistry (Royal Vitoria Infirmary, Newcastle upon Tyne). EDTA plasma concentrations of β-CTx, PINP, and PTH were measured using electrochemiluminescence immunoassay (ECLIA) on a Cobas e601 analyser (Roche Diagnostics, Germany), with inter-assay coefficient of variation (CV) ≤3% within the analytical range 0.2-1.5 µg/L, 20-600 µg/L and 0.127-530 pmol/L, respectively. Serum total calcium, phosphate, and albumin were measured using standard spectrophotometric methods performed on the Roche Cobas c501 analyser, with inter-assay CVs ≤2% within the analytical range of 0.05-5.00 mmol/L, 0.10-6.46 mmol/L and 10-70 g/L, respectively. Serum albumin values were used to calculate the albumin-adjusted calcium (ACa) using the equation ACa=(0.8×[Albumin-4])+[Total calcium]. The analyses took place at the Bioanalytical Facility (University of East Anglia, UK).

Statistical analysis

Participants' characteristics were tabulated as frequencies and percentages (%) for qualitative variables and means±standard deviations (SD) for quantitative variables. A mixed model (Time*Group) repeated measures ANOVA with Tukey post-hoc analysis was performed for absolute values and baseline adjusted percentage change values a) to assess the effect of moderate-intensity physical exercise on bone turnover markers over time; b) to compare the overall effect of exercise on bone turnover markers in people with type 1 diabetes and healthy controls; and c) to examine the difference in the effect of exercise on bone turnover markers over time between people with type 1 diabetes and healthy individuals. Resting counts were assessed by independent t-test. Data were assessed for normality and outliers by Shapiro-Wilk test and boxplots, with skewed data transformed. Relationships were assessed by Pearson's or Spearman's correlation.

Sample size was estimated from available data;(35, 36) in order to detect a difference of at least 5% in the changes in β-CTx, P1NP, and PTH with exercise between groups a sample size of 15 (excluding dropout) per group was needed to test the null hypothesis that the population means are equal with a probability of 0.8. The type 1 error associated with this test is 0.05. Data are presented as means±standard deviation (SD). A P-value <0.05 was considered statistically significant.

Results

Fifteen people with type 1 diabetes (age: 38.7±13.3, diabetes duration: 19.3±11.4 years, method of control: MDI n=6 and CSII n=9) and age-, fitness-, anthropometric- and gender-matched controls were included in this case-control study (Table 1). No participant reported having a dietary pattern or restrictions that would suggest an abnormal habitual dietary calcium intake. All participants completed the protocol without any adverse events or missed samples. The two groups exercised at a matched intensity of their VO_{2peak} (type 1 diabetes: 60.4±4.4 % vs. control: 60.8±5.1 %, p=0.734).

Table 1: Characteristics of participants at baseline. Data are presented as mean ± SD.

Variable	**Type 1 diabetes (n=15)**	**Control (n=15)**	***P*-Value**
Age (years)	38.7 ± 13.3	41.6 ± 12.4	0.546
Gender, n (%)			
Female	7 (46.7)	7 (46.7)	--
Male	8 (53.3)	8 (53.3)	
Ethnicity, n (%)			
White British	14 (93.3)	14 (93.3)	
Indian	1 (6.7)	0 (0)	
African	0 (0)	1 (6.7)	
BMI (kg/m^2)	24.2 ± 2.1	23.9 ± 3.2	0.775
VO_{2peak} (ml/kg/min)	39.1 ± 9.3	44.7 ± 11.6	0.153

HbA_{1c} (mmol/mol)	60.5 ± 6.7	34.0 ± 2.2	<0.001
HbA_{1c} Range (mmol/mol)	53 to 74	30 to 39	

BMI: body mass index; HbA_{1c}: glycated hemoglobin; VO_{2peak}: body weight relativized peak oxygen uptake. Independent sample t-tests were performed to compare quantitative variables between groups.

The absolute data are presented in

Figure **1**, with percentage change from baseline plotted in

Figure 2. The P values for main effects of time, group, and group*time interaction are included with each parameter within Figures 1 and 2. Baseline concentrations for all markers are presented in Table 2.

*****INSERT FIGURES 1 AND 2 HERE*****

There were main effects of time for β-CTx, PTH, albumin, phosphate, and calcium (Figure 1). There were group effects for albumin, and ionized calcium with controls having higher concentrations of both measures at rest and at all time points, when compared with type 1 diabetes (Figure 1.D + 1.H, Table 2.). There were time*condition effects for β-CTx only, with the control group only having a reduced concentration at 60 minutes compared to baseline (Figure 1.A). When expressed as a percentage change from baseline, there were time*group interactions for β-CTx, with reduction in concentrations of 16±12 % versus 25±8 % in the type 1 diabetes group and controls, respectively (p=0.018; Figure 2.A).

HbA1c was inversely related to albumin (r=-0.657, p< 0.001) in the fasted state. VO_{2peak} was related to fasting levels of β-CTx (r=0.478, p=0.008) and P1NP (r= 0.417, p=0.022). Duration of type 1 diabetes was inversely associated with fasted concentrations of albumin (r= -0.636, p<0.001) and β-CTx (r=-0.535, p=0.045). Within the type 1 diabetes group, neither HbA1c, BMI nor VO2peak predicted any percentage change with exercise of the measured variables (p>0.05). An older age and longer duration of diabetes was associated with a greater decrease in albumin (r=-0.757, p=0.001; r=-0.635, p=0.011), calcium (r=-0.764, p=0.001; r=-0.574, p=0.025) and adjusted calcium (r=-0.748, p=0.001; r=-0.555, p=0.032) at 30 minutes post exercise, respectively.

Variable	**Type 1 diabetes (n=15)**	**Control (n=15)**	***P*-Value**
β-CTx (μg/L)	0.32 ± 0.17	0.40 ± 0.16	0.174
P1NP (μg/L)	44.58 ± 22.06	54.40 ± 25.57	0.235
PTH (pmol/L)	2.37 ± 1.03	2.89 ± 0.90	0.149
Albumin (g/L)	39.44 ± 2.03	43.11 ± 2.47	<0.001
Phosphate (mmol/L)	1.05 ± 0.19	1.02 ± 0.18	0.675
Calcium (mmol/L)	2.26 ± 0.08	2.27 ± 0.8	0.652
Adjusted Calcium (mg/dL)	2.27 ± 0.08	2.24 ± 0.07	0.394
Ionised Calcium (mmol.L)	1.16 ± 0.04	1.20 ± 0.01	0.011

Independent sample t-tests were performed to compare quantitative variables between groups.

Table 2: Resting concentrations of biochemical markers of bone turnover. Data are presented as mean ± SD.

Discussion

The aim of the present study was to assess whether the bone turnover response to acute exercise in type 1 diabetes differs compared to that in matched controls free from diabetes. Previous

research has largely studied the bone turnover response to acute exercise in healthy individuals.(19, 28-30, 35) This was the first study, to the best of our knowledge, to assess this acute response in humans with type 1 diabetes, with previous research having been carried out in diabetic rat models (20, 21) or exploring exercise training over several months in children or adolescents with type 1 diabetes.(22, 23) The key findings were: 1) baseline P1NP concentrations were comparable between groups and unaffected by moderate-intensity exercise, 2) baseline β-CTx concentrations were also comparable between groups and fell with exercise (more so in controls), 3) PTH and phosphate levels both rose with exercise, with no difference between groups, and 4) those with type 1 diabetes had overall lower levels of albumin and ionised calcium. Our data provide a clinically relevant insight into the interaction.

In the present study moderate-intensity exercise caused no change in P1NP, a fall in β-CTx, and a rise in PTH in both participants with type 1 diabetes and controls. While there was no statistically significant differences in resting concentration of P1NP and β-CTx in the current study (Table 2.), the baseline β-CTx concentration of 0.8 μg/L lower in type 1 diabetes group compared to the controls is similar to the significantly reduced β-CTx levels (−0.10 μg/L, 95% confidence intervals −0.18 to −0.01 μg/L) seen in a recent meta-analysis.(4) While a lack of studies have investigated P1NP in type 1 diabetes specifically, the resting concentration in this study (-9.82 μg/L in the type 1 diabetes group in comparison to the controls) is reduced a similar amount as individuals with type 2 diabetes compared to controls.(4)

P1NP

Moderate intensity exercise training has previously been demonstrated to alter resting P1NP concentration. Indeed, Adami et al. (37) found that P1NP increased after an exercise program of a month's duration in pre-menopausal women, which was associated with increased bone mineral density. In comparison, Maggio et al. (22) did not find a significant change in P1NP in

either children with or without type 1 diabetes competing 9 months of two 90-min plyometric exercise sessions per week compared to non-exercising controls. This is despite the type 1 diabetes group having significantly lower resting concentration. In comparison, Elhabashy et al. (23) demonstrated that P1NP significantly increased by 40% after 3 months of 3 60-min aerobic exercise per week in adolescent with type 1 diabetes. However, without a control group it is unclear if this was a normal response. While both studies found increases in bone mineral density, differences in biochemical markers of bone turnover are likely due to differing populations, the frequency and type of exercise used. The acute P1NP response to exercise is less clear with Rantalainen et al. (28) finding a non-significant increase in P1NP 24 hours, but not immediately, after a jumping exercise carried out to exhaustion. Hammond et al. (38) and Townsend et al. (35) found an immediate increase in P1NP post exercise in both fasted and fed states before a subsequent decline below baseline by an hour post 8 × 5 min running at 85% VO_{2peak} HIT or running at 75% VO_{2max} till exhaustion. It is possible that our protocol was not strenuous enough to induce an increase in P1NP, or that we did not take measurements over a sufficient time period to detect such a rise. Never the less, Scott et al. (30) reported an acute increase in P1NP levels when healthy males ran at 55%, 65%, and 75% of VO_{2max}, but there was no effect of exercise intensity on P1NP response. It is possible the amount of mechanical strain, rather than the exercise intensity, is key to inducing P1NP secretion.(25) It would thus be interesting to repeat our study using a running protocol.

β-CTx

The fall in β-CTx observed implies that there was a lower rate of bone resorption at the end of exercise. Scott et al. (30) also found that β-CTx fell in their study both during and for three hours after treadmill running at 55% and 65% of VO_{2max}. Other studies have however observed β-CTx to increase with acute exercise or be unaffected. Indeed, Guillemant et al. (29) found elevated β-CTx levels in male athletes after 60 minutes of cycling at 80% VO_{2max}. The acute

response of β-CTx to exercise may depend on the exercise type, duration, and intensity as well as the individual's age, sex, habitual loading of bones.(19, 39, 40) Importantly, prior consumption of calcium or carbohydrate, as administered in this study, appears to result in greater suppression of β-CTx during and after exercise compared to fasting conditions.(29, 35, 38) Additionally, β-CTx is influenced by the circadian rhythm, peaking at 5:30am, with the nadir at 1:30pm.(41) After 9 months of exercise training, Maggio et al. (20) found both children with and without type 1 diabetes had reduced levels of β-CTx, however the reduction was less than both the type 1 diabetes and control groups who did not go through the exercise intervention. In the present study baseline β-CTx levels were (not significantly) lower, and when adjusted for baseline the percentage decrease with exercise in those with type 1 diabetes was less than the controls. This suggests that bone in type 1 diabetes may have a lower basal rate of turnover and remodels less in response to a bout of fed moderate intensity exercise. Further investigation is needed to understand if the acute difference seen in β-CTx around exercise are clinically relevant in the long term.

PTH

The exercise-induced rise in PTH that we observed was in keeping with previous work (26) and was likely due mainly to a fall in ionised calcium.(36) As with P1NP and β-CTx, there may be an exercise intensity threshold for PTH secretion in endurance exercise.(26) Whilst high basal levels of PTH have a catabolic effect, intermittent PTH secretion, such as that observed here, has an anabolic effect on bone,(13) stimulating proliferation and inhibiting apoptosis in osteoblasts.(26) It is difficult to prove without a longer-term study, but tempting to suggest, that our exercise protocol had a positive net effect on bone turnover in both the healthy controls and the type 1 diabetes group, even if the β-CTx resorption response may be slightly attenuated in the latter.

Bone turnover associated metabolites

In the current study, those with type 1 diabetes had lower absolute levels of albumin compared to controls. A longer duration of type 1 diabetes and poorer glycemic control were both also associated with lower fasted levels of albumin. This is unsurprising, as hepatic albumin production is stimulated by insulin and is therefore decreased in type 1 diabetes.(42) We also found lower levels of ionised calcium in participants with type 1 diabetes compared with controls. This has been reported previously, as a result of reduced intestinal absorption and increased urinary excretion of calcium, as well as the dysregulated PTH secretion observed in type 1 diabetes.(43) Whilst still within normal ranges, the lower levels of albumin and ionised calcium in the type 1 diabetes group compared to the age-, gender-, anthropometry- and cardiorespiratory fitness-matched controls may be an indicator of future clinical implications such as oedema formation, neuromuscular irritability, and cardiac complications of hyoocalcemia. In the present study the response of calcium, ionised calcium, adjusted calcium, and albumin to exercise was in line with previous studies (26, 44) and the same in both groups: falling 30 minutes post-exercise and returning to baseline by 60 minutes post. The response of phosphate to exercise was also comparable between groups: increasing immediately after and returning to baseline by 30 minutes post-exercise. This reflects previous studies carried out in healthy individuals.(36, 44) As inorganic phosphate is a major component of bone mineral, its post-exercise rise may be evidence of bone resorption having occurred during the protocol.(36) In type 1 diabetes calcium, albumin, and phosphate homeostasis is deranged,(42, 43) and yet we found that moderate-intensity exercise could still induce appropriate changes in their profiles. Bone in type 1 diabetes is thus seemingly still responsive to acute exercise.

Type 1 diabetes and bone health

As healthcare continues to improve, people with type 1 diabetes are living longer.(14, 15) As most are diagnosed during their youth / young adulthood,(45) they are exposed for many years to the effects of hyperglycemia, and do not fully benefit from the anabolic effects of endogenous insulin on bone during the period of peak acquisition of bone mass.(10) Chronic hyperglycemia results in the formation of advanced glycation end products, which suppress bone formation, increases bone brittleness and impairs fracture healing.(46) Long-term hyperglycemia also compromises bone vasculature, resulting in decreased bone remodelling.(1) This results is poor bone mineral density and quality, and a high rate of osteoporotic fractures in an ageing type 1 diabetes population.(1, 6) This ultimately may leads to increased morbidity and mortality.(3) Exercise can improve glycemic control (17) and sensitivity to exogenous insulin,(18) and was shown to reduce the risk of falls and fractures in the elderly through a Cochrane review,(47) a complication that individuals with type 1 diabetes are highly vulnerable to.(48) Yet exercise rates are lower in those with type 1 diabetes, with reasons including fear of hypoglycaemia, uncertainty about how to control blood sugars around exercise, and diabetes complications.(17) Our data show that those with type 1 diabetes still benefit from the acute effects of exercise on bone turnover. This further underlines the benefits of exercise in type 1 diabetes and that it may be a viable strategy to reducing the osteoporotic fracture rate in older individuals with type 1 diabetes.

Strengths, limitations and future work

Comparing our exercising type 1 diabetes group to a control group who rested, or having participants perform the protocol in a fasted state would have provided useful further data. However, we were most interested in studying the bone turnover response in type 1 diabetes to exercise under real world conditions: moderate intensity, moderate duration exercise as recommended by the ADA,(16) whilst following international guidelines for glycemic management around exercise.(17) We believe this makes the research more generalizable and

is a strength. A limitation of the current study was blood samples not being taken during exercise or beyond one hour afterwards, which would have given us information on any mid-exercise or delayed post-exercise response of bone turnover markers. Additionally, measuring bone mineral density at rest would have been interesting to explore if this was associated with the bone turnover response to acute exercise. As baseline samples were fasted, changes in bone turnover markers likely have been impacted by the consumption of the carbohydrate snack.(40) Whilst our type 1 diabetes group had a range of ages and HbA1c levels, our study was not designed to explore the influence of age and HbA1c on the bone turnover response to exercise. Future studies should explore how individuals with differing HbA1c respond to exercise in order to translate this research to the wider type 1 diabetes population. As individuals with type 1 diabetes are at increased risk of fractures due to trabecular and cortical bone density defects,(6) understanding if exercise improves bone quality at sites most at risk of fractures would be beneficial. Further understanding how clinical factors, such as age and sex differences, influences the bone turnover response to exercise would also be beneficial. Indeed, as C-peptide infusion improved bone quality in a type 1 diabetic rat model (49) and endogenous C-peptide secretion partially predicts post exercise glycaemic control, residual β-cell function may influence bone turnover in individuals with type 1 diabetes.(33)

Conclusion

We present novel data on the response of bone turnover markers to acute exercise in type 1 diabetes. Whilst bone remodelling is dysregulated in type 1 diabetes, we found fed moderate-intensity walking to have a similarly positive impact on biochemical markers of bone turnover in those with type 1 diabetes compared to controls, evidenced by the rise in PTH and the fall in β-CTx. Studies carried out in healthy individuals suggest that exercise protocols that maximise mechanical strain on the bone may be superior for bone health,(13, 50) and that commencing exercise at a young age is important.(25) Further research should aim to decipher

the optimal exercise type, duration, and intensity to maximise bone turnover in the long-term in type 1 diabetes. This will be key to reducing the incidence of fractures and osteoporosis in this patient group, and ultimately improving morbidity and mortality.

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Conflict of interest: None to declare.

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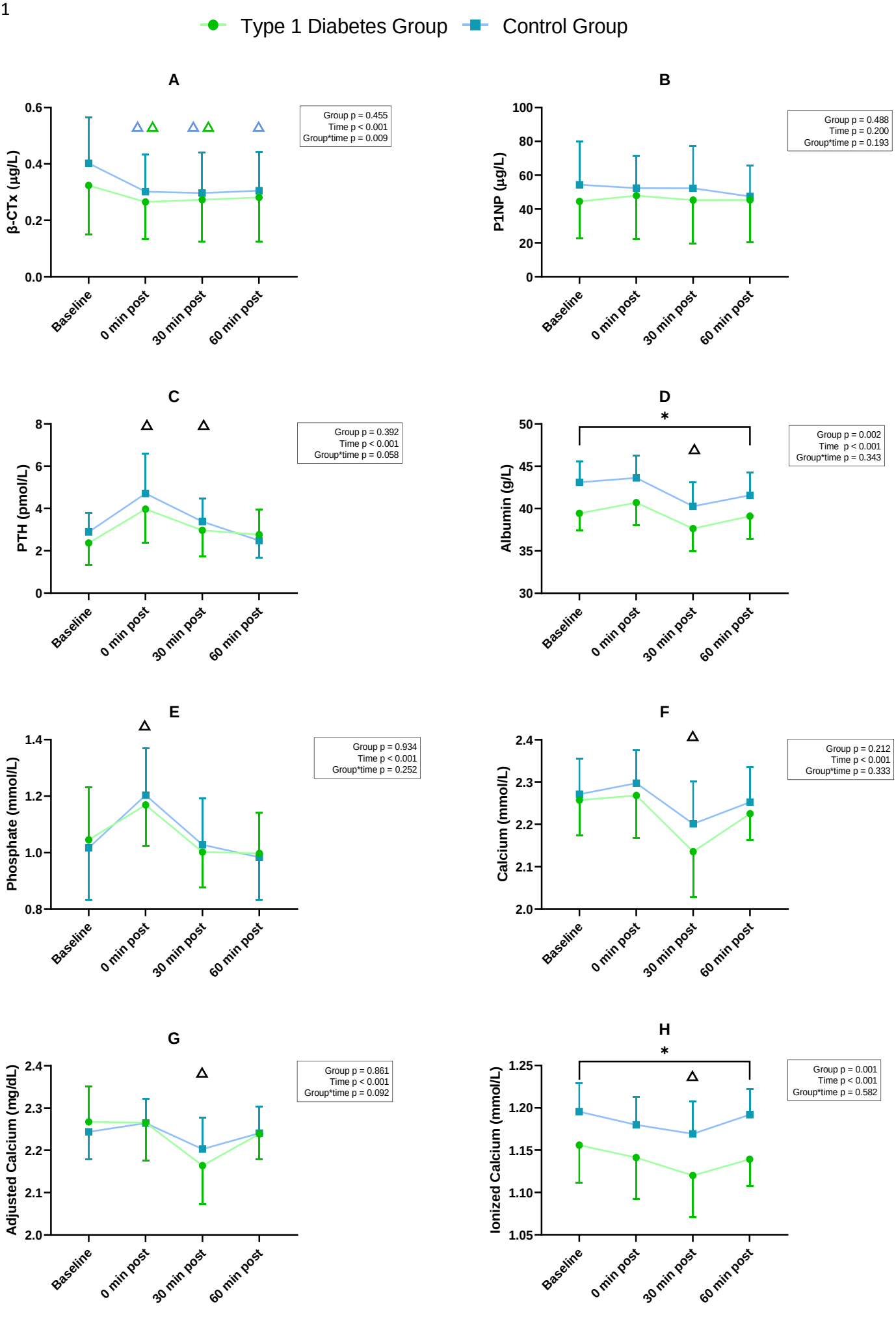
Type 1 Diabetes Group
Control Group
A
β-CTx (µg/L)
Group p = 0.455
Time p < 0.001
Group*time p = 0.009
B
P1NP (µg/L)
Group p = 0.488
Time p = 0.200
Group*time p = 0.193
C
PTH (pmol/L)
Group p = 0.392
Time p < 0.001
Group*time p = 0.058
D
Albumin (g/L)
Group p = 0.002
Time p < 0.001
Group*time p = 0.343
E
Phosphate (mmol/L)
Group p = 0.934
Time p < 0.001
Group*time p = 0.252
F
Calcium (mmol/L)
Group p = 0.212
Time p < 0.001
Group*time p = 0.333
G
Adjusted Calcium (mg/dL)
Group p = 0.861
Time p < 0.001
Group*time p = 0.092
H
Ionized Calcium (mmol/L)
Group p = 0.001
Time p < 0.001
Group*time p = 0.582
Baseline
0 min post
30 min post
60 min post

Figure 1: Absolute change of β-CTx (A), P1NP (B), PTH (C), albumin (D), phosphate (E), calcium (F), adjusted calcium (G) and ionised calcium (H) in response to a single bout of moderate-intensity exercise in those with type 1 diabetes and healthy controls. β-CTx: crosslinking telopeptides of type 1 collagen C-terminal; P1NP: procollagen type 1 propeptides amino-terminal; PTH: parathyroid hormone. Blood samples were collected 30 minutes before exercise (baseline), and immediately (0 min post), 30, and 60 minutes after cessation of exercise.

#[- Significant main effect of group differences, △ – Significant main effect of time difference from baseline, *| - Significant group differences at time point, △ (green triangle) - Significant time difference from baseline in the type 1 diabetes group, △ (blue triangle) - Significant time difference from baseline in the control group

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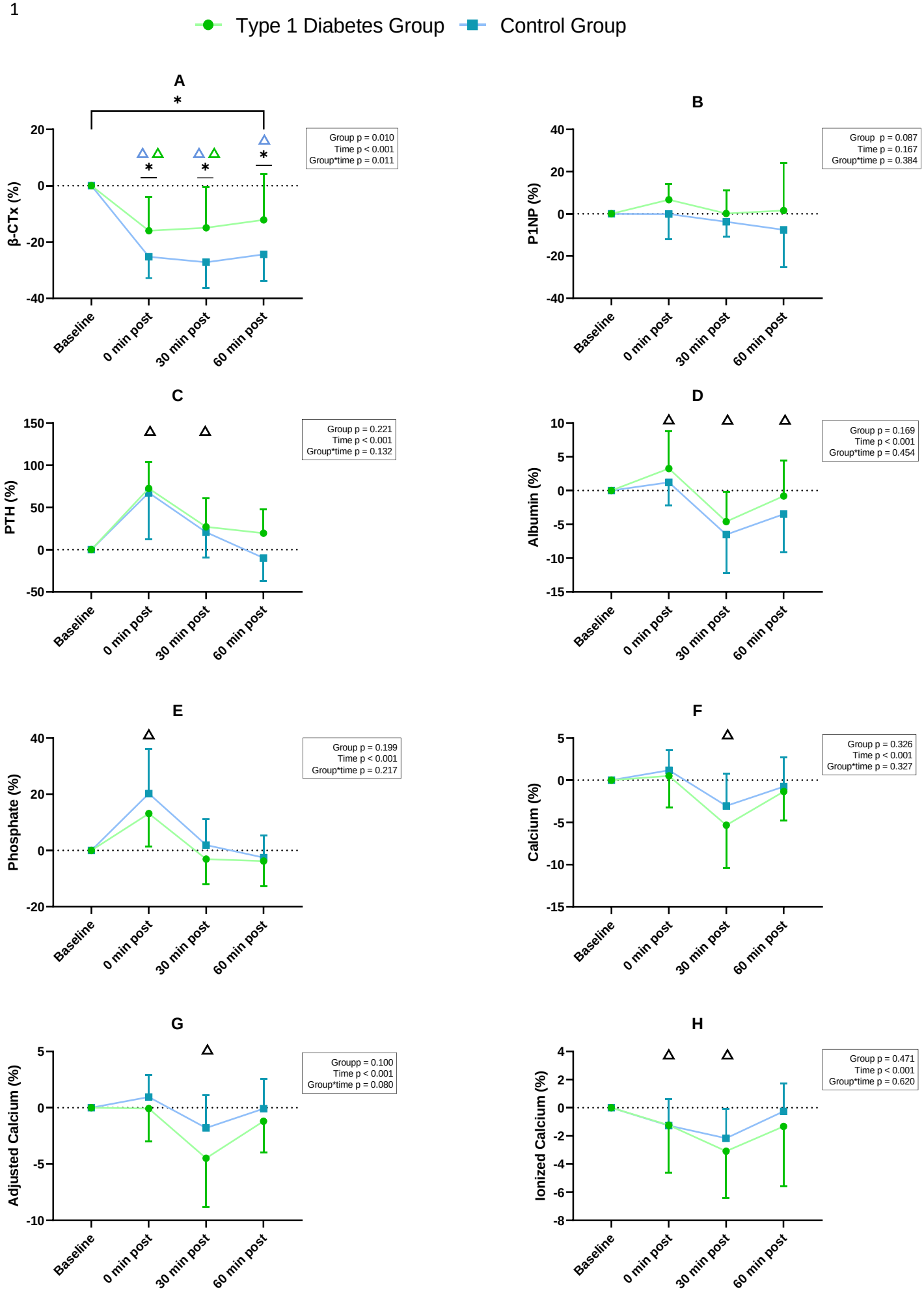
Type 1 Diabetes Group
Control Group
A
β-CTx (%)
Group p = 0.010
Time p < 0.001
Group*time p = 0.011
B
P1NP (%)
Group p = 0.087
Time p = 0.167
Group*time p = 0.384
C
PTH (%)
Group p = 0.221
Time p < 0.001
Group*time p = 0.132
D
Albumin (%)
Group p = 0.169
Time p < 0.001
Group*time p = 0.454
E
Phosphate (%)
Group p = 0.199
Time p < 0.001
Group*time p = 0.217
F
Calcium (%)
Group p = 0.326
Time p < 0.001
Group*time p = 0.327
G
Adjusted Calcium (%)
Groupp = 0.100
Time p < 0.001
Group*time p = 0.080
H
Ionized Calcium (%)
Group p = 0.471
Time p < 0.001
Group*time p = 0.620
Baseline
0 min post
30 min post
60 min post

Figure 2: Percentage change from baseline of β-CTx (A), P1NP (B), PTH (C), albumin (D), phosphate (E), calcium (F), adjusted calcium (G) and ionised calcium (H) in response to a single bout of moderate-intensity exercise in those with type 1 diabetes and healthy controls. β-CTx: crosslinking telopeptides of type 1 collagen C-terminal; P1NP: procollagen type 1 propeptides amino-terminal; PTH: parathyroid hormone. Blood samples were collected 30 minutes before exercise (baseline), and immediately (0 min post), 30, and 60 minutes after cessation of exercise.

#[- Significant main effect of group differences, △ – Significant main effect of time difference from baseline, *| - Significant group differences at time point, △ (green triangle) - Significant time difference from baseline in the type 1 diabetes group, △ (blue triangle) - Significant time difference from baseline in the control group