

REVIEW OPEN ACCESS

Dietary Carbohydrate Intake and Skeletal Muscle Glycogen: A Systematic Review and Meta-Analysis to Determine Optimal Carbohydrate Loading Strategies

R. O. Jones^{1,2}  | J. L. Areta¹  | H. O. Fortis¹  | J. Pugh¹  | J. B. Louis¹ 

¹Research Institute for Sports and Exercise Sciences (RISES), Liverpool John Moores University, Liverpool, UK | ²College of Medicine and Public Health, Flinders University, Bedford Park, South Australia, Australia

Correspondence: J. B. Louis (j.b.louis@ljmu.ac.uk)

Received: 10 February 2026 | **Revised:** 23 September 2026 | **Accepted:** 25 September 2026

ABSTRACT

A high dietary carbohydrate intake in the days pre-competition is recommended to optimize glycogen stores for endurance exercise performance. However, previous reports show high variability in muscle glycogen concentrations between individuals following similar CHO intakes. Therefore, the amount of CHO, duration, and amount of exercise required to maximize glycogen stores pre-competition remains unclear. The aim of this systematic review and meta-analysis was to determine the relationship between dietary CHO availability and skeletal muscle glycogen, whilst identifying the effect of other covariates on muscle glycogen concentration. The analysis included trials (randomized or non-randomized from five online databases) published until June 2026, that reported CHO intake and exercise for ≥ 24 h prior to collection of muscle samples using biopsies to measure skeletal muscle glycogen. Included studies were assessed descriptively and quantitatively using linear and non-linear regression, and generic inverse-variance random-effects meta-analyses, including subgroups to investigate heterogeneity. Descriptive synthesis included 63 trials ($n = 641$ participants), 18 were included within the meta-analysis. Exploratory linear regression indicated a significant relationship between relative CHO intake and whole muscle glycogen for recreationally active ($r^2 = 0.332$, $p < 0.001$) and endurance trained individuals ($r^2 = 0.490$, $p < 0.001$). Meta-analyses revealed increased dietary CHO intake from a low-moderate ($< 6.5 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$) to a high-very high intake ($> 6.5 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$) significantly increased muscle glycogen concentrations ($+191.9 \text{ mmol} \cdot \text{kg}^{-1} \text{ DM}$, 95% CI from 142.8 to 241.1 $\text{mmol} \cdot \text{kg}^{-1} \text{ DM}$, $p < 0.0001$) but displayed significant heterogeneity ($p < 0.0001$, $I^2 = 94\%$). The delta CHO intake between study conditions (expressed as relative $\text{g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$ and absolute $\text{g} \cdot \text{day}^{-1}$) and duration of CHO loading interventions suggested a significant subgroup effect ($p < 0.05$) but failed to explain the heterogeneity in muscle glycogen concentration. Glycogen concentration primarily depends on quantity of CHO intake, with period of increased CHO intake, participant training status, and exercise all being key effectors. Based on current findings, to significantly increase pre-competition muscle glycogen stores athletes should consume $> 8 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$ for 36–48 h pre-competition, with prior glycogen depleting exercise combined with consumption of high glycaemic index CHO allowing more rapid enhancement of stores (24–36 h). However, significant heterogeneity and confounding variables in previous research introduce uncertainty in these recommendations, and further well-controlled studies are required to determine the true optimal pre-exercise CHO intake to maximize glycogen concentrations.

This is an open access article under the terms of the [Creative Commons Attribution](https://creativecommons.org/licenses/by/4.0/) License, which permits use, distribution and reproduction in any medium, provided the original work is properly cited.

© 2026 The Author(s). *Scandinavian Journal of Medicine & Science in Sports* published by John Wiley & Sons Ltd.

1 | Introduction

Endogenous stores of dietary carbohydrate (CHO), in the form of muscle glycogen, are the primary fuel source during moderate-to-high-intensity endurance exercise [1, 2]. Limited stores within skeletal muscle and the liver (~400 and ~100–120 g, respectively, at rest under “normal” physiological conditions) are rapidly depleted during endurance sports competitions, leading to a reduction in physical work capacity due to reduced substrate availability for high-intensity exercise [3–5]. As such, endurance athletes increase dietary CHO intake and decrease training load in the days pre-competition to maximize glycogen storage within skeletal muscle, a strategy termed CHO loading [3, 6, 7].

CHO loading was first introduced by Scandinavian researchers in the 1960s [6], shortly followed by the introduction of the “classic” strategy, where two exhaustive exercise sessions separated by 72 h of low dietary CHO intake were followed by 72 h of rest and a high CHO intake (~575 g per day [g·day⁻¹]). This resulted in above-normal muscle glycogen stores (~800 mmol kg⁻¹ of dry muscle mass [DM]) [3], a phenomenon now termed “muscle glycogen supercompensation.” CHO loading strategies have since been modified and updated, suggesting that shorter periods (24–48 h) of very high CHO intakes, without exhaustive exercise, can be equally effective in achieving enhancement of muscle glycogen storage [8–10]. As such contemporary nutritional guidelines recommend 10–12 g of CHO per kg of body mass per day (g·kg⁻¹·day⁻¹) for 36–48 h pre-endurance competition, to maximize muscle glycogen concentrations and enhance subsequent endurance exercise performance [11].

Despite over half a century of research, and the importance of a high CHO intake for supercompensation of muscle glycogen stores being well-established [3, 8–11], until recently, a systematic review and meta-analysis that objectively quantified the effectiveness of nutritional interventions to maximize muscle glycogen stores had never been conducted [12]. This is particularly relevant, because even though CHO loading is a practice well-established and commonly advocated [11], its effectiveness in maximizing muscle glycogen concentration is still not completely clear (particularly for CHO loading durations of 24–72 h, which was beyond the inclusion criteria of the previous review [12]). For example, studies that utilized similar CHO loading protocols (~8 g·kg⁻¹·day⁻¹ for 72 h) in endurance trained participants have reported high variability of muscle glycogen concentration, with values ranging from 450 to 800 mmol·kg⁻¹ DM [3, 13–15]. The reasons for such variability are unclear, as well-established effectors of glycogen synthesis (CHO quantity, loading duration and training status) were matched [16–18]. Therefore, a better understanding of the effectors which influence glycogen stores is required to determine optimal CHO loading strategies. As such, this systematic review and meta-analysis aimed to examine the relationship and effect of dietary CHO intake on muscle glycogen, and to characterize covariates of muscle glycogen concentration.

2 | Methods

This systematic review and meta-analysis was registered on the Open Science Framework in December 2023 (<https://osf.io/bdasy/>).

The Preferred Reporting Items for Systematic Reviews and Meta-analysis Statement (PRISMA) and Cochrane handbook [19, 20] were consulted throughout the review protocol. Peer-reviewed, English-language journal articles from any time period were included, whilst conference proceedings were excluded due to insufficient information. The inclusion criteria were as follows:

- i. Population—Healthy human, male and/or female participants (aged 18–60 years).
- ii. Intervention—Carbohydrate intake (relative to body mass [g·kg⁻¹·day⁻¹] or calculable as such) and exercise status reported for ≥ 24 h pre-biopsy.
- iii. Comparisons—Different carbohydrate intakes, loading durations (e.g., muscle biopsies following 24 vs. 72 h), participants or conditions.
- iv. Outcome—Whole skeletal muscle glycogen reported in dry mass or calculable as such, analyzed from muscle samples obtained using muscle biopsies and biochemical analysis.
- v. Study design—Randomized, counterbalanced, or non-randomized controlled trials.

Any studies that failed to meet the above criteria were excluded. As there is limited data to determine how age influences the ability to achieve glycogen supercompensation, the population age range was estimated. Children/youth participants were excluded due to hormonal and metabolic differences versus adults, which may influence glycogen storage. No exclusion criteria were specified for muscle biopsy location, yet only *vastus lateralis* and *gastrocnemius* biopsies surpassed the screening process. Non-randomized trials were included to incorporate all relevant information within qualitative descriptive synthesis.

2.1 | Search Strategy and Yield

Five online databases (PubMed, Web of Science, Cochrane, SportDiscus and Scopus) were independently searched by two researchers in February 2022 and December 2024 (R.O.J. and H.O.F., respectively), using the following combination of key words: (CHO OR carbohydrate OR maltodextrin OR glucose OR sucrose OR fructose) AND (Load* OR supercompensat*) AND (muscle OR glycogen OR stores OR storage) AND (concentration OR content OR synthesis OR resynthesis OR utilization OR accumulation). The search was updated again in June 2026, to ensure all relevant studies were captured. No database filters or limits were used to include all potential studies within the screening process. The keyword algorithm from each database for the latest search is reported within [Supporting Information](#). Full search records were exported to Endnote citation manager (version 21, Philadelphia, USA) for title and abstract screening. Any discrepancies were discussed until agreement.

2.2 | Data Extraction

Articles which qualified following title and abstract screening were read in full, with relevant study data extracted onto a custom Microsoft Excel spreadsheet (Microsoft, California,

USA). In line with the PICO terms (Population, Intervention, Comparison, Outcome), data surrounding participants (sample size, sex, age, training level, $\dot{V}O_{2max}$), CHO loading interventions (glycogen depletion/exercise protocol, relative and absolute CHO intake, CHO type [food versus supplements and glycaemic index], CHO loading protocol duration, exercise conducted during loading days), study outcome (whole muscle glycogen across multiple timepoints) and study design were all extracted. Other potentially important variables, such as muscle biopsy method (Bergström, Weil-Blakesley conchotome or gun) and location, biochemical analysis technique, and study dietary control method were also extracted. Participant training status/performance level was categorized as untrained, recreationally active, trained, well-trained, and elite using criteria outlined by De Pauw et al. [21] and Decroix et al. [22] for males and females, respectively. Decisions were made based on available information reported within published articles. If limited information was available, provided ≥ 2 of the performance level criteria were met, participants were categorized accordingly. If no information was available to provide a judgment, author description was accepted. It was noted whether studies reported $\dot{V}O_{2max}$ or $\dot{V}O_{2peak}$, however, for training status classification, both were considered as $\dot{V}O_{2max}$ [23].

All muscle glycogen values were presented as means $\pm$ SD or SE, either in text, tables or figures, apart from Jensen et al. [24] where glycogen was reported as median and interquartile range. To quantify data points presented exclusively in figures ($n=17$ studies [10, 13, 15, 16, 25–37]), an online plot digitizer was used (Plot Digitizer, POrbital, Mumbai, India), which was tested for accuracy using studies containing data presented in text and figures, which generated a mean difference of 2.2 ± 5.5 and 5.5 ± 7.5 mmol·kg⁻¹ DM for studies originally presented as mmol·kg⁻¹ DM and those converted from mmol·kg⁻¹ wet mass to DM, respectively [9, 10, 15, 25, 38–42]. Data not presented as mmol·kg⁻¹ DM were converted using previously established conversion factors [43].

2.3 | Descriptive Methods

The literature search and descriptive synthesis aimed to collect robust evidence within the topic, whilst describing different participant and methodological characteristics between studies. As such, descriptive analysis used tallies of extracted data, which were conducted in two stages by an individual researcher. The first stage focused on study and participant characteristics whilst the second focused on differences between study exercise interventions, CHO loading protocols and muscle biopsy measures. Tallies were totaled and repeated by the same researcher to ensure accuracy, with any discrepancies repeated a third time. To compare differences in CHO intake a low, moderate, high and very high relative CHO intake was defined as <3 , $3-6.5$, $>6.5-9$ and >9 g·kg⁻¹·day⁻¹ (adapted from Areta and Hopkins [43]).

2.4 | Exploratory Linear Regression

To visually describe the relationship between dietary CHO intake (relative and absolute) and skeletal muscle glycogen,

exploratory simple linear and nonlinear 2nd degree polynomial regressions were conducted using information included within the descriptive synthesis. Individual study groups/conditions were plotted based on CHO intake during the intervention and muscle glycogen, separated by participant training status (recreationally active, endurance trained and well-trained). Study conditions that received additional interventions that significantly influenced muscle glycogen concentrations, such as multiple biopsies in close anatomical proximity [44], creatine ingestion [45], delayed CHO refeed [27] or repeated glycogen depletion [46] were excluded, whilst control/placebo conditions were included provided inclusion criteria were satisfied. Studies with multiple timepoints were included as separate groups, as such all observations were not statistically independent. Sensitivity analyses were conducted to determine effects of including all types of study design within the same regression model, the best fit nonlinear polynomial model and to confirm results were robust to the violated statistical assumption of independence of observations (see [Supporting Information](#)).

2.5 | Meta-Analysis Methods

To quantify meaningful differences between dietary CHO intake and skeletal muscle glycogen refined inclusion criteria were introduced to primarily include robust study protocols with low risk of bias (≤ 1 risk of bias domain judged as some concerns; see Section 2.6) within the meta-analysis. As such, studies were required to be randomized controlled trials where a low-moderate CHO intake control was compared against an intervention group which consumed a high-very high relative CHO intake ($0-6.5$ vs. >6.5 g·kg⁻¹·day⁻¹, respectively). CHO intake was required to be consistent throughout the loading period with total loading duration matched between conditions (e.g., 3 g·kg⁻¹·day⁻¹ for 48 h vs. 8 g·kg⁻¹·day⁻¹ for 48 h).

The meta-analysis was conducted using a generic inverse-variance random-effects model on RevMan (version 5.4.1, Copenhagen, Denmark), where a mean difference and SE of the mean difference were calculated for each pairwise comparison. As magnitude of treatment effect can be influenced by experimental design, parallel group and crossover trials were analyzed separately [47]. For crossover trials ($n=12$) dependency was accounted for using previously described methods [48]. Three studies reported individual participant data within original articles [49–51], remaining authors were contacted, and a further two provided individual participant data [14, 52]. For the remaining seven crossover trials, a pooled correlation coefficient was used based on the available calculated correlation data [14, 49, 50, 52]. Crossover trials with multiple eligible comparisons (i.e., four conditions which allowed two comparisons of a low-moderate vs. high-very high CHO intake) were included separately, and sensitivity analyses were conducted to confirm this did not impact results in practice (see [Supporting Information](#), Table S3; [19]). As Jones et al. [51] contained three conditions (moderate, high and very high CHO), the high and very high CHO groups were combined for the overall effect estimate, to include a single pairwise comparison [19, 48]. Two mixed design trials were included within the quantitative synthesis, where two separate

groups (males and females) were compared across repeated measures of manipulated CHO intake. Groups within the same defined category of CHO intake were combined using previously described methods (see [Supporting Information; \[19\]](#)) and pairwise comparisons were then treated as crossover [53] or parallel group trials where appropriate [54]. Sensitivity analyses were conducted to determine the effects of choosing a fixed versus random effects model and ensure borrowed correlation coefficients did not significantly impact results ([Supporting Information; Table S4](#)).

Authors pre-identified the differences in CHO intake between conditions (expressed as relative and absolute intake), duration of manipulated CHO intake (loading duration), participant training status and sex, and muscle biopsy location as variables that could explain predicted study heterogeneity, which was explored with subgroup analyses exclusively in randomized crossover design studies. Pre-analysis, subgroup comparisons between different defined levels of CHO intake (e.g., low versus high, moderate versus high, moderate versus very high) were planned, however this analysis was not deemed appropriate due to vastly uneven covariate distributions (2, 3, 5, and 11 comparisons included within each subgroup) and impaired sensitivity. Therefore, subgroups for the delta (Δ) in relative and absolute CHO intake between low-moderate versus high-very high CHO loading interventions were explored, with subgroups specified at $1\text{ g}\cdot\text{kg}^{-1}\cdot\text{day}^{-1}$ increments from $1.5\text{ g}\cdot\text{kg}^{-1}\cdot\text{day}^{-1}$ and 100 g increments from $100\text{ g}\cdot\text{day}^{-1}$ of CHO, respectively. These increments were pre-determined based on the possibility to result in a small but worthwhile change in glycogen concentration. An analysis for both relative and absolute CHO was conducted to explore whether differences occurred based on how CHO intake was expressed, and whether an absolute CHO threshold for glycogen supercompensation existed. For subgroups of ΔCHO intake, Jones et al. [51] was entered as multiple comparisons across separate subgroups (moderate vs. high and moderate vs. very high) to explore the differences between a moderate vs. high and moderate vs. very high CHO intake. The number of subgroups was determined post hoc as this was dependent on study interventions. Statistical significance was set at $p < 0.10$ for heterogeneity (i.e., variability in the intervention effect between studies) determined using the Cochran's Q test, to account for limited power when relatively few studies are included. The variability in the effect estimate caused by heterogeneity was described using the I^2 statistic, with scores of 0%–40%, 30%–60%, 50%–90%, and 75%–100% corresponding to potentially unimportant, moderate, substantial and considerable heterogeneity, respectively [19].

2.6 | Risk of Bias Assessment

All trials were assessed for risk of bias using the Cochrane risk of bias tools for randomized crossover and parallel group trials (RoB 2 [55]), and non-randomized trials of interventions (ROBINS-I [56]). Assessments were conducted on a whole study level across six domains using both tools (RoB 2 included randomization, carryover effects, deviations from intended interventions, missing outcome data, measurement of the outcome and selection of reported results; ROBINS-I included

confounding, classification of interventions, participant selection, missing data, measurement outcome and selection of reported results). Each domain contained a series of signaling questions answered “yes,” “probably yes,” “no,” “probably no,” or “no information.” The risk of bias categorized for each domain (“low risk,” “some concern,” or “high risk” for RoB 2 and “low,” “moderate,” “serious,” and “critical” risk for ROBINS-I) was judged by researchers and later confirmed by a pre-determined algorithm. Overall, risk of bias was based on the highest risk score across all domains. Assessments were completed by two researchers independently (R.O.J. and J.B.L.) with results compared, and any disagreements discussed until agreement.

3 | Results

3.1 | Descriptive Synthesis

3.1.1 | Study Selection and Characteristics

Figure 1 displays the latest search yield and each stage of the search in a PRISMA flow chart. The search discovered 13 503 potential articles across five electronic databases. Following removal of duplicates and title and abstract screening, 91 articles were read in full, with a further 46 excluded due to failure to meet the inclusion criteria. Sixteen additional studies were identified through citation searching of all included full texts and existing narrative reviews within the area [8, 57, 58]. Therefore, 61 published reports containing 63 experiments were included in the qualitative and descriptive analysis (henceforth terms studies, trials or experiments encompass all 63 included protocols; experiments from the same journal article are differentiated by Experiment 1 and 2 corresponding to the order presented within original articles [13, 16]). Forty-two experiments were randomized, 26 of which used a repeated measures or crossover design, with the remaining 11 and five parallel groups or mixed study designs, respectively. The remaining 21 trials were not randomized (17 repeated measures, two parallel groups and two mixed design). Only eight experiments incorporated blinding, as three studies blinded participants only [27, 28, 30] and five were double-blinded [45, 53, 59–61]. Study and participant characteristics are reported in Table 1.

3.1.2 | Participants

The descriptive synthesis included 641 participants in total, primarily aged 18–30 years (83% of experiments), with only one study reporting participant mean age > 35 years old (37 ± 7 years; [68]). Only 5% of experiments included solely female participants [15, 50, 77] whilst 16% recruited both males and females either as combined [18, 51, 68, 71, 80] or separate groups [31, 36, 38, 53, 54], with the remaining 79% including males exclusively. In total, only 74 females were included across all trials (12% of participants). Participants were primarily endurance trained or well trained as 40% and 22% of trials recruited participants at performance levels 3 and 4, respectively [21, 22]. Twenty-seven percent recruited recreationally active individuals, whilst the remaining 13%

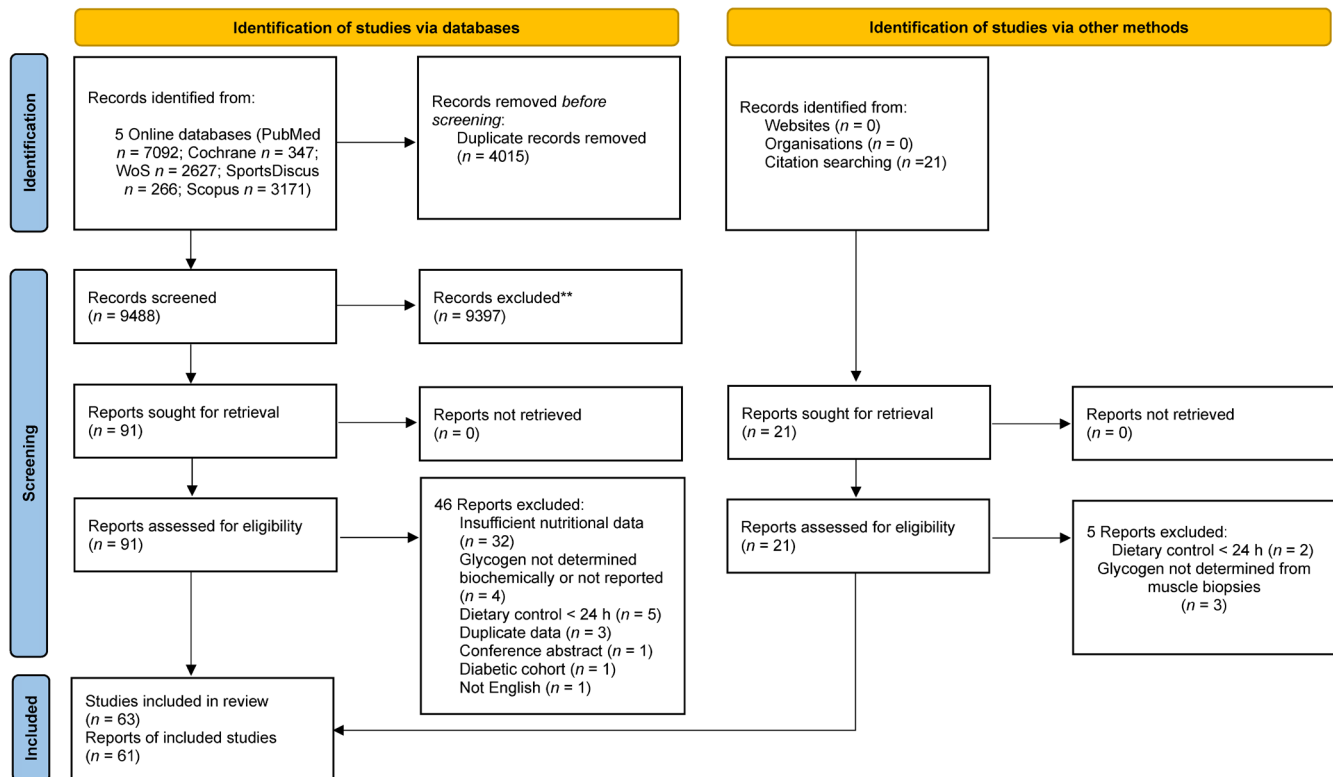


FIGURE 1 | PRISMA flowchart displaying each stage of the literature search conducted to attain articles included within the current systematic review and meta-analysis.

had sedentary participants [32, 33], elite team sport athletes from rugby and ice hockey [40, 62], or compared untrained vs. recreationally active, trained or well-trained participants [13, 17, 18] (Experiment 1).

3.1.3 | CHO Loading Interventions (Dose, Duration, and Type)

Twenty-six studies compared experimental conditions of different defined levels of CHO intake (2 compared low vs. moderate [71, 81], 4 low vs. high [3, 14, 29, 32], 1 compared low vs. very high [2], 11 moderate vs. high [10, 15, 25, 36, 50, 59, 62, 63, 74, 79, 80], 7 compared moderate vs. very high [9, 42, 45, 46, 49, 52, 68] and 1 compared high vs. very high CHO [65]). A further eight studies [13, 34, 37, 38, 40, 70, 73, 77] (Experiment 1) compared different CHO intakes, however expressed relative to body mass ($\text{g}\cdot\text{kg}^{-1}\cdot\text{day}^{-1}$) conditions fell within the same defined category (i.e., moderate). Six trials included multiple conditions of varied CHO intake in a dose-response type study design [16, 24, 51, 53, 54, 75] (Experiment 2). Twenty-one studies compared the exact same relative CHO intake across conditions, with intakes of 6–7 [27, 28, 31, 44, 67, 76] and 9–10 $\text{g}\cdot\text{kg}^{-1}\cdot\text{day}^{-1}$ [17, 18, 26, 60, 61, 64, 66, 69] most common. Manipulation of CHO intake/CHO loading was not an aim of these studies, but rather a controlled factor. Trials were included within descriptive synthesis as all inclusion criteria were met and information was relevant for between-study comparisons, provided the primary study intervention did not significantly impact glycogen concentrations (if this was the case, only the control conditions were considered).

The most common CHO loading/dietary control durations were 48 and 72 h (13 and 21 trials, respectively; Table 1). Six and seven trials used 24 and 96 h loading periods, respectively, whilst others used less common durations of 36, 70, or 84 h. A further five experiments had periods ≥ 5 days [34, 50, 70–72] whilst five studies used different periods of dietary control between conditions, where a control/baseline period of moderate CHO intake and a muscle biopsy was followed by experimental conditions [35, 46, 68, 80].

CHO type was poorly described, as 28 out of 63 trials did not provide enough information regarding CHO consumed to judge glycaemic index. Twenty-eight studies provided mixed diets containing both high and low glycaemic index foods, whilst five trials provided exclusively high glycaemic index CHO sources [9, 29, 39, 65, 66]. Only two trials directly compared different types of CHO sources [16, 64] (Experiment 1).

Overall, dietary interventions were well controlled, as 84% of studies provided participants with food items and meals. However, only 10% of these studies measured and reported actual dietary compliance. The remaining nine experiments (16%) either did not specifically state how dietary intake was controlled [16, 33, 35, 63, 71] (Experiment 1 and 2) or provided participants with meal plans [28, 37, 42, 77].

3.1.4 | Exercise Interventions

Exercise pre-loading was very common (50 out of 63 studies), with some studies allowing participants to continue

TABLE 1 | Summary of studies included within systematic review of CHO loading literature (*n* = 63).

Study	Experimental condition	Participants		CHO intake			Duration	Biopsy location	Whole muscle glycogen (mmol·kg ⁻¹ DM)	Significance between conditions
		<i>n</i> and sex	Training status	Glycogen depletion exercise	Relative (g·kg ⁻¹ ·day ⁻¹)	Absolute (g·day ⁻¹)				
Adamo et al. [25]	LCHO vHCHO	9M	3	Cycling	3.4 8.2	262 640	48 h	VL	294.0 480.0	<i>p</i> < 0.05 between conditions
Akermark et al. [62]	MCHO HCHO	16M 16M	5 5	Other	6.2 8.4	498 675	72 h	VL	352.4 ± 30.5 430.7 ± 30.5	<i>p</i> < 0.05 between conditions
Arkinstall et al. [2]	LCHO LCHO vHCHO vHCHO	7M	3	Cycling	0.7 0.7 10.0 10.0	56 56 803 803	48 h	VL	180.0 ± 17.0 223.0 ± 25.0 591.0 ± 34.0 601.0 ± 52.0	<i>p</i> < 0.001 between low and very high CHO conditions
Arnall et al. [26]	vHCHO +3d vHCHO +5d vHCHO +7d (+ days investigated ability to maintain enhanced stores)	6M 6M 5M	3 3 3	Cycling	9.1 9.1 9.1	720 720 720	72 h	VL	752.6 ± 130.5 791.7 ± 60.9 596 ± 43.5	<i>p</i> < 0.05 vs. baseline
Bergström et al. [3]	High fat + PRO HCHO	9M	3	Cycling	0.0 8.3	0 575	72 h	VL	152.2 ± 24.2 799.4 ± 72.5	<i>p</i> < 0.001 between conditions
Blom et al. [13] Experiment 1	HCHO untrained HCHO well trained	6M 6M	1 4	Running	7.9 9.0	600 600	70 h	G	287.1 ± 37.8 583.3 ± 132.7	<i>p</i> < 0.05 between conditions
Blom et al. [13] Experiment 2	HCHO (exercise) HCHO (rest) (days pre-depletion)	6M	3	Running	8.3 8.3	600 600	70 h	G	654.2 ± 94.8 711.7 ± 104.4	<i>p</i> > 0.05 between conditions
Bosch et al. [63]	MCHO HCHO	7M 9M	2 2	No	4.0 8.1	303 600	72 h	VL	539.4 ± 30.5 843.9 ± 17.4	NC

(Continues)

TABLE 1 | (Continued)

Study	Experimental condition	Participants		CHO intake			Biopsy location	Whole muscle glycogen (mmol·kg ⁻¹ DM)	Significance between conditions
		n and sex	Training status	Glycogen depletion exercise	Relative (g·kg ⁻¹ ·day ⁻¹)	Absolute (g·day ⁻¹)			
Bradley et al. [40]	MCHO	7M	5	Other	3.0	264	VL	449.0 ± 19.3 ^a	Unclear differences between conditions (magnitude-based statistics)
	MCHO	7M	5		6.0	528		444.0 ± 30.6 ^a	
Bradley et al. [27]	MCHO (immediate)	8M	2	Cycling	6.0	524	VL	416 ± 56.9 ^a	NC
	MCHO (delayed) (acute post-depletion refeed strategy)	7M	2		6.0	524		362 ± 54.0 ^a	
Burke et al. [64]	vHCHO low GI	5M	4	Cycling	10.0	687	VL	462.8 ± 33.5	p < 0.05 between conditions
	vHCHO high GI				10.0	687		575.9 ± 32.6	
Burke et al. [65]	HCHO	8M	3	Cycling	7.0	518	VL	522.9 ± 41.8	p > 0.05 between conditions
	HCHO + fat +PRO				7.0	518		452.4 ± 16.5	
	vHCHO Energy match				11.8	873		519.4 ± 25.2	
Burke et al. [66]	vHCHO Gorging	8M	3	Cycling	10.0	702	VL	565.1 ± 44.4	p > 0.05 between conditions
	vHCHO Nibbling				10.0	702		628.6 ± 53.1	
Burke et al. [59]	MCHO	7M	4	Cycling	5.8	419	VL	485.0 ± 48.4 ^a	p < 0.05 between conditions
	vHCHO				9.0	646		572 ± 40.4 ^a	
Bussau et al. [9]	Baseline	8M	4	No	5.8	447	VL	417.6 ± 17.4	p < 0.05 vs. baseline
	vHCHO				10.2	785		848.3 ± 74.0	
Costill et al. [16] Experiment 1	HCHO complex	6M	3	Running	9.0 + 5.8	648 + 415	G	725.1 ± 41.3	p < 0.05 between conditions
	HCHO simple				9.0 + 5.8	648 + 415		642.1 ± 32.6	

(Continues)

TABLE 1 | (Continued)

Study	Experimental condition	Participants		CHO intake			Duration	Biopsy location	Whole muscle glycogen (mmol·kg ⁻¹ DM)	Significance between conditions
		n and sex	Training status	Glycogen depletion exercise	Relative (g·kg ⁻¹ ·day ⁻¹)	Absolute (g·day ⁻¹)				
Costill et al. [16] Experiment 2	LCHO	4M	3	Running	2.4	188	24 h	G	289.7 ± 33.9	<i>p</i> < 0.05 between high conditions vs. LCHO and MCHO
	MCHO				4.7	375			322.8 ± 17.0	
	HCHO				6.6	525			440.2 ± 90.9	
	HCHO				6.6	525			546.4 ± 47.4	
Costill et al. [44]	HCHO control	8M	3	Cycling	6.6	500	48 h	VL	604.2	<i>p</i> > 0.05 between conditions
	HCHO distal (multiple biopsies)	(4M)			6.6	500			551.1	
De Bock et al. [67]	MCHO	8M	2	No	6.1	458	72 h	VL	455.0 ± 40.0	<i>p</i> > 0.05 between conditions
	MCHO				6.1	458			466.0 ± 19.0	
Doering et al. [68]	Baseline	6M 1F	3	No	5.2	396	24 h	VL	583.6 ± 45.3 ^a	Conditions vs. baseline <i>p</i> = 0.04 <i>p</i> = 0.01
	vHCHO			Cycling	10.6	807	96 h		835.1 ± 46.1 ^a	
				Cycling	10.6	807	96 h		848.3 ± 45.5 ^a	
	vHCHO 2nd depletion									
Duhamel et al. [32]	LCHO	10M	1	Cycling	1.1	94	96 h	VL	365.7 ± 26.6	<i>p</i> < 0.05 between conditions
	HCHO				7.5	657			568.5 ± 44.9	
Fairchild et al. [39]	Baseline	7M	3	No	~5.6	~430	96 h	VL	474.6 ± 35.7	<i>p</i> < 0.05 vs. baseline
	vHCHO			Cycling	10.3	793	24 h		862.2 ± 57.0	
Fell et al. [41]	vHCHO	8M	3		12.0	883	36 h	VL	698.0 ± 34.6 ^a	<i>p</i> = 0.14 between conditions
	vHCHO			Cycling	12.0	883			742.0 ± 38.5 ^a	
	vHCHO				12.0	883			767.0 ± 30.8 ^a	
	HCHO	8M	3	Cycling	6.9	500	48 h	VL	794.3 ± 28.7	
Flynn et al. [28]	HCHO				6.9	500			778.7 ± 47.4	<i>p</i> > 0.05 between conditions
	HCHO				6.9	500			824.3 ± 41.8	
	HCHO				6.9	500			836.1 ± 65.3	

(Continues)

TABLE 1 | (Continued)

Study	Experimental condition	Participants		Glycogen depletion exercise	CHO intake		Duration	Biopsy location	Whole muscle glycogen (mmol·kg ⁻¹ DM)	Significance between conditions
		n and sex	Training status		Relative (g·kg ⁻¹ ·day ⁻¹)	Absolute (g·day ⁻¹)				
Fogelholm et al. [37]	vHCHO half marathon	6 M	4	Running	9.1	598	96 h	VL	281.0 ± 25.4 ^a	<i>p</i> > 0.05 between conditions
Foskett et al. [60]	vHCHO Fartlek								343.5 ± 27.6 ^a	
	vHCHO	6 M	2	Running	9.5	626				
	vHCHO				10.0	750	48 h	VL	512.0 ± 41.6 ^a	<i>p</i> > 0.05 between conditions
Galbo et al. [29]	Fat + PRO	7 M	3	Cycle + run	10.0	750			533.0 ± 31.4 ^a	
	HCHO				0.0	0	96 h	VL	195.5 ± 26.1	<i>p</i> < 0.05 between conditions
Goforth et al. [69]	vHCHO	14 M	2	Cycling	7.3	559			487.1 ± 65.2	
					9.3	720	72 h	VL	729.0 ± 83.9 ^a	<i>p</i> < 0.05 vs. baseline
Greive et al. [18]	vHCHO untrained	2 M 4 F	1	Cycling	10.0	630	48 h	VL	428.5 ± 85.7	<i>p</i> < 0.05 between conditions
	vHCHO 10wk trained		2		10.0	630			795.6 ± 124.9	
Hawley et al. [42]	MCHO	6 M	4	No	5.9	426	72 h	VL	459.0 ± 33.9 ^a	<i>p</i> < 0.05 between conditions
	vHCHO				9.3	661			565.0 ± 25.3 ^a	
Helge et al. [70]	7wk high Fat +1wk HCHO	7 M	2	Cycling	6.9	595	7 d	VL	872.0 ± 59.0	<i>p</i> < 0.05 between conditions
Hickner et al. [17]	8wk HCHO	6 M	2		7.0	599			688.0 ± 43.0	
	vHCHO trained	3 M	3	Cycling	10.0	736	48 h	VL	635.1 ± 167.5	<i>p</i> < 0.05 between trained vs. untrained
	vHCHO trained	3 M			10.0		72 h		792.1 ± 90.5	<i>p</i> = 0.46 for 48
	vHCHO untrained	3 M	1		10.0	688	48 h		275.8 ± 31.3	vs. 72 h trained
	vHCHO untrained	3 M			10.0		72 h		584.6 ± 37.4	<i>p</i> < 0.01 for 48 vs. 72 h untrained
Hill et al. [30]	vHCHO + PRO	6 M	3	No	8.0 + 10.0	592 + 740	14 d + 48 h	VL	633.0 ± 82.0	<i>p</i> > 0.05 between conditions
	vHCHO				8.0 + 10.0	592 + 740			695.0 ± 61.0	

(Continues)

TABLE 1 | (Continued)

Study	Experimental condition	Participants		CHO intake			Duration	Biopsy location	Whole muscle glycogen (mmol·kg ⁻¹ DM)	Significance between conditions
		<i>n</i> and sex	Training status	Glycogen depletion exercise	Relative (g·kg ⁻¹ ·day ⁻¹)	Absolute (g·day ⁻¹)				
Impey et al. [31]	MCHO for all, post-load running exercise manipulated	11M	2	Running	6.0	457	48 h	VL	446.0 ± 74.0	<i>p</i> < 0.001 for males vs. females in G <i>p</i> > 0.05 between all other conditions.
					6.0	457		VL	421.0 ± 74.0	
					6.0	457		VL	409.0 ± 91.0	
					6.0	457		G	504.0 ± 157.0	
					6.0	457		G	498.0 ± 207.0	
					6.0	457		G	542.0 ± 117.0	
		10F	2		6.0	369		VL	367.0 ± 148.0	
					6.0	369		VL	430.0 ± 128.0	
					6.0	369		VL	373.0 ± 95.0	
					6.0	369		G	433.0 ± 117.0	
					6.0	369		G	296.0 ± 117.0	<i>p</i> > 0.05 between conditions <i>p</i> < 0.05 vs. baseline for all <i>p</i> < 0.05 between conditions
					6.0	369		G	438.0 ± 140.0	
James et al. [38]	vHCHO	6M	3	Cycling	10.5	872	72 h	VL	796.1 ± 44.4 ^a	
	vHCHO pre-menses	6F	3		9.9	684			878.7 ± 35.5 ^a	
	vHCHO post-menses				9.9	684			839.6 ± 24.9 ^a	
Jansson and Kaijser, [71]	LCHO	4 M 3 F	2	No	0.3	21	120 h	VL	239.0 ± 25.7 ^a	
	MCHO				5.9	383			445.0 ± 43.5 ^a	
Jensen et al. [24]	LCHO	11M	3	Cycling	0.2	15	72 h	VL	257 (233–360)	
	MCHO				4.7	361	24 h		488 (460–532)	
	HCHO				8.0	616	72 h		499 (472–568)	
									Median (IQR)	

(Continues)

TABLE 1 | (Continued)

Study	Experimental condition	Participants		CHO intake			Whole muscle glycogen (mmol·kg ⁻¹ DM)	Significance between conditions
		n and sex	Training status	Glycogen depletion exercise	Relative (g·kg ⁻¹ ·day ⁻¹)	Absolute (g·day ⁻¹)		
Jones et al. [61]	HCHO	10M	2	Cycling	9.0	654	355.0 ± 20.6 ^a	p < 0.05 between conditions
	HCHO + Blackcurrant supplement				9.0	654	493.0 ± 26.9 ^a	
Jones et al. [51]	MCHO	11M	3	Cycling	5.9	419	460.9 ± 30.5 ^a	p < 0.05 vs. vHCHO
	HCHO				7.8	560	506.1 ± 37.4 ^a	
	vHCHO				9.7	691	635.5 ± 23.5 ^a	
Kavouras et al. [14]	LCHO	12M	3	Cycling	1.4	100	314.1 ± 24.4	p < 0.05 between conditions
	vHCHO				8.2	600	454.6 ± 40.9	
Kirwan et al. [72]	MCHO + HCHO	10M	4	Running	6.2 ± 8.0	413 + 533	524.6 ± 30.9	p < 0.05 between conditions
	MCHO + MCHO				6.2 ± 3.9	413 + 260	355.0 ± 41.8	
Kochan et al. [33]	vHCHO no depletion	6M	1	No	11.3	823	502.3 ± 67.6	p < 0.05 between conditions
	vHCHO depletion (one leg model)			Cycling	11.3	823	912.9 ± 77.3	
Lamb et al. [73]	vHCHO pasta	7M	4	Running	11.3	809	566.4 ± 55.2	p > 0.05 between conditions
	vHCHO supplement	7M	4		11.7	800	653.4 ± 51.3	
MacDougall et al. [74]	MCHO	3M	2	Cycling	3.2	252	347.6 ± 10.0	p < 0.05 vs. LCHO only
	HCHO	3M	2		7.7	617	367.6 ± 17.4	
Maunder et al. [75]	LCHO	13M	2	Running	2.4	169	236.2 ± 38.2 ^a	p < 0.05 vs. LCHO only
	MCHO				5.0	322	360.2 ± 27.2 ^a	
	MCHO				6.5	460	355.0 ± 26.1 ^a	

(Continues)

TABLE 1 | (Continued)

Study	Experimental condition	Participants		CHO intake			Duration	Biopsy location	Whole muscle glycogen (mmol·kg ⁻¹ DM)	Significance between conditions
		n and sex	Training status	Glycogen depletion exercise	Relative (g·kg ⁻¹ ·day ⁻¹)	Absolute (g·day ⁻¹)				
McInerney et al. [46]	Baseline	6M	3	No	6.0	435	24 h	VL	435.0 ± 57.0	p < 0.01 between conditions and vs. baseline
	vHCHO			Cycling	12.0	870	48 h		713.0 ± 60.0	
	vHCHO 2nd depletion			Cycling	12.0	870			409.0 ± 40.0	
McLay et al. [15]	MCHO midfollicular	9F	3	Cycling	5.2	359	72 h	VL	575.0 ± 47.7 ^a	p = 0.02 for moderate CHO vs. all other conditions
	MCHO midluteal				5.2	359			771.0 ± 45.7 ^a	
	HCHO midfollicular				8.4	551			728.0 ± 47.0 ^a	
	HCHO midluteal				8.4	551			756.0 ± 37.3 ^a	
Nelson et al. [76]	HCHO depletion	12M	2	Cycling	6.6	NC	72 h	VL	602.0 ± 39.5 ^a	p < 0.05 vs. baseline p > 0.05 between conditions
	HCHO no depletion (one leg model)			No	6.6	NC			488.0 ± 29.4 ^a	
Nicklas et al. [77]	MCHO midluteal	6F	2	Cycling	3.9	237	72 h	VL	451.1 ± 23.9	p > 0.05 between conditions
	MCHO midfollicular				4.2	254			404.6 ± 25.7	
Palmer et al. [78]	MCHO steady state	6M	4	Cycling	5.4	398	72 h	VL	678.6 ± 60.9	p > 0.05 between conditions
	MCHO variable intensity exercise				5.4	398			643.8 ± 100.1	
Rauch et al. [49]	MCHO	8M	4	No	6.2	439	72 h	VL	454.0 ± 34.8	p < 0.0001 between conditions
	vHCHO				10.5	750			664.5 ± 39.2	
Roberts et al. [34]	HCHO	7M	2	Cycling	8.1	637	6 days	VL	789.0 ± 38.0	p < 0.01 between conditions
	HCHO + Creatine	7M			8.2	645			948.0 ± 75.0	
Sasaki et al. [79]	MCHO	5M	2	Cycling	3.4	214	7 days	VL	260.8 ± 45.9	p < 0.05 between conditions
	HCHO				8.0	499			840.2 ± 94.4	
Schytz et al. [52]	MCHO	22M	2	Cycling	4.0	309	48 h	VL	367.0 ± 14.1 ^a	p < 0.0001 between conditions
	HCHO				10.0	773			525.0 ± 14.4 ^a	

(Continues)

TABLE 1 | (Continued)

Study	Experimental condition	Participants		CHO intake			Biopsy location	Whole muscle glycogen (mmol·kg ⁻¹ DM)	Significance between conditions
		n and sex	Training status	Glycogen depletion exercise	Relative (g·kg ⁻¹ ·day ⁻¹)	Absolute (g·day ⁻¹)			
Sherman et al. [10]	MCHO	6M	4	Running	5.1	353	G	693.4 ± 28.7	p < 0.05 between MCHO and HCHO conditions
	3d LCHO 3d HCHO				7.8	542		903.9 ± 50.9	
	3d MCHO 3d HCHO				7.8	542		884.4 ± 32.6	
Sherman et al. [35]	HCHO exercise	5M	4	Running	12.7 ± 7.1	800 + 450	G	474.6 ± 30.5	p < 0.05 for both conditions vs. pre-marathon
	HCHO rest	5M			11.6 ± 6.5	800 + 450		613.8 ± 26.1	
	Pre-marathon	(10M)			8.2	542		852.6 ± 33.1	
Tarnopolsky et al. [36]	MCHO	6F	4	No	5.4	317	VL	438.0 ± 105.6	p > 0.05 between conditions
	HCHO	6M	4		7.1	478		471.4 ± 28.3	
Tarnopolsky et al. [54]	MCHO	8F	3	No	4.8	274	VL	407.3 ± 27.6 ^a	p < 0.01 between male HCHO vs. male LCHO
	MCHO				6.4	370		407.8 ± 20.2 ^a	
	HCHO	7M	3		6.6	492		401.5 ± 29.0 ^a	
	HCHO				8.2	614		565.4 ± 33.3 ^a	
Tarnopolsky et al. [53]	HCHO	6M	3	Cycling	7.9	585	VL	660.2 ± 49.0 ^a	p < 0.05 for males vHCHO and HCHO vs. MCHO
	vHCHO				10.5	778		741.1 ± 76.6 ^a	
	MCHO (habitual)				6.1	452		537.2 ± 32.1 ^a	
	MCHO	7F	3		6.4	380		650.6 ± 71.0 ^a	
Tomcik et al. [45]	HCHO				8.8	523		737.7 ± 74.9 ^a	p < 0.05 between MCHO and vHCHO conditions
	MCHO (habitual)				5.1	303		629.1 ± 87.3 ^a	
	MCHO + Creatine	9M	4	Cycling	6.0	469	VL	589.0 ± 31.3 ^a	
	vHCHO + Creatine				12.0	938		692.0 ± 32.3 ^a	
Vandenbergh et al. [80]	MCHO	9M			6.0	469		581.0 ± 42.3 ^a	p < 0.05 between conditions
	vHCHO				12.0	938		724.0 ± 36.0 ^a	
	MCHO	10 NC	2	Cycling	4.6	313	VL	364.0 ± 23.0	
	HCHO				7.7	525		568.0 ± 35.0	

(Continues)

TABLE 1 | (Continued)

Study	Experimental condition	Participants		CHO intake			Whole muscle glycogen (mmol·kg ⁻¹ DM)	Significance between conditions
		n and sex	Training status	Glycogen depletion exercise	Relative (g·kg ⁻¹ ·day ⁻¹)	Absolute (g·day ⁻¹)		
Walker et al. [50]	MCHO	6F	4	Cycling	4.7	266	625.2 ± 50.1	p < 0.05 between conditions
	HCHO				8.2	464	709.0 ± 44.8	
Widrick et al. [81]	LCHO	8M	3	Cycling	2.3	181	434.1 ± 26.1	p < 0.05 between LCHO and MCHO conditions
	LCHO				2.3	181	477.2 ± 23.1	
	MCHO				6.5	511	783.9 ± 42.2	
	MCHO				6.5	511	740.4 ± 45.2	

Note: Included studies reported exercise status and CHO intake for at least 24 h pre-muscle biopsy. Table outlines experimental conditions, participant characteristics, CHO loading interventions (CHO intake [relative and absolute] and duration), muscle biopsy location and post-load muscle glycogen concentration. Data presented as means ± SE unless otherwise stated. LCHO, MCHO, HCHO, and vHCHO for low, moderate, high, and very high carbohydrate intake as previously defined (< 3, 3–6.5, > 6.5–9, and > 9 g·kg⁻¹·day⁻¹ respectively). *p* values indicate significance between conditions with bold values the threshold for significance < 0.05.

Abbreviations: F, females; G, *gasotrocentius*; h, hours; M, males; NC, not confirmed; PRO, protein; VL, *vastus lateralis*.
aIndicates calculated SE from SD presented in original article. Training status was classified according to De Pauw et al. [21] and Decroix et al. [22] with 1, 2, 3, 4, and 5 corresponding to sedentary, recreationally active, endurance trained, well trained and elite, respectively.

habitual training, which was standardized between conditions [40, 59, 62, 70, 72, 78], however, most trials utilized specific glycogen depleting exercise (44 out of 48 studies). Cycling was the primary exercise mode as 72% of protocols were in cycling models. The remaining 22% and 6% of studies were in running or other mixed protocols, respectively. Only 22 studies allowed exercise during the loading period (35%); these sessions were primarily light-to-moderate-intensity steady state, short duration sessions (≤ 60 min) designed to mimic a pre-competition taper or described as “habitual” training. In the remaining 65%, participants were rested throughout the loading period.

3.2 | Exploratory Linear Regression

Recreationally active individuals demonstrated a significant linear relationship between relative CHO intake and whole muscle glycogen ($F_{1,42}=20.91$, $r^2=0.332$, $p<0.001$; Figure 2a). The strength of the linear relationship increased in endurance trained individuals ($F_{1,75}=72.14$, $r^2=0.490$, $p<0.001$; Figure 2b), with results consistent across second-degree polynomial models ($r^2=0.332$, $p<0.001$ and $r^2=0.515$, $p<0.001$, for recreationally active and endurance trained, respectively). In well-trained participants, simple linear regression indicated that relative CHO intake only predicted 8.6% of the variation in muscle glycogen ($F_{1,24}=2.24$, $r^2=0.086$, $p=0.147$; Figure 2c), which was also consistent in a second-degree polynomial model ($r^2=0.092$, $p=0.331$; Figure 2f).

Regression results were consistent for absolute CHO intake, as recreationally active ($F_{1,40}=16.14$, $r^2=0.288$, $p<0.001$; Figure 3a) and endurance trained individuals ($F_{1,75}=65.02$, $r^2=0.464$, $p<0.001$; Figure 3b) showed a significant linear relationship between absolute CHO intake and muscle glycogen. Results were consistent across second-degree polynomial regression models ($r^2=0.297$, $p<0.001$ and $r^2=0.497$, $p<0.001$, respectively). For well-trained participants, absolute CHO intake only predicted 4.7% of the variance in glycogen concentration ($F_{1,23}=1.13$, $p=0.298$; Figure 3c), which was maintained in a second-degree polynomial model ($r^2=0.047$, $p=0.587$; Figure 3f).

3.3 | Effectors of Muscle Glycogen Concentration

3.3.1 | CHO Intake

Eighteen studies were included within the meta-analyses (13 repeated measures and 5 parallel groups; Figure 4). Increasing relative CHO intake from a low-moderate (< 6.5 g·kg⁻¹·day⁻¹) to high-very high intake (> 6.5 g·kg⁻¹·day⁻¹) increased muscle glycogen by 191.9 mmol·kg⁻¹ DM (95% CI = 142.8 to 241.1 mmol·kg⁻¹ DM, $n=111$, $Z=7.66$, $p<0.0001$) and by 107.6 mmol·kg⁻¹ DM (95% CI = -10.5 to 225.7 mmol·kg⁻¹ DM, $n=81$, $Z=1.79$, $p=0.07$) for repeated measures and parallel group trials, respectively. There was considerable and statistically significant heterogeneity for both repeated measures ($\text{Chi}^2=271.78$, $\text{df}=16$, $p<0.0001$, $I^2=94\%$) and parallel groups designs ($\text{Chi}^2=49.87$, $\text{df}=4$, $p<0.0001$, $I^2=92\%$), which was expected at this stage of the analysis, as sources of heterogeneity were yet to be explored.

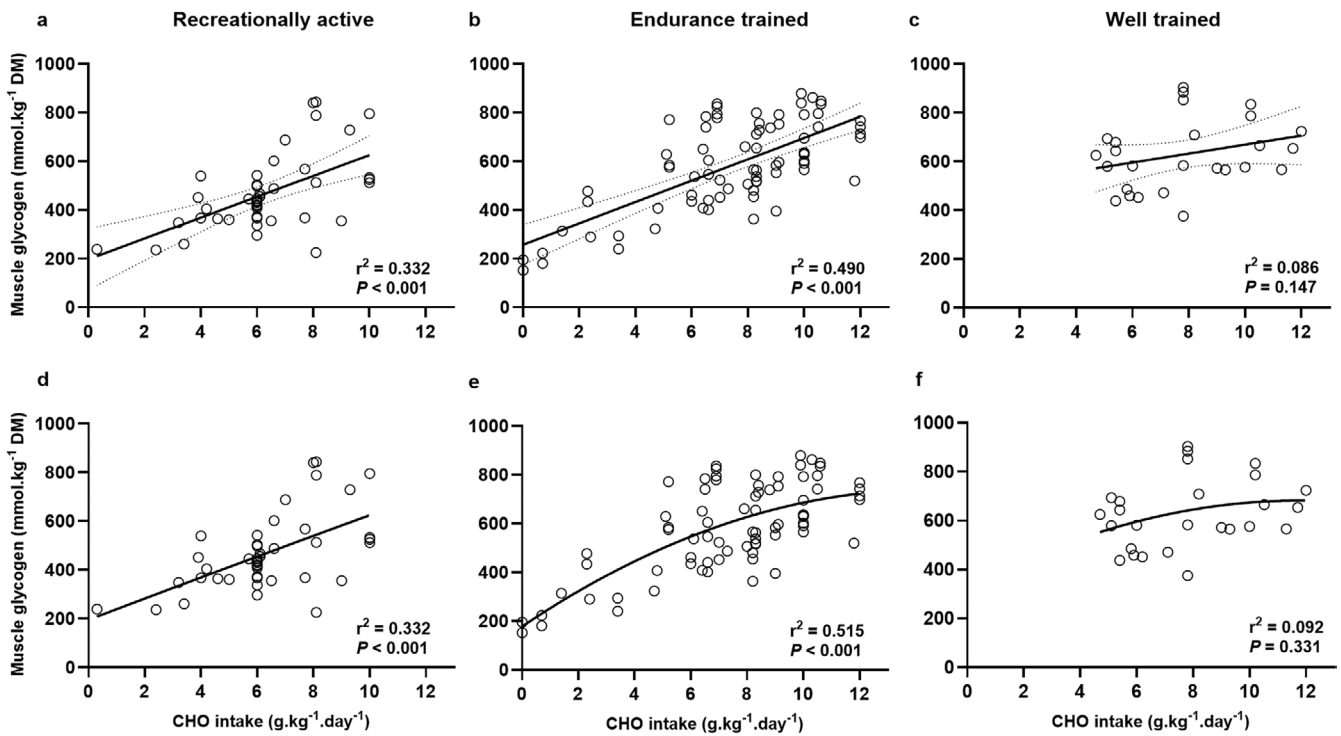


FIGURE 2 | Exploratory simple linear (a–c) and nonlinear second-degree polynomial regression analyses (d–f) of relative CHO intake and skeletal muscle glycogen in recreationally active (a, d), endurance trained (b, e), and well-trained individuals (c, f). Each data point represents a study group/condition. Included study groups reported exercise status and CHO intake for at least 24 h pre-muscle biopsy, and studies which implemented further interventions which may have impacted glycogen concentrations were excluded. Dotted lines display 95% confidence interval bands.

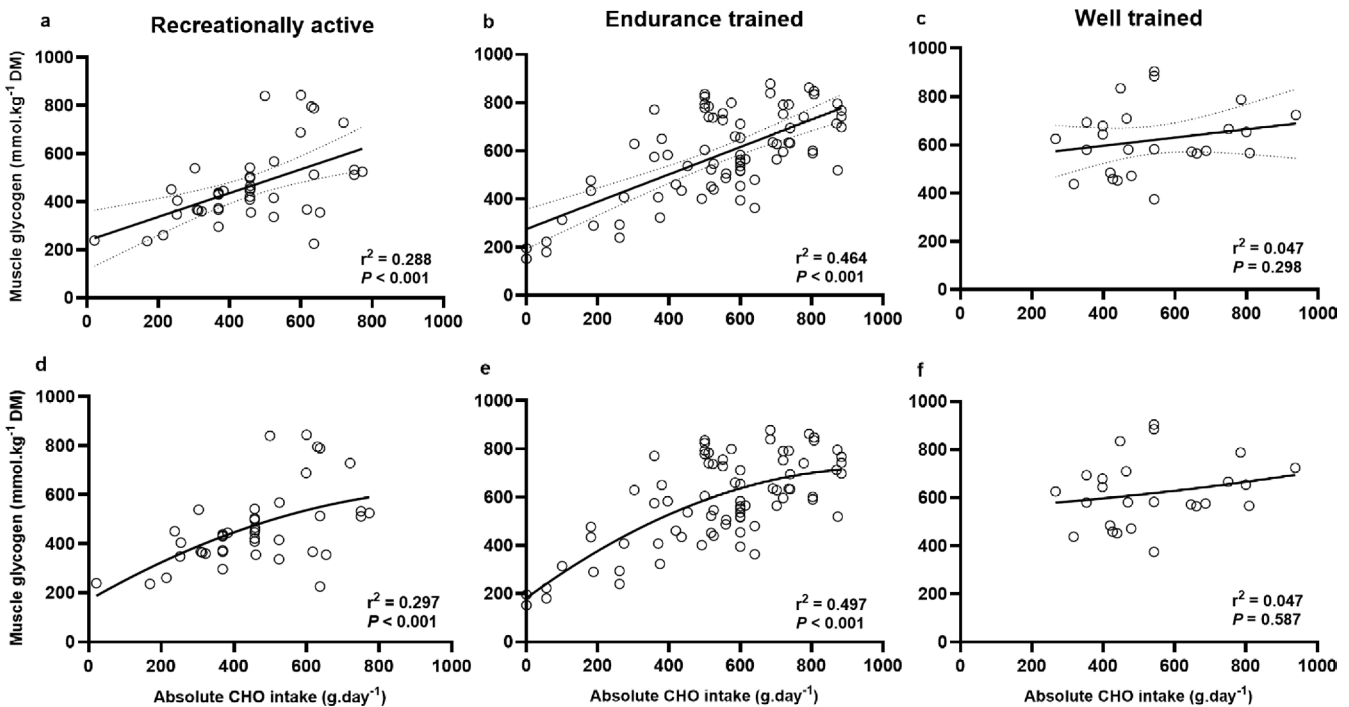


FIGURE 3 | Exploratory simple linear (a–c) and nonlinear second-degree polynomial regression analyses (d–f) of absolute CHO intake and skeletal muscle glycogen in recreationally active (a, d), endurance trained (b, e) and well-trained individuals (c, f). Each data point represents a study group/condition. Included study groups reported exercise status and CHO intake for at least 24 h pre-muscle biopsy, and studies that implemented further interventions which may have impacted glycogen concentrations were excluded. 95% confidence interval bands are displayed as dotted lines.

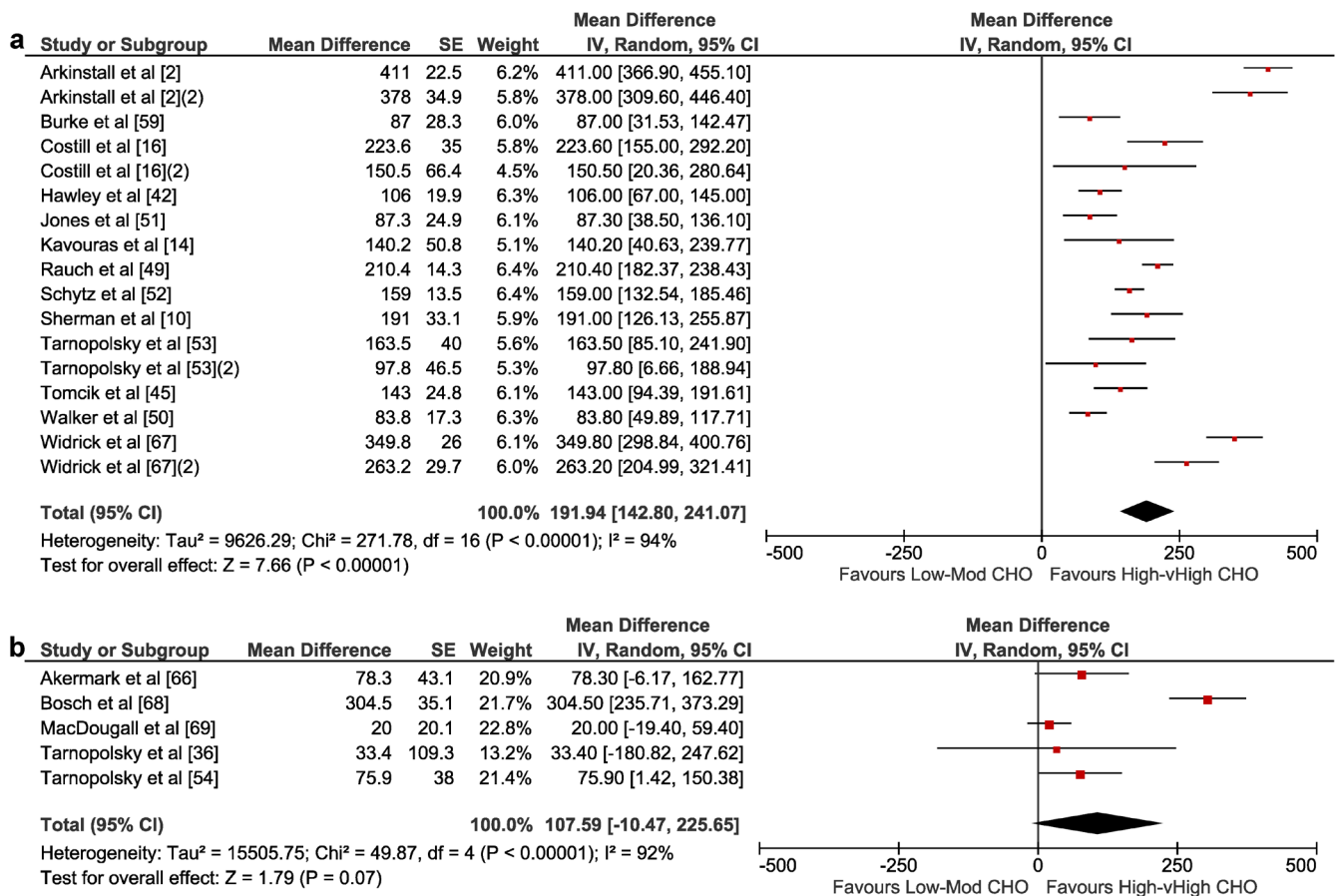


FIGURE 4 | Forest plot of general inverse-variance random-effects meta-analyses conducted for randomized crossover (a) and parallel groups (b) study designs that investigated the effects of a low-moderate versus high-very high CHO intake (Low-MCHO $< 6.5 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$ vs. high-vHCHO $> 6.5 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$) on whole skeletal muscle glycogen obtained from muscle biopsy samples. Included study groups reported exercise status and CHO intake for at least 24 h pre-muscle biopsy.

The Δ relative CHO intake between low-moderate versus high-very high conditions was assessed as incremental $1 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$ subgroups from 1.5 to $> 5.5 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$, however the 4.5 – $5.5 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$ subgroup was not included due to only containing one pairwise comparison [52]. There was a significant quantitative subgroup effect ($p = 0.01$), where increased Δ between relative CHO intakes resulted in an increase in muscle glycogen content across subgroups (Figure 5). However, effect estimates should be interpreted with caution, as despite an improvement in heterogeneity, all subgroups still displayed significant heterogeneity ($I^2 = 93\%$, $p = 0.0001$; $I^2 = 54\%$, $p = 0.06$; $I^2 = 88\%$, $p < 0.0001$; $I^2 = 96\%$, $p < 0.0001$, for Δ relative CHO intakes of 1.5 – 2.5 , 2.5 – 3.5 , 3.5 – 4.5 and $> 5.5 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$, respectively). Covariate distribution was similar for most subgroups (apart from 1.5 – $2.5 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$), as 4–6 study pairwise comparisons contributed to each subgroup with a similar number of participants ($n = 28$ – 38 participants per condition, per subgroup; Figure 5).

Subgroups for Δ absolute CHO intake between conditions were 100 – 200 , 200 – 300 , 300 – 400 , 400 – 500 and $> 500 \text{ g}$, as the $< 100 \text{ g}$ subgroup only contained one pairwise comparison [16] (Experiment 2). There was a significant subgroup effect ($p < 0.0001$), as the magnitude of the significant effect of CHO intake on muscle glycogen increased in line with increases in Δ absolute CHO intake (Figure 6). The 200 – 300 , 400 – 500

and $> 500 \text{ g}$ subgroups presented no significant heterogeneity ($I^2 = 21\%$, $p = 0.29$; $I^2 = 0\%$, $p = 0.82$; $I^2 = 0\%$, $p = 0.43$, respectively), whilst the 100 – 200 g and 300 – 400 g groups also saw decreased heterogeneity compared to the overall effect estimate for crossover trials; however, heterogeneity was still considerable and significant ($I^2 = 83\%$, $p < 0.0001$ and $I^2 = 88\%$, $p < 0.0001$, respectively). Moreover, the number of studies contributing to each subgroup was low, particularly in those with low heterogeneity, as only 4, 3 and 2 pairwise comparisons contributed to the 200 – 300 , 400 – 500 and $> 500 \text{ g}$ subgroups, respectively.

3.3.2 | CHO Loading Duration

There was a significant subgroup effect ($p = 0.01$) as the 24 and 48-h loading periods had a greater impact on muscle glycogen compared to 72 and 96 h. The 48 and 72 h subgroups presented considerable and statistically significant heterogeneity ($p < 0.0001$ for both), whereas the 24 h and 96 h subgroups did not. However, effect estimates should be cautiously interpreted due to an uneven covariate distribution across loading durations and vastly different numbers of participants contributing to each subgroup (Table 2). Bussau et al. [9] reported no benefit in terms of glycogen content following a longer loading period (24 vs. 72 h), whereas others have shown a continued enhancement

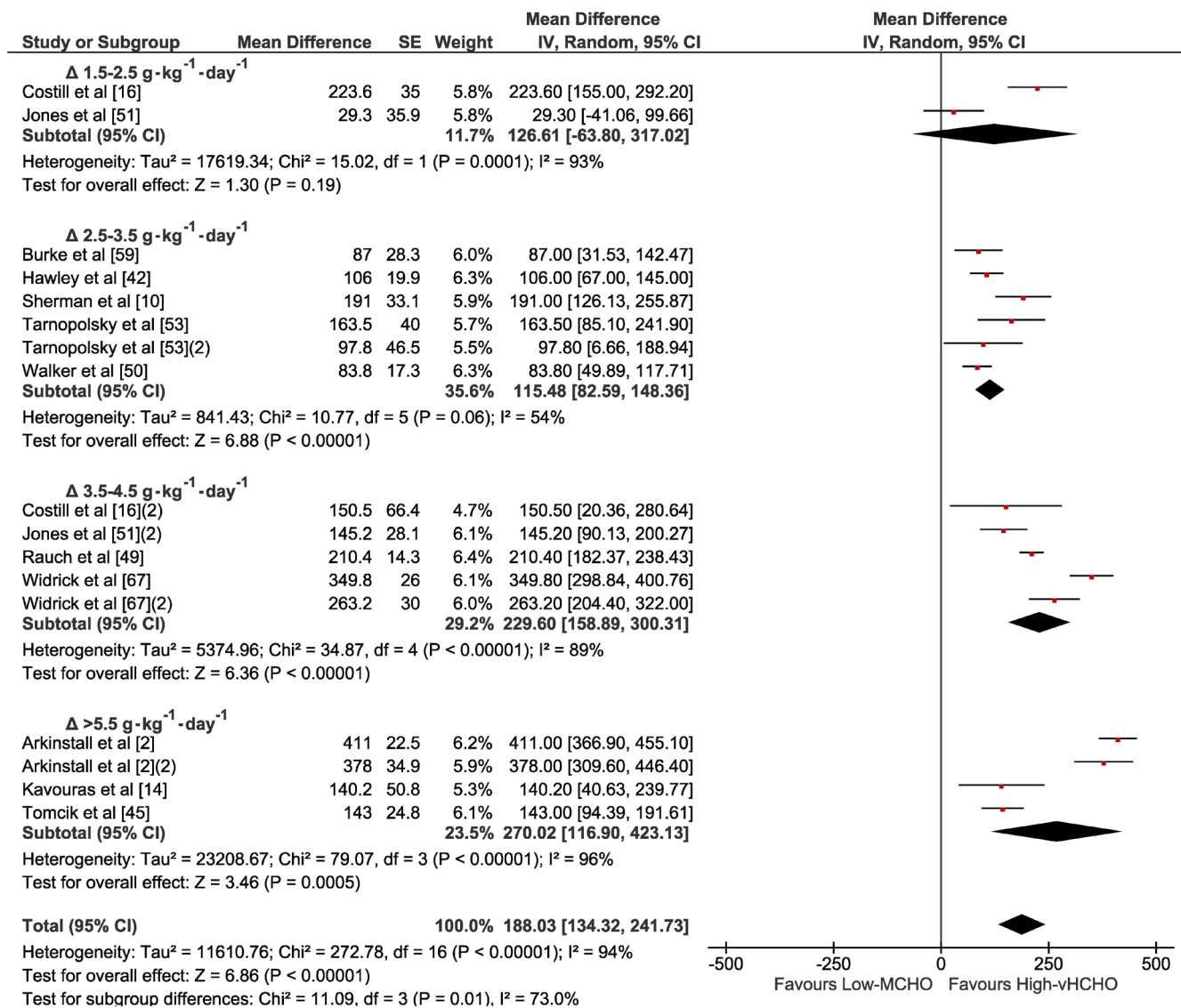


FIGURE 5 | Forest plot of general inverse-variance random-effects meta-analysis conducted in crossover design studies with subgroups for the Δ relative CHO intake between defined low-moderate and high-very high CHO conditions (Low-MCHO $< 6.5 \text{ g}\cdot\text{kg}^{-1}\cdot\text{day}^{-1}$ vs. high-vHCHO $> 6.5 \text{ g}\cdot\text{kg}^{-1}\cdot\text{day}^{-1}$) on whole skeletal muscle glycogen obtained from muscle biopsy samples at subgroups of 1.5–2.5, 2.5–3.5, 3.5–4.5 and $> 5.5 \text{ g}\cdot\text{kg}^{-1}\cdot\text{day}^{-1}$ of CHO. Included study groups reported exercise status and CHO intake for at least 24 h pre-muscle biopsy. Total effect estimates differ due to omission of subgroups with only 1 comparison.

of muscle glycogen stores following high CHO intake over periods of 48 [25], 72 [34] and 96 h [33] versus 24 h.

3.3.3 | Training Status

Subgroup analysis was attempted for participant training status; however, only trained and well-trained subgroups contained more than one pairwise comparison. There was no significant subgroup difference ($p = 0.06$), and CHO intake between study groups in the trained versus well-trained subgroup was similar (mean ΔCHO between comparisons of 4.9 vs. $4.0 \text{ g}\cdot\text{kg}^{-1}\cdot\text{day}^{-1}$, respectively). However, both subgroups presented significant heterogeneity ($p < 0.0001$ and $I^2 \geq 88\%$, for both; Table 2), and the covariate distribution was uneven, as an additional 24 participants contributed to the endurance trained subgroup. Individual studies that investigated differences in glycogen concentrations

following 72 h of a very high CHO intake in trained versus untrained participants [13, 17] (Experiment 1) showed significantly greater glycogen stores in trained versus untrained individuals (583.3 ± 132.7 vs. 287.1 ± 37.8 and 792.1 ± 90.5 vs. $584.6 \pm 37.4 \text{ mmol}\cdot\text{kg}^{-1} \text{ DM}$, respectively). Further, Greiwe et al. [18] reported significantly greater muscle glycogen from the same untrained participants following a repeated CHO loading protocol pre and post 10-week endurance cycling training program (428.5 ± 85.7 and $795.6 \pm 124.9 \text{ mmol}\cdot\text{kg}^{-1} \text{ DM}$, respectively).

3.3.4 | Exercise Pre- and During Loading

Kochan et al. [33] and Nelson et al. [76] compared depleting exercise versus resting conditions specifically using one-legged models in untrained participants. Both studies reported

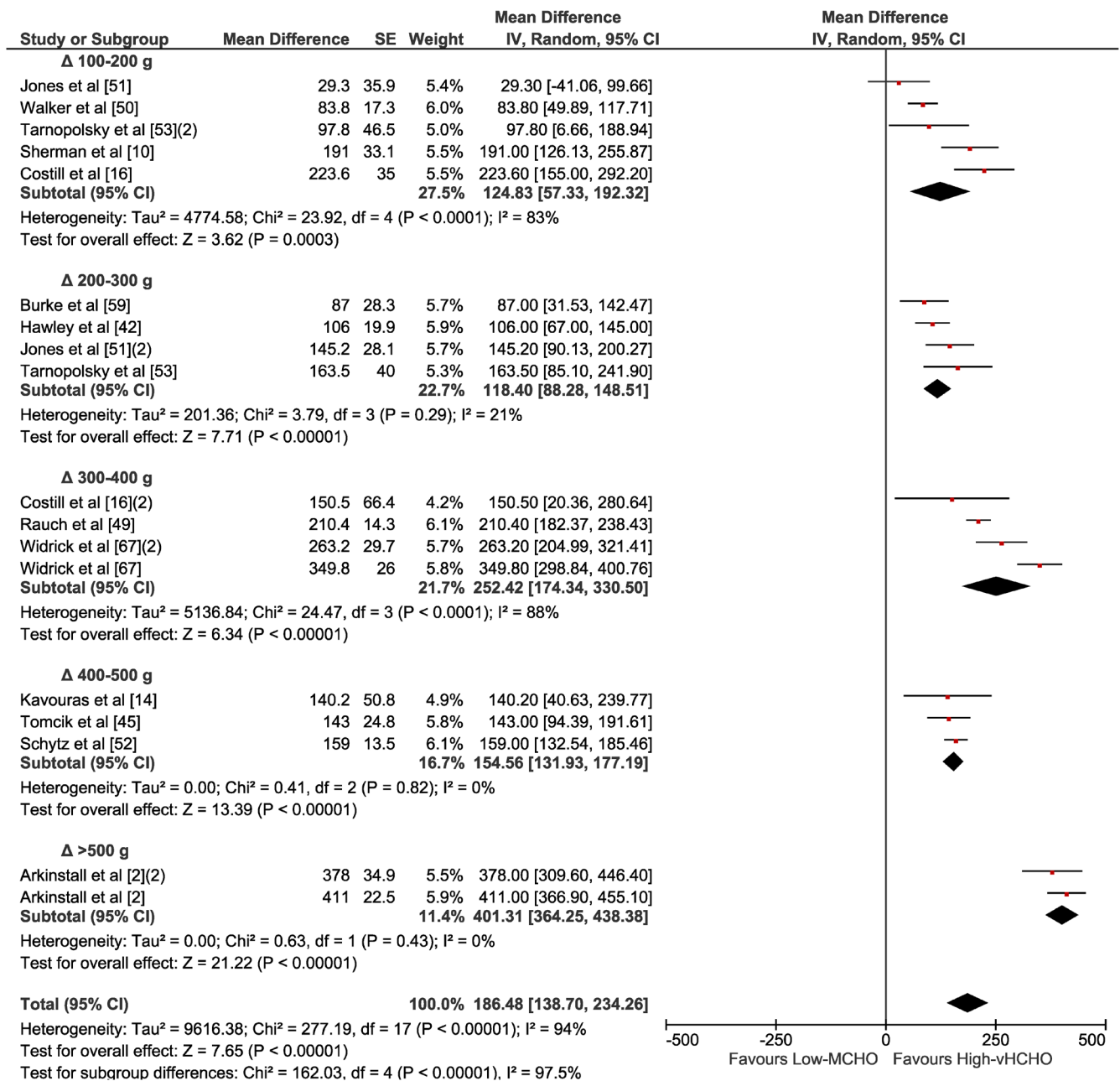


FIGURE 6 | Forest plot of general inverse-variance random-effects meta-analysis conducted in crossover design studies with subgroups for the Δ absolute CHO intake between defined low-moderate and high-very high CHO intake conditions (Low-MCHO $< 6.5 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$ vs. high-vHCHO $> 6.5 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$) on whole skeletal muscle glycogen obtained from muscle biopsy samples in $100 \text{ g} \cdot \text{day}^{-1}$ increments from 100 to $> 500 \text{ g} \cdot \text{day}^{-1}$ of CHO. Included study groups reported exercise status and CHO intake for at least 24 h pre-muscle biopsy. Total effect estimates differ due to omission of subgroups including only 1 comparison.

significantly higher muscle glycogen concentrations in the exercised leg compared to the non-exercised control. Whereas numerous others have reported similar increases in muscle glycogen concentration when a high or very high CHO intake ($8.1\text{--}10.5 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$) was not preceded by specific glycogen depleting exercise [9, 30, 42, 49, 54, 63], however, these were trained individuals and likely completed habitual training in the 24–36 h pre-intervention. Between study comparisons suggest exercise during the loading period does not negatively impact glycogen content, provided exercise is light-to-moderate intensity and relatively short duration (≤ 60 min), replicating

a pre-competition taper [53, 54, 68] and CHO intake is very high [51].

3.3.5 | Other Considered Covariates

No subgroup analysis was attempted for participant sex or muscle biopsy location due to substantially uneven covariate distributions between categorized subgroups (1 study recruited females only, 13 recruited males only and 4 recruited both; 2 studies biopsied the *gastrocnemius* whilst the remaining 16 took

TABLE 2 | Summary statistics for subgroup analyses of loading duration and training status in the random effects meta-analysis of randomized crossover design CHO loading studies, which reported exercise status and CHO intake for at least 24 h pre-muscle biopsy.

Variable	Subgroups	Mean Δ CHO (g·kg ⁻¹ ·day ⁻¹)	n of comparisons (participants)	Mean difference (mmol·kg ⁻¹ DM)	Low 95% CI	High 95% CI	Overall effect	Heterogeneity <i>I</i> ² (p)
Loading duration	24 h	3.1	2 (4)	207.7	147.0	268.4	<i>p</i> < 0.0001	0% (<i>p</i> = 0.33)
	48 h	6.0	7 (56)	255.0	160.7	349.2	<i>p</i> < 0.0001	97% (<i>p</i> < 0.0001)
	72 h	4.1	5 (38)	148.0	90.0	206.0	<i>p</i> < 0.0001	85% (<i>p</i> < 0.0001)
	96 h	3.4	3 (13)	106.4	59.0	153.7	<i>p</i> < 0.0001	40% (<i>p</i> = 0.19)
	Subgroup differences						<i>p</i> = 0.01	
Training status	Trained	4.9	10 (47)	229.9	147.1	312.6	<i>p</i> < 0.0001	94% (<i>p</i> < 0.0001)
	Well trained	4.0	6 (42)	136.6	86.5	186.8	<i>p</i> < 0.0001	88% (<i>p</i> < 0.0001)
	Subgroup differences						<i>p</i> = 0.06	

Note: Subgroups with only one comparison were not pooled and are not shown. Subgroup analyses for Δ relative and Δ absolute CHO intake are presented in Figures 5 and 6. Sex and biopsy-location subgroup analyses were not performed due to uneven covariate distributions. *p* values indicate significance between conditions with bold values the threshold for significance ≤ 0.01 .

Abbreviations: CHO, carbohydrates; CI, confidence interval; h, hours; n, number of pairwise comparisons/participants in each subgroup; wk, week.

muscle samples from the *vastus lateralis*). When CHO intake was matched, males and females appear to have the same capacity to increase muscle glycogen stores [50, 53]. Impey et al. [31] reported lower glycogen concentrations in the *gastrocnemius* of female participants compared to males; however, no differences occurred in the *vastus lateralis*. When other covariates were similar (CHO intake, loading duration and participant training status), studies that collected biopsies from the *gastrocnemius* [10, 13, 16, 35, 72] have reported comparable glycogen content to those collecting samples from the *vastus lateralis* [3, 26, 65].

3.4 | Risk of Bias Results

Of the 42 randomized experiments, 24 studies posed some concerns (Table 3), primarily due to lack of detail regarding randomization and allocation concealment (Domain 1), as methods were inadequately described. The remaining 18 randomized trials were deemed low risk. Of the 21 non-randomized trials assessed using the ROBINS-I (Table 4), 13 studies posed a serious risk of bias due to confounding variables (Domain 1) as participant training status, exercise status ≤ 24 h pre- and during the intervention or menstrual cycle phase were not controlled (or the control was inadequately described). The remaining eight trials posed the lowest overall risk of bias, except for concerns regarding uncontrolled confounding associated with the observational/non-randomized nature of these studies.

4 | Discussion

Muscle glycogen is recognized as critical to optimize exercise capacity in most endurance sport disciplines and strategies to increase glycogen storage have been developed over half a century of research; however the relationship between dietary CHO

intake and muscle glycogen concentration is not well understood. Therefore, this review and meta-analysis aimed to describe and quantify the relationship between pre-exercise dietary CHO intake and skeletal muscle glycogen, whilst identifying key covariates of glycogen concentration. Exploratory regression analyses indicated a linear dose-response relationship between dietary CHO intake and muscle glycogen concentration, with a hierarchy regarding covariates. Quantity of CHO ingested, training status, loading duration and exercise (pre- and during loading) were all effectors of muscle glycogen. Meta-analyses revealed considerable and significant heterogeneity, which resulted from confounding of important covariates across studies. More research is required to truly determine whether an optimal CHO loading strategy for muscle glycogen supercompensation exists.

This systematic review and meta-analysis, for the first time, suggests a significant linear dose-response relationship between dietary CHO intake and skeletal muscle glycogen, as relative CHO intake predicted 49% of the variation in muscle glycogen in endurance trained individuals (Figure 2b), which, in agreement with other reviews within this topic [8, 12, 43], suggests increased quantity of CHO ingested is one of the most important factors to achieve supercompensation of muscle glycogen stores. In line with this, subgroup analyses revealed Δ relative CHO intake subgroups increased muscle glycogen in a stepwise manner, with a possible saturation point, as muscle glycogen only marginally increased in the > 5.5 g·kg⁻¹·day⁻¹ subgroup (Figure 5). Considering habitual CHO intakes of trained individuals (~ 3 – 6 g·kg⁻¹·day⁻¹ dependent on training demands), a 5.5 g·kg⁻¹·day⁻¹ increase would achieve an intake of 8.5 – 11.5 g·kg⁻¹·day⁻¹ of CHO, which corresponds with contemporary CHO loading guidelines [11] and a previously suggested saturation point for muscle glycogen (7 – 10 g·kg⁻¹·day⁻¹) [8, 11, 65]. However, in contrast, subgroup analysis for Δ absolute CHO intake and regression analyses suggested higher CHO intakes are

TABLE 3 | Cochrane risk of bias tool for randomized trials (RoB 2) included within systematic review descriptive synthesis.

Study	D1	DS	D2	D3	D4	D5	Overall
<i>Repeated measures</i>							
Adamo et al. [25]	⚠	+	+	+	+	+	⚠
Arkinwall et al. [2]	+	+	+	+	+	+	+
Burke et al. [64]	⚠	+	+	+	+	+	⚠
Burke et al. [65]	⚠	+	+	+	+	+	⚠
Burke et al. [66]	⚠	+	+	+	+	+	⚠
Burke et al. [59]	+	+	+	+	+	+	+
Costill et al. [16] (Experiment 1)	⚠	+	+	+	+	+	⚠
Costill et al. [16] (Experiment 2)	⚠	+	+	+	+	+	⚠
De Bock et al. [67]	+	+	+	+	+	+	+
Fell et al. [41]	⚠	+	+	+	+	+	⚠
Flynn et al. [28]	⚠	+	+	+	+	+	⚠
Foskett et al. [60]	+	+	+	+	+	+	+
Goforth et al. [69]	⚠	+	+	+	+	+	⚠
Hawley et al. [42]	+	+	+	+	+	+	+
Hill et al. [30]	+	+	+	+	+	+	+
Jones et al. [61]	+	+	+	+	+	+	+
Jones et al. [51]	+	+	+	+	+	+	+
Kavouras et al. [14]	⚠	+	+	+	+	+	⚠
Kirwan et al. [72]	⚠	+	+	+	+	+	⚠
Maunder et al. [75]	⚠	+	+	+	+	+	⚠
McLay et al. [15]	⚠	+	+	+	+	+	⚠
Palmer et al. [78]	⚠	+	+	+	+	+	⚠
Rauch et al. [49]	⚠	+	+	+	+	+	⚠
Schytz et al. [52]	+	+	+	+	+	+	+
Sherman et al. [10]	⚠	+	+	+	+	+	⚠
Tomcik et al. [45]	+	+	+	+	+	+	+
Walker et al. [50]	+	+	+	+	+	+	+
Widrick et al. [81]	⚠	+	+	+	+	+	⚠
<i>Parallel groups or mixed</i>							
Akermark et al. [62]	⚠	+	+	+	+	⚠	⚠
Arnall et al. [26]	+	+	+	+	+	+	+
Bosch et al. [63]	⚠	+	+	+	+	+	⚠
Bradley et al. [40]	⚠	+	+	+	+	⚠	⚠
Bradley et al. [27]	+	+	+	+	+	+	+
Helge et al. [70]	+	+	+	+	+	+	+
Impey et al. [31]	⚠	+	+	+	+	+	⚠
Lamb et al. [73]	⚠	+	+	+	+	+	⚠
MacDougall et al. [74]	⚠	+	+	+	+	+	⚠
Roberts et al. [34]	+	+	+	+	+	+	+
Sherman et al. [35]	+	+	+	+	+	+	+
Tarnopolsky et al. [54]	+	+	+	+	+	+	+
Tarnopolsky et al. [53]	+	+	+	+	+	+	+
Vandenbergh et al. [80]	⚠	+	+	+	+	+	⚠

Note: Included studies reported exercise status and CHO intake for at least 24 h pre-muscle biopsy. ⚠, ⚠, and + correspond to high risk, some concerns and low risk of bias, respectively.
Abbreviations: D1, randomization; D2, deviation from intended intervention; D3, missing data; D4, measurement of outcome; D5, selection of reported results; DS, period and carryover effects.

superior for maximization of glycogen stores pre-competition, as the $> 500 \text{ g} \cdot \text{day}^{-1}$ subgroup had the greatest enhancement of glycogen stores ($+401.3 \text{ mmol} \cdot \text{kg}^{-1} \text{ DM}$), and both simple linear and second-degree polynomial regression models consistently suggested a linear dose–response relationship (Figures 2 and 3), with no apparent saturation point or plateau. The linearity of the relationship weakened somewhat as CHO intake increased $> 10 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$ (Figure 2e) suggesting the true saturation point may be higher than previously suggested [8], at least in endurance trained individuals. However, it should be considered only five study groups contributed to CHO intakes of $\sim 12 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$; therefore, this slight plateau could also be due to lack of information at higher CHO intakes (Figures 2b and 3b).

The discrepancy between subgroup analyses of the Δ CHO consumed highlights the challenging nature of interpreting CHO loading research, particularly as both analyses display considerable and statistically significant heterogeneity across multiple subgroups, uneven covariate distributions and small numbers of study pairwise comparisons contributing to each subgroup (below the $n = 10$ recommended by statisticians; Figures 5 and 6; [19]). As such, the exact magnitude of the relationship between dietary CHO intake and muscle glycogen remains unclear. Conversely, exploratory regression analyses indicate a dose–response relationship, and it remains to be determined whether the previously suggested “ceiling” effect truly exists [65]. Ultimately, irrespective of how CHO intake was expressed (relative or absolute), a marked increase in CHO ingestion appears the most important factor to attain a significant enhancement or supercompensation of muscle glycogen stores (Figures 2 and 3).

Meta-analysis results suggest 24–48 h was sufficient to achieve muscle glycogen supercompensation, whereas longer loading periods of 72 and 96 h provided no further benefit (Table 2). Bussau et al. [9] also claimed 24 h was adequate when CHO intake was very high ($10.2 \text{ g} \cdot \text{kg}^{-1}$), as 72 h provided no further increase in muscle glycogen. However, the true loading period in the previous study was $\sim 36 \text{ h}$ (as the final habitual training session was $\sim 12 \text{ h}$ pre-CHO loading), whilst the current review quantitative effect estimates are untrustworthy due to considerable and statistically significant heterogeneity, small numbers of studies contributing to each subgroup and confounding (mean Δ CHO dose differed across subgroups); therefore, glycogen concentration cannot be attributed to loading duration alone (Table 2), and an optimal loading duration cannot be determined from this analysis. Studies included within the descriptive synthesis showed a continued increase of glycogen stores with 48–72 versus 24 h when CHO intakes were $> 8 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$ [10, 16, 25, 33, 34] (Experiment 1). Additionally, others have only reported replenishment of glycogen to expected baseline concentrations ($\sim 400 \text{ mmol} \cdot \text{kg}^{-1} \text{ DM}$) following 24 h of increased CHO intake post-glycogen depletion [6, 33, 74], however, these participants were not endurance trained. Fairchild et al. [39] reported one of the highest glycogen values ($862.2 \pm 57.0 \text{ mmol} \cdot \text{kg}^{-1} \text{ DM}$) following $10.3 \text{ g} \cdot \text{kg}^{-1}$ of CHO for 24 h in endurance trained participants, suggesting when all factors are optimal (very high CHO intake, high glycaemic index CHO, heightened post-exercise glycogen resynthesis, endurance trained participants), it is possible to achieve glycogen supercompensation within 24 h. Nevertheless, in most cases ~ 1.5 -fold enhancement of stores occurs within 24 h

TABLE 4 | Cochrane risk of bias tool for non-randomized trials (ROBINS-I) included within systematic review descriptive synthesis.

Study	D1	D2	D3	D4	D5	D6	Overall
<i>Repeated measures</i>							
Bergström et al. [3]	!	+	+	+	+	+	!
Blom et al. [13] (Experiment 2)	-	+	+	+	+	+	-
Bussau et al. [9]	-	+	+	+	+	+	-
Costill et al. [44]	-	+	+	+	+	+	-
Doering et al. [68]	!	+	+	+	+	+	!
Duhamel et al. [32]	!	+	+	+	+	+	!
Fairchild et al. [39]	!	+	+	+	+	+	!
Fogelholm et al. [37]	-	+	+	+	+	+	-
Galbo et al. [29]	-	+	+	+	+	+	-
Greiwe et al. [18]	-	+	+	+	+	+	-
Jansson and Kaijser [71]	-	+	+	+	+	+	-
Jensen et al. [24]	!	+	+	+	+	+	!
Kochan et al. [33]	-	+	+	+	+	+	-
McInerney et al. [46]	-	+	+	+	+	+	-
Nelson et al. [76]	-	+	+	+	+	+	!
Nicklas et al. [77]	-	+	+	+	+	+	-
Sasaki et al. [79]	!	+	+	+	+	+	!
<i>Parallel groups or mixed</i>							
Blom et al. [13] (Experiment 1)	-	+	+	+	+	+	-
Hickner et al. [17]	-	+	+	+	+	+	-
James et al. [38]	-	+	+	+	+	+	-
Tarnopolsky et al. [36]	!	+	+	+	+	+	!

Note: Included studies reported exercise status and CHO intake for at least 24 h pre-muscle biopsy. In line with the ROBINS-I criteria, -, !, and + correspond to serious, moderate and low risk of bias. For D1 and overall risk of bias, ! refers to low risk except for concerns about uncontrolled confounding associated with observational and non-randomized study designs.

Abbreviations: D1, confounding; D2, classification of interventions; D3, selection of participants; D4, missing data; D5, measurement of outcome; D6, selection of reported results.

[64–66]. Therefore, 36–48 h of a high to very high CHO intake ($> 8 \text{ g} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$) can be considered a practical recommendation to significantly increase pre-competition glycogen stores.

It is well-established that high glycaemic index CHO sources are superior for achieving a greater enhancement of glycogen synthesis and concentration during shorter recovery periods

(<24h) [64, 82]. High glycaemic index CHO sources provide readily available glucose molecules for absorption within the gut, resulting in higher blood glucose and insulin responses, greater glucose uptake by the muscle, and increased glycogen synthesis compared to low glycaemic index CHO sources, which require longer to digest and absorb, and do not elicit the same blood glucose response [8, 64, 82]. Costill et al. [16] (Experiment 1) suggested there was no effect of consuming simple compared to complex CHO sources over a 24h loading period; however, the classification of complex and simple CHO sources does not correspond to high and low glycaemic index, as complex CHO can still be high glycaemic index [83]. This likely contributed to the lack of differences between conditions reported in the previous study, and unfortunately foods consumed by participants were not adequately described to judge glycaemic index [8, 16] (Experiment 1). Based on existing evidence, athletes implementing shorter duration loading periods pre-competition (<36h) should primarily consume high glycaemic index CHO sources and take advantage of the acute phase of increased post-exercise resynthesis. For athletes using longer loading periods (>36h), a mixed healthy diet with a combination of low and high glycaemic index foods would likely be adequate, as longer recovery periods appear to minimize the importance of CHO type [8, 84]. However, as achieving very high CHO intakes can be challenging due to potential gastrointestinal discomfort [53, 73], athletes should supplement nutritional intake with low fiber, high glycaemic index CHO sources to support tolerability.

Training status did not significantly impact glycogen concentration; however, subgroup effect estimates are untrustworthy due to high subgroup heterogeneity and unbalanced covariate distributions (Table 2). A previous meta-analysis of CHO intake and muscle glycogen reported that under periods of normal and high CHO availability for every $10\text{ mL}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ increase in $\dot{V}\text{O}_{2\text{max}}$, individuals have 67 ± 15 and $123\pm 42\text{ mmol}\cdot\text{kg}^{-1}\text{ DM}$ greater glycogen concentrations at rest [43]. In agreement, previous studies have consistently shown endurance trained individuals have a greater ability to store muscle glycogen post-exercise with increased dietary CHO intake [13, 17, 18], due to higher glycogen synthesis rates post-exercise, primarily due to greater GLUT4 and glycogen synthase activity [13, 17, 18]. Furthermore, endurance trained individuals have a greater composition of type I muscle fibers, and considering glycogen is preferentially stored in the intermyofibrillar space of type I fibers following a high CHO intake for 72h [85], theoretically trained individuals have greater capacity for glycogen storage. Interestingly, regression data showed no significant linear relationship between CHO intake and muscle glycogen in well-trained participants (Figures 2c and 3c). One potential reason behind this could be as training status increases, required CHO intake to achieve maximization of glycogen stores diminishes due to upregulation of post-exercise CHO storage via greater glycogen synthase and GLUT4 activity post-exercise. However, results were likely affected by the smaller number of study groups in the well-trained regression analysis, particularly at low and very high CHO intakes, where no studies fed <4.7 or $>12.0\text{ g}\cdot\text{kg}^{-1}\cdot\text{day}^{-1}$ of CHO (Figure 2c), limiting the ability to detect potential linearity through a smaller spread across CHO intakes compared to other regression models.

Conducting exercise pre-CHO loading, either as a specific glycogen depleting session or a high-intensity exercise stimulus

increases muscle glycogen synthesis rates compared to rested conditions as glycogen utilization drives localized glycogen re-synthesis in the exercised muscle through increased glycogen synthase activity [6, 33, 76, 86]. However, it should be noted that previous studies were conducted in relatively untrained participants, as others have shown a very high CHO intake ($9\text{--}10\text{ g}\cdot\text{kg}^{-1}\cdot\text{day}^{-1}$) without challenging exercise pre-CHO loading to increase glycogen concentrations in endurance trained individuals [9, 30, 49]. However, considering the beneficial mechanisms of an exercise stimulus for glycogen synthesis, athletes should strategically implement CHO loading around pre-competition training schedules, where elite athletes typically complete their final high-intensity training sessions 2–3 days prior (unpublished observations from the field), which would provide a strong stimulus to drive glycogen resynthesis. In combination with an aggressive acute post-exercise refeed of CHO ($1\text{--}1.2\text{ g}\cdot\text{kg}^{-1}\cdot\text{h}^{-1}$ for the first 4h [11]) and a high-very high CHO intake for the subsequent 36–48h, in theory, this would optimize glycogen synthesis and maximize pre-competition glycogen stores (provided no further moderate-to-high-intensity exercise was conducted).

Exercise during the loading period itself also requires consideration, as training at a moderate-to-high intensity in the days before competition would utilize glycogen stores, potentially leading to suboptimal glycogen concentrations for endurance performance. Between study comparisons suggest, provided CHO intake is very high (e.g., $10\text{ g}\cdot\text{kg}^{-1}$) and training intensity is low-to-moderate (e.g., pre-competition taper), there is no negative effect of training during the loading period on muscle glycogen concentration [10, 51, 53, 68], which is common practice for endurance athletes to maintain training adaptations [87]. Interestingly, 65% of studies rested participants during periods of increased CHO intake, which does not represent the real-world pre-competition practices of athletes. Furthermore, those who included light-to-moderate-intensity exercise sessions as a simulated pre-competition taper, collected no measurements during exercise to describe physiological or metabolic responses [10, 53, 68]. Considering increased glycogen stores augment the contribution of glycogen to CHO oxidation and exercise energy expenditure at the same relative exercise intensity [1, 2], glycogen utilization during exercise conducted during the loading period would be increased (provided intensity and duration were consistent). Alternatively, these sessions also provide an exercise stimulus, with augmented post-exercise synthesis rates providing an opportunity to replenish glycogen stores (provided post-exercise CHO ingestion was adequately increased and other effectors [timing and type] were optimal), which could explain the high glycogen concentrations reported in the previous studies [10, 51, 53, 68]. Nevertheless, these suggestions are speculative and require further investigation.

Tarnopolsky et al. [54] suggested participant biological sex was a covariate of muscle glycogen, as females were shown to have an impaired ability to store muscle glycogen. However, differences were likely due to variable CHO intakes between conditions as opposed to biological sex (6.4 vs. $8.2\text{ g}\cdot\text{kg}^{-1}\cdot\text{day}^{-1}$ CHO for females and males, respectively). When relative or absolute CHO intake ($\text{g}\cdot\text{kg}^{-1}\cdot\text{day}^{-1}$ or $\text{g}\cdot\text{day}^{-1}$, respectively) was similar between males and females, either within the same study [31, 38, 53] or between trials [15, 50, 77], females had the same capacity for muscle

glycogen storage (Table 1), suggesting CHO guidelines are applicable to both sexes. However, effects of female menstrual cycle phase on muscle glycogen storage are inconclusive, as some reports suggest greater glycogen stores for females in the mid-luteal compared to mid-follicular phase [15, 77]. However, when CHO intake was 8–10 g·kg⁻¹·day⁻¹, no differences were apparent between menstrual cycle phases [15, 38]. These mixed results justify further investigation; however, the practical relevance is debatable due to alignment of competition days and menstrual cycle phase being beyond the control of female athletes.

A limitation of the quantitative analysis is the observational nature of subgroup analyses [19]; however, caution has been suggested throughout regarding interpretation of effect estimates, with transparency in terms of covariate distribution (number of pairwise comparisons and participants) and heterogeneity. It could be argued meta-regression may have been more statistically effective in assessing effects of covariates; however, due to the confounded nature of covariates that contributed to heterogeneity in muscle glycogen (CHO intake, loading duration, and training status were not classifiable due to inconsistency across the different levels of these factors), a meta-regression was deemed inappropriate. Another limitation of the current systematic review and meta-analysis is that beyond heterogeneity, no other quality of evidence criteria were assessed (e.g., indirectness, imprecision or publication bias).

This systematic review and meta-analysis highlights a linear dose–response relationship between dietary CHO intake and muscle glycogen, with the saturation point remaining unclear due to a lack of information at higher CHO intakes (≥ 12 g·kg⁻¹·day⁻¹). A multifaceted heterogeneity exists, as the quantity of CHO ingested (expressed as g·kg⁻¹·day⁻¹ or g·day⁻¹), loading duration, training status, and exercise (pre- and during the loading period) were all effectors of glycogen concentration. Therefore, achieving an optimization of pre-competition glycogen stores is a balancing act of what is optimal for glycogen synthesis and what is practical for athletes to implement in the real world, which is achievable with different combinations of these key effectors. Findings reveal a hierarchy, as dietary CHO intake (> 8 g·kg⁻¹·day⁻¹ for 36–48 h) was the most important factor to significantly increase muscle glycogen stores. Despite specific depleting exercise pre-loading not being a pre-requisite for endurance trained individuals, athletes should strategically implement loading strategies around their training sessions pre-competition to efficiently maximize glycogen stores. However, significant heterogeneity in past research introduces uncertainty in these recommendations, and further well-controlled studies are required to determine the true optimal pre-exercise CHO intake to maximize glycogen concentrations, and what effect this has on endurance exercise performance.

5 | Perspectives

Future research should aim to discover the upper limit of CHO ingestion during loading with intakes ≥ 12 g·kg⁻¹·day⁻¹ to identify the saturation point for both muscle glycogen and gastrointestinal tolerance in endurance trained individuals. The descriptive element of this systematic review collates the characteristics of existing CHO loading research, which identifies key trends and

potential avenues for new research within this topic. Examples include the limited replication of athlete pre-competition training practices and poor representation of females and master athletes (no study groups aged > 37 years old). As such, the effects of menstrual cycle and age on the ability to achieve glycogen supercompensation remain unclear. It should be noted that this review and meta-analysis focused on “optimizing” CHO loading in the context of maximizing muscle glycogen stores; however, in many sports (e.g., team sports) and some endurance activities (e.g., race-walking < 20 km), this is unnecessary, as adequate fuelling through the normalization of muscle glycogen stores can be achieved with intakes from 6 to 10 g·kg⁻¹·day⁻¹.

Acknowledgments

Authors would like to thank Dr. Theodoros Bampouras for his support and guidance with statistical methods and analyses.

Funding

The authors have nothing to report.

Conflicts of Interest

The 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.

References

1. J. A. Romijn, E. F. Coyle, L. S. Sidossis, et al., “Regulation of Endogenous Fat and Carbohydrate Metabolism in Relation to Exercise Intensity and Duration,” *American Journal of Physiology* 265 (1993): E380–E391.
2. M. J. Arkinstall, C. R. Bruce, S. A. Clark, C. A. Rickards, L. M. Burke, and J. A. Hawley, “Regulation of Fuel Metabolism by Preexercise Muscle Glycogen Content and Exercise Intensity,” *Journal of Applied Physiology* 97, no. 6 (2004): 2275–2283.
3. J. Bergstrom, L. Hermansen, E. Hultman, and B. Saltin, “Diet, Muscle Glycogen and Physical Performance,” *Acta Physiologica Scandinavica* 71, no. 2 (1967): 140–150.
4. E. F. Coyle, A. R. Coggan, M. K. Hemmert, and J. L. Ivy, “Muscle Glycogen Utilization During Prolonged Strenuous Exercise When Fed Carbohydrate,” *Journal of Applied Physiology* 61, no. 1 (1986): 165–172.
5. M. A. Hearris, K. M. Hammond, J. M. Fell, and J. P. Morton, “Regulation of Muscle Glycogen Metabolism During Exercise: Implications for Endurance Performance and Training Adaptations,” *Nutrients* 10, no. 3 (2018): 298.
6. J. Bergstrom and E. Hultman, “Muscle Glycogen Synthesis After Exercise: An Enhancing Factor Localized to the Muscle Cells in Man,” *Nature* 210, no. 5033 (1966): 309–310.
7. J. Karlsson and B. Saltin, “Diet, Muscle Glycogen, and Endurance Performance,” *Journal of Applied Physiology* 31, no. 2 (1971): 203–206.
8. L. M. Burke, L. J. C. van Loon, and J. A. Hawley, “Postexercise Muscle Glycogen Resynthesis in Humans,” *Journal of Applied Physiology* 122, no. 5 (2017): 1055–1067.
9. V. A. Bussau, T. J. Fairchild, A. Rao, P. Steele, and P. A. Fournier, “Carbohydrate Loading in Human Muscle: An Improved 1 Day Protocol,” *European Journal of Applied Physiology* 87, no. 3 (2002): 290–295.

10. W. M. Sherman, D. L. Costill, W. J. Fink, and J. M. Miller, "Effect of Exercise-Diet Manipulation on Muscle Glycogen and Its Subsequent Utilization During Performance," *International Journal of Sports Medicine* 2, no. 2 (1981): 114–118.
11. D. T. Thomas, K. A. Erdman, and L. M. Burke, "American College of Sports Medicine Joint Position Statement. Nutrition and Athletic Performance," *Medicine and Science in Sports and Exercise* 48, no. 3 (2016): 543–568.
12. K. Solem, M. Clauss, and J. Jensen, "Glycogen Supercompensation in Skeletal Muscle After Cycling or Running Followed by a High Carbohydrate Intake the Following Days: A Systematic Review and Meta-Analysis," *Frontiers in Physiology* 16 (2025): 1620943.
13. P. C. Blom, D. L. Costill, and N. K. Vollestad, "Exhaustive Running: Inappropriate as a Stimulus of Muscle Glycogen Supercompensation," *Medicine and Science in Sports and Exercise* 19, no. 4 (1987): 398–403.
14. S. A. Kavouras, J. P. Troup, and J. R. Berning, "The Influence of Low Versus High Carbohydrate Diet on a 45-Min Strenuous Cycling Exercise," *International Journal of Sport Nutrition and Exercise Metabolism* 14, no. 1 (2004): 62–72.
15. R. T. McLay, C. D. Thomson, S. M. Williams, and N. J. Rehrer, "Carbohydrate Loading and Female Endurance Athletes: Effect of Menstrual-Cycle Phase," *International Journal of Sport Nutrition and Exercise Metabolism* 17, no. 2 (2007): 189–205.
16. D. L. Costill, W. M. Sherman, W. J. Fink, C. Maresh, M. Witten, and J. M. Miller, "The Role of Dietary Carbohydrates in Muscle Glycogen Resynthesis After Strenuous Running," *American Journal of Clinical Nutrition* 34, no. 9 (1981): 1831–1836.
17. R. C. Hickner, J. S. Fisher, P. A. Hansen, et al., "Muscle Glycogen Accumulation After Endurance Exercise in Trained and Untrained Individuals," *Journal of Applied Physiology* 83, no. 3 (1997): 897–903.
18. J. S. Greiwe, R. C. Hickner, P. A. Hansen, S. B. Racette, M. M. Chen, and J. O. Holloszy, "Effects of Endurance Exercise Training on Muscle Glycogen Accumulation in Humans," *Journal of Applied Physiology* 87, no. 1 (1999): 222–226.
19. J. P. T. T. J. Higgins, J. Chandler, M. Cumpston, T. Li, M. J. Page, and V. A. Welch, eds., *Cochrane Handbook for Systematic Reviews of Interventions Version 6.5 (Updated August 2024)* (Cochrane, 2024), www.training.cochrane.org/handbook.
20. M. J. Page, J. E. McKenzie, P. M. Bossuyt, et al., "The PRISMA 2020 Statement: An Updated Guideline for Reporting Systematic Reviews," *BMJ* 29, no. 372 (2021): n71.
21. K. De Pauw, B. Roelands, S. S. Cheung, B. de Geus, G. Rietjens, and R. Meeusen, "Guidelines to Classify Subject Groups in Sport-Science Research," *International Journal of Sports Physiology and Performance* 8, no. 2 (2013): 111–122.
22. L. Decroix, K. De Pauw, C. Foster, and R. Meeusen, "Guidelines to Classify Female Subject Groups in Sport-Science Research," *International Journal of Sports Physiology and Performance* 11, no. 2 (2016): 204–213.
23. J. R. Day, H. B. Rossiter, E. M. Coats, A. Skasick, and B. J. Whipp, "The Maximally Attainable $\dot{V}O_2$ During Exercise in Humans: The Peak vs. Maximum Issue," *Journal of Applied Physiology* 95, no. 5 (2003): 1901–1907.
24. R. Jensen, N. Ortenblad, M. H. Stausholm, et al., "Heterogeneity in Subcellular Muscle Glycogen Utilisation During Exercise Impacts Endurance Capacity in Men," *Journal of Physiology* 598, no. 19 (2020): 4271–4292.
25. K. B. Adamo, M. A. Tarnopolsky, and T. E. Graham, "Dietary Carbohydrate and Postexercise Synthesis of Proglycogen and Macroglycogen in Human Skeletal Muscle," *American Journal of Physiology* 275, no. 2 (1998): E229–E234.
26. D. A. Arnall, A. G. Nelson, J. Quigley, S. Lex, T. Dehart, and P. Fortune, "Supercompensated Glycogen Loads Persist 5 Days in Resting Trained Cyclists," *European Journal of Applied Physiology* 99, no. 3 (2007): 251–256.
27. W. J. Bradley, M. P. Hannon, V. Benford, et al., "Metabolic Demands and Replenishment of Muscle Glycogen After a Rugby League Match Simulation Protocol," *Journal of Science and Medicine in Sport* 20, no. 9 (2017): 878–883.
28. M. G. Flynn, D. L. Costill, J. A. Hawley, et al., "Influence of Selected Carbohydrate Drinks on Cycling Performance and Glycogen Use," *Medicine and Science in Sports and Exercise* 19, no. 1 (1987): 37–40.
29. H. Galbo, J. J. Holst, and N. J. Christensen, "The Effect of Different Diets and of Insulin on the Hormonal Response to Prolonged Exercise," *Acta Physiologica Scandinavica* 107, no. 1 (1979): 19–32.
30. K. M. Hill, C. G. Stathis, E. Grinfeld, A. Hayes, and A. J. McAinch, "Co-Ingestion of Carbohydrate and Whey Protein Isolates Enhance PGC-1 α mRNA Expression: A Randomised, Single Blind, Cross Over Study," *Journal of the International Society of Sports Nutrition* 10, no. 1 (2013): 8.
31. S. G. Impey, E. Jevons, G. Mees, et al., "Glycogen Utilization During Running: Intensity, Sex, and Muscle-Specific Responses," *Medicine and Science in Sports and Exercise* 52, no. 9 (2020): 1966–1975.
32. T. A. Duhamel, J. G. Perco, and H. J. Green, "Manipulation of Dietary Carbohydrates After Prolonged Effort Modifies Muscle Sarcoplasmic Reticulum Responses in Exercising Males," *American Journal of Physiology. Regulatory, Integrative and Comparative Physiology* 291, no. 4 (2006): R1100–R1110.
33. R. G. Kochan, D. R. Lamb, S. A. Lutz, C. V. Perrill, E. M. Reimann, and K. K. Schlender, "Glycogen Synthase Activation in Human Skeletal Muscle: Effects of Diet and Exercise," *American Journal of Physiology* 236, no. 6 (1979): E660–E666.
34. P. A. Roberts, J. Fox, N. Peirce, S. W. Jones, A. Casey, and P. L. Greenhaff, "Creatine Ingestion Augments Dietary Carbohydrate Mediated Muscle Glycogen Supercompensation During the Initial 24 h of Recovery Following Prolonged Exhaustive Exercise in Humans," *Amino Acids* 48, no. 8 (2016): 1831–1842.
35. W. M. Sherman, D. L. Costill, W. J. Fink, F. C. Hagerman, L. E. Armstrong, and T. F. Murray, "Effect of a 42.2-km Footrace and Subsequent Rest or Exercise on Muscle Glycogen and Enzymes," *Journal of Applied Physiology: Respiratory, Environmental and Exercise Physiology* 55, no. 4 (1983): 1219–1224.
36. L. J. Tarnopolsky, J. D. MacDougall, S. A. Atkinson, M. A. Tarnopolsky, and J. R. Sutton, "Gender Differences in Substrate for Endurance Exercise," *Journal of Applied Physiology* 68, no. 1 (1990): 302–308.
37. G. M. Fogelholm, H. O. Tikkanen, H. K. Naveri, L. S. Naveri, and M. H. Harkonen, "Carbohydrate Loading in Practice: High Muscle Glycogen Concentration Is Not Certain," *British Journal of Sports Medicine* 25, no. 1 (1991): 41–44.
38. A. P. James, M. Lorraine, D. Cullen, et al., "Muscle Glycogen Supercompensation: Absence of a Gender-Related Difference," *European Journal of Applied Physiology* 85, no. 6 (2001): 533–538.
39. T. J. Fairchild, S. Fletcher, P. Steele, C. Goodman, B. Dawson, and P. A. Fournier, "Rapid Carbohydrate Loading After a Short Bout of Near Maximal-Intensity Exercise," *Medicine and Science in Sports and Exercise* 34, no. 6 (2002): 980–986.
40. W. J. Bradley, J. C. Morehen, J. Haigh, et al., "Muscle Glycogen Utilisation During Rugby Match Play: Effects of Pre-Game Carbohydrate," *Journal of Science and Medicine in Sport* 19, no. 12 (2016): 1033–1038.
41. J. M. Fell, M. A. Hearris, D. G. Ellis, et al., "Carbohydrate Improves Exercise Capacity but Does Not Affect Subcellular Lipid Droplet

- Morphology, AMPK and p53 Signalling in Human Skeletal Muscle," *Journal of Physiology* 599, no. 11 (2021): 2823–2849.
42. J. A. Hawley, G. S. Palmer, and T. D. Noakes, "Effects of 3 Days of Carbohydrate Supplementation on Muscle Glycogen Content and Utilisation During a 1-h Cycling Performance," *European Journal of Applied Physiology and Occupational Physiology* 75, no. 5 (1997): 407–412.
 43. J. L. Areta and W. G. Hopkins, "Skeletal Muscle Glycogen Content at Rest and During Endurance Exercise in Humans: A Meta-Analysis," *Sports Medicine* 48, no. 9 (2018): 2091–2102.
 44. D. L. Costill, D. R. Pearson, and W. J. Fink, "Impaired Muscle Glycogen Storage After Muscle Biopsy," *Journal of Applied Physiology* 64, no. 5 (1988): 2245–2248.
 45. K. A. Tomcik, D. M. Camera, J. L. Bone, et al., "Effects of Creatine and Carbohydrate Loading on Cycling Time Trial Performance," *Medicine and Science in Sports and Exercise* 50, no. 1 (2018): 141–150.
 46. P. McInerney, S. J. Lessard, L. M. Burke, et al., "Failure to Repeatedly Supercompensate Muscle Glycogen Stores in Highly Trained Men," *Medicine and Science in Sports and Exercise* 37, no. 3 (2005): 404–411.
 47. S. B. Morris and R. P. DeShon, "Combining Effect Size Estimates in Meta-Analysis With Repeated Measures and Independent-Groups Designs," *Psychological Methods* 7, no. 1 (2002): 105–125.
 48. D. R. Elbourne, D. G. Altman, J. P. Higgins, F. Curtin, H. V. Worthington, and A. Vail, "Meta-Analyses Involving Cross-Over Trials: Methodological Issues," *International Journal of Epidemiology* 31, no. 1 (2002): 140–149.
 49. L. H. Rauch, I. Rodger, G. R. Wilson, et al., "The Effects of Carbohydrate Loading on Muscle Glycogen Content and Cycling Performance," *International Journal of Sport Nutrition* 5, no. 1 (1995): 25–36.
 50. J. L. Walker, G. J. Heigenhauser, E. Hultman, and L. L. Spriet, "Dietary Carbohydrate, Muscle Glycogen Content, and Endurance Performance in Well-Trained Women," *Journal of Applied Physiology* 88, no. 6 (2000): 2151–2158.
 51. R. O. Jones, J. L. Areta, S. Bennett, J. Pugh, and J. B. Louis, "Dose-Response of Dietary Carbohydrate Intake on Skeletal Muscle Glycogen, Gastrointestinal Comfort and Body Composition in Endurance-Trained Individuals in Simulated Preparation for Competition," *Scandinavian Journal of Medicine & Science in Sports* 36, no. 6 (2026): e70312.
 52. C. T. Schytz, N. Ortenblad, T. A. Birkholm, et al., "Lowered Muscle Glycogen Reduces Body Mass With no Effect on Short-Term Exercise Performance in Men," *Scandinavian Journal of Medicine & Science in Sports* 33, no. 7 (2023): 1054–1071.
 53. M. A. Tarnopolsky, C. Zawada, L. B. Richmond, et al., "Gender Differences in Carbohydrate Loading Are Related to Energy Intake," *Journal of Applied Physiology* 91, no. 1 (2001): 225–230.
 54. M. A. Tarnopolsky, S. A. Atkinson, S. M. Phillips, and J. D. MacDougall, "Carbohydrate Loading and Metabolism During Exercise in Men and Women," *Journal of Applied Physiology* 78, no. 4 (1995): 1360–1368.
 55. J. A. C. Sterne, J. Savovic, M. J. Page, et al., "RoB 2: A Revised Tool for Assessing Risk of Bias in Randomised Trials," *BMJ* 366 (2019): 14898.
 56. J. A. Sterne, M. A. Hernan, B. C. Reeves, et al., "ROBINS-I: A Tool for Assessing Risk of Bias in Non-Randomised Studies of Interventions," *BMJ* 355 (2016): i4919.
 57. L. M. Burke, B. Kiens, and J. L. Ivy, "Carbohydrates and Fat for Training and Recovery," *Journal of Sports Sciences* 22, no. 1 (2004): 15–30.
 58. M. Hargreaves, J. A. Hawley, and A. Jeukendrup, "Pre-Exercise Carbohydrate and Fat Ingestion: Effects on Metabolism and Performance," *Journal of Sports Sciences* 22, no. 1 (2004): 31–38.
 59. L. M. Burke, J. A. Hawley, E. J. Schabort, A. St Clair Gibson, I. Mujika, and T. D. Noakes, "Carbohydrate Loading Failed to Improve 100-km Cycling Performance in a Placebo-Controlled Trial," *Journal of Applied Physiology* 88, no. 4 (2000): 1284–1290.
 60. A. Foskett, C. Williams, L. Boobis, and K. Tsintzas, "Carbohydrate Availability and Muscle Energy Metabolism During Intermittent Running," *Medicine and Science in Sports and Exercise* 40, no. 1 (2008): 96–103.
 61. L. A. Jones, L. Leung, J. S. Barrett, H. O. Fortis, J. A. Strauss, and S. O. Shepherd, "New Zealand Blackcurrant Extract Augments Muscle Glycogen Storage at Rest and Enhances Intramuscular Triglyceride Degradation During Prolonged Exercise," *European Journal of Applied Physiology* 126, no. 3 (2026): 1857–1872.
 62. C. Akermark, I. Jacobs, M. Rasmusson, and J. Karlsson, "Diet and Muscle Glycogen Concentration in Relation to Physical Performance in Swedish Elite Ice Hockey Players," *International Journal of Sport Nutrition* 6, no. 3 (1996): 272–284.
 63. A. N. Bosch, S. M. Weltan, S. C. Dennis, and T. D. Noakes, "Fuel Substrate Kinetics of Carbohydrate Loading Differs From That of Carbohydrate Ingestion During Prolonged Exercise," *Metabolism* 45, no. 4 (1996): 415–423.
 64. L. M. Burke, G. R. Collier, and M. Hargreaves, "Muscle Glycogen Storage After Prolonged Exercise: Effect of the Glycemic Index of Carbohydrate Feedings," *Journal of Applied Physiology* 75, no. 2 (1993): 1019–1023.
 65. L. M. Burke, G. R. Collier, S. K. Beasley, et al., "Effect of Coingestion of Fat and Protein With Carbohydrate Feedings on Muscle Glycogen Storage," *Journal of Applied Physiology* 78, no. 6 (1995): 2187–2192.
 66. L. M. Burke, G. R. Collier, P. G. Davis, P. A. Fricker, A. J. Sanigorski, and M. Hargreaves, "Muscle Glycogen Storage After Prolonged Exercise: Effect of the Frequency of Carbohydrate Feedings," *American Journal of Clinical Nutrition* 64, no. 1 (1996): 115–119.
 67. K. De Bock, W. Derave, M. Ramaekers, E. A. Richter, and P. Hespel, "Fiber Type-Specific Muscle Glycogen Sparing due to Carbohydrate Intake Before and During Exercise," *Journal of Applied Physiology* 102, no. 1 (2007): 183–188.
 68. T. M. Doering, G. R. Cox, J. L. Areta, and V. G. Coffey, "Repeated Muscle Glycogen Supercompensation With Four Days' Recovery Between Exhaustive Exercise," *Journal of Science and Medicine in Sport* 22, no. 8 (2019): 907–911.
 69. H. W. Goforth, Jr., D. A. Arnall, B. L. Bennett, and P. G. Law, "Persistence of Supercompensated Muscle Glycogen in Trained Subjects After Carbohydrate Loading," *Journal of Applied Physiology* 82, no. 1 (1997): 342–347.
 70. J. W. Helge, P. W. Watt, E. A. Richter, M. J. Rennie, and B. Kiens, "Partial Restoration of Dietary Fat Induced Metabolic Adaptations to Training by 7 Days of Carbohydrate Diet," *Journal of Applied Physiology* 93, no. 5 (2002): 1797–1805.
 71. E. Jansson and L. Kaijser, "Effect of Diet on Muscle Glycogen and Blood Glucose Utilization During a Short-Term Exercise in Man," *Acta Physiologica Scandinavica* 115, no. 3 (1982): 341–347.
 72. J. P. Kirwan, D. L. Costill, J. B. Mitchell, et al., "Carbohydrate Balance in Competitive Runners During Successive Days of Intense Training," *Journal of Applied Physiology* 65, no. 6 (1988): 2601–2606.
 73. D. R. Lamb, A. C. Snyder, and T. S. Baur, "Muscle Glycogen Loading With a Liquid Carbohydrate Supplement," *International Journal of Sport Nutrition* 1, no. 1 (1991): 52–60.
 74. J. D. MacDougall, G. R. Ward, and J. R. Sutton, "Muscle Glycogen Repletion After High-Intensity Intermittent Exercise," *Journal of Applied Physiology: Respiratory, Environmental and Exercise Physiology* 42, no. 2 (1977): 129–132.
 75. E. Maund, H. E. Bradley, C. S. Deane, et al., "Effects of Short-Term Graded Dietary Carbohydrate Intake on Intramuscular and Whole Body Metabolism During Moderate-Intensity Exercise," *Journal of Applied Physiology* 131, no. 1 (2021): 376–387.

76. A. G. Nelson, D. A. Arnall, J. Kokkonen, R. Day, and J. Evans, "Muscle Glycogen Supercompensation Is Enhanced by Prior Creatine Supplementation," *Medicine and Science in Sports and Exercise* 33, no. 7 (2001): 1096–1100.
77. B. J. Nicklas, A. C. Hackney, and R. L. Sharp, "The Menstrual Cycle and Exercise: Performance, Muscle Glycogen, and Substrate Responses," *International Journal of Sports Medicine* 10, no. 4 (1989): 264–269.
78. G. S. Palmer, L. B. Borghouts, T. D. Noakes, and J. A. Hawley, "Metabolic and Performance Responses to Constant-Load vs. Variable-Intensity Exercise in Trained Cyclists," *Journal of Applied Physiology* 87, no. 3 (1999): 1186–1196.
79. H. Sasaki, N. Hotta, and T. Ishiko, "Comparison of Sympatho-Adrenal Activity During Endurance Exercise Performed Under High- and Low-Carbohydrate Diet Conditions," *Journal of Sports Medicine and Physical Fitness* 31, no. 3 (1991): 407–412.
80. K. Vandenbergh, P. Hespel, B. Vanden Eynde, R. Lysens, and E. A. Richter, "No Effect of Glycogen Level on Glycogen Metabolism During High Intensity Exercise," *Medicine and Science in Sports and Exercise* 27, no. 9 (1995): 1278–1283.
81. J. J. Widrick, D. L. Costill, W. J. Fink, M. S. Hickey, G. K. McConell, and H. Tanaka, "Carbohydrate Feedings and Exercise Performance: Effect of Initial Muscle Glycogen Concentration," *Journal of Applied Physiology* 74, no. 6 (1993): 2998–3005.
82. P. C. Blom, A. T. Hostmark, O. Vaage, K. R. Kardel, and S. Maehlum, "Effect of Different Post-Exercise Sugar Diets on the Rate of Muscle Glycogen Synthesis," *Medicine and Science in Sports and Exercise* 19, no. 5 (1987): 491–496.
83. D. J. Jenkins, T. M. Wolever, R. H. Taylor, et al., "Glycemic Index of Foods: A Physiological Basis for Carbohydrate Exchange," *American Journal of Clinical Nutrition* 34, no. 3 (1981): 362–366.
84. R. Jentjens and A. Jeukendrup, "Determinants of Post-Exercise Glycogen Synthesis During Short-Term Recovery," *Sports Medicine* 33, no. 2 (2003): 117–144.
85. R. Jensen, N. Ortenblad, M. H. Stausholm, et al., "Glycogen Supercompensation Is due to Increased Number, Not Size, of Glycogen Particles in Human Skeletal Muscle," *Experimental Physiology* 106, no. 5 (2021): 1272–1284.
86. J. Bergstrom, E. Hultman, and A. E. Roch-Norlund, "Muscle Glycogen Synthetase in Normal Subjects. Basal Values, Effect of Glycogen Depletion by Exercise and of a Carbohydrate-Rich Diet Following Exercise," *Scandinavian Journal of Clinical and Laboratory Investigation* 29, no. 2 (1972): 231–236.
87. I. Mujika and S. Padilla, "Detraining: Loss of Training-Induced Physiological and Performance Adaptations. Part II: Long Term Insufficient Training Stimulus," *Sports Medicine* 30, no. 3 (2000): 145–154.

Supporting Information

Additional supporting information can be found online in the Supporting Information section. **Table S1:** Sensitivity analysis to assess goodness of fit of nonlinear regression models used to describe the relationship between dietary CHO intake and muscle glycogen using studies included within systematic review descriptive synthesis. **Table S2:** Linear and nonlinear sensitivity analysis to describe the relationship between dietary CHO intake and muscle glycogen, using only independent observations. **Table S3:** Sensitivity analysis summary statistics for multiple vs. combined pairwise comparisons within meta-analysis which quantified the relationship between dietary CHO intake and muscle glycogen. Included studies reported exercise status and CHO intake for at least 24h pre-muscle biopsy. **Table S4:** Sensitivity analysis summary statistics for fixed versus random effects meta-analytic models using imputations with different borrowed correlation coefficients. Meta-analysis quantified the relationship between dietary CHO intake

and muscle glycogen using studies which reported exercise status and CHO intake for at least 24h pre-muscle biopsy.