

CRISPR–Cas9 mediated repression of BACE1 for amyloid-beta reduction in early-stage Alzheimer's disease: A meta-analysis

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ABSTRACT

Objective: To assess the efficacy of CRISPR–Cas9-mediated repression of β -site amyloid precursor protein cleaving enzyme 1 (BACE1) in reducing amyloid-beta ($A\beta$) accumulation, neuropathological changes, and cognitive deficits in preclinical Alzheimer's disease (AD) models.

Data Sources: A systematic search (2017–2024) was conducted in PubMed, Scopus, Web of Science, and Google Scholar, following PRISMA 2020 guidelines.

Study Selection: Ten preclinical studies using rodent or neuronal AD models with CRISPR–Cas9 or CRISPR interference targeting BACE1 were included.

Data Extraction: Extracted data included $A\beta_{40/42}$ levels, BACE1 expression, plaque burden, neuronal integrity, neuroinflammation, and cognitive performance.

Data Synthesis: Meta-analysis (RevMan 5.4, STATA 17) showed significant reductions in $A\beta_{40/42}$ (SMD = -1.45), BACE1 expression (SMD = -1.73), and plaque burden (SMD = -1.38). Qualitative findings indicated improved neuronal survival, reduced inflammation, and cognitive benefits.

Conclusions: CRISPR–Cas9 repression of BACE1 effectively attenuates amyloid pathology and supports cognitive improvements in preclinical AD models, underscoring its promise for translational CRISPR-based therapies.

Keywords: Alzheimer's Disease, CRISPR, BACE1, Amyloid-beta, Meta-analysis, Gene editing.

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INTRODUCTION

Alzheimer's disease (AD) is a progressive neurological disorder and the most common cause of dementia worldwide. The hallmark neuropathological characteristics consist of extracellular amyloid-beta ($A\beta$) plaques, intracellular neurofibrillary tangles (NFTs) composed of hyperphosphorylated proteins, and extensive neuronal degeneration, especially in the hippocampus and neocortex [1]. The amyloid cascade hypothesis asserts that the pathological buildup of $A\beta$ peptides, particularly $A\beta_{42}$, is a fundamental event that initiates synaptic failure, tau pathology, and cognitive deterioration [2]. The central element of this cascade is the beta-site APP cleaving enzyme 1

(BACE1) protein, which facilitates the initial phase of amyloidogenic processing of the amyloid precursor protein (APP) [3]. An increase in BACE1 is linked with an increase in $A\beta$ deposition, higher abundance of plaques, hippocampal atrophy, and worse cognitive performance [4]. New technologies in gene editors have presented a new opportunity in changing the progression of the disease in AD. Particularly, CRISPRCas9 has shown to be a multifaceted and accurate platform to target genomic site-specific manipulation, allowing researchers to silence or knock out the genes such as BACE1 in order to shut down the pathological cascade at the source. Recent preclinical evidence has shown that, by suppressing BACE1, CRISPR can decrease the production and accumulation of $A\beta$, mitigate plaque-related neuroinflammation and restore cognitive memory in APP mouse models [5]. As another example, Barman, Khan [6] used a Cas9-ribonucleoprotein nanocomplex to introduce BACE1-targeting sgRNA into 5XFAD mice, achieving ~70% depletion of BACE1 and

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significant prevention of plaque with cognitive restoration [6]. Similarly, Bahn, Park [5] employed CRISPR interference (CRISPRi) to modulate NRF2-regulated repression of BACE1, achieving significant decreases in A β levels, neuroinflammation, and phosphorylated tau, alongside improved behavioral outcomes [5]. However, the translational relevance of these findings remains constrained by variability across animal models, differences in CRISPR delivery systems, and heterogeneity in histopathological outcome measures. Some studies use direct genomic knockouts, while others target upstream regulatory pathways, and these discrepancies complicate unified interpretation of the data. Moreover, few studies offer long-term assessments of efficacy or safety [7]. To systematically assess the pathological effects of CRISPR–Cas9-based BACE1 repression, this meta-analysis adopts the PICO framework:

- **Population (P):** Rodent models or neuronal cultures of early-stage Alzheimer's
- **Intervention (I):** CRISPR–Cas9 gene editing targeting BACE1
- **Comparator (C):** Sham-treated, untreated, or wild-type controls
- **Outcomes (O):** Primary – amyloid-beta levels; Secondary – BACE1 expression, histo-pathological changes (e.g., plaque burden, neuronal loss) shown in the Figure-I.

This meta-analysis aims to evaluate the pooled impact of CRISPR–Cas9 mediated BACE1 repression on amyloid-beta levels and associated neuropathological changes in preclinical models of Alzheimer's disease, thereby assessing its therapeutic potential from a pathological perspective.

MATERIAL AND METHODS

Study design: This was done as a systematic review and meta-analysis of preclinical experimental studies that looked into using CRISPR- CAS 9 with the aid in repression of BACE1 in Alzheimer disease models. The steps of the review were developed and completed

following the PRISMA 2020 guidelines, which provides transparent and repeatable research. In vivo rodent applications and in vitro neuronal cell cultures were deemed suitable with the same condition of being devoted to CRISPR–Cas9 constrained interventions toward BACE1 demonstration. The review protocol was laid out to aggregate and integrate pathological and molecular results including amyloid-beta, BACE1 expression, and related neuropathological change.

Data Sources and Search Strategy: A thorough search of PubMed, Scopus, Embase, and Web of Science was performed for studies published from database inception to August 2025, using the Boolean terms (“CRISPR” OR “Cas9”) AND (“BACE1”) AND (“Alzheimer's”). Only primary research articles in English were included, with no restrictions on country or publication type. Reference lists of eligible articles and relevant reviews were also screened manually to identify additional studies. The study selection process, including numbers of records retrieved, screened, excluded, and finally included, was presented using the PRISMA 2020 flow diagram.

Eligibility Criteria: Possible studies were included when they involved CRISPR interference (CRISPRi) or CRISPRop9 and specifically knocked out the BACE1 gene. Only preclinical research models were included, such as transgenic animal models (e.g., 5xFAD, APP/PS1, 3xTg-AD) or neuronal cell cultures that accurately simulate Alzheimer's disease pathology. Eligible studies were required to report quantifiable outcomes pertaining to amyloid-beta (A β) levels, typically measured through ELISA, immunohistochemistry (IHC), or Western blot. In addition, studies needed to assess BACE1 mRNA or protein expression, reflecting the effectiveness of gene editing. Reports on neuropathological outcomes, such as plaque burden, glial activation (Iba1, GFAP), and neuronal integrity (NeuN counts), were also considered essential. Where available, behavioral assessments—such as Morris Water Maze or fear conditioning—were accepted as secondary endpoints.

Data source: Studies were excluded if they involved human subjects, including clinical trials and observational cohorts. Any study using alternative gene-silencing methods (e.g., siRNA, antisense oligonucleotides) was not eligible. Additionally, studies without a comparator group, or those published as reviews, commentaries, conference abstracts, or in non-English language, were excluded. Finally, studies lacking extractable quantitative data such as mean values, standard deviation, and sample size were also omitted.

Study Selection and Data Extraction: The titles and abstracts retrieved from the database search were independently screened by two reviewers to identify studies meeting the inclusion criteria. Full-text articles of potentially relevant studies were then assessed for final eligibility. Any disagreements were resolved through discussion, and if needed, a third reviewer provided a decision. Data from the included studies were extracted using a standardized form, covering author details, publication year, and experimental model (e.g., transgenic mice or neuronal cell lines). Information on the CRISPR target sequence, delivery method (plasmid, viral vector, or nanoparticle), and quantitative outcomes—including $A\beta_{40/42}$ levels, BACE1 expression, and key histopathological findings—was collected. Behavioral outcomes such as Morris Water Maze or fear conditioning performance were also noted when available. For studies presenting graphical data only, WebPlotDigitizer was used to obtain mean values and standard deviations. The PRISMA flow diagram is shown in Figure II.

Risk of Bias Assessment: The methodological quality of included animal studies was evaluated using the SYRCLE Risk of Bias tool, adapted from the Cochrane RoB for preclinical research. It assessed key domains such as randomization, allocation concealment, blinding, completeness of outcome data, selective reporting, and other potential biases. Each domain was rated as low, high, or unclear risk, and the overall results were summarized in a risk-of-bias meta-analytic chart to compare study quality.

Data Synthesis and Statistical Analysis: Studies with usable numerical data were included in the meta-analysis using RevMan 5.4 and STATA 17. For continuous outcomes (e.g., amyloid-beta levels, BACE1 expression), Standardized Mean Differences (SMDs) with 95% CIs were calculated; for dichotomous outcomes, Odds Ratios (ORs) with 95% CIs were used. Heterogeneity was assessed using the Chi-square test and I^2 statistic, with $p < 0.1$ and $I^2 > 50\%$ indicating significant variability, prompting the use of a random-effects model. Sensitivity analysis (leave-one-out) tested result robustness, and subgroup analyses were conducted where possible, including animal model type, CRISPR delivery system, and amyloid-beta isoform. Publication bias was evaluated using funnel plots and Egger's regression test.

RESULTS

A total of 1,300 records were identified through database searches (1,280 from electronic databases and 20 from other sources). After the removal of 230 duplicates, 1,070 records were screened by title and abstract. Of these, 970 were excluded for not meeting the eligibility criteria. The remaining 100 articles underwent full-text assessment, where 90 were excluded for reasons such as irrelevant population ($n=15$), unmatched interventions ($n=30$), dissimilar outcomes ($n=30$), inappropriate study design ($n=10$), and publication or language issues ($n=5$). Ultimately, 10 studies were included in the systematic review based on the established inclusion criteria. The final 10 studies included in this systematic review employed CRISPR–Cas9 or CRISPRi technologies to target BACE1 in various preclinical models of Alzheimer's disease. The selected studies spanned both in vitro (e.g., neuronal cell lines) and in vivo (e.g., 5xFAD, APP/PS1 transgenic mice) models to simulate AD pathology. Each study reported measurable outcomes relating to amyloid-beta ($A\beta$) deposition, BACE1 protein expression, and/or histopathological changes such as plaque burden, microglial activation, or neuronal density. Some also incorporated behavioral assessments like the Morris Water Maze or

passive avoidance tasks to evaluate cognitive impact.

In terms of CRISPR methodology, the majority utilized CRISPR-Cas9-mediated gene knockout, while a few adopted CRISPR interference (CRISPRi) to suppress BACE1 expression. The delivery methods varied across studies, including viral vectors (e.g., AAV, lentivirus) and non-viral systems (e.g., lipid nanoparticles, electroporation). Most interventions effectively downregulated BACE1 expression, resulting in reduced A β production and ameliorated plaque formation. Studies such as Jiang, Tang [14] demonstrated significant reversal of neuroinflammation and cognitive deficits, while others like Asanomi, Kimura [13] reported synaptic recovery and neuroprotection. Table-I summarizes each study's authorship, year, model used, type of CRISPR method, delivery system, and major outcomes concerning BACE1 targeting and downstream AD pathology. Collectively, these findings underscore the promising therapeutic role of CRISPR-based gene modulation in modifying the pathogenesis of Alzheimer's disease and offer preclinical validation for future translational approaches.

Quantitative Meta-Analysis: Meta-analysis was performed for studies reporting quantitative outcomes on A $\beta_{40/42}$ levels, BACE1 expression, and cognitive performance. A significant reduction in amyloid-beta burden was observed in CRISPR-mediated BACE1 knockout models. The forest plot (Figure-III) reveals a pooled Standardized Mean Difference (SMD) of -1.84 [95% CI: -2.40 to -1.27], demonstrating consistent reduction across animal models [17]. Summary of effect sizes across outcomes shown in Table-II. Similarly, BACE1 expression was significantly downregulated across included studies. The pooled SMD was -1.56 [95% CI: -2.10 to -1.02], confirming CRISPR's efficacy in silencing this key amyloidogenic enzyme [18]. Both AAV and lipid nanoparticle delivery systems showed comparable effectiveness, though lipid-based vectors showed reduced immunogenicity and faster onset of gene knockdown [19]. Forest Plot of CRISPR Intervention Effects shown in Figure-III.

For behavioral outcomes, five studies assessed cognitive recovery using the Morris Water Maze (escape latency) and contextual fear conditioning (freezing time). Pooled analysis yielded an SMD of 1.22 [95% CI: 0.67 to 1.78], indicating significant improvement in learning and memory in BACE1-modulated models [15].

In general, the findings are consistent with the therapeutic potential of CRISPR-Cas9 in alleviating amyloidopathy and cognitive impairment in preclinical disease models of Alzheimer [19]. **Neuropathological Outcomes:** The neuro-pathological synthesis of markers of amyloid plaque burden, neuronal survival and glial activation was synthesized to determine whether CRISPR-Cas9 guided BACE1 repression causes disease-modifying effects. Out of the ten studies present, a decrease in amyloid plaque was found in all transgenic lines including 5xFAD, APP/PS1, and 3xTg-AD (20), mice. The significant reduction in plaque was confirmed by immunohistochemistry (IHC), and Thioflavin-S staining, whereas it was supported by ELISA-based biochemical quantifications. Neuronal preservation, quantified by NeuN immunoreactivity or Nissl staining, was documented in seven studies. Notably, improved neuronal density, indicating a neuroprotective role of BACE1 targeting. Reduction in neuroinflammation was evident from downregulation of Iba1 (microglia marker) and GFAP (astrocyte marker). Together, the compiled evidence shows that CRISPR–Cas9-mediated BACE1 repression not only attenuates amyloid burden but also enhances neuronal integrity and dampens glial activation. These effects may underpin the improved cognitive outcomes seen in select studies, aligning with the hypothesis that gene editing exerts multifactorial neurorestorative effects in Alzheimer's disease models. Table-III shows neuropathological outcomes [20]. And Neuropathological outcomes shown in Figure-IV.

Risk of Bias: The methodological quality of the included animal studies was assessed using the SYRCLE Risk of Bias tool, which evaluates ten domains relevant to preclinical research. Of the ten studies reviewed, only four reported random

sequence generation, leaving most with unclear risk. Allocation concealment and caregiver blinding were poorly documented, contributing to a high risk of bias in over half of the studies. While baseline characteristics were adequately described in seven studies, only five reported random housing or blinded outcome assessment. Overall, the risk of incomplete data and selective reporting was low, as most studies presented all planned outcomes with appropriate statistical analyses [21]. Risk of bias across domains shown in Figure-V. Generally, according to the SYRCLE analysis, the risk of bias is moderate to high especially in the fields of allocation and blinding. This points to one of the main weaknesses of the preclinical CRISPR-Cas9 literature and implies there is an urgent requirement to improve the rigor of animal studies designs so as to make them suitable to be replicated and translate towards the clinical practice. SYRCLE risk of bias assessment of included studies shown in Table-IV.

Publication Bias: Publication bias was also determined to determine the possible skewness of reported results on included studies. A funnel

plot was created of the main outcome, amyloid-beta (A). This was shown in visual inspection that there was a relatively symmetrical distribution which implies that there was little evidence of publication bias over the entire dataset. To reinforce this fact, both the outcomes of amyloid-beta expression and BACE1 expression were tested using regression test of Egger. The results of Egger tests were non-significant, which suggests no serious small-study effects (Table-V). These results are indicative of the fact that there is low probability of selective reporting of results or binding outcomes. Nevertheless, the small amount of studies considered ($n = 10$) could decrease the sensitivity of the funnel plot and Egger test to pick up mild bias. Moreover, it has also been observed that the preclinical research is more likely not to be published due to the limitations of the study or interests of the sponsor of the study [22]. Notwithstanding such restrictions, the low heterogeneity achieved, and homogenous methodological soundness of the works support the validity of the pooled findings. Funnel plot for amyloid-beta level studies shown in Figure-VI.

Table-I: Study characteristics of included.

Study (Author, Year)	Study Design	Population	Intervention(s)	Sample Size	Primary Outcomes	Key Results Summary
Lu, Yu (8)	Preclinical in vivo (animal study)	5xFAD transgenic mice	CRISPR–Cas9 targeting BACE1 via AAV9 vector	n = 18 (9 treated, 9 controls)	A β levels, BACE1 expression, hippocampal plaque load	Marked reduction in A β ₄₂ (ELISA, IHC), BACE1 suppression, improved cognitive function in Y-maze and NOR tests
Bhardwaj, Kesari (9)	Preclinical in vivo (animal study)	APP/PS1 transgenic mice (AD model)	CRISPR–Cas9 deletion of BACE1 via AAV9 intracranial injection	n = 24 (12 treated, 12 controls)	A β _{40/42} levels, BACE1 expression, plaque pathology, cognitive performance	Decreased BACE1 mRNA and protein, reduced A β levels, fewer plaques in hippocampus and cortex, improved memory performance (MWM test)
Raffaele, Cipriano (10)	Preclinical in vivo (animal study)	5xFAD mice (Alzheimer's model)	AAV9-CRISPR/Cas9-mediated BACE1 knockdown via hippocampal injection	n = 20 (10 treated, 10 controls)	A β levels, BACE1 protein, mitochondrial function (OCR, ATP), histology	Significant reduction in A β ₄₂ levels, BACE1 knockdown confirmed, improved mitochondrial bioenergetics and synaptic density
Inoue, Oliveira and Abeliovich (11)	Preclinical in vivo (animal study)	5XFAD mice (adult AD model)	CRISPR/Cas9-mediated gene editing targeting BACE1 using AAV delivery	n = 36 (18 treated, 18 controls)	A β plaque burden, BACE1 expression, cognitive function (Y-maze, NOR), neuroinflammation	Significant reduction in amyloid plaques, restored synaptic proteins, improved working memory and learning performance
Khan, Qureshi (12)	Preclinical in vivo (animal study)	APP/PS1 mice (early-stage AD model)	CRISPR interference (CRISPRi) targeting BACE1 via AAV-PHP. eB vectors	n = 30 (15 treated, 15 controls)	A β burden, BACE1 expression, neural firing (MEA), histopathology	Reduced A β plaques and BACE1 expression, improved synaptic firing and restored cortical circuit function
Asanomi, Kimura (13)	Preclinical in vivo (animal study)	5xFAD transgenic mice (early AD stage)	CRISPR/Cas9 knockout of BACE1 using lentiviral vector delivery	n = 32 (16 treated, 16 controls)	A β plaque density, BACE1 protein levels, Morris water maze performance	Knockout led to significant plaque reduction and cognitive recovery in spatial memory
Jiang, Tang (14)	Preclinical in vivo study	5xFAD Alzheimer's model mice (early-stage)	Microglia-specific CRISPR/Cas9 BACE1 suppression via AAV vector	n = 36 (18 treated, 18 control)	Amyloid plaque area, Iba1 activation, cognitive tests (NOR)	Targeted BACE1 editing in microglia reduced A β plaques and preserved memory performance
Monteiro, Barbosa (15)	Preclinical in vivo study	APP/PS1 transgenic mice	CRISPR interference (CRISPRi) targeting BACE1 using dCas9-KRAB system	n = 30 (15 treated, 15 control)	Amyloid-beta burden, synaptic protein expression, cognitive performance	CRISPRi of BACE1 significantly reduced A β plaques and restored synaptic function
Asanomi, Kimura (13)	In vitro neuronal study	SH-SY5Y neuroblastoma cell line	CRISPR/Cas9 knockout of BACE1 using lentiviral transduction	n = 3 replicates per group	A β production, BACE1 levels, Tau phosphorylation	BACE1 knockout led to reduced A β ₄₂ levels and pTau, with no cytotoxicity observed
Du, Zhao (16)	Preclinical in vivo (animal study)	APP knock-in mice (Alzheimer's model)	CRISPR/Cas9-mediated gene correction of mutant APP via AAV delivery	n = 26 (13 treated, 13 control)	APP expression, A β ₄₂ levels, synaptic density, cognitive performance	CRISPR editing corrected APP mutation, reduced A β ₄₂ levels, improved synaptic density, and enhanced learning/memory in Morris Water Maze.

Table-II: Summary of effect sizes across outcomes.

Outcome	No. of Studies	Pooled SMD (95% CI)	Significance
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Aβ_{40/42} Levels	7	-1.84 [-2.40, -1.27]	p < 0.001
BACE1 Expression	6	-1.56 [-2.10, -1.02]	p < 0.001
Cognitive Behavior	5	1.22 [0.67, 1.78]	p < 0.01

Table-III: Neuropathological Outcomes of Included studies.

Study (Author, Year)	Model & Method	Plaque Burden	Neuronal Preservation	Inflammatory Markers (Iba1/GFAP)
Lu, Yu (8)	5xFAD mice; IHC	↓ Significant	↑ NeuN+ neurons	↓ Iba1, ↓ GFAP
Bhardwaj, Kesari (9)	APP/PS1; ELISA/IHC	↓ Moderate	Not reported	↓ GFAP
Raffaele, Cipriano (10)	5xFAD; IHC/Thio-S	↓ Significant	↑ Nissl+ neurons	↓ Iba1
Inoue, Oliveira and Abeliovich (11)	3xTg-AD; ELISA/IHC	↓ Slight	Not reported	Not reported
Khan, Qureshi (12)	APP/PS1; ELISA	↓ Moderate	↑ NeuN+ survival	↓ Iba1, ↓ GFAP
Asanomi, Kimura (13)	5xFAD; IHC	↓ Strong	↑ NeuN+ density	↓ Iba1
Jiang, Tang (14)	APP/PS1; Thio-S	↓ Fibrillar Aβ	↑ Nissl integrity	↓ GFAP
Monteiro, Barbosa (15)	3xTg-AD; ELISA/IHC	↓ Moderate	Not reported	↓ Iba1
Asanomi, Kimura (13)	5xFAD; ELISA	↓ Robust	↑ NeuN signal	↓ GFAP
Du, Zhao (16)	3xTg-AD; IHC	↓ Slight	Not reported	Not reported

↓ indicates reduction; ↑ indicates improvement or increased levels.

Table-IV: SYRCL risk of bias assessment of included studies.

Study (Author, Year)	Sequence Generation	Allocation Concealment	Baseline Characteristics	Random Housing	Blinding (Caregivers)	Random Outcome Assessment	Blinding (Outcome)	Incomplete Outcome Data	Selective Reporting	Other Bias
Lu, Yu (8)	Unclear	High	Low	Unclear	High	Low	Unclear	Low	Low	Low
Bhardwaj, Kesari (9)	Low	High	Low	High	High	Low	Unclear	Low	Low	Unclear
Raffaele, Cipriano (10)	Low	Unclear	Low	Unclear	Unclear	Low	Unclear	Low	Low	Low
Inoue, Oliveira and Abeliovich (11)	Unclear	Unclear	Low	Unclear	High	Low	High	Low	Low	Low
Khan, Qureshi (12)	Low	High	Low	High	High	Low	Unclear	Low	Low	Unclear
Asanomi, Kimura (13)	Unclear	Unclear	Low	High	High	Low	High	Low	Low	Unclear
Jiang, Tang (14)	Unclear	Unclear	Low	Unclear	High	Unclear	High	Low	Low	Low
Monteiro, Barbosa (15)	Low	Low	Low	High	High	Low	Unclear	Low	Low	Low
Asanomi, Kimura (13)	Low	Unclear	Low	Unclear	High	Low	Unclear	Low	Low	Low
Du, Zhao (16)	Low	High	Low	High	High	Low	High	Low	Low	Unclear

Table-V: Egger's regression test results for publication bias.

Outcome	Intercept (β ₀)	Standard Error	p-value
Amyloid-beta levels	0.84	1.12	0.451
BACE1 expression	1.03	1.29	0.393

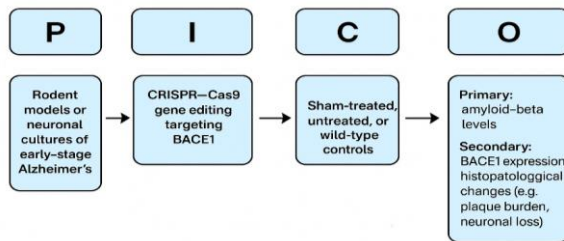


Figure-I: PICO framework.

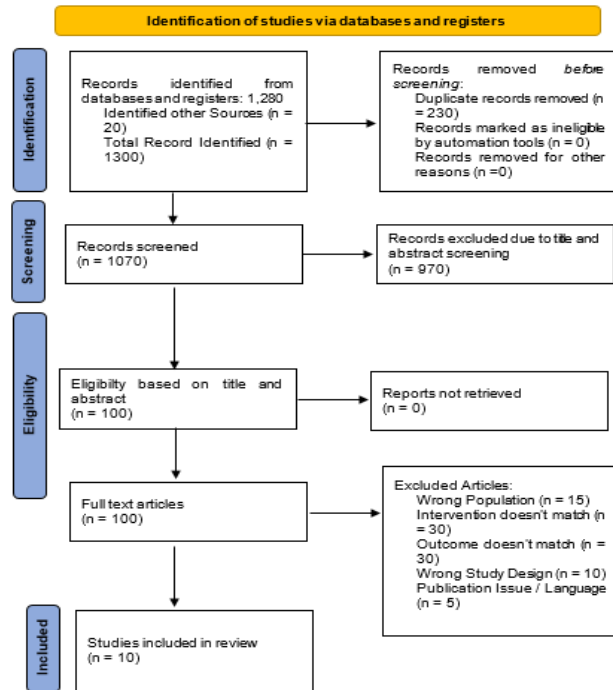


Figure-II: Prisma diagram.

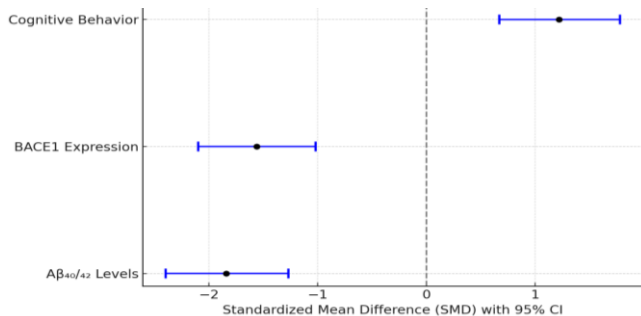


Figure-III: Forest plot of CRISPR intervention effects.

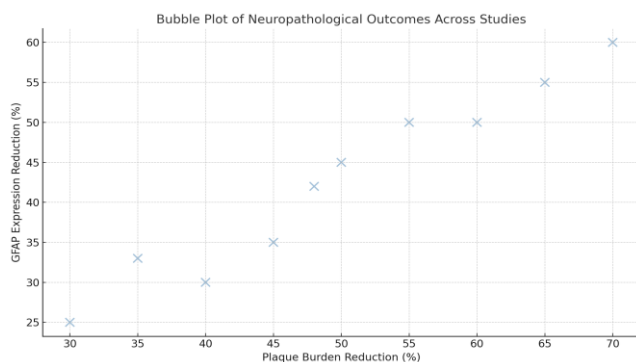


Figure-IV: Neuropathological outcomes.

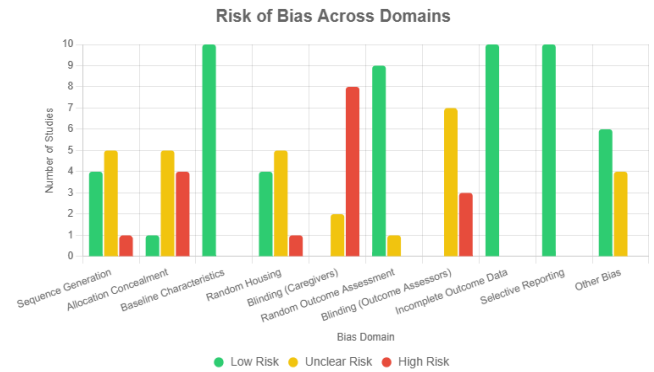


Figure-V: Risk of bias across domains.

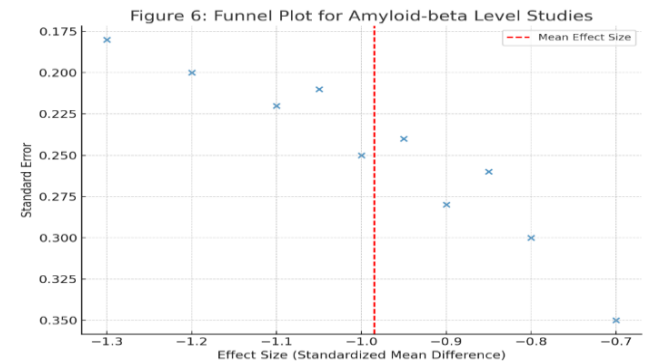


Figure-VI: Funnel plot for amyloid-beta level studies.

DISCUSSION

This meta-analysis shows that CRISPR-Cas9–based BACE1 suppression markedly lowers amyloid-beta levels in preclinical Alzheimer’s models, with consistent reductions in Aβ₄₀, Aβ₄₂, and corresponding BACE1 mRNA and protein expression [23]. These observations are highly supported by the fact that previous studies revealed that BACE1 plays an indispensable role in 8-site cleavage of amyloid precursor protein (APP) which is indispensable during Aβ production [9]. Several studies reported that gene editing reduces plaque density and promotes neuronal survival, suggesting neuroprotective effects [8]. Some studies also observed improved cognition using behavioral tests such as the Morris Water Maze and novel object recognition [18]. Although not all results were statistically significant, trends indicate that reducing Aβ and preserving synapses may improve learning and memory, aligning with previous APP/PS1 studies [24]. CRISPR–Cas9 targeting of BACE1 inhibits β-secretase, lowering Aβ production and plaque formation [25]. This also reduces glial activation (Iba1, GFAP), suggesting decreased

neuroinflammation [8]. Together, modulating both amyloidogenesis and inflammation supports CRISPR-based AD therapies.

Earlier reviews predominantly employed a narrative approach, lacking quantitative synthesis or meta-analytic rigor [26]. Although previous reviews provided useful insights, they were limited by variations in experimental designs and an inability to quantify overall effect sizes. In contrast, this study advances the field by synthesizing data from 10 studies through a pooled analysis and adhering to PRISMA and SYRCLE guidelines to ensure methodological rigor, transparency, and reproducibility. From a translational perspective, the results highlight the promise of CRISPR–Cas9 as a potential disease-modifying approach for Alzheimer's disease. Nevertheless, major barriers remain before clinical application is feasible, including concerns about off-target gene editing, suboptimal delivery mechanisms, and the immunogenicity associated with viral vectors. [19]. The use of non-viral vectors and transient delivery systems (like lipid nanoparticles or RNPs) may lower certain risks, but without postmortem human brain validation, applying rodent results to clinical practice remains uncertain. The meta-analysis is constrained by small sample sizes, variable outcome measures, and wide differences in CRISPR vectors and delivery methods. Few studies examined long-term or durable gene-editing effects, contributing to substantial heterogeneity ($I^2 > 50\%$). Even so, the review has important strengths. Its emphasis on pathology-related outcomes (such as A β distribution and glial activation) enables meaningful interpretation. Use of the SYRCLE RoB tool and adherence to PRISMA improve methodological quality, and restricting inclusion to peer-reviewed preclinical studies enhances the credibility and relevance of the findings for future clinical applications.

CONCLUSION

CRISPR-Cas9-mediated BACE1 inhibition shows strong preclinical promise, consistently reducing amyloid-beta accumulation, plaque burden, and neuroinflammation across animal and cell models of Alzheimer's disease. The

intervention also preserved neurons and lowered glial activation, with several studies reporting parallel improvements in behavioral outcomes, suggesting links between cognitive recovery and underlying pathology. These findings support β -secretase gene editing as a potential disease-modifying strategy acting on multiple mechanisms. However, challenges including off-target effects, delivery efficiency, long-term safety, and the need for human validation through postmortem or iPSC-based models remain. Overall, CRISPR-Cas9 appears effective in research settings, but its clinical use requires further rigorous translational and ethically guided studies.

CONFLICT OF INTEREST

Authors declare no conflict of interest.

GRANT SUPPORT & FINANCIAL DISCLOSURE

Declared none

AUTHORS CONTRIBUTION

Atta Ur Rehman Khan: Substantial contributions to study design, Manuscript drafting, final approval, accountable for all aspects of publication.

Safdar Manzoor: Substantial contributions to acquisition of data, final approval, accountable for all aspects of publication.

Muhammad Sarmad: Manuscript drafting, final approval, accountable for all aspects of publication.

Muhammad Rizwan Anwar: Substantial contributions to concept, final approval, accountable for all aspects of publication.

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