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Exercise, Physical Activity, and Dietary Protein in Relation to Gestational Diabetes and Preeclampsia: A Literature Review and Meta-Analysis

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Abstract

Background/Objectives: We re-estimated effects of prenatal exercise on gestational diabetes mellitus (GDM) and preeclampsia, and cohort associations of activity and protein intake with GDM. **Methods:** This reanalysis (not a systematic review) pooled, once each, primary studies of 13 meta-analyses, partially updated. Unvalidated artificial intelligence (AI) replaced registered dual human review, with human re-screening of two update searches (κ 0.86; 1 record missed by the AI). The 7160 registered-search records were unscreened; registration was retrospective. Random-effects models pooled trials as risk ratios, cohorts within one measure. **Results:** Main analyses included 106 studies (85 trials; 21 cohorts). The evidence is very uncertain about the effect of exercise on GDM (41 trials; risk ratio 0.77, 95% CI 0.66–0.90) and preeclampsia (16 trials; 0.63, 0.39–1.02). Post hoc cohort odds ratios (highest vs. lowest): pre-pregnancy activity 0.91 (0.71–1.15; 11 cohorts); total protein before pregnancy 1.24 (0.80–1.94; 3 cohorts), at the earliest in-pregnancy assessment 1.70 (0.76–3.78; 4 cohorts), latest 1.86 (1.03–3.36; 5 cohorts). Certainty was very low throughout. **Conclusions:** Trials alone cannot support exercise guidance; cohort associations, open to confounding and reverse causation, support no advice on activity or protein. These results describe selected evidence, not all eligible studies. Registration: osf.io/9nhj8; parent review PROSPERO CRD420261486377.

Keywords: gestational diabetes mellitus; preeclampsia; exercise; physical activity; dietary protein; body mass index; pregnancy; GRADE; meta-analysis; reanalysis

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1. Introduction

Gestational diabetes mellitus (GDM) affects about one pregnancy in seven worldwide [1], under International Association of Diabetes and Pregnancy Study Groups (IADPSG) criteria [2]. Hypertensive disorders of pregnancy are a leading cause of maternal death [3], and preeclampsia has no curative treatment other than delivery [4]. Physical activity and diet are modifiable exposures for either condition, and both can act before conception [5].

Randomised trials and an individual-participant-data (IPD) meta-analysis indicate that lifestyle interventions in pregnancy, and physical activity in particular, reduce GDM [6]. Observational meta-analyses associate higher protein intake, particularly animal protein, with a higher risk of GDM [7–9]. These syntheses share three problems. First, they overlap: the same trials appear in several meta-analyses, so their agreement is not independent confirmation [10,11]. Second, they mix exposure windows: intake measured before conception, in early pregnancy and close to GDM screening is often pooled in one overall estimate [8,9]. Third, they mix effect measures and outcome definitions. Odds ratios and risk ratios are combined although GDM is not rare, and the IPD meta-analysis estimated a smaller effect under National Institute for Health and Care Excellence (NICE) criteria than under IADPSG criteria [6].

Body mass index (BMI) complicates both bodies of evidence, because baseline GDM risk, habitual activity and diet may all differ by BMI. In trials, BMI may modify the effect of exercise; in cohorts, it may confound the association of activity or protein intake with GDM and, for activity, partly mediate it (Sections 3.6 and 4.4).

We therefore pooled the primary studies of the published meta-analyses so that each study counted once, kept randomised and observational evidence apart, used one effect measure for the principal trial analysis, pooled cohort estimates only within one effect measure, stated the assessment window of each cohort estimate, and examined BMI post hoc. GDM and preeclampsia were registered outcomes of the parent review from which this work derives; the observational analyses (the cohort layers; Section 2.2) are post hoc.

2. Materials and Methods

2.1. Design, Registration and Deviations

This article reports a reanalysis and partial update of the primary studies included in published meta-analyses. It is not a systematic review. Most studies were identified through these meta-analyses (the sources, Section 2.2); the records of the registered search were never screened (below). Update searches covered only some evidence layers (Section 2.2); their records were screened by artificial intelligence (AI) and, in revision 2, those of two of the three searches also by an author (Section 2.4). The work derives from a parent review registered in PROSPERO (CRD420261486377) on 12 September 2026, after its searches of 23 August 2026. These searches retrieved 11 of the 13 source meta-analyses. For GDM, preliminary pooled estimates preceded the PROSPERO record (below).

The PROSPERO record (version 1.1) lists screening and extraction as completed. However, the 7160 records that the registered search retrieved were never formally screened. Before registration, a script had sorted these records into priority lanes by title and abstract. Also before registration, effect estimates had been extracted from abstracts and four preliminary pools had been computed, all for GDM (carbohydrate, protein and physical activity on two contrasts) and none for preeclampsia. No record was removed, and no screening decision is recorded for any record (Table S16, item (cc)). The record carries the title of another team's review (CRD420251001515), and its exposure field does not match this reanalysis.

The reanalysis itself was registered on the Open Science Framework (OSF; https://osf.io/9nhj8/) on 12 October 2026. This registration was retrospective, made after searching, selection, extraction and analysis, and it contains statements that this article corrects (Table S16, item (dd)); these were corrected in updates of the registration approved on 11 and 20 November 2026. Table S16 lists the deviations identified from the PROSPERO record and protocol. They include the replacement of the registered dual human review by AI. Section 4.7 discusses their implications for the findings. Reporting follows PRISMA 2020 [12] (File S1) and, where applicable, PRISMA-S [13]; Text S1 gives methodological detail.

Terms are used as follows. Prespecified means named as a main outcome in the PROSPERO record and its protocol, which differ in places (Table S16, item (cc)); exploratory means an outcome that was not so named. Registered means stated in that record or its protocol, not in the retrospective OSF registration. Post hoc means an analysis that is not stated in that record or protocol and was defined after data extraction. Section 2.3 gives the status of the outcomes and analyses. A record is a search hit, and a report is a publication. A study is a trial or a cohort, which may have several reports. A row is one study-level estimate as transferred from a source.

2.2. Study Identification and Currency of Each Evidence Layer

Primary studies were identified through 13 published meta-analyses, hereafter referred to as the sources (Table 1). These were an IPD meta-analysis [6], meta-analyses of exercise trials [10,11,14–18], a meta-analysis of pre-pregnancy activity [19], three meta-analyses of protein-intake cohorts [7–9] and a Cochrane review of protein–energy supplementation [20]. An earlier meta-analysis of physical activity before and during pregnancy [21], not one of the 13, served as an additional route to pre-pregnancy cohorts. Eleven of the 13 sources were the physical-activity and protein syntheses cited in an early, unscreened draft of the parent review. Two were added for specific needs: trial-level biomarker data [17], and preeclampsia and gestational hypertension by trial [18]. No eligibility criteria for choosing the sources were recorded, and the sources were not appraised (Table S16, item (z)). Other syntheses retrieved by the searches were not listed, and published results were already available when the sources were chosen. The choice of sources may therefore have introduced selection bias (Text S1, Section S2.3; Table S16, item (gg)).

Because studies were identified through existing meta-analyses, each source's published pooled estimate could be re-computed from the transferred rows before use. This served as a check on the transfer (the acceptance test, Section 2.4). The same design inherits the sources' study selection, including any tendency towards published and positive studies. It cannot show what a screened de novo search would have added.

The update search strategies are given in Table S6. The exercise-trial update searched PubMed, Europe PMC and ClinicalTrials.gov from 1 January 2017 to 5 October 2026. The pre-pregnancy activity cohort update searched PubMed only, from 1 August 2015 to 30 September 2026; because the screening decisions of that run were not recorded, it was rerun on 7 October 2026 (Text S1, Section S2.3). The protein–energy supplementation trial update covered 1 February 2015 to 7 October 2026. Protein-intake cohorts were not searched beyond the sources; the most recent of these searched the literature to August 2025 [9]. In revision 2, two prospective cohorts, found after the results were known among the unscreened records of the parent review's protein search (23 August 2026), were added post hoc to the total-protein pools [22,23] (Section 3.5; Table S16, item (pp)). The evidence layers therefore differ in completeness and currency, and Table 1 states the status of each.

Table 1. Evidence layers: (A) sources, update searches, currency and analysis status; (B) provenance and verification of the data.

(A)

Layer	Sources	Update Search and Databases	Search End Date	Studies Added by the Update	Status
Exercise trials: GDM, preeclampsia	IPD meta-analysis and meta-analyses of exercise trials [6,10,11,14] ([18] in sensitivity analyses only)	From 1 January 2017; PubMed, Europe PMC, ClinicalTrials.gov	5 October 2026	7 trials (4 in the GDM pool, one of these also in the preeclampsia and gestational-hypertension pools; 3 in neonatal pools only)	Prespecified outcomes; analysis rules and effect measure set after data transfer
Exercise trials: gestational hypertension	As above	As above	5 October 2026	As above	Not prespecified; exploratory
Pre-pregnancy activity cohorts	[19] and one cohort cited by the registered review; extended with [21]	From 1 August 2015; PubMed only	30 September 2026 (rerun 7 October 2026)	4 cohorts from the update search (a further 5 from [21])	Post hoc (extended set defined after extraction)
Protein-intake cohorts	[7–9,22,23]	None	Latest source search August 2025	2 cohorts found among the unscreened records of the parent review's search (revision 2; total protein)	Post hoc (windows defined after extraction; the 2 added cohorts after the results were known)
Protein–energy supplementation trials	Cochrane review [20]	From 1 February 2015; PubMed, Europe PMC, ClinicalTrials.gov	7 October 2026	16 trials	Prespecified outcomes: GDM not reported, preeclampsia in two trials; other outcomes exploratory; Text S2
Neonatal outcomes of exercise trials	[16] and the exercise update; [15] in sensitivity analyses only	Exercise update only	Source closed January 2017	From the exercise update	Exploratory; Text S2
Maternal glycaemic biomarkers	[17]	PubMed search for a source	5 October 2026	None	Not prespecified; Text S2

(B)

Layer	Screening and Eligibility	Data Transfer or Extraction	Risk of Bias	Weight Supported by a Primary Report or Abstract	Authors' Check
Exercise trials: GDM, preeclampsia	Sources: as published. Update search: two instances of one AI model (39 records by one instance); disagreements resolved by a third AI assessment. Revision 2: every record re-screened by one author, blinded, with adjudication	Source rows transferred once by AI-written code; update trials extracted twice by two AI instances, quotations checked by script	Sources' published judgements mapped to RoB 2 by the AI; RoB 2 of new trials drafted by the AI (four trials not as-	GDM 43%; preeclampsia 37%	Update-search eligibility and counts, risk of bias and GRADE reviewed unblinded by two authors; transferred rows and the four trials added on 12 October 2026 not checked

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			sessed); not used for certainty (rated very serious)		
Exercise trials: gestational hypertension	As above	As above	As above	11%	As above
Pre-pregnancy activity cohorts	Sources: as published. Update search: two AI instances (re-run of 67 records). Revision 2: re-screened by one author, blinded, with adjudication	Source estimates transferred; update cohorts recorded once and matched to the report by script	Newcastle–Ottawa scores from the sources; 97% of the weight of the OR pool not assessable	41%	Eligibility and Newcastle–Ottawa judgements reviewed; cohort estimates not
Protein-intake cohorts	Sources: as published; no update search; 2 cohorts from the unscreened records of the parent review's search added in revision 2	Transferred from the sources; two estimates replaced from the primary reports; the 2 added cohorts from their primary reports	Newcastle–Ottawa scores from the sources (the 2 added cohorts not assessed); not assessable: 64%, 57% and 42% of the weight of the before-pregnancy, earliest and latest OR pools	100%, 24% and 27%	Newcastle–Ottawa judgements reviewed; transferred estimates not
Neonatal outcomes of exercise trials	As for exercise trials	As for exercise trials	As for exercise trials	Preterm birth 57%; macrosomia 44%; low birthweight 51%	As for exercise trials
Supplementation trials; glycaemic biomarkers	Supplementation update: two AI instances (258 records)	Cochrane and biomarker rows transferred; update trials extracted twice by two AI instances	As published, or drafted by the AI for new trials (Text S2)	Not compared	Update-search decisions reviewed; transferred rows not

Notes: Status: outcome and analysis status as defined in Section 2.1; the registered analyses, the registered analyses not performed as registered and the post hoc analyses are listed in Section 2.3. Study counts: 26 supplementation trials in the main analyses (15 of the 16 from the update search, the sixteenth in sensitivity analyses only; 11 from the Cochrane review). The Nurses' Health Study II is counted once, so 14 activity and 8 protein cohorts make 21 cohorts; one protein cohort without diagnosed GDM is outside the main analyses (Section 2.3). Part B: AI, instances of Claude Opus 5.5 (Section 2.4). Weight supported by a primary report or abstract: the share of the pool's random-effects weight carried by rows confirmed against a primary report or its abstract, taken directly from a report, or later read in one. For the cohort layers, this share refers to the revision-1 pools, which combined effect measures. The authors' check and the independent human screening of revision 2 are described in Section 2.4. The full notes of revision 1 are in Text S1, Section S3.13. BMI, body mass index; GDM, gestational diabetes mellitus; RoB 2, revised Cochrane risk-of-bias tool for randomised trials.

2.3. Eligibility, Outcomes and Analysis Status

Participants were women before conception or during pregnancy, without restriction on age, parity, BMI or setting. Women with pregestational diabetes or chronic hypertension were excluded. Trials tested exercise without a dietary or other lifestyle co-intervention. Exercise covers every intervention pooled as exercise or physical activity by the sources or, for the update search, by this review (Table S3): aerobic, resistance, aquatic and mixed programmes; walking and cycling; yoga; and pelvic-floor and stabilising training, including in women with pelvic girdle pain. Two further trials were retained as exercise trials: one of exercise within smoking-cessation support and one of reduced sedentary behaviour that did not increase steps (Table S16, item (r)). The update search required a usual-care or other non-exercise control. Three trials from the sources compared exercise with another active intervention or with a different dose of exercise; they were kept because their sources had included them (Table S16, item (r)). Cohorts compared the highest with the lowest category of activity or protein intake, for GDM only; preeclampsia was not assessed in the observational layers.

In revision 2, two eligibility criteria were applied strictly. Trials had to be randomised, so one trial included by the sources, which allocated participants by alternation, was excluded from the trial pools (Table S16, item (nn)). The cohort outcome had to be diagnosed GDM, so one protein cohort, which reported a positive glucose challenge test, was excluded from the protein pools (Table S16, item (oo)). Both studies enter sensitivity analyses (Tables S18 and S20).

The status of the outcomes and analyses, in the terms defined in Section 2.1, is as follows. The prespecified outcomes are GDM and preeclampsia, as each study defined them; gestational hypertension was not prespecified and is exploratory. Several choices for the two prespecified trial pools are post hoc, set after the data had been transferred from the sources (Table S16): the rules on which rows to exclude, the removal of duplicate reports of one trial, the modified Hartung–Knapp intervals and the retention of the three comparator trials. The risk ratio was chosen as their single measure after risk-ratio, odds-ratio and combined pools had been computed. Four analyses were registered and performed: trim-and-fill, Egger's test, leave-one-out analysis and the funnel plot. Leave-one-out analysis and the funnel plot were first run after the other analyses had been reported. The pooling of cohort estimates within one measure was also registered; it was first done in revision 2 (Section 2.6; Table S16, items (aa) and (kk)).

The dose–response analysis, primary in the registered protocol, was not done (Table S16, item (aa)). Exposure window and pre-pregnancy BMI category were registered subgroups but were not analysed as registered. The following are post hoc: the extended set of activity cohorts (beyond [19] and one cohort cited by the registered review; Table 1), the three protein windows and the addition of two protein cohorts found among the unscreened records of the parent review's search (revision 2); every study-level BMI analysis, including the BMI strata and the within-cohort BMI comparison; and the small-study methods other than the four registered analyses. The rule of Section 2.5 that rates down risk of bias two levels for every trial pool, and every other sensitivity and subgroup analysis, are also post hoc. In the cohort layers, the outcome (GDM) is therefore prespecified and the within-measure pooling registered, whereas the sets of cohorts and windows analysed are post hoc.

2.4. Data Transfer, Use of AI and Human Checks

Estimates were transferred from the sources at the most granular level available, with event counts preferred to printed ratios. Before any transferred row was used, each source's rows were re-pooled with the source's own method. They had to reproduce its published pooled estimate to two decimals (the acceptance test). A discrepancy traceable to the rounding of printed values was accepted and is reported. A source that failed the test contributed to sensitivity analyses only. The test is consistent with faithful transfer of the rows from each meta-analysis, to the precision of its pooled estimate. It does not show that the meta-analysis had extracted them correctly from the primary study.

Each trial or cohort was entered once. Reports were matched by PubMed identifier and then by registration number or by authors, programme, site and recruitment period. One estimate per cohort and window was taken from the most adjusted model (Text S1, Section S2.5).

A separate AI step, outside the authors' check described below, compared 37 transferred rows from 23 studies with their primary reports. The rows were chosen for influence rather than sampled. In each pool as first transferred, they were the rows that together carried at least half of the weight, plus every row that carried 15% or more. Eleven rows were compared with full texts and the rest with abstracts. Overall, eleven matched, four matched within rounding, one was contradicted and 21 could not be verified from the accessible text. This check, full texts obtained later, and other checks together led to 11 corrections (Table S8). They included event counts that contradicted a trial's abstract, a control denominator, a per-protocol row replaced by intention-to-treat counts, two protein estimates found in neither primary report, and three duplicate rows. Seven known discrepancies remain uncorrected because primary data were not available. Because the checked rows were chosen for influence, their error rate cannot be generalised to the transferred rows that were never compared.

In the GDM pool, rows confirmed against a primary report or its abstract carried 29% of the weight, rows checked but not verifiable 20%, rows never compared 48% and rows taken directly from primary reports 3%; in the preeclampsia pool, the shares were 21%, 38%, 39% and 2%. Rows later read in a primary report raise the share supported by a primary report or abstract to 43% for GDM and 37% for preeclampsia (Section 4.7). Across the protein pools of revision 1, which combined effect measures, rows never compared carried up to 58% of the weight (animal protein, earliest assessment).

Generative AI (Claude Opus 5.5, Anthropic, San Francisco, CA, USA), run in an agent framework, wrote and ran the code for extraction, transfer and analysis. The model and version are as recorded by the authors; they were not logged per step. In the update searches, two instances of the same model screened records, assessed eligibility and extracted trial outcome data (Text S1, Section S2.9). In the exercise search, 37 records already in the source data and 295 reviews, protocols and editorials, removed by publication type or title, were set aside before screening. The 39 records of this search found only in Europe PMC (28) or ClinicalTrials.gov (11) were screened by one instance. Disagreements on eligibility were resolved by a third AI assessment. One instance drafted the risk-of-bias and trustworthiness judgements for most new trials.

The two instances agreed on 98% of the title-and-abstract decisions for the 392 PubMed records of the exercise search (κ 0.94). Of the 73 reports assessed for eligibility in this search, some from the abstract alone where the full text could not be obtained, 72 were assessed by both instances, which agreed on 90.3% of the decisions (κ 0.85). The remaining report, a companion of an excluded trial, was decided by a third AI assessment alone. This agreement measures the consistency of one model, not independent validation. For trials from the update searches, each extracted value except inferred denominators is accompanied by a verbatim quotation from the report, which a script checked against the source

text. The script confirms that the quoted string occurs in the source, not that the correct arm, denominator or analysis population was selected. The estimates of the update cohorts were recorded once, and a script matched each recorded value to its report.

Two authors (V.G.-O., I.G.) reviewed the AI-generated decisions and their rationale on update-search eligibility and counts, risk of bias and GRADE ratings. This step is called the authors' check. The two authors did not make independent decisions from the source texts. This unblinded verification of AI output is open to automation bias [24]. No item-level log is available, so the number of items checked and an agreement statistic cannot be reported. According to a corresponding author's statement (File S2), one of the two raised 9 queries and the other 14, none in common. K.S., who ran the pipeline, adjudicated all 23 queries and retained every decision; V.G.-O. had built the analysis framework. Neither the content of the queries nor the reasons for retaining the decisions were recorded. No check is recorded for the four exercise trials added on 12 October 2026, after the check. The inclusion of one of them and the no-overlap decisions for two others are recorded as confirmed by an author, outside the authors' check. In revision 2, two included trials of one research group with separate registrations [25,26] were found to possibly share participants: a further report of the first [27] and a correction of the second both give 124 women, 70 and 54 per arm. They are pooled together only for preterm birth; without the first, preterm birth gave 1.01 (0.82–1.24) in 30 trials (post hoc; Table S16, item (qq)).

Later AI steps were also outside the authors' check: the BMI classification of the trials, the cohort estimates from models without BMI (Section 2.6), the final GRADE ratings, the ICEMAN judgement of effect modification (Section 2.6) and the quotation of Tables 8 and 9 of the IPD meta-analysis [6] (Text S1, Section S2.9). On 11 November 2026, before resubmission, the same two authors reviewed three of these items and recorded each decision. Each author classified the 42 GDM trials of revision 1 by BMI eligibility from the trial reports, mainly the PubMed abstracts; these were the 41 trials of the revision 2 pool and the quasi-randomised trial (Section 2.3). Both agreed with the AI class of every trial. Both also reviewed the six certainty ratings of the summary-of-findings table of revision 1 and the ICEMAN judgement, with the AI output in view, and agreed with each. The ratings of revision 2 follow the rule of Section 2.5. The certainty ratings of the outcomes outside the summary of findings, the BMI classes of the 10 trials pooled only for preeclampsia or gestational hypertension, the cohort estimates without BMI and the quotation from [6] were not checked.

Independent human screening (revision 2): One author who had not built or run the pipeline (C.D.) screened every record that the AI had screened, blinded to the AI decisions. These were the 431 records of the exercise-trial update search and the 67 records of the activity-cohort update search; the 295 records removed by the publication-type and title filter were offered as an optional set (Text S1, Section S2.9). Screening used the title and abstract or the registry record, against the eligibility criteria of Text S1 (Sections S2.2 and S2.3), and the records were presented in random order. Every record forwarded by the screener or by the AI was then assessed in full text by the screener. Disagreements between the screener's final decision (after full-text assessment where applicable) and the final AI decision were resolved by a second author (K.S., who took part in revising the analyses of revision 2 but did not screen the records), who recorded each decision and its reason (File S2). Agreement is reported as Cohen's κ, and the recall of the AI screening as the proportion of the records eligible by the adjudicated decision that the AI had forwarded, with an exact 95% confidence interval (Text S1, Section S3.14; Table S21). The supplementation update search was not re-screened.

2.5. Risk of Bias and Certainty of the Evidence

Risk-of-bias judgements were obtained in four ways. For studies from the sources, the AI compiled a first-pass table from the sources' published judgements and mapped those of trials to the RoB 2 domains [28] (Figure S1). The derivation of the overall judgement is given in Text S1, Section S2.7. Four trials and one cohort without a published judgement were judged by the AI from the full text. Other cohorts carried the sources' Newcastle–Ottawa scores (7–9 low, 5–6 moderate, 0–4 high risk of bias). The two protein cohorts added in revision 2 (Section 2.2) have no risk-of-bias assessment and count as not assessable. Trials from the update searches were assessed with RoB 2 from their primary reports, as AI drafts reviewed in the authors' check. The four exercise trials added on 12 October 2026 (Section 2.4) have no recorded risk-of-bias assessment or trustworthiness screen. Studies without an accessible or recorded judgement were counted as not assessable. Because exercise cannot be blinded, lack of blinding alone was judged as 'some concerns', a declared deviation from the registered review (Table S16, item (k)). A second AI assessment is deposited in File S2 (Section 4.7). These judgements are reported to describe the trials, but they were not used to set the certainty of trial evidence (below).

Certainty was rated with GRADE [29,30], starting high for randomised trials and low for cohorts. In revision 2, risk of bias was rated down two levels (very serious) for every pool of randomised trials, whatever the risk-of-bias judgements of its trials. The reason is that none of these judgements came from a standardised, human-led assessment, and a second AI assessment agreed poorly with the first (Section 4.7). This rule was set after the results were known, and it leaves every trial outcome at very low certainty. For cohort pools, risk of bias was rated with the coded rule used in revision 1. It was rated down two levels when studies at high risk carried 50% or more of the random-effects weight. It was rated down one level when they carried 25% or more, when studies at high risk or with some concerns (for cohorts, moderate risk) together carried 50% or more, or when studies not assessable carried 50% or more. The revision-1 ratings of the trial pools, which used the same rule, are in File S2. For the latest-assessment protein windows, risk of bias was rated down one level by reasoned override for possible reverse causation (Section 3.5).

The other domains were rated with rules recorded in code after results were available (File S2; Text S1, Section S2.7). Inconsistency was rated down one level at I^2 of 50% or more (three or more studies). It was rated down two levels at I^2 of 75% or more with a prediction interval extending below 0.80 and above 1.25, which no rated ratio pool reached. Indirectness was rated down where the outcome was a proxy or the population did not match the question. Imprecision was rated down two levels where the confidence interval included both a ratio below 0.75 and one above 1.25, and one level where it included no effect together with either. Otherwise, imprecision was rated down one level where three or fewer studies were pooled or, for binary trial outcomes, where the participants fell short of the optimal information size. This is the number of participants that a single adequately powered trial would need to detect a 25% relative reduction at the pooled control risk of the trials with counts (two-sided α 0.05, power 0.80). Publication bias was rated down, with at least ten studies, where a small-study test gave $p < 0.10$: Harbord's or Peters' test for binary trial outcomes and Egger's test for other pools. It was also rated down where the pooled estimate excluded no effect but both the precision-effect test (PET) estimate and the pool of the most precise quarter of the studies included it. This second criterion was first set for GDM after results were available and was later applied to every rated pool. Findings are worded as GRADE informative statements [31]; for preeclampsia, the statement is supplemented by its sensitivity results (Section 3.3).

2.6. Statistical Analysis

Random-effects models (restricted maximum likelihood, REML) were fitted within layer and outcome. They used modified Hartung–Knapp 95% confidence intervals (CIs) [32–35], with 95% prediction intervals for pools of five or more studies [36]. The Hartung–Knapp interval uses a t distribution with $k - 1$ degrees of freedom (k, the number of studies). It can be narrower than the conventional interval when the studies agree more closely than expected by chance, as in 174 of the 211 random-effects pools of the revision-1 analysis (Text S1, Section S2.6). The modification, $q^* = \max(1, q)$, prevents this, so no random-effects interval reported here is narrower than the conventional one. Prediction intervals use $k - 2$ degrees of freedom and nearly coincide with the CI where τ^2 is estimated at zero. Wald-type intervals for pools of three or four studies are in Table S10.

A trial with no event in one arm entered with 0.5 added to each cell, and a trial with no event in either arm was not pooled. Mantel–Haenszel risk ratios without continuity correction (common-effect, with z-based intervals) were computed as a rare-event sensitivity analysis. All intervals are two-sided 95% intervals, and inference rests on the confidence and prediction intervals rather than on significance thresholds. Only the small-study tests are reported with p-values; their threshold for rating down publication bias is given in Section 2.5.

The principal GDM measure was the risk ratio (RR). Trials with event counts contributed RRs computed from their counts. Seven trials had no counts and were available only as odds ratios (ORs) adjusted in the IPD meta-analysis. These were converted to RRs with the Zhang–Yu method [37], using the median control risk of the trials with counts (7.5%). The conversion is not strictly valid for adjusted ORs and may give intervals that are too narrow [38]. Absolute effects and the number needed to treat to prevent one diagnosis (NNT) were computed on the RR scale at the same median control risk.

Cohort estimates were reported as risk ratios (often termed relative risks), ORs or hazard ratios (HRs). In revision 2, the AI recorded the measure of each cohort estimate from its primary report, with the regression model and a verbatim quotation, and an author checked each recorded row against the report (Table S20). An estimate called a relative risk but obtained by logistic regression was classed as an OR. Cohort estimates were pooled only within one measure, and only where at least three cohorts reported it. Other estimates are shown individually, and no estimate was pooled across measures.

Where at least ten studies were pooled, small-study effects were assessed with Egger's, Harbord's and Peters' tests [39,40], PET and the precision-effect estimate with standard error (PEESE) [41], and trim-and-fill [42]. They were also assessed by restriction to the most precise half and quarter of the trials (smallest standard errors). For this restriction, numbers of trials were rounded to the nearest whole number, halves to the even number; the quarter was 10 of 41 trials. For GDM, a leave-one-out analysis (Table S18) and a contour-enhanced funnel plot (Figure S3) were also produced. Section 2.3 gives the registration status of each method.

BMI could not be entered as a participant-level covariate, because the reanalysis uses study-level data; its study-level analogues are post hoc (Section 2.3). Trial pools were stratified by whether a trial enrolled only women with overweight or obesity (restricted trials) or had no such criterion (unrestricted trials). Each stratum was pooled with its own τ^2. Strata were compared as ratios of RRs with Wald 95% CIs, which understate uncertainty, and no p-values were reported. The credibility of any effect modification was judged with ICEMAN [43,44]. For cohorts, estimates with and without BMI in the model were compared within the same cohort where such a model had been extracted.

Analyses used Python 3.11.15 (submitted analyses), 3.13.9 (revision 1) and 3.12.10 (revision 2) (Python Software Foundation, Wilmington, DE, USA). The principal pools

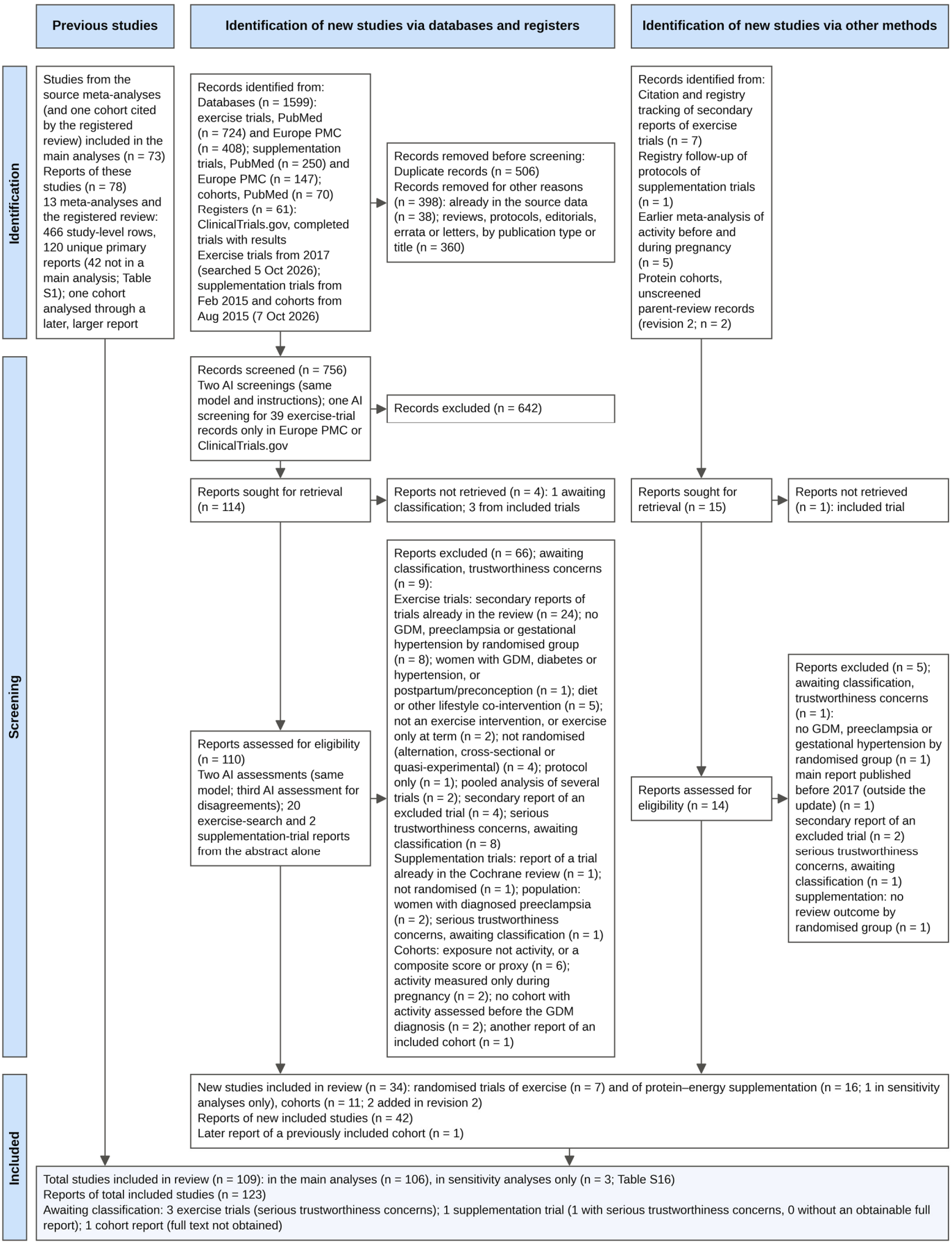
and the within-measure cohort pools of revision 2 were re-computed in R 4.6.1 (R Foundation for Statistical Computing, Vienna, Austria; metafor 5.2.1).

3. Results

3.1. Study Flow and Characteristics

The 13 source meta-analyses contributed 466 study-level rows, which after de-duplication corresponded to 120 unique primary reports (Figure 1; Tables S1–S3) [6–11,14–20]. Every source used in a main analysis passed the acceptance test for the pooled estimates used here; overall estimates that three sources print only in their text could not be reproduced and were not used [8,10,19]. Two other sources failed the test wholly or in part and contribute to sensitivity analyses only [15,18].

Thirty-three studies not in the sources entered the main analyses: 7 exercise and 15 supplementation trials from the update searches (a sixteenth supplementation trial enters sensitivity analyses only; Figure 1), 9 activity cohorts (4 from the update search and 5 from an earlier meta-analysis [21]) and 2 protein cohorts found among the unscreened records of the parent review's search and added post hoc in revision 2 [22,23]. The exercise update search screened 431 records, assessed 73 reports and added seven trials (9 reports) [25,26,45–49]. Three trials with serious trustworthiness concerns await classification [50–52]; four further reports whose full texts could not be obtained belong to included trials, among them [27], which shares its registration (NCT02420288) with an included trial [25], and [53] (Table S13). For the cohort search, the screening decisions of the first run were not recorded; the search was therefore re-run after selection; its 70 records (67 screened twice) yielded the same five reports, with one further report awaiting classification (Tables S13–S15). In the independent human screening of revision 2, the screener and the AI agreed on 96% of the title-and-abstract decisions for the 431 exercise records (κ 0.86) and on 85% of the 67 cohort records (κ 0.53). After full-text assessment and adjudication of 46 disagreements, 15 exercise and 8 cohort records were relevant (eligible studies, further reports of included studies, or studies awaiting classification). The AI had forwarded 100% (15 of 15; 95% CI 78–100%) of the exercise records and 88% (7 of 8; 95% CI 47–100%) of the cohort records, and one relevant record had been excluded by the AI at title and abstract (37826849); it reports the Nurses' Health Study II, which already enters the activity analyses once (Section 3.4), and was not added, because each cohort is counted once (Table S21; Text S1, Section S3.14). For five trials in the main pools, the adjudicated decision differed from the AI's: three were judged not eligible because the report examined gave no arm-specific GDM, preeclampsia or gestational hypertension results (SPRING, the PEACH vascular report and Brik 2019 [26]; for SPRING, the AI had recorded these results by arm from the same supplementary table), and two were judged to await classification (Bacchi 2018 [49] and Barakat 2018 [25]). Three of these five decisions were made from abstracts (PEACH vascular, Bacchi 2018 [49] and Barakat 2018 [25]), whereas the AI had assessed the authors' full texts. The main analyses keep the AI-selected set. With the adjudicated decisions applied (post hoc), GDM gave 0.77 (0.66–0.90) (39 trials), preeclampsia 0.60 (0.37–0.99) (15 trials), gestational hypertension 0.57 (0.39–0.84) (18 trials), preterm birth 1.02 (0.83–1.26) (27 trials), macrosomia 0.71 (0.51–1.01) (15 trials) and low birthweight 0.94 (0.72–1.23) (14 trials) (Table S21); the preeclampsia interval then excluded no effect and the macrosomia interval included it, for outcomes whose certainty is very low. One trial of the sources (quasi-randomised) and one protein cohort (outcome not diagnosed GDM) were excluded in revision 2 (Sections 3.2 and 3.5). Overall, 106 studies entered the main analyses: 85 trials (59 of exercise, 26 of supplementation) and 21 cohorts (14 of pre-pregnancy activity and 8 of protein, with the Nurses' Health Study II (NHS II) counted in both).



Two instances of one AI model screened and assessed (consistency, not validation); two authors reviewed their decisions and rationale unblinded; another author (K.S.) adjudicated (Section 2.4).

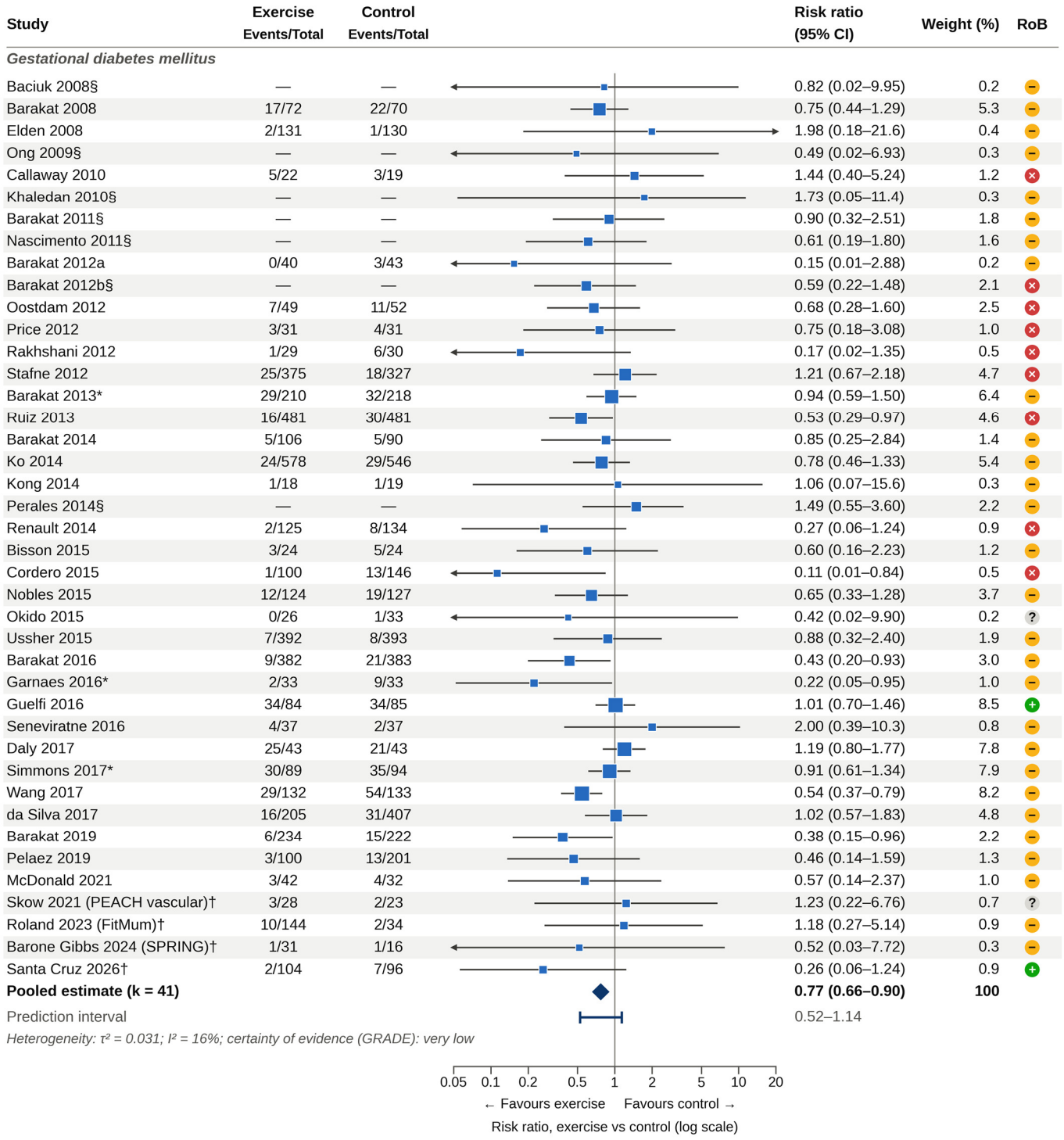
Figure 1. PRISMA 2020 flow diagram for updated reviews. Previous studies: primary studies of the 13 source meta-analyses and one cohort cited by the registered review. New studies: update searches for exercise trials (from January 2017), protein–energy supplementation trials (from February 2015) and pre-pregnancy activity cohorts (from August 2015 to 30 September 2026; re-run 7 October 2026), and other methods (citation and registry tracking, an earlier meta-analysis of physical activity before and during pregnancy and, in revision 2, two protein cohorts found among the unscreened records of the parent review's search). Screening and eligibility were performed by two instances of the same artificial intelligence (AI) model (39 exercise-search records by one instance); in revision 2, one author re-screened the exercise and activity-cohort searches blinded to the AI decisions, with adjudication by a second author (Section 2.4). In revision 2, one quasi-randomised trial and one cohort whose outcome was not diagnosed GDM moved from the main to the sensitivity analyses (Sections 3.2 and 3.5; Table S16, items (nn) and (oo)), and two protein cohorts found among the unscreened records of the parent review's search were added to the main analyses (Section 3.5; Table S16, item (pp)). The 7160 records of the registered search received no eligibility screening; they were sorted by script before registration (Table S16, item (cc)).

Exercise characteristics (frequency, duration, intensity, supervision, adherence, start week) were neither synthesised nor filled in from full texts where the sources left them blank (Table S3). Twelve GDM trials came from one research group in Madrid, hereafter named the Madrid group; several recruited at the same hospitals over overlapping periods and may share participants (Table S9).

3.2. Exercise and GDM

The evidence is very uncertain about the effect of exercise during pregnancy on GDM (very low certainty). The sources and the update search provided 42 trials [45–48,54–93]; one of them [72] allocated participants by alternation of entry order, which makes it quasi-randomised, and in revision 2 it was excluded from the trial pools (with it, 0.75 (0.65–0.88); Table S18). The remaining 41 trials gave an RR of 0.77 (0.66–0.90), prediction interval 0.52–1.14, I^2 16%, τ^2 0.031 (Figure 2; Table 2). The 34 trials with event counts recorded 807 GDM diagnoses among 9373 participants; their median control risk was 75 per 1000 (range 8–488). The optimal information size, 4052 participants at the pooled control risk of these trials (99 per 1000), was met. At the median control risk, the estimate corresponds to 17 fewer (25 to 8 fewer) GDM diagnoses per 1000 women, a number needed to treat of 59 (40 to 120). With every trial as an OR (sensitivity analysis), the estimate was 0.72 (0.60–0.85) (Table S18).

Small-study effects were examined on both scales, and the methods disagreed. On the RR scale, Egger's test gave $p = 0.055$. Two of the five adjusted estimates included no effect: the PET estimate, 0.95 (0.76–1.20), and the pool of the most precise quarter of the trials, 0.86 (0.70–1.07). The other three did not: PEESE 0.84 (0.72–0.97), trim-and-fill 0.81 (0.70–0.95) and the pool of the most precise half of the trials 0.81 (0.68–0.95) (Table S18). On the OR scale, Egger's test gave $p = 0.294$, and the PET estimate, 0.84 (0.61–1.16), again included no effect. Harbord's ($p = 0.31$) and Peters' ($p > 0.99$) tests, which are computed on the OR scale from the 34 trials with event counts, gave no evidence of asymmetry (Figure S3).



* Row corrected (Supplementary Table S8); Barakat 2013 checked against the trial abstract only (full report not obtained).
† Trial found by the update search (Supplementary Table S7).
§ No event counts available: adjusted odds ratio from individual participant data converted to a risk ratio (Zhang–Yu method at the median control risk, 7.5%).
RoB, overall risk-of-bias judgement (RoB 2; Section 2.5): + low; − some concerns; × high; ? not assessable; not used to rate certainty (risk of bias rated very serious for every trial pool, Section 2.5).

Figure 2. Exercise during pregnancy and gestational diabetes mellitus: randomised trials (k = 41), risk ratios. Squares: trial estimates, area proportional to the random-effects weight; diamond: pooled estimate (restricted maximum likelihood (REML), modified Hartung–Knapp 95% CI); bar: 95% prediction interval. § odds ratio adjusted in the individual-participant-data (IPD) meta-analysis, converted to a risk ratio (Zhang–Yu, control risk 7.5%); * row corrected (Supplementary Table S8); Barakat 2013 [70] checked against the trial abstract only; other unmarked rows were checked against abstracts only or not compared with primary reports (Section 2.4; File S2), so absence of a mark does

not indicate verification; Barakat 2016 [82]: full report not obtained; † trial from the update search. RoB, overall risk of bias (+ low; − some concerns; × high; ? not assessable). Citations in Section 3.2 include companion reports of these trials. Trials shown: [45–48,54–71,73–78,80–89,91–93].

Three post hoc exclusions gave estimates closer to 1. Without the 12 trials of the Madrid group, which carry 31% of the weight, the remaining 29 trials gave 0.83 (0.69–0.99); without the trials at high risk of bias, 32 trials gave 0.80 (0.67–0.94); and without both groups, 23 trials gave 0.82 (0.67–1.01). The 34 trials with event counts gave 0.76 (0.64–0.89), whereas the seven converted trials alone, which carry 8% of the weight, gave 0.86 (0.47–1.56). Conversion at control risks of 2% and 25% instead of 7.5% gave 0.77 (0.66–0.89) and 0.78 (0.67–0.90), so the choice of control risk for the conversion changes the estimate little (Table S18).

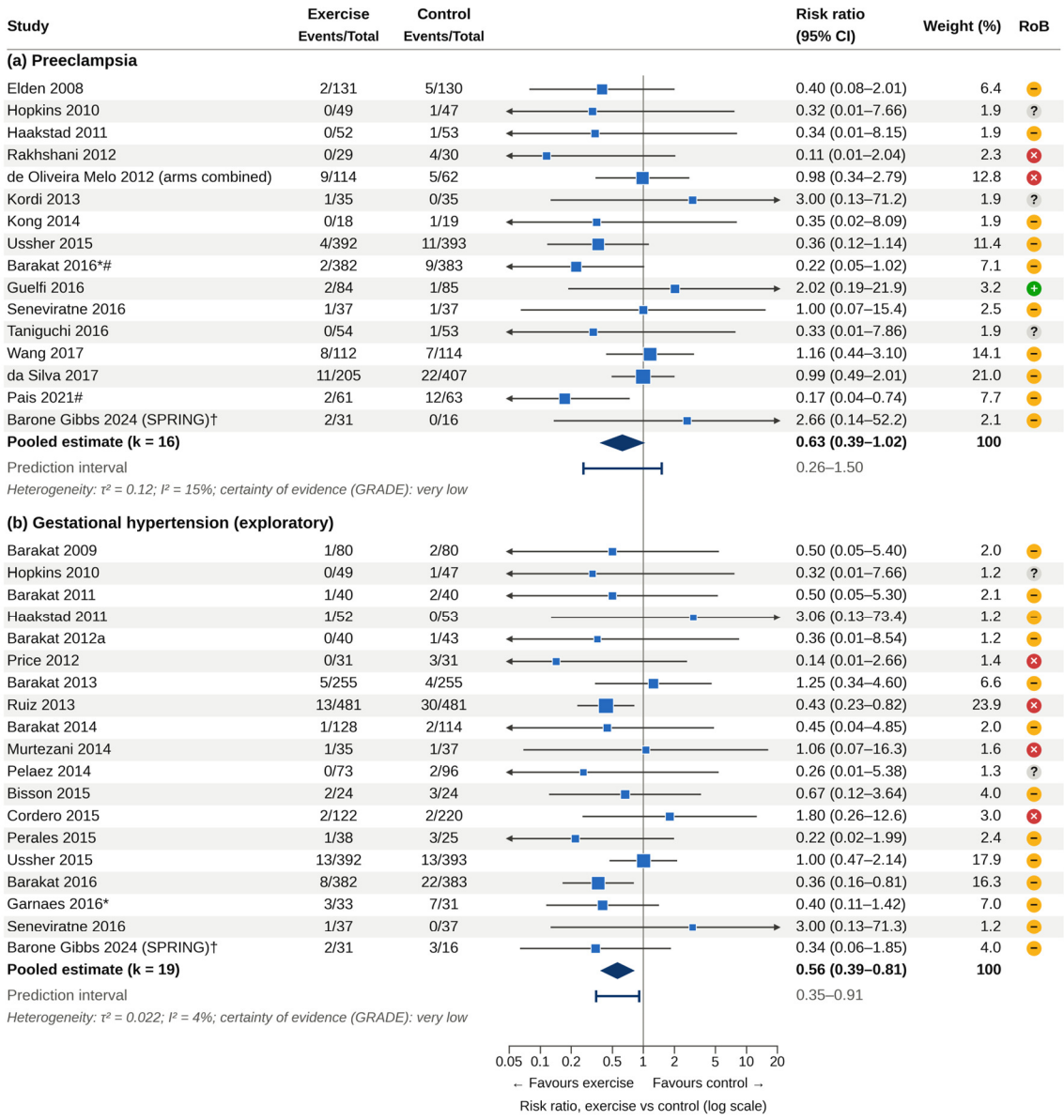
The 37 trials identified through the sources gave 0.77 (0.66–0.90), the four trials found only by the update search 0.69 (0.17–2.85), and the 43 trials including those awaiting classification 0.76 (0.66–0.88) (Table S18). Adding the four trials found only by the update search therefore changed the GDM estimate little, but they are few and imprecise, and this comparison cannot show what a screened de novo search, or a search before 2017, would have added.

3.3. Exercise, Preeclampsia and Gestational Hypertension

The evidence is very uncertain about the effect of exercise on preeclampsia (very low certainty): the estimate is compatible with a reduction and with no effect (upper confidence limit 1.02), and it is not robust. Sixteen trials [45,55,58,68,76,82,84,85,88,89,94–99] gave 0.63 (0.39–1.02), prediction interval 0.26–1.50, I^2 15% (Figure 3a; Table 2); with the quasi-randomised trial [72], the 17 trials gave 0.63 (0.39–1.00). With 3713 participants and 125 events, the optimal information size of 10,064 was not met. At the median control risk of 28 per 1000, the estimate corresponds to 10 fewer (17 fewer to no difference) cases per 1000 women.

Without the two influential trials whose full reports could not be checked [82,98], the remaining 14 trials gave 0.80 (0.51–1.25). These two trials were singled out post hoc because of their large effects, and they are not the only ones without a checked report: two further rows also lack one [55,99], and without all four trials, 12 trials gave 0.88 (0.52–1.50). Ten of the 16 single omissions put the upper limit above 1, and two more at 1.00. The PET estimate, 0.93 (0.41–2.14), and the most precise quarter, 0.87 (0.41–1.83), include no effect; because the pooled interval itself includes no effect, the publication-bias criterion of Section 2.5 does not apply (Table 2).

Gestational hypertension, an exploratory outcome, gave 0.56 (0.39–0.81) in 19 trials [45,55,62,64,67,70,71,73,74,78,82,83,85,94,95,100–103] (Figure 3b; very low certainty). The Madrid group's reports carried 61% of the weight; without them, nine trials gave 0.72 (0.39–1.33).



* Row corrected (Supplementary Table S8); Barakat 2016 control denominator corrected from the trial abstract, events as printed by the sources (full report not obtained).
† Trial found by the update search (Supplementary Table S7).
# Full report could not be checked; without these two trials 0.80 (0.51–1.25), k = 14.
RoB, overall risk-of-bias judgement (RoB 2; Section 2.5): + low; − some concerns; × high; ? not assessable; not used to rate certainty (risk of bias rated very serious for every trial pool, Section 2.5).

Figure 3. Exercise during pregnancy and hypertensive disorders: randomised trials, risk ratios from event counts. (**a**) Preeclampsia (k = 16, prespecified outcome); (**b**) gestational hypertension (k = 19, exploratory, not prespecified). No estimate is pooled across panels. Squares, diamonds, bars and risk-of-bias symbols as in Figure 2; * row corrected (Supplementary Table S8); † trial from the update search (Supplementary Table S7). # full report could not be checked. Barakat 2016 [82]: control denominator corrected from the trial abstract, events as printed by the sources; its report was also not obtained for its panel (**b**) row. Pais 2021 [98]: events as printed by one source, because the abstract gives no counts. Without these two trials, the preeclampsia estimate was 0.80 (0.51–1.25). They were selected post hoc for their large effects and are not the only rows lacking a checked report (Section 3.3). Two trials tested yoga ([68], against walking, and [98]); without both, preeclampsia 0.76 (0.49–1.18) (post hoc, defined after the effects were seen; [98] is also one of the two influential trials whose full reports could not be checked). Trials shown: [45,55,58,62,64,67,68,70,71,73,74,76,78,82–85,88,89,94–103].

Table 2. Summary of findings: principal outcomes of the randomised trials and the observational layers.

Outcome	Studies (Participants)	Relative Effect (95% CI)	Absolute Effect per 1000 (95% CI)	Certainty (GRADE)	Reasons for Rating Down	Analysis Status
Exercise in pregnancy → GDM	41 trials (9373 in 34)	RR 0.77 (0.66–0.90)	17 fewer (25 to 8 fewer) at 75 per 1000 [a]	Very low	Risk of bias (very serious); publication bias [b]	Prespecified outcome (parent protocol); measure, conversion and analysis rules set after data transfer
Exercise in pregnancy → preeclampsia	16 trials (3713)	RR 0.63 (0.39–1.02)	10 fewer (17 fewer to no difference) at 28 per 1000 (median control risk)	Very low [c]	Risk of bias (very serious); imprecision [c]	Prespecified outcome (parent protocol); analysis rules set after data transfer
Pre-pregnancy activity, highest vs. lowest, and GDM	11 cohorts (OR); 2 (RR) and 1 (HR), not pooled	OR 0.91 (0.71–1.15); RR 0.54 to 0.90 and HR 0.81, not pooled	not estimated	Very low	Risk of bias (most weight not assessable); imprecision [d]	Post hoc (defined after extraction)
Total protein before pregnancy and GDM	3 cohorts (OR); 1 (HR)	OR 1.24 (0.80–1.94); HR 1.59 (0.76–3.39)	not estimated	Very low	Risk of bias (most weight not assessable); imprecision [e]	Post hoc (defined after extraction)
Total protein, earliest assessment in pregnancy and GDM	4 cohorts (OR); 1 (HR); 1 (RR)	OR 1.70 (0.76–3.78); HR 0.89 (0.43–1.86); RR 1.00 (0.72–1.38)	not estimated	Very low	Risk of bias (most weight not assessable) [f]; inconsistency; imprecision	Post hoc (defined after extraction)
Total protein, latest assessment before the GDM test and GDM	5 cohorts (OR); 1 (HR)	OR 1.86 (1.03–3.36); HR 1.92 (1.10–3.14)	not estimated	Very low	Inconsistency; risk of bias (possible reverse causation) [g]	Post hoc (defined after extraction)

Notes: Certainty (GRADE): very low, the true effect is probably substantially different from the estimate. Random-effects REML with modified Hartung–Knapp intervals. Risk of bias was rated very serious for every trial pool, because the risk-of-bias judgements were not made by a standardised, human-led assessment (Section 2.5); with the coded rule of revision 1, GDM and preeclampsia would each have been rated low. The trial pools exclude one quasi-randomised trial (Section 3.2). Cohort estimates are pooled only within one effect measure (OR, odds ratio; HR, hazard ratio; RR, risk ratio; Table S20), with categories as each report defined them; ORs overstate the corresponding risk ratios where GDM is common (4–26% in the in-pregnancy cohorts). Indirectness was not rated down: GDM and preeclampsia were accepted as each study defined them. Cohorts start at low certainty; protein cohorts were not searched beyond the sources (search to August 2025), and two cohorts found among the unscreened records of the parent review's search were added post hoc in revision 2. The full notes of revision 1 are in Text S1, Section S3.13. [a] Median control risk of the trials with counts; NNT 59 (40 to 120). At baseline risks of 50, 140 and 250 per 1000, the absolute effects would be 11 (17 to 5), 32 (47 to 14) and 57 (84 to 26) fewer, assuming that the risk ratio applies at each baseline risk. [b] The PET estimate, 0.95 (0.76–1.20), and the most precise quarter of the trials, 0.86 (0.70–1.07), include no effect; Egger's test $p = 0.055$. At low risk of bias: 2 of 41 trials (9% of the weight). All-OR sensitivity analysis: 0.72 (0.60–0.85); with the quasi-randomised trial, 0.75 (0.65–0.88). [c] Optimal information size (10,064) not met. The confidence interval includes no effect, so publication bias was not rated down; the PET estimate and the most precise quarter also include no effect. Without the two influential trials whose full reports

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could not be checked, 0.80 (0.51–1.25); with the quasi-randomised trial, 0.63 (0.39–1.00). At low risk of bias: 1 of 16 trials (3% of the weight). [d] Cohorts not assessable for risk of bias carry 97% of the weight of the OR pool. The risk ratios and the hazard ratio were not pooled (fewer than three cohorts per measure). [e] Cohorts not assessable for risk of bias carry 64% of the weight of the OR pool. One cohort measured intake before conception (USA), two recalled it at 9–11 weeks (China, Malaysia), and one, added in revision 2 (Omoto 2025 [23], Japan), recalled intake from the year before conception to the first trimester; its estimate was re-referenced from the middle quintile (interval approximate). [f] Cohorts not assessable for risk of bias carry 57% of the weight; the pool includes a mid-pregnancy assessment (29.8% of the weight). [g] Possible reverse causation: intake was measured in mid-pregnancy in three of the five cohorts, within weeks of GDM screening. Without the cohort whose report does not document that the dietary assessment preceded the glucose test, 1.90 (0.87–4.14); with the cohort whose outcome was a positive glucose challenge test (outside the pools), 1.83 (1.15–2.90). Other abbreviations as in the list after the Conflicts of Interest.

3.4. Pre-Pregnancy Physical Activity

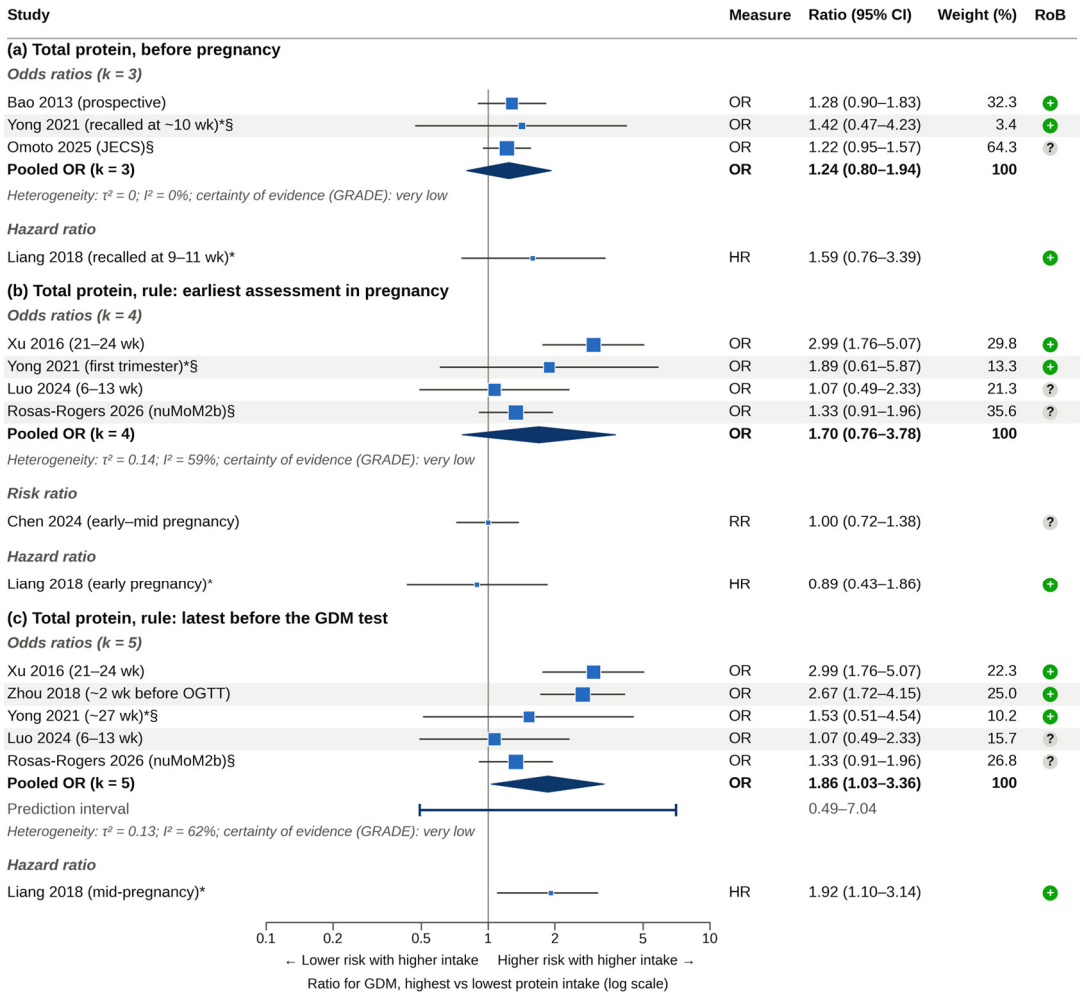
Fourteen cohorts [104–117] compared the highest with the lowest category of pre-pregnancy physical activity in relation to GDM. This extended set adds four cohorts from the update search and five from an earlier meta-analysis [21] to the five cohorts of the source set and was defined post hoc (Table 1; Figure S4). Eleven cohorts reported ORs, which gave 0.91 (0.71–1.15), prediction interval 0.56–1.47, I^2 32% (very low certainty). Two reported risk ratios (0.54 (0.32–0.91) and 0.90 (0.50–1.60)), which were not pooled (fewer than three cohorts). One reported a hazard ratio (Zhang 2006 [105], 0.81 (0.68–1.01)), which was not pooled. One OR was corrected to the most adjusted model of its report (Chasan-Taber 2014 [112], 1.34 (0.55–3.27); Table S8). Cohorts whose risk of bias could not be assessed carried 97% of the weight of the OR pool. In the source-restricted set of five cohorts (four from [19] and the cohort cited by the registered review [104–107,118]), the three ORs gave 0.63 (0.18–2.15), and the risk ratio of Dempsey 2004 [118], 0.26, and the hazard ratio of Zhang 2006 [105], 0.81, were not pooled. Restricted to the cohorts whose reference period lay wholly before pregnancy, the ORs gave 0.92 (0.72–1.19) (10 cohorts), and without the cohort that recalled activity at the GDM screening visit 0.96 (0.74–1.23) (10 cohorts) (post hoc; Table S20).

3.5. Dietary Protein

Twelve publications from eleven cohorts reported protein intake and GDM [22,23,119–128]; two of these cohorts were found among the unscreened records of the parent review's search and added post hoc in revision 2 (Table S16, item (pp)). One cohort [126] reported a positive glucose challenge test, not diagnosed GDM; in revision 2 it was excluded from the window pools and entered a sensitivity analysis (Table S20). Four cohorts assessed intake before pregnancy, two of them by recall at 9–11 weeks of gestation and one by a first-trimester questionnaire covering the year before conception to the first trimester [23], and seven assessed it during pregnancy; two cohorts are in both groups, having assessed intake at both times. Of the remaining two cohorts, one gave continuous estimates only (per 1% of total protein from animal or vegetable sources) [122] and one measured intake at the visit at which GDM was diagnosed [125]; neither enters a window pool. Estimates were grouped in three assessment windows, defined by selection rules: intake before pregnancy, the earliest assessment in pregnancy, and the latest assessment before the GDM test. The two in-pregnancy rules select an assessment, not a period of gestation: the earliest-assessment window includes an assessment at 21–24 weeks (Xu 2016 [127]), and the latest-assessment window includes assessments at 6–13 weeks (Luo 2024 [124]; Rosas-Rogers 2026 [22], whose single assessment enters both in-pregnancy windows). The window estimates are ORs from logistic regression, except those of Liang 2018 [123] (HRs, Cox model) and Chen 2024 [121] (risk ratios, Poisson model); Bao 2013 [120] reports ORs from logistic regression under the name relative risk (Table S20). All three windows were defined post hoc and have equal status (Figure 4; very low certainty).

For total protein before pregnancy, three cohorts reported ORs, which gave 1.24 (0.80–1.94), I^2 0% (1.28, 0.90–1.83; 1.42, 0.47–4.23; and 1.22, 0.95–1.57, re-referenced from the middle quintile, interval approximate [23]), and one an HR (1.59, 0.76–3.39). In pregnancy, ORs gave 1.70 (0.76–3.78) at the earliest assessment (four cohorts, three in Asia and one in the USA; I^2 59%) and 1.86 (1.03–3.36) at the latest assessment before the GDM test (5 cohorts; I^2 62%). The cohorts with other measures gave an HR of 0.89 (0.43–1.86) at the earliest and 1.92 (1.10–3.14) at the latest assessment (Liang 2018 [123]) and a risk ratio of 1.00 (0.72–1.38) at the earliest assessment (Chen 2024 [121]). Only the latest-assessment OR pool has an interval that excludes 1; in three of its five cohorts, intake was assessed in mid-pregnancy, within weeks of the test (Table 2, note g; Section 4.5). GDM occurred in 4% to 26% of the women in these cohorts, so their ORs overstate the corresponding risk ratios.

The two in-pregnancy OR pools share three cohorts that contribute the same estimate to both (Luo 2024 [124], Xu 2016 [127], Rosas-Rogers 2026 [22]); they differ in Yong 2021 [119] (1.89 at the earliest and 1.53 at the latest assessment) and in Zhou 2018 [128] (2.67), which enters only the latest pool. Larger shifts were seen outside the OR pools: in Liang 2018 [123] the HR moved from 0.89 to 1.92, and the Tongji cohort contributes a risk ratio of 1.00 through one publication [121] at the earliest assessment and an OR of 2.67 through another [128] at the latest, a change that mixes measures and publications. For animal and for vegetable protein, the later ratio was numerically higher in both cohorts assessed twice (Liang 2018 [123], Yong 2021 [119]); Luo 2024 [124] contributes one assessment to both windows. For vegetable protein, whose ratios were below 1, higher means closer to or above 1. The windows also overlap in time: one cohort assessed intake only in mid-pregnancy and three only in early pregnancy, so neither window isolates early or late intake.



* Estimate of the primary report for that window (Supplementary Text S1).
§ Highest vs lowest category re-derived from estimates against the middle category (tertiles; Omoto 2025, quintiles), interval approximate; Rosas-Rogers 2026, reciprocal of the printed lowest-vs-highest estimate. Omoto 2025 and Rosas-Rogers 2026 were added post hoc in revision 2; risk of bias not assessed.
RoB, overall risk-of-bias judgement (Newcastle–Ottawa Scale; Section 2.5): + low; ? not assessable.
Estimates are pooled only within one effect measure and only when at least three cohorts report it; no estimate is pooled across measures.
OR, odds ratio; RR, risk ratio; HR, hazard ratio.
All three windows were defined after data extraction and have equal status; Chen 2024 and Zhou 2018 report the same cohort.
wk, week of gestation of the dietary assessment.

Figure 4. Total protein intake (highest vs. lowest category) and gestational diabetes mellitus (GDM) in cohorts, by assessment window, each defined by a selection rule: (**a**) before pregnancy; (**b**) earliest assessment in pregnancy; (**c**) latest assessment before the GDM test. Within each window, estimates are grouped by effect measure (OR, odds ratio; HR, hazard ratio; RR, risk ratio) and pooled only within one measure when at least three cohorts report it; no estimate is pooled across measures

(Table S20). The rules select an assessment, not a period of gestation (week of each assessment in the row labels). * estimate of the primary report for that window; § highest vs. lowest category re-derived from estimates against the middle category (tertiles; Omoto 2025 [23], quintiles), interval approximate, or, for Rosas-Rogers 2026 [22], the reciprocal of the printed lowest-vs-highest estimate; both cohorts were found among the unscreened records of the parent review's search and added post hoc in revision 2, and their risk of bias was not assessed. Chen 2024 [121] (panel (**b**)) and Zhou 2018 [128] (panel (**c**)) report the same cohort. Tajima 2017 [126] (positive glucose challenge test, not diagnosed GDM) is not shown (Section 3.5). Risk of bias is the Newcastle–Ottawa judgement published by the sources for each publication (+ low; ? not assessable). Restricted maximum likelihood (REML) with modified Hartung–Knapp 95% confidence interval (CI); prediction intervals for $k \geq 5$. All windows were defined post hoc and have equal status. That the dietary assessment preceded the glucose test is not documented for Yong 2021 [119] and Liang 2018 [123] (panel (**c**); Text S1, Section S3.12). Animal and vegetable protein are shown in Figure S2. Cohorts shown: [22,23,119–121,123,124,127,128].

For animal and vegetable protein, ORs could be pooled only at the latest assessment, where three cohorts reported them: animal protein gave 2.30 (1.09–4.86) and vegetable protein 0.66 (0.16–2.74); in the other windows, fewer than three cohorts reported ORs (Figure S2). Three studies of other designs [129–131] and the cohort that measured intake at diagnosis are in Text S1.

3.6. Body Mass Index

The IPD meta-analysis of diet- and physical-activity-based interventions [6] (23,698 women, 54 trials) examined BMI as a participant-level effect modifier and reported "no differences by maternal body mass index, age, parity or ethnicity"; its interaction odds ratios are quoted in Table 3.

At trial level, 13 of the 41 GDM trials were restricted (enrolling only women with overweight or obesity) and 28 were unrestricted (Table 3). In the restricted trials, exercise gave 0.73 (0.52–1.03), and in the unrestricted trials 0.80 (0.68–0.94). The ratio of RRs, restricted against unrestricted, was 0.91 (0.64–1.29). The relative effect therefore showed no evidence of a difference between the two kinds of trial, although the interval is wide enough to include important differences in either direction. The comparison is post hoc and between trials, and by ICEMAN criteria the credibility of any effect modification is low [43]. Trial BMI category was assigned by the AI from the stated enrolment criteria; two authors agreed with it for all 41 GDM trials (Section 2.4). Reassigning any one unrestricted trial to the restricted stratum gave ratios from 0.84 (0.60–1.18) to 1.02 (0.74–1.40); classing Nobles 2015 [80], in which 96.8% of participants had a BMI of 25 or more, as restricted gave 0.90 (0.65–1.24) (post hoc). The median control risk was 208 per 1000 in the restricted trials and 68 per 1000 in the others; the difference may reflect BMI but also differences in diagnostic criteria. With similar relative effects and a higher control risk, the modelled absolute reduction was numerically larger in the restricted trials, 57 fewer (101 fewer to 6 more) per 1000 against 14 fewer (22 to 4 fewer) (Table 3). For preeclampsia and for exploratory gestational hypertension only three trials each were restricted, too few for a useful comparison.

In cohorts, same-cohort estimates with and without BMI in the model were extracted for three activity pairs and two protein pairs, and pools in which an estimate was replaced by its model without BMI resembled the pools as used (Table 3, Part B). A meta-regression on mean baseline BMI was not done: mean baseline BMI was not extracted, and a search of abstracts and open-access full texts did not establish how many trials report it. Such a regression would in any case be ecological: it would compare trial averages and could not show whether the effect differs between women of different BMI within a trial [132,133].

Table 3. Body mass index (BMI): (A) participant-level evidence from the IPD meta-analysis, and randomised trials stratified by BMI eligibility; (B) cohort estimates with and without BMI in the model.

(A)

Outcome	Stratum	Trials	Risk Ratio (95% CI)	Median Control Risk per 1000	Absolute Effect per 1000 (95% CI)	Ratio of Risk Ratios (95% CI)
GDM, participant level (IPD meta-analysis [6], diet- and physical-activity-based interventions combined)	BMI as a participant-level effect modifier	54 (23,361 women in the GDM analysis)	No differences by maternal BMI reported	not reported	not reported	Interaction OR vs. normal weight, any criteria: overweight 0.98 (0.74–1.29), obese 0.90 (0.72–1.11), per BMI unit 1.00 (0.98–1.02); NICE criteria: overweight 1.11 (0.67–1.83), obese 1.07 (0.69–1.68), per BMI unit 1.00 (0.99–1.02) (trial and participant numbers per contrast: Table S19)
GDM, trial level	Overweight or obesity only	13	0.73 (0.52–1.03)	208	57 fewer (101 fewer to 6 more)	0.91 (0.64–1.29)
	Not restricted to overweight or obesity	28	0.80 (0.68–0.94)	68	14 fewer (22 to 4 fewer)	reference
Preeclampsia	Overweight or obesity only	3	1.04 (0.15–7.27)	53	2 more (45 fewer to 330 more)	1.88 (0.67–5.23)
	Not restricted to overweight or obesity	13	0.55 (0.31–0.98)	23	10 fewer (16 fewer to no difference)	reference
Gestational hypertension (exploratory)	Overweight or obesity only	3	0.57 (0.07–4.74)	125	54 fewer (116 fewer to 467 more)	1.02 (0.36–2.87)
	Not restricted to overweight or obesity	16	0.56 (0.37–0.85)	26	11 fewer (16 to 4 fewer)	reference

(B1)

Same-Cohort Pair	Exposure	Model in the Main Analysis	Estimate in the Main Analysis (95% CI)	Model without BMI	Estimate without BMI (95% CI)
Whitaker 2018, CARDIA [114]	Pre-pregnancy activity	Adjusted for waist circumference (not BMI), HOMA-IR and HDL-C	1.19 (0.76–1.88)	Partially adjusted, without BMI or waist circumference	1.22 (0.79–1.90)
McDonald 2021, NICHD Fetal Growth Studies [115]	Pre-pregnancy activity	Age, race/ethnicity and BMI	0.90 (0.50–1.60)	Crude	0.80 (0.50–1.50)
Slapgaard 2025, HUNT4 [104]	Pre-pregnancy activity	Includes BMI	0.55 (0.16–1.94)	Partially adjusted	0.41 (0.12–1.39)

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Liang 2018 [123]		Total protein, before pregnancy	Includes BMI	1.59 (0.76–3.39)	Crude (Model 1; intake as energy-adjusted residuals)	1.17 (0.61–2.27)
Liang 2018 [123]		Total protein, mid-pregnancy	Includes BMI	1.92 (1.10–3.14)	Crude (Model 1; intake as energy-adjusted residuals)	2.11 (1.15–2.96)

(B2)

Pool	Cohorts	As Used, within One Measure (95% CI)	Cohorts with a Model without BMI Extracted	Estimates without BMI Where Extracted (95% CI)	BMI-Adjusted Cohorts Only (95% CI)	Cohorts Not Adjusted for BMI or Adjustment Unclear (95% CI)
Pre-pregnancy activity	14 (11 OR, 2 RR, 1 HR)	OR 0.91 (0.71–1.15)	1 of 11 OR cohorts	0.90 (0.71–1.14)	5 OR cohorts: 1.02 (0.72–1.43)	6 OR cohorts: 0.87 (0.58–1.32)
Total protein, before pregnancy	4 (3 OR, 1 HR)	OR 1.24 (0.80–1.94)	1 of 4 (Liang 2018 [123], HR; see notes)	—	3 OR cohorts: 1.24 (0.80–1.94)	none
Total protein, earliest assessment	6 (4 OR, 1 HR, 1 RR)	OR 1.70 (0.76–3.78)	not extracted	—	4 OR cohorts: 1.70 (0.76–3.78)	Chen 2024 [121] (not classified): not pooled
Total protein, latest assessment	6 (5 OR, 1 HR)	OR 1.86 (1.03–3.36)	1 of 6 (Liang 2018 [123], HR)	OR pool unchanged	5 OR cohorts: 1.86 (1.03–3.36)	none

Notes: Part A: interaction odds ratios from the two-stage IPD analysis as printed in [6]; they compare the intervention odds ratio between BMI groups, are on the odds-ratio scale, are not comparable with the ratios of risk ratios, and were not re-analysed here; [6] assessed effect modifiers only for all lifestyle interventions combined. Trial-level rows: risk ratios from random-effects REML pools with modified Hartung–Knapp intervals; the ratio of risk ratios compares the restricted with the unrestricted stratum (Wald intervals, which understate uncertainty); post hoc, without *p*-values. Absolute effects at each stratum's median control risk apply the stratum's own risk ratio. Trial BMI category was assigned by the AI from stated enrolment criteria; two authors agreed with it for all 41 GDM trials, but the 10 trials pooled only for preeclampsia or gestational hypertension were not checked (Section 2.4). Unrestricted trials also enrolled women with overweight or obesity (Section 4.4). Part B: the models without BMI are crude or partially adjusted and omit other covariates too, so they differ from the published models in more than BMI; they were extracted by the AI and were not reviewed in the authors' check. Pools are within one effect measure (OR, odds ratio; HR, hazard ratio; RR, risk ratio); a pool "without BMI" replaces an estimate with the estimate from its model without BMI where one was extracted. Whitaker 2018 [114], adjusted for waist circumference, is counted with the cohorts not adjusted for BMI. The adjusted and not-adjusted pools compare different cohorts. Tajima 2017 [126] (positive glucose challenge test, not diagnosed GDM; adjustment for BMI unclear) is outside the protein pools in revision 2 (Section 2.3). Details, including the revision-1 notes, in Table S19 and Text S1, Section S3.13. AI, artificial intelligence; CARDIA, Coronary Artery Risk Development in Young Adults; CI, confidence interval; GDM, gestational diabetes mellitus; HDL-C, high-density lipoprotein cholesterol; HOMA-IR, homeostasis model assessment of insulin resistance; HUNT4, fourth Trøndelag Health Study; IPD, individual participant data; NICE, National Institute for Health and Care Excellence; NICHD, Eunice Kennedy Shriver National Institute of Child Health and Human Development; REML, restricted maximum likelihood.

3.7. Other Evidence Layers

No randomised trial of protein–energy supplementation reported GDM. Two reported preeclampsia: one in the Cochrane review [20] and one among the 16 trials added by the update search [134–149]; the latter recorded one event in each arm [137]. For the exploratory neonatal outcomes of the exercise trials [16,25,26,49,150–158], preterm birth gave 1.01 (0.82–1.24) in 31 trials, macrosomia 0.72 (0.53–0.97) in 18 trials (0.97 (0.74–1.27) without the Madrid group's reports, which carry 32% of the weight), and low birthweight 0.93 (0.72–1.20) in 16 trials, all at very low certainty. Supplementation, glycaemic biomarkers and the neonatal outcomes are reported in detail in Text S2.

4. Discussion

4.1. Principal Findings

The evidence is very uncertain about the effect of exercise during pregnancy on GDM; its effect on preeclampsia is also very uncertain, with an estimate compatible with benefit and with no effect that is not robust. The relative effect on GDM showed no evidence of a difference by BMI eligibility (post hoc; ratio of RRs 0.91, 0.64–1.29, an interval compatible with important differences in either direction). The post hoc associations of pre-pregnancy activity and of protein intake with GDM are very uncertain. All of these findings rest on studies selected by existing meta-analyses and, for the update searches, on AI-assisted selection and extraction, with the limitations set out in Section 4.7.

Our GDM estimates were closer to 1 than the published pools, although the intervals overlapped: on the OR scale 0.72 here against 0.62 [11] and 0.64 [6] in the sources, and on the RR scale 0.77 against 0.66 [14]. The published pools of protein intake during pregnancy, 2.00 [8] and 2.24 [7], resembled our latest-assessment OR pool. Pooled across windows with one estimate per cohort, as the sources did, but within one measure (post hoc), the eight cohorts that reported ORs gave 1.65 (1.18–2.31) (Table S20). These comparisons cannot attribute the differences to counting each study once, because the trial sets, the rows selected, the pooling methods and the effect measures also differed.

4.2. Interpreting the Effect on GDM

Combining ORs and RRs gives a quantity without a single interpretation when the outcome is common. The RR is collapsible (without confounding, the overall RR is a weighted average of covariate-specific RRs, which does not hold for the OR) and answers the clinical question more directly; the cost of using it here is the approximate conversion of seven ORs [37,38], which carry 8% of the weight (Section 3.2).

Because the PET estimate includes no effect on both scales, as does the pool of the most precise quarter of trials on the RR scale (at the boundary on the OR scale; Table S18), the true reduction may be smaller than the pooled estimate. Identifying trials through existing meta-analyses may add to this, because it inherits any tendency of the sources towards published and positive studies (Section 2.2). The prediction interval also includes no effect, so in some settings the benefit may be absent.

Diagnostic criteria, recorded in our data for 3 of the 41 trials, were not extracted from full texts, so no subgroup analysis by criteria was done, although relative effects may depend on them [6]. The consequences are visible in the control-group risks: they ranged from 8 to 488 per 1000 across the 34 trials with counts, a range that reflects populations and diagnostic criteria together, and the median was 208 per 1000 in the restricted trials against 68 in the others (Table 3). The trial carrying the most weight (9%), in women with previous GDM, gave 1.01 (0.70–1.46). Indirectness was not rated down for this mixture of definitions and populations, because GDM was accepted as each trial defined it; rating it

down would not change the very low certainty. One-step screening diagnoses about twice as many women as two-step screening [159], and GDM frequency under IADPSG criteria varies widely across centres [160], so absolute effects and the NNT depend on the criteria through the baseline risk (Table 2, note a).

4.3. Preeclampsia

The estimate is compatible with benefit and with no effect, depends on two influential trials whose full reports could not be checked, falls short of the optimal information size, and pools preeclampsia as each trial defined it. Those definitions were not extracted, and most trials predate the 2018 definition of the International Society for the Study of Hypertension in Pregnancy [161]. Ten of the 16 single omissions put the upper confidence limit above 1, and two more at 1.00, and certainty is very low (Section 3.3). The evidence is therefore insufficient to establish a preventive effect, and these data provide no basis for using exercise in place of low-dose aspirin in women at high risk [162–164].

4.4. Interpreting the Body Mass Index Analyses

In randomised trials, randomisation balances BMI between arms in expectation, so BMI does not confound the exercise–control comparison. Across trials, BMI may modify the effect, but the stratum comparison is observational and can be confounded by other trial features, such as diagnostic criteria. Unrestricted trials also enrolled women with overweight or obesity (about 29% in one trial, from the counts tabulated by [15]; Text S1, Section S3.8), so the comparison is not of overweight with normal weight, and any true difference would tend to be attenuated. Because the classes rest mainly on abstracts, the trial-level comparison remains tentative. In cohorts, pre-pregnancy BMI is a confounder for protein intake during pregnancy. For pre-pregnancy activity, and for protein intake before pregnancy, it is both a confounder and a possible partial mediator, so adjustment can remove part of the effect of interest [165]. Pools with and without BMI gave similar estimates, but models without BMI were extracted for few cohorts, and the BMI-adjusted and the unadjusted activity pools compare different cohorts (Table 3, Part B2).

4.5. Pre-Pregnancy Activity and Dietary Protein

The association of pre-pregnancy physical activity with GDM, an OR of 0.91 (0.71–1.15) in 11 cohorts, with two unpooled risk ratios from 0.54 to 0.90 and one unpooled hazard ratio of 0.81, is very uncertain. Activity was recalled during pregnancy in several cohorts, so recall and social-desirability bias are possible, as they are for self-reported diet. In the source-restricted set, the three ORs gave 0.63 (0.18–2.15); its risk ratio is an early report of the Omega cohort (0.26), whose later report (0.54) is in the extended set. Categories were cohort-specific, so no dose or threshold of activity can be inferred.

The protein associations are observational and very uncertain. Pooling within one measure removed the combination of ratio measures, but the OR pools still combine different adjustment sets, and because GDM was common in several cohorts their ORs overstate the corresponding risk ratios. The difference between the earliest and the latest assessment was not formally tested; within the OR pools it was small (1.70 (0.76–3.78) against 1.86 (1.03–3.36)), and larger shifts appeared within two cohorts that reported both assessments, one with HRs and one across two publications with different measures (Section 3.5). Such shifts may have three explanations, which these data cannot separate (Table 2). First, reverse causation: women at metabolic risk may raise their protein intake before GDM screening or, where it is not documented that the dietary assessment preceded the glucose test, after the diagnosis. Second, a true effect of intake close to the test. Third, selection: the two windows take different publications or assessments from the same cohorts (Section 3.5).

Residual confounding is also likely. The cohort with the largest in-pregnancy estimate [127] lists no energy term in its model, so its contrast may partly reflect total intake; it contributes the same estimate to both windows. Without that cohort, the latest-assessment OR pool gave 1.63 (0.78–3.39) in four cohorts, which are also the cohorts whose energy term is confirmed; at the earliest assessment, the three remaining cohorts gave 1.32 (0.64–2.72) (post hoc; File S2). The other cohorts adjusted for different sets of macronutrients, so each estimate answers a different question about what protein replaces in the diet; where fat and carbohydrate were both adjusted for, no substitution is defined (Table S17, Panel C) [166]. Pre-pregnancy BMI is a confounder; gestational weight gain is a possible mediator, and at least one cohort adjusted for it, which may bias towards no association [128,165]. Animal protein may mark a dietary pattern rather than a causal nutrient, and the estimates describe associations, not the effects of changing intake.

4.6. Implications by Layer

Preventive exercise programmes: The very uncertain randomised evidence on GDM is consistent with guidance that women without contraindications be active in pregnancy [167,168], but it cannot by itself support that guidance, which rests on wider evidence. These estimates identify no programme, dose, start time or target group (Sections 2.3 and 3.1). Trial populations were not synthesised beyond BMI eligibility, so applicability to women with pregnancy complications was not assessed. Preventing a diagnosis of GDM is not the same as preventing its complications [159]. The exploratory neonatal outcomes did not establish an effect on complications, and they showed no clear excess of preterm birth; caesarean delivery, stillbirth and small for gestational age were not analysed (Text S2; Table S16).

Habitual activity and protein intake: The very uncertain observational evidence provides no basis for advice on activity before conception or on protein intake to prevent GDM, nor for restricting protein in pregnancy. Two randomised trials of a Mediterranean-style diet, which changes protein sources together with fats and other foods, gave discordant results for GDM: fewer diagnoses with the diet in women with metabolic risk factors (84/477 against 124/497) [169], and more in women at high risk of a small-for-gestational-age infant, where GDM was an exploratory outcome (48/392 against 30/401) [170]. These trials test a dietary pattern, not protein intake, and were not pooled here.

Protein–energy supplementation: Trials of protein–energy supplementation, mostly in undernourished women in low- and middle-income countries, did not report GDM, and the two that reported preeclampsia are too few to inform it (Text S2).

4.7. Strengths and Limitations

The main strengths are that no trial or cohort is counted more than once within a pool, except that trials from the Madrid group may share participants; that randomised and observational evidence are kept apart, with one effect measure for the principal trial analyses and cohort pools within one effect measure and assessment window; that each source's transferred rows were tested against its published pools; and that the data, the recorded decisions and the code are available (File S2). The limitations below, the first five of which concern how the evidence was assembled, mean that this work is a reanalysis of selected evidence, not a systematic review, and that its estimates should not be read as summaries of all eligible studies.

Study selection: The 7160 records retrieved by the registered search were not formally screened (Section 2.1). Primary studies were instead identified through existing meta-analyses chosen without recorded criteria, and update searches covered only some layers and omitted Embase, CENTRAL and Scopus. The comprehensiveness and reproducibility of study selection were therefore not established: eligible studies retrieved by

the registered search but absent from the sources may have been missed, and any tendency of the sources towards published and positive studies is inherited, which may bias the estimates away from no effect.

Unvalidated use of artificial intelligence: AI screened records, assessed eligibility and extracted data in the update searches, transferred the source rows and drafted risk-of-bias judgements, and in the original analysis none of these steps were validated by humans: the authors' check was an unblinded review of AI output without an item-level log, and all 23 queried decisions were retained without recorded reasons (Section 2.4). In revision 2, the eligibility decisions of the exercise and activity-cohort update searches were checked by an independent human screener with adjudication: agreement at title and abstract was κ 0.86 for exercise trials and κ 0.53 for cohorts, the recall of the AI screening against the adjudicated human decisions was 100% (15 of 15; 95% CI 78–100%) for exercise trials, and one relevant record had been excluded by the AI at title and abstract (37826849); it reports the Nurses' Health Study II, which already enters the activity analyses once (Zhang 2006 [105]), and was not added, because each cohort is counted once. The adjudicated decisions would remove five trials from the main pools; three of these decisions rested on abstracts, and applying them changed the pooled estimates little (Section 3.1). This check does not cover the supplementation update, the transfer of source rows, the extraction of event counts or the risk-of-bias judgements, so errors in those steps, and a selection or extraction bias of unknown size and direction, cannot be excluded.

Deviation from the registered protocol: The registered protocol specified dual independent human review; its replacement by AI is a substantial deviation. Other deviations add to it: the sources were not appraised with AMSTAR 2, cohorts were not rated with ROBINS-E, and the registered dose–response analysis and the observational preeclampsia layer were not done (Table S16). The methods actually used therefore differ materially from those registered, the reliability of the selection and extraction steps rests on checks made after the fact (Section 2.4), and post hoc choices of measure, analysis rules and cohort sets may have influenced the estimates; no comparison made after the results were seen is interpreted as confirmatory.

Incomplete update: Exercise trials and pre-pregnancy activity cohorts were updated, the latter in PubMed only, but protein-intake cohorts were not searched beyond the sources, whose latest search ended in August 2025 (Table 1). Two cohorts found after the results were known, among the unscreened records of the parent review's search, were added post hoc to the total-protein pools in revision 2; one of them was published within the search period of the latest source but is not among the cohorts transferred from it. Because their addition did not follow a search, other eligible cohorts may have been missed, and the protein layer may be incomplete and less current than the exercise layer (see Not undertaken, below).

Retrospective registration: The reanalysis was registered after searching, selection, extraction and analysis, and preliminary GDM pools preceded the PROSPERO record of the parent review (Section 2.1). Prespecified methodological decisions therefore cannot be distinguished reliably from post hoc ones. Apart from the two prespecified outcomes, four registered analyses (trim-and-fill, Egger's test, leave-one-out analysis and the funnel plot) and the registered within-measure pooling of cohort estimates are labelled as registered; every other analysis is labelled post hoc or exploratory (Table 1; Section 2.3), and the choice of the principal effect measure, the analysis rules and the certainty rules were all made after results were available.

Data and verification: Only the most influential transferred rows were checked by the AI and mostly against abstracts. Rows that were confirmed against a primary report or abstract, taken directly from a report or later read in one carry 43% of the GDM weight

and 37% of the preeclampsia weight; the remainder could not be verified or were never compared, so residual extraction error cannot be excluded [171].

Risk of bias and certainty: Risk-of-bias judgements came from heterogeneous pathways (Section 2.5), could not be assessed for 11 of the 85 trials and 15 of the 21 cohorts, and were not reassessed by humans in a standardised way; a second AI assessment agreed poorly with the first (overall κ 0.20 in 9 trials). We therefore did not let these judgements support any trial certainty rating: risk of bias was rated very serious for every trial pool; cohort pools were rated with the coded rule of revision 1 (Section 2.5), and every outcome is at very low certainty. With the coded rule of revision 1, which used these judgements, GDM and preeclampsia would each have been rated low. At low risk of bias were 2 of 41 trials (9% of the weight) for GDM and 1 of 16 trials (3% of the weight) for preeclampsia. The trustworthiness screen covered only trials from the update search, not the source trials. Of the seven exercise trials screened, three were included, and three others, among them the only trial of the Madrid group that was screened, raised serious concerns; the seventh was excluded on eligibility.

Not undertaken: We did not run a full update search for protein cohorts, extract diagnostic criteria, mean BMI, exercise dose or substitution estimates from full texts, re-assess risk of bias uniformly or synthesise trial populations beyond BMI eligibility. A limited AI-assisted PubMed check of records entered from 15 August 2025 to 11 November 2026, not reviewed by the authors and not a substitute for an update search, found one further prospective cohort [22], which was added to the total-protein pools in revision 2 (Section 3.5), and one report that cannot be expressed as a highest-versus-lowest estimate [172], which was not pooled. A similar AI-assisted check of the GDM trials found diagnostic criteria stated in fewer than half of the reports that could be read in full; for most trials, no open-access full text was available to the check.

5. Conclusions

In this reanalysis and partial update of the primary studies included in published meta-analyses, the evidence is very uncertain about the effect of exercise during pregnancy on GDM, and no effect remains possible: some small-study analyses include no effect, most transferred rows were not verified against primary reports, trials from the Madrid group may share participants (without them, 0.83, 0.69–0.99), and the risk-of-bias judgements could not support a higher rating. The relative effect showed no evidence of a difference by BMI eligibility (post hoc); absolute effects depend on baseline risk and diagnostic criteria. The evidence on preeclampsia is very uncertain: the estimate is compatible with benefit and with no effect, and it is not robust to omitting two influential trials whose full reports could not be checked. The associations of pre-pregnancy activity and protein intake with GDM are very uncertain; protein estimates differed by the assessment selected, reverse causation cannot be excluded for later assessments, and the estimates do not support advice on protein intake. Because studies were identified through existing meta-analyses rather than through screening of the registered search, AI replaced the registered dual human review, the update was partial and the registration retrospective, these results describe the selected evidence and should not be taken as a systematic summary of all eligible studies.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/nu18233986/s1. Text S1: Detailed methods and results; Text S2: Secondary and exploratory evidence layers; Table S1: Primary studies of the source meta-analyses excluded from the main analyses, with reasons; Table S2: Source meta-analyses and other sources of transferred rows: role, rows transferred, reports and acceptance test (466 study-level rows in all); Table S3: Characteristics of the 106 studies in the main analyses and of 7 reports used in

sensitivity analyses only; Table S4: Maternal glycaemic biomarkers: trial-level estimates (Supplementary Text S2, Section 2); Table S5: Protein–energy supplementation trials of the Cochrane review [20]: rows transferred from its analyses of balanced, high-protein and isocaloric protein supplementation (45 rows from 11 trials); Table S6: Searches (PRISMA-S): update search for randomised trials of exercise, update search for protein–energy supplementation trials, search for cohorts of pre-pregnancy activity, and search for a source of maternal biomarker data; Table S7: Trials added by the update search, and neonatal outcomes of included trials published since 2017: event counts from two extractions by two instances of the same AI model, with verbatim source quotations, trial characteristics, risk of bias and trustworthiness screen; Table S8: Transferred rows corrected against the primary reports; Table S9: Reports from one research group (Madrid): trial-level map used for the de-duplication sensitivity analysis; Table S10: Main and sensitivity analyses (REML; 95% CI by the modified Hartung–Knapp method, with the ad hoc variance correction $q^* = \max(1, q)$; 95% prediction interval with $k - 2$ degrees of freedom, not shown for fewer than five studies); Table S11: Small-study effects, exercise and GDM (randomised trials; risk-ratio scale, both scales in Supplementary Table S18); Table S12: GRADE domains, information size and absolute effects; Table S13: Update search: full-text decisions, with reasons (two assessments by two instances of the same AI model and the final decision), and decisions on records found only in Europe PMC or ClinicalTrials.gov; Table S14: Search for cohorts of pre-pregnancy activity published from 1 August 2015 (PubMed, re-run on 7 October 2026): screening decisions, with reasons (two screenings by two instances of the same AI model and the final decision); Table S15: Protein–energy supplementation trials published from February 2015 (update search): full-text decisions, characteristics, risk of bias and outcome data from two extractions by two instances of the same AI model, with verbatim source quotations; Table S16: Deviations from the registered review (PROSPERO CRD420261486377), with reasons; Table S17: Body mass index (BMI) as a possible effect modifier: trials by BMI eligibility and cohorts by adjustment for pre-pregnancy BMI (post hoc; added at the request of the Academic Editor); Table S18: Exercise during pregnancy and gestational diabetes mellitus (GDM) analysed in one effect measure; Table S19: Body mass index (BMI): trials stratified by BMI eligibility, and cohort estimates with and without adjustment for BMI; Table S20: Cohort estimates by effect measure (revision 2); Table S21: Independent human screening of the update searches (revision 2); Figure S1: Risk of bias of the randomised trials in the main pools; Figure S2: Animal (a–c) and vegetable (d–f) protein intake (highest vs. lowest category) and gestational diabetes mellitus (GDM) in cohorts, by assessment window: before pregnancy, earliest assessment in pregnancy, latest assessment before the GDM test; Figure S3: Contour-enhanced funnel plot of the principal GDM pool (41 randomised trials of exercise, risk-ratio scale; revision 2, without the quasi-randomised trial), drawn from the deposited rows (File S2, r1_new/r1_funnel.py); Figure S4: Pre-pregnancy physical activity (highest vs. lowest category) and gestational diabetes mellitus (GDM) in cohorts: study-level estimates grouped by effect measure; File S1: PRISMA 2020 checklist and PRISMA 2020 for Abstracts checklist; File S2: Data, code and decisions.

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Data Availability Statement: The data, code and recorded decisions are provided in File S2 (a README lists its contents): the transferred rows and acceptance-test outputs; the update-search records with screening, eligibility and extraction decisions and verbatim quotations; the independent human screening of revision 2, with the adjudication record; the row-level checks against primary reports, and the second AI assessment of risk of bias and certainty; the written instructions deposited for the AI instances (Text S1, Section S2.9; the other AI steps applied the eligibility criteria of Section 2.3 and the RoB 2 and Newcastle–Ottawa tools); and the analysis code in Python and R (metafor; versions in Section 2.6), including the re-computation of the principal pools in R. Not provided: full texts of the primary reports, raw record downloads and screening batches containing titles and abstracts (copyright); the records of the parent review's search; scripts run by the AI in its session folders (among them, those behind the quotation checks of the risk-of-bias and trustworthiness judgements and the kappa statistics of the second AI assessment). Also not provided: the agent runtime and the instructions identified as not deposited in Text S1, Section S2.9.

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Abbreviations

The following abbreviations are used in this manuscript: AI, artificial intelligence; AMSTAR 2, A MeaSurement Tool to Assess systematic Reviews 2; BMI, body mass index; CI, confidence interval; GDM, gestational diabetes mellitus; GRADE, Grading of Recommendations Assessment, Development and Evaluation; HDL-C, high-density lipoprotein cholesterol; HOMA-IR, homeostasis model assessment of insulin resistance; HR, hazard ratio; IADPSG, International Association of Diabetes and Pregnancy Study Groups; ICEMAN, Instrument for assessing the Credibility of Effect Modification Analyses; IPD, individual participant data; NHS II, Nurses' Health Study II; NICE, National Institute for Health and Care Excellence; NNT, number needed to treat; OGTT, oral glucose tolerance test; OR, odds ratio; PEESE, precision-effect estimate with standard error; PET, precision-effect test; PRISMA, Preferred Reporting Items for Systematic Reviews and Meta-Analyses; PRISMA-S, PRISMA extension for reporting literature searches; PROSPERO, International Prospective Register of Systematic Reviews; REML, restricted maximum likelihood; RoB 2, revised Cochrane risk-of-bias tool for randomised trials; ROBINS-E, Risk Of Bias In Non-randomized Studies of Exposures; RR, risk ratio.

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