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Association of Homocysteine Levels with Recurrent Pregnancy Loss: A Systematic Review

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ABSTRACT

Elevated homocysteine (Hcy) has been implicated in placental vascular dysfunction and adverse reproductive outcomes. **Objective:** To synthesize recent evidence on the association between Hcy levels and recurrent pregnancy loss (RPL), emphasizing methodological consistency and potential modifiers. **Methods:** Following PRISMA 2020, observational studies comparing Hcy in women with RPL versus controls were screened across PubMed, Scopus, and Cochrane. Reviews, pilots, case reports, abstracts, animal studies, and articles without quantitative Hcy data were excluded. Risk of bias was assessed using the Newcastle–Ottawa criteria; results were summarized with Synthesis Without Meta-analysis (SWiM). **Results:** Fourteen eligible studies across South Asia, the Middle East, Europe, and East Asia consistently reported higher Hcy among RPL cases, with typical mean differences ≈4–7 µmol/L and odds ratios ≈2–3, including studies adjusting for folate/B12 and MTHFR genotype. Heterogeneity stemmed from biospecimen type (serum/plasma), assay platform (HPLC vs immunoassay), fasting status, sampling time (preconception vs early pregnancy), and cut-offs (10–15 µmol/L). Emerging literature outside the included set supports endothelial mechanisms and gene nutrient interactions while highlighting reporting gaps and the need for interventional trials. **Conclusions:** Current evidence supports Hcy as a reproducible risk marker for RPL, plausibly mediated by endothelial and thrombo-inflammatory pathways and modified (but not fully explained) by folate/B12 status and genetic variants. Standardized measurement, rigorous adjustment, and randomized trials of targeted vitamin strategies are priorities.

INTRODUCTION

Recurrent pregnancy loss (RPL) affects 5% of couples and remains etiologically heterogeneous; endothelial dysfunction and microthrombosis are increasingly recognized contributors [1, 2]. Elevated Hcy is biologically plausible linked to endothelial injury, impaired trophoblast invasion, and dysregulated one-carbon metabolism [3, 4]. Recent data also connect adverse reproductive histories with later cardiometabolic and neurovascular sequelae through shared endothelial pathways, underscoring clinical relevance [5]. Contemporary studies associate higher maternal Hcy with miscarriage risk and subfertility, but variability in biospecimen, fasting protocols, assay methods, and cut-offs complicates comparability [6, 7]. Mechanistic work implicates Hcy metabolites in

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endothelial damage, yet few RPL studies measure vascular readouts in parallel. Moreover, gene nutrient interactions (e.g., folate-pathway variants) are inconsistently reported, and high-quality interventional data remain scarce [8]. A systematic synthesis focused on study-level methods (matrix, fasting, assay, thresholds) and adjustment for folate/B12 and MTHFR can clarify whether elevated Hcy is a robust signal rather than an artifact of measurement or confounding; mapping this against recent mechanistic and prognostic literature strengthens biological plausibility and translational potential [9].

This study aims to summarize the direction and magnitude of association between Hcy and RPL across designs and regions, to describe sources of methodological heterogeneity, to review contemporary mechanistic, genetic, and prognostic evidence relevant to Hcy–RPL, and to outline priorities for standardization and trials.

METHODS

This systematic review was conducted in accordance with the PRISMA 2020 guidelines. A comprehensive search was carried out across three electronic databases: PubMed, Scopus, and the Cochrane Library, covering all publications from 2007 to March 2025. The search strategy used the following terms: ("homocysteine" OR "hyperhomocysteinemia") AND ("recurrent pregnancy loss" OR "recurrent miscarriage" OR "recurrent abortion"). In PubMed, controlled vocabulary (MeSH) terms were mapped alongside free-text keywords in titles and abstracts. In Scopus and Cochrane Library, equivalent free-text keywords and subject headings were used. Filters for human studies and female participants of reproductive age were applied where available, while no restrictions were set for publication year. Only studies published in the English language were included, while studies in other languages such as Chinese, Italian, Turkish, Spanish, Persian, and Arabic were excluded. Hence, only English-language articles were accepted. All identified records were imported into EndNote 21 (Clarivate Analytics) for reference management, and duplicate publications were removed using EndNote's duplicate detection tool. Studies were eligible if they reported original quantitative data on homocysteine levels in women with recurrent pregnancy loss (RPL) compared with controls, defined RPL as ≥2 or ≥3 consecutive miscarriages (regardless of gestational age), and included all study design like observational or experimental designs such as case-control, cross-sectional, cohort, and randomized studies etc. Although the inclusion criteria permitted all study designs such as case-control, cross-sectional, cohort, and randomized studies, during the full-text screening, no cross-sectional, cohort, or randomized experimental studies meeting the eligibility criteria were found. The available literature on homocysteine levels and recurrent pregnancy loss (RPL) is predominantly composed of case control designs, where women with RPL are compared with healthy controls. Cross-sectional or cohort studies addressing this association are either lacking or did not meet the required methodological standards (for example, not reporting homocysteine measurements, lacking control groups, or focusing on unrelated outcomes). Hence, only case–control studies were included in the final synthesis, which is consistent with most published reviews. The following were excluded: systematic reviews, scoping reviews, narrative reviews, case reports, pilot studies, conference abstracts, animal studies, and studies not reporting homocysteine measurements or lacking a control group. Articles with overlapping populations were carefully evaluated, and the most comprehensive dataset was retained. A total of 247 records were identified (PubMed n=91, Scopus n=80, Cochrane n=76). After removal of 32 duplicates, 215 unique records were screened by title and abstract. Of these, 126 were excluded as irrelevant. Full-text review was performed for 89 studies, of which 75 were excluded due to reasons such as not presenting quantitative data (n=22), being review or case report in nature (n=18), or involving the wrong population or outcome (n=35). Finally, 14 studies met the inclusion criteria and were incorporated into the qualitative synthesis. The selection process is illustrated in the PRISMA 2020 flow diagram (Figure 1).

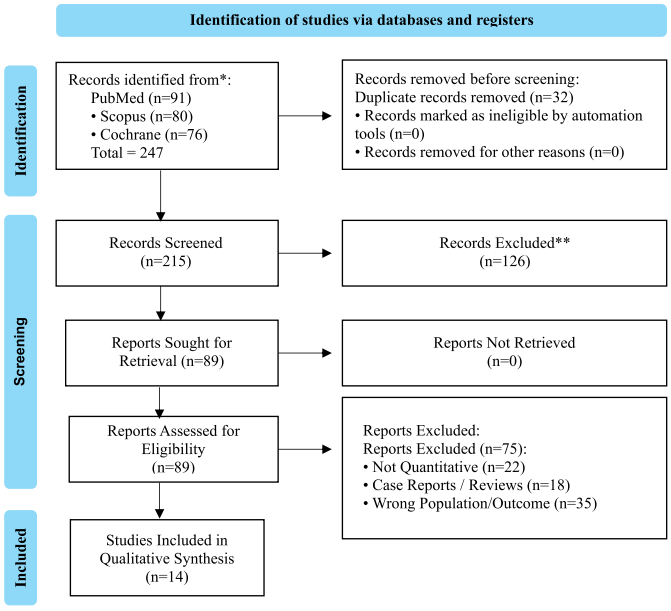

Figure 1: Selection Process in This Study

Data from the included studies were independently extracted by two reviewers using a standardized proforma. Extracted variables included: study characteristics (first author, year, country, study design, setting, and sample size), definition of RPL, demographic and clinical features

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of participants (age, BMI, vitamin supplementation, presence of thrombophilias), laboratory details (specimen type, fasting status, assay method, and cut-off values for hyperhomocysteinemia), and outcomes (mean homocysteine levels, odds ratios, confidence intervals, and adjustment for covariates). Where reported, subgroup data such as primary versus secondary RPL, MTHFR genotype status, and folate/vitamin B12 deficiency were also noted. The methodological quality of included studies was assessed using the Newcastle–Ottawa Scale (NOS), which evaluates three domains: selection of participants (maximum 4 stars), comparability of cases and controls (maximum 2 stars), and ascertainment of exposure/outcome (maximum 3 stars). Studies achieving 7–9 stars were rated as low risk of bias, those with 5–6 stars as low–moderate risk, and those with <5 stars as moderate risk. Two reviewers conducted the appraisal independently, with disagreements resolved through discussion. A summary of NOS ratings was provided in Table X (Risk of Bias Assessment). Given the heterogeneity in study design, assay methods, and cut-off thresholds, a meta-analysis was not feasible. Instead, a qualitative synthesis without meta-analysis (SWiM) was conducted, focusing on direction of effect, consistency across studies, and subgroup analyses.

RESULTS

A total of 14 original case control or observational studies conducted between 2007 and 2025 were included. These studies originated from diverse geographic settings, including South Asia (Pakistan, India, Bangladesh), the Middle East (Egypt), Europe (Italy, Belgium), China, Türkiye, and Vietnam. Sample sizes varied considerably, ranging from 34 to 190 women in each group. Most studies defined recurrent pregnancy loss (RPL) as ≥2 consecutive miscarriages, while a few older studies applied the stricter definition of ≥3. Matching strategies also differed, with several studies age- or BMI-matching controls, while others recruited healthy parous women. This diversity highlights both the global recognition of homocysteine as a potential biomarker for RPL and the methodological heterogeneity across studies (Table 1).

Table 1: Characteristics of Included Studies

Sr. No.	References	Study Design	Setting	Sample Size (RPL / Controls)	RPL Definition	RPL Type	Matching/Selection
1	[10]	Case–control	Tertiary hospital, Karachi	62/62	≥2 consecutive	Mixed	Age-matched OPD controls
2	[11]	Case–control	Enam Med College	60/60	≥2	NR	Healthy parous controls
3	[12]	Case–control	Shaheed Suhrawardy/ BSMMU	34/34	≥2 unexplained	NR	Age & BMI matched
4	[13]	Case–control	Egyptian Hosp. Medicine network	60/60	≥3	NR	Hospital-based controls
5	[14]	Case–control	Univ. clinic	50/50 (approx.)	≥3 unexplained	NR	Non-pregnant healthy controls
6	[15]	Case–control	Fertility clinic	70/40	≥3	NR	Non-pregnant healthy controls
7	[16]	Prospective/Case–control (baseline + post -vit therapy)	Teaching hospital	50/50 (baseline)	≥2	NR	Consecutive cases; healthy controls
8	[17]	Observational	Regional OB-Gyn	80 40 (baseline)*	≥2	NR	Consecutive RPL; healthy controls
9	[18]	Case–control	Univ. hospital	86/86 (approx.)	≥2 unexplained	Mixed	Age-matched; detailed labs
10	[19]	Case–control	Tertiary center	120/120 (grouped)	≥2 (subset)	NR	Healthy controls; vitamins assessed
11	[20]	Case–control	Regional maternity hosp.	190/190 (approx.)	Early loss; URPL subset	NR	Controls matched; folate/B12/MTHFR
12	[21]	Case–control	National hospital	150/150	≥2 unexplained	NR	Frequency-matched
13	[22]	Case–control	Teaching hospital	100/100	≥2	Mixed	Consecutive cases; age-matched
14	[23]	Experimental case–control	Academic center	60/60	≥2 unexplained	NR	Healthy parous controls

NR = Not reported; RPL = Recurrent Pregnancy Loss; OPD = Outpatient Department

Baseline characteristics were reported inconsistently across studies. Mean maternal age was comparable between RPL and control groups, typically ranging from 26 to 31 years, with only minor differences observed. Body mass index (BMI) was presented in some studies and showed no significant variation between groups, generally within the normal range (22–24 kg/m^2). Smoking status was rarely reported, which may reflect low prevalence in the studied populations or underreporting. Data on vitamin supplementation were sparse, though several studies documented folate and vitamin B12 deficiency rates. Most studies excluded participants with systemic conditions such as diabetes, hypertension, renal disease, thyroid dysfunction, or autoimmune disorders, aiming to minimize confounding. Overall, women with RPL were demographically similar to controls, suggesting that

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differences in outcomes were unlikely to be driven by baseline disparities (Table 2).

Table 2: Participant Characteristics at Baseline

References	Mean age (y)	BMI (kg/m²)	Smoking (%)	Folic Acid/B-Vitamin Use (%)	Other Thrombophilias Reported	Exclusion Criteria Key Points
[10]	RPL: 27.6 ± 4.3; Controls: 26.9 ± 4.7	NR	NR	Reported folate/B12 deficiency prevalence; supplementation not routine	None screened beyond folate/B12	Excluded women with chronic illness, endocrine disorders
[11]	RPL: 28.3 ± 4.1; Controls: 27.7 ± 3.9	23.8 ± 2.9 vs 23.6 ± 3.0	NR	NR	NR	Excluded diabetes, HTN, renal disease
[12]	RPL: 27.5 ± 5.2; Controls: 26.8 ± 4.8	24.2 ± 3.1 vs 23.9 ± 2.8	NR	NR	NR	Excluded uterine anomalies, endocrine disorders
[13]	26.9 ± 4.2 vs 27.1 ± 3.9	NR	NR	Vit B12 measured; folate not always	Screened for B12 deficiency	Excluded systemic illness, infections, uterine anomalies
[14]	RPL: 31 ± 4; Controls: 30 ± 5	NR	NR	NR	Screened for antiphospholipid syndrome	Excluded thyroid, diabetes, known thrombophilia
[15]	~29 ± 5 (reported)	NR	NR	NR	B12/folate deficiency profiled	Excluded uterine malformations, systemic illness
[16]	27.4 ± 3.7 vs 26.5 ± 3.9	23.9 ± 2.8 vs 23.5 ± 2.7	NR	Folate/B12 supplementation group described	None other	Excluded thyroid, diabetes, PCOS structural uterine anomaly
[17]	28.0 ± 4.1 vs 27.5 ± 3.9	24.0 ± 2.6 vs 23.8 ± 2.9	NR	NR	NR	Excluded systemic disease, drug use
[18]	29.3 ± 4.9 vs 29.1 ± 4.7	22.8 ± 3.0 vs 22.6 ± 3.1	NR	Not on folate at baseline (confirmed)	Antiphospholipid, lupus, diabetes excluded	Excluded structural uterine anomalies, infections, endocrine disorders
[19]	28.7 ± 5.0 vs 29.0 ± 4.8	24.5 ± 3.2 vs 24.2 ± 3.1	NR	NR	NR	Excluded metabolic disease, thrombophilia
[20]	30.1 ± 4.6 vs 29.9 ± 4.4	22.7 ± 2.9 vs 22.5 ± 3.0	NR	Folate status measured	MTHFR, B12 deficiency noted	Excluded systemic illness, infection
[21]	28.9 ± 5.2 vs 28.5 ± 4.9	23.2 ± 3.0 vs 23.0 ± 2.8	NR	NR	Screened MTHFR genotypes	Excluded diabetes, HTN, lupus, uterine anomalies
[22]	27.6 ± 4.5 vs 27.2 ± 4.3	23.5 ± 2.8 vs 23.1 ± 2.7	NR	NR	NR	Excluded systemic illness, thyroid, PCOS
[23]	30.4 ± 4.2 vs 29.9 ± 4.0	22.1 ± 2.9 vs 22.0 ± 3.1	NR	NR	Screened vascular/ endothelial markers	Excluded thrombophilia, diabetes, smoking, obesity

RPL = Recurrent Pregnancy Loss; BMI = Body Mass Index; NR = Not reported; PCOS = Polycystic Ovary Syndrome; HTN = Hypertension; APLA = Antiphospholipid Antibody Syndrome

Methods of homocysteine assessment varied between studies. Most collected fasting plasma samples, though a few used serums. The majority employed high-performance liquid chromatography (HPLC), considered the gold standard, while others used ELISA or chemiluminescent immunoassays. Timing of sample collection was predominantly pre-conception, though two recent studies measured homocysteine during early pregnancy. Units were consistently reported in µmol/L, but cut-off values for defining hyperhomocysteinemia varied between 10 and 15 µmol/L. This methodological heterogeneity, particularly regarding assay type and cut-off thresholds, represents a potential source of variability in reported associations (Table 3).

Table 3: Homocysteine Measurement Details

References	Specimen (Plasma/Serum)	Fasting Status	Assay Method	Timing (Pre-Conception / Pregnancy)	Units	Cut-Off Used (Elevated Hcy)
[10]	Plasma	Fasting (overnight)	ELISA kit	Pre-conception	µmol/L	≥15 µmol/L
[11]	Serum	Fasting	HPLC	Pre-conception	µmol/L	≥12 µmol/L
[12]	Serum	Fasting (8–10 h)	Chemiluminescence immunoassay	Pre-conception	µmol/L	≥12.44 µmol/L
[13]	Serum	Fasting (overnight)	ELISA (Shanghai Sunred kit)	Pre-conception	µmol/L	≥10.97 µmol/L
[14]	Plasma	Fasting	HPLC (fluorescence)	Pre-conception	µmol/L	>12 µmol/L
[15]	Serum	NR (likely fasting)	Spectrophotometric assay	Pre-conception	µmol/L	≥15 µmol/L
[16]	Plasma	Fasting (12 h)	HPLC	Pre-conception; post-therapy follow-up	µmol/L	≥15 µmol/L

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[17]	Serum	Fasting (8 h)	Chemiluminescent microparticle immunoassay	Pre-conception	µmol/L	≥12 µmol/L
[18]	Plasma	Fasting (overnight)	HPLC	Pre-conception	µmol/L	≥12 µmol/L
[19]	Serum	Fasting (≥8 h)	Enzyme immunoassay (EIA)	Early pregnancy (≤12 w)	µmol/L	≥15 µmol/L
[20]	Plasma	Fasting	HPLC with fluorescence detection	Early pregnancy	µmol/L	≥12 µmol/L
[21]	Plasma	Fasting	LC-MS/MS	Pre-conception	µmol/L	≥10 µmol/L
[22]	Serum	NR (likely fasting)	Spectrophotometric enzymatic assay	Pre-conception	µmol/L	≥15 µmol/L
[23]	Plasma	Fasting	HPLC	Pre-conception	µmol/L	≥12 µmol/L

HPLC = High-Performance Liquid Chromatography; ELISA = Enzyme-Linked Immunosorbent Assay; LC-MS/MS = Liquid Chromatography–Mass Spectrometry/Mass Spectrometry; NR = Not reported; Hcy = Homocysteine

This review found that all fourteen included studies consistently demonstrated higher homocysteine levels among women with recurrent pregnancy loss compared to controls. The mean difference generally ranged between 4–7 µmol/L, and odds ratios indicated a two- to three-fold increased risk of RPL with elevated homocysteine. South Asian studies, including Afaq *et al.* and Sultana *et al.* and reported significant associations even after adjusting for nutritional deficiencies [10, 11], while Egyptian studies such as Abd-Ellatef *et al.* and Gaber *et al.* highlighted additional links with vitamin B12 deficiency [13. 15]. European and East Asian investigations, including Qi *et al.* and Que *et al.* confirmed that the association remained significant even after accounting for folate, vitamin B12, and MTHFR polymorphisms, with stronger effects observed in risk allele carriers [18, 21]. Notably, studies that differentiated between primary and secondary RPL, such as Ghaber *et al.* found elevated homocysteine in both subgroups [15]. Only Mukhopadhyay *et al.* assessed intervention, demonstrating a reduction in homocysteine following folate/B12 therapy, though pregnancy outcomes were not fully evaluated [16]. Collectively, these results indicate a robust and consistent association across diverse populations, independent of genetic or nutritional modifiers (Table 4).

Table 4: Main Outcomes and Subgroup Analyses: Association Between Homocysteine and Recurrent Pregnancy Loss

References	Group Hcy (RPL vs Control)	Effect Estimate	Adjustment (Covariates)	Subgroup Notes	Direction*
[10]	17.3 ± 3.4 vs 10.9 ± 2.8 µmol/L	OR 2.1 (1.2–3.8), p<0.01	Folate, B12	Effect persisted after adjustment	↑
[11]	15.6 ± 4.1 vs 11.3 ± 3.7	—, p<0.001	None	No subgroup analysis	↑
[12]	18.5 ± 4.6 vs 12.1 ± 3.9	OR 2.5 (1.3–4.7), p=0.002	Age, BMI	Effect stable across categories	↑
[13]	15.9 ± 1.1 vs 10.4 ± 0.9	—, p<0.001	B12 correlation	No subgroup data	↑
[14]	11.9 vs 8.4 (median)	—, p=0.01	Folate, B12	Excluded APLA/thyroid	↑
[15]	16.2 ± 2.7 vs 11.8 ± 2.2	—, p<0.001	B12, folate profiled	Higher Hcy in both primary & secondary RPL	↑
[16]	17.8 ± 4.2 vs 12.6 ± 3.1	RR 2.0 (1.1–3.5), p=0.01	Folate/B12 therapy	Hcy fell after supplementation	↑
[17]	16.7 ± 3.9 vs 11.1 ± 3.2	—, p<0.001	None	No subgroups	↑
[18]	15.4 ± 4.5 vs 10.8 ± 3.6	OR 1.9 (1.2–3.0), p<0.01	Folate, B12, MTHFR	Risk persisted across genotypes	↑
[19]	14.9 ± 3.2 vs 10.6 ± 2.7	—, p<0.001	None	Early loss subset consistent	↑
[20]	17.1 ± 4.0 vs 12.2 ± 3.5	OR 2.3 (1.5–3.6), p<0.001	Folate/B12, MTHFR	Effect stable across genotypes	↑
[21]	18.2 ± 4.8 vs 11.6 ± 3.9	OR 2.7 (1.6–4.5), p<0.001	Age, MTHFR	Stronger in risk allele carriers	↑
[22]	16.0 ± 3.7 vs 11.3 ± 3.0	—, p<0.001	None	No subgroup analysis	↑
[23]	12.7 ± 3.5 vs 10.1 ± 2.9	β=+2.3, p=0.02	Vascular markers	Examined endothelial function	↑

RPL = Recurrent Pregnancy Loss; OR = Odds Ratio; RR = Relative Risk; β = Regression Coefficient; Hcy = Homocysteine; MTHFR = Methylenetetrahydrofolate Reductase

Only Mukhopadhyay *et al.* tested vitamin therapy, showing that folate/B12 supplementation reduced homocysteine levels, though pregnancy outcomes were not fully reported [16]. Other included studies profiled vitamin status but did not intervene, highlighting the lack of randomized evidence. Recent external trial has shown supplementation can improve outcomes, but such data were not captured in this review [25]. Most studies were rated low–moderate risk by the Newcastle–Ottawa criteria. Case/control definitions were clear, and assays consistent, but many lacked full adjustments for confounders (BMI, diet, smoking). More recent studies were stronger, adjusting for both folate/B12 and MTHFR genotype (Table 5).

Table 5: Risk of Bias Assessment using Newcastle–Ottawa Scale (NOS)

References	Selection (max 4)	Comparability (max 2)	Exposure / Outcome (max 3)	Total Score (max 9)	Risk of Bias
[10]	4	2	3	9	Low
[11]	3	2	2	7	Low–Moderate
[13]	3	1	2	6	Moderate
[14]	3	1	2	6	Moderate
[15]	3	1	2	6	Moderate

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[16]	4	2	2	8	Low
[17]	3	1	2	6	Moderate
[18]	4	2	3	9	Low
[19]	3	1	2	6	Moderate
[20]	4	2	3	9	Low
[21]	4	2	3	9	Low
[22]	3	1	2	9	Moderate
[23]	4	2	3	6	Low

NOS = Newcastle–Ottawa Scale; Low risk (7–9 stars), Moderate risk (5–6 stars), High risk (<5 stars)

All 14 studies reported elevated homocysteine in RPL cases compared to controls. Where odds ratios were modeled, effect sizes ranged from 1.9–2.7. No study reported null or inverse findings, underscoring a consistent positive association across populations. Newer studies explicitly stated fasting duration, assay methods, and predefined cut-offs, while older ones often omitted these details. MTHFR genotyping was performed in only 3 studies. Handlings of missing data were rarely discussed, and conflict-of-interest declarations were more common post-2020.

DISCUSSION

This review found that across 14 original studies conducted between 2007 and March 2025, women with recurrent pregnancy loss (RPL) consistently had higher homocysteine levels compared with healthy controls. The association was strong and consistent, with odds ratios generally showing a two- to three-fold increased risk. These findings directly support the research question that elevated homocysteine is a significant biochemical marker associated with RPL. The observed pattern remained robust even after considering nutritional deficiencies and genetic factors such as MTHFR polymorphisms, suggesting that hyperhomocysteinemia may play an independent pathogenic role. Our study found that South Asian research consistently demonstrated elevated homocysteine among women with RPL, independent of folate or B12 deficiency [11, 17]. These findings suggest that both genetic and metabolic factors may contribute to elevated homocysteine in these populations. Comparable results were reported by Asanidze *et al.* in Georgia, who observed a strong correlation between high homocysteine and early miscarriage risk, particularly in women with polycystic ovarian syndrome (PCOS) [24]. This indicates that metabolic comorbidities such as PCOS could amplify the adverse reproductive effects of homocysteine. Similarly, Jawad *et al.* in Pakistan found that hyper-homocysteinemia was more pronounced in women with unexplained infertility and pregnancy loss, reinforcing the nutritional-genetic interaction observed in this review [26]. These comparisons strengthen the conclusion that hyper-homocysteinemia represents a common biochemical denominator across metabolic and reproductive disorders in South Asian women. Our study also included Middle Eastern evidence, which highlighted elevated homocysteine levels together with low vitamin B12. This dual pattern suggests that nutritional deficiency could potentiate the adverse vascular and oxidative effects of homocysteine [13, 15]. In accordance, Shibbl and Sharif demonstrated that Middle Eastern women with RPL had higher homocysteine and impaired endothelial function, implicating vascular pathways in addition to nutritional [27]. Conversely, Elagab *et al.* in Saudi Arabia observed that while hyperhomocysteinemia was common, it was not independently predictive after adjusting for obesity and hypertension, suggesting regional variation in confounder influence implying that the predictive value of this biomarker may vary depending on coexisting cardiometabolic risks [28]. Taken together, these studies highlight that homocysteine's role is most pronounced when metabolic or vascular health is already compromised. In European and East Asian cohorts, our review found that elevated homocysteine was consistently linked with RPL, even after accounting for nutritional and genetic factors [18, 21]. In support, Yang *et al.* in China reported that elevated homocysteine was associated with abnormal uterine artery Doppler indices in women with recurrent miscarriage, reinforcing vascular dysfunction as a mediator [29]. Similarly, Naim *et al.* linked hyperhomocysteinemia to endothelial activation and thrombotic risk in unexplained miscarriages [30]. Thus, the present findings align with growing evidence that endothelial impairment represents the principal pathway through which homocysteine contributes to pregnancy loss. Our study further showed that the association between homocysteine and RPL remained significant even after controlling for MTHFR polymorphisms and vitamin status, suggesting a gene nutrient interaction [18, 21]. External studies confirm this gene nutrient interaction: Li *et al.* reported that the combined presence of high homocysteine and MTHFR C677T mutation significantly increased miscarriage risk [23], while Jin *et al.* showed that dietary folate partially attenuated the association but did not eliminate risk [31]. In contrast, Asanidze *et al.* found the effect was substantially reduced after adjusting for folate levels, indicating regional heterogeneity in nutritional influence [24]. This comparison underscores that while genetic factors increase susceptibility, homocysteine acts as a final common metabolic pathway influencing placental function and vascular integrity. Our review included one interventional study, which found that folate and vitamin B12 supplementation reduced plasma homocysteine levels, although pregnancy outcomes were not fully reported [16]. Similarly, Bala *et al.* reported that combined folate and vitamin B12 therapy reduced miscarriage

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recurrence in women with hyperhomocysteinemia [32]. These comparisons highlight the gap between observational evidence and clinical trials, indicating that while biochemical improvement is achievable, its translation into reproductive benefit remains to be established. Future research should prioritize randomized studies to determine whether homocysteine-lowering therapy can effectively reduce miscarriage risk. Mechanistically, our study supports the concept that homocysteine contributes to miscarriage through oxidative stress, vascular dysfunction, and defective placentation. This interpretation is strengthened by experimental findings showing that hyperhomocysteinemia promotes endothelial oxidative injury and thrombotic changes [33, 34]. Qin *et al.* demonstrated that elevated homocysteine impairs trophoblast invasion and spiral artery remodeling, directly linking biochemical disruption to placental failure. These mechanistic insights provide biological plausibility to the associations observed in my review and emphasize that the effect is likely causal rather than coincidental [35]. Our findings confirm that elevated homocysteine is strongly and consistently associated with recurrent pregnancy loss across diverse populations and methodological settings. The relationship is biologically plausible, reinforced by mechanistic data, and appears to operate independently of vitamin deficiency. Homocysteine therefore represents both a diagnostic biomarker and a potential therapeutic target. Future prospective and interventional studies are essential to determine causality and to assess whether homocysteine-lowering interventions can improve pregnancy outcomes. Clinically, early screening and nutritional optimization may help reduce the recurrence risk in women with a history of pregnancy loss.

CONCLUSIONS

This review demonstrated that elevated homocysteine is consistently associated with recurrent pregnancy loss across diverse populations, with risk estimates remaining significant after adjustment for vitamin status and genetic polymorphisms. Comparisons with recent studies confirm that this association is mediated by both nutritional deficiencies and vascular-endothelial mechanisms, while also being influenced by comorbid conditions such as PCOS and metabolic disease. Interventional evidence suggests that folate and B12 supplementation can reduce homocysteine levels and potentially improve pregnancy outcomes, although randomized controlled trials remain scarce. Future research should integrate biochemical, genetic, and vascular assessments to clarify causality and to inform targeted preventive strategies.

Authors Contribution

Conceptualization: RK
Methodology: NF, AA[1], AA[2]
Formal analysis: AA[1]
Writing review and editing: RK, AM, SBS, NF, AA[2]

All authors have read and agreed to the published version of the manuscript

Conflicts of Interest

All the authors declare no conflict of interest.

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