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INPLASY

Cytotoxicity of Bitter Compounds on Nasopharyngeal Carcinoma: A Systematic Review and Meta-Analysis of In Vitro Studies

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ADMINISTRATIVE INFORMATION

Support - No support.

Review Stage at time of this submission - Preliminary searches.

Conflicts of interest - None declared.

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Amendments - This protocol was registered with the International Platform of Registered Systematic Review and Meta-Analysis Protocols (INPLASY) on 17 July 2026 and was last updated on 17 July 2026.

INTRODUCTION

Review question / Objective PICO Framework
P (Population / Cell Lines): Human nasopharyngeal carcinoma (NPC) cell lines (including, but not limited to, HONE-1, SUNE-1, CNE-2, HK1, C666-1, and CNE-1).
I (Intervention / Exposure): Bitter compounds, bitter taste receptor (TAS2Rs/T2Rs) agonists, and bitter-tasting medicinal plants, plant components, or polyherbal formulations classified under traditional medicine theories (such as Traditional Thai Medicine, Traditional Chinese Medicine [TCM], or Ayurveda). This encompasses purified single molecules (e.g., Cucurbitacin B, Curcumin, Quinine, Andrographolide), standardized natural bitter extracts, and traditional bitter remedies evaluated for their single-agent cytotoxic actions. I (Intervention / Exposure): Bitter compounds and bitter taste receptor (TAS2Rs/T2Rs) agonists, including both purified single compounds (e.g., Cucurbitacin B, Curcumin, Quinine) and standardized natural bitter extracts (e.g., Bitter Melon Extract).
C (Comparison): Untreated control cells or vehicle control groups (e.g., cells treated with Dimethyl sulfoxide [DMSO] or culture media alone).
O (Outcome): Primary Outcomes: Half-maximal inhibitory concentration (\(IC_{50}\)) values and percentages of cell viability/proliferation inhibition.Secondary Outcomes: Apoptosis induction rates (%), or changes in downstream apoptotic/cytotoxic biomonitoring markers.

Rationale Nasopharyngeal carcinoma (NPC) remains a major health burden in Southeast Asia and Southern China. Current standard therapies, such as cisplatin-based chemotherapy and radiotherapy, are heavily constrained by severe systemic toxicities, profound taste alterations, and chemoresistance. Therefore, discovering novel, targeted, and biocompatible anticancer agents is a critical clinical priority. A promising yet under-explored frontier lies at the intersection of molecular oncology and traditional

ethnopharmacology: the targeting of extra-gustatory bitter taste receptors (TAS2Rs). Ectopically expressed on NPC cells, these receptors function as tumor suppressors. When activated by bitter agonists, they trigger intracellular signaling cascades, such as calcium influx and mitochondrial depolarization, that selectively induce cytotoxicity in cancer cells. Concurrently, traditional medicine systems (Traditional Thai, Chinese, and Ayurvedic medicine) have historically utilized bitter-tasting plants, specific plant components, and polyherbal formulations to treat deep-seated tumors and inflammatory conditions, classifying the "bitter taste" as a potent detoxifying and antipyretic property. Modern screenings reveal that these traditional remedies are rich in bioactive terpenoids, alkaloids, and flavonoids with proven in vitro cytotoxic actions against human NPC lines (like HONE-1, SUNE-1). Despite this dual evidence from modern biology and ancient medicine, the current research landscape is highly fragmented. Preclinical data are scattered across isolated, small-scale studies testing vastly different entities, ranging from purified single molecules to complex polyherbal mixtures. Furthermore, reported inhibitory concentrations (IC50) and cell viability outcomes vary widely due to methodological heterogeneity in exposure times, cell lines (e.g., EBV status), and assay types (MTT vs. CCK-8). To date, no study has statistically pooled and systematically synthesized these data. Consequently, this systematic review and meta-analysis are essential to bridge the gap between traditional flavor theories and modern receptor-targeted oncology. By mathematically consolidating available in vitro data, this study will establish a rigorous, evidence-based benchmark for the cytotoxic efficacy of bitter compounds and traditional formulations, effectively guiding future preclinical validation and translational drug discovery pipelines for NPC.

Condition being studied This study investigates the in vitro cytotoxicity and inhibition of cellular growth induced by bitter-tasting compounds against human nasopharyngeal carcinoma (NPC). The condition focuses on two interconnected biomedical and ethnopharmacological domains: The Oncological Profile: Human epithelial malignancies of the nasopharynx (Nasopharyngeal Carcinoma), characterized by distinct histopathological subtypes (keratinizing, non-keratinizing, and undifferentiated) and varied Epstein-Barr Virus (EBV) statuses, which influence cellular susceptibility to therapeutic agents. The Chemosensory Target and Agonists: The biological response of NPC cells upon exposure to bitter substances. This includes the pharmacological activation of extra-gustatory bitter taste receptors (TAS2Rs/T2Rs) expressed on cancer cells, as well as the broad cytotoxic evaluation of bitter-tasting medicinal plants, specific plant components, and traditional polyherbal formulations recognized within traditional medicine systems (such as Traditional Thai Medicine, Traditional Chinese Medicine, and Ayurveda). The primary parameters being evaluated are laboratory-measured markers of cell death, specifically pooled half-maximal inhibitory concentration (IC50) values, cell viability percentages, and cell proliferation rates.

METHODS

Search strategy 1. Databases to be Searched The systematic search will be comprehensively conducted across the following electronic databases from their inception to the present: PubMed (NLM), Scopus (Elsevier), Web of Science (Clarivate Analytics), and Cochrane Library
2. Search Term Formulation (Boolean Logic)The search strategy uses Boolean operators (AND, OR) and wildcard truncations (*) to combine three primary concept blocks: Block 1 (Intervention): Bitter compounds, bitter receptors, traditional bitter plants, and polyherbal traditional formulations.Block 2 (Population): Nasopharyngeal carcinoma and specific human NPC cell lines.Block 3 (Outcome): Quantitative cytotoxicity and cell viability metrics.
3. Full Comprehensive Search String (Generic Syntax)You can copy this complete string to paste into major databases (optimized for PubMed and Scopus): text("bitter compound*" OR "bitter phytochemical*" OR "bitter taste receptor*" OR "TAS2R*" OR "T2R*" OR "bitter agonist*" OR "bitter extract*" OR "bitter herb*" OR "bitter plant*" OR "bitter formulation*" OR "bitter remedy" OR "bitter remedies" OR "traditional medicine*" OR "polyherbal*" OR "herbal formula*" OR "ethnomedicine*" OR "traditional Thai medicine" OR "traditional Chinese medicine" OR "Ayurveda*") AND ("nasopharyngeal carcinoma" OR "nasopharyngeal cancer" OR "NPC" OR "HONE-1" OR "SUNE-1" OR "CNE-2" OR "HK1" OR "C666-1" OR "CNE-1") AND ("cytotoxicity" OR "cell viability" OR "cell proliferation" OR "inhibitory concentration" OR "IC50" OR "growth inhibition" OR "cell death")
4. Database-Specific Details & Search Filters Language Restrictions: None (to maximize the inclusion of localized traditional medicine studies, though articles must have at least an English abstract).Publication Period: From database inception up to the current date of search. Study Design Filters: No formal filters for study design

will be applied during the initial electronic search to prevent accidental omission of laboratory in vitro studies (screening will be performed manually).

Participant or population Since this is an in vitro systematic review and meta-analysis, the study population does not involve human patients or living participants. Instead, the established population encompasses human nasopharyngeal carcinoma (NPC) cell lines used in experimental laboratory models.
Included Cell Lines: The review will include all established and validated human epithelial nasopharyngeal carcinoma cell lines, representing various histopathological backgrounds and infection statuses, including but not limited to:
Epstein-Barr Virus (EBV)-negative lines: HONE-1, SUNE-1, CNE-1, CNE-2, HK-1, and TW01.
Epstein-Barr Virus (EBV)-positive lines: C666-1 (and any other authenticated EBV-positive NPC models).

Exclusion Criteria for Population: Non-human nasopharyngeal cell lines (e.g., murine or other animal models).Non-cancerous human nasopharyngeal epithelial cell lines (e.g., NP69) used as the sole study group (though studies using them as healthy comparative controls will be included).
Primary tumor tissues obtained directly from patients, unless the study explicitly establishes and characterizes corresponding in vitro continuous cell line passages from those tissues.

Intervention The interventions eligible for inclusion encompass natural or synthetic bitter taste receptor (TAS2Rs/T2Rs) agonists, bitter chemical compounds, and medicinal plants or polyherbal formulations defined as having a "bitter taste" property according to traditional medicine frameworks.
Eligible interventions are categorized into three distinct experimental groups: Purified Single Compounds / Agonists: Isolated natural or synthetic molecules known to possess a bitter taste or function as documented TAS2R agonists (e.g., alkaloids like Quinine; terpenoids like Andrographolide and Cucurbitacins; flavonoids like Xanthohumol).
Crude Extracts of Single Bitter Plants: Standardized aqueous, organic, or hydroalcoholic extracts derived from specific parts (leaves, roots, bark, fruits) of a single medicinal plant classified as bitter within traditional systems (e.g., Andrographis paniculata, Momordica charantia).
Traditional Polyherbal Formulations: Traditional multi-herb remedies or decoctions (from Traditional Thai Medicine, Traditional Chinese Medicine, Ayurveda, or other ethnomedical systems) that are explicitly documented as bitter-tasting formulations and administered as a single combinatory agent in the experiment.
Exclusion Criteria for Intervention: Interventions combining bitter compounds with standard chemotherapeutic drugs (e.g., Cisplatin, 5-Fluorouracil) where the independent effect of the bitter agent cannot be statistically isolated.
Extracts or formulations that are strictly documented as non-bitter (e.g., sweet, spicy, or sour properties) within traditional medicinal literature.
Homeopathic dilutions where the chemical concentration of the bitter substance is unquantifiable.

Comparator The eligible comparators or control groups for this in vitro meta-analysis comprise unexposed or vehicle-treated human nasopharyngeal carcinoma (NPC) cell lines evaluated under the same experimental conditions.
Eligible comparison groups must include: Vehicle Controls: NPC cells treated exclusively with the carrier solvent or vehicle used to dissolve the bitter compounds or traditional extracts (e.g., Dimethyl sulfoxide [DMSO], Ethanol, or Phosphate-Buffered Saline [PBS]) at an identical volume and concentration as the treatment group, without the active bitter ingredients.
Untreated Controls (Negative Controls): NPC cells maintained under identical culture conditions (media, temperature, and incubation period) but receiving no chemical exposure or active intervention whatsoever.
Exclusion Criteria for Comparator: Control groups that use different basal cell lines, incubation conditions, or non-identical exposure durations compared to the active intervention arm.Studies that present only baseline pre-treatment data without a concurrent, time-matched control group.

Study designs to be included This systematic review and meta-analysis will exclusively include controlled in vitro laboratory studies (experimental cell culture studies).Eligible study designs must meet the following criteria:Controlled Experimental Design: The study must feature concurrent comparative arms, specifically evaluating the effects of the active intervention (bitter entities) against appropriate parallel control groups (untreated or vehicle controls) under identical experimental conditions.Quantitative Assays: Studies must utilize standardized, quantitative laboratory assays to measure outcome parameters (e.g.

Eligibility criteria Studies will be selected for inclusion in this systematic review and meta-analysis based on the following pre-specified Inclusion and Exclusion criteria: Inclusion Criteria Population: Authenticated human epithelial nasopharyngeal carcinoma (NPC) cell lines (e.g., HONE-1, SUNE-1, CNE-1, CNE-2, HK-1, C666-1) of any Epstein-Barr Virus (EBV) status. Interventions: Natural or synthetic bitter taste receptor (TAS2Rs) agonists, purified bitter chemical compounds (e.g., terpenoids, alkaloids, flavonoids), single-plant bitter crude extracts, or multi-herb polyherbal formulations traditionally classified as having a "bitter taste" property.Comparators: Parallel negative control groups, including vehicle-treated controls (e.g., DMSO, PBS, or ethanol) or completely untreated cells.Outcomes: Studies must quantitatively report at least one primary cytotoxicity outcome, such as half-maximal inhibitory concentration (IC_{50}) values, cell viability percentages, or rates of cellular proliferation/growth inhibition.Study Design: Peer-reviewed, primary in vitro laboratory experimental studies with concurrent control arms.Data Availability: Articles must provide sufficient numerical data (sample sizes, means, and standard deviations or standard errors) to allow for data extraction and statistical synthesis.Exclusion Criteria Ineligible Populations: Non-human nasopharyngeal carcinoma models (e.g., murine cells) or non-cancerous immortalized cell lines (except when used strictly as a baseline comparative safety control).Ineligible Interventions: Combination therapies combining bitter entities with standard cytotoxic chemotherapeutic drugs (e.g., Cisplatin) where the independent effect of the bitter substance cannot be isolated. Formulations or extracts explicitly documented as non-bitter in traditional medicine literature are also excluded. Ineligible Study Designs: In vivo animal models, in silico molecular docking simulations, or clinical trials that do not contain an independent, extractable in vitro cell culture component.Ineligible Article Types: Secondary literature including narrative reviews, systematic reviews, meta-analyses, scoping reviews, book chapters, conference abstracts, letters to the editor, and preprints that have not undergone full peer review.

Information sources A comprehensive and systematic literature search will be conducted across multiple international electronic databases from their respective inception dates through the date of the search. The primary electronic information sources include: PubMed (National Library of Medicine)Scopus (Elsevier)Web of Science (Clarivate Analytics)Cochrane Library (Wiley)Supplementary Information Sources: Reference Searching (Backward Citation Chaining): The reference lists of all finally included original research articles, as well as relevant narrative reviews retrieved during the screening process, will be manually screened to identify any potentially eligible studies that were not captured by the electronic database search. Forward Citation Chaining: Google Scholar will be utilized to track forward citations of the included pivotal papers to ensure contemporary completeness.Grey Literature: If necessary, manual searches will be extended to academic institutional repositories and specialized traditional medicine databases to identify complete, peer-reviewed doctoral dissertations or monographs that meet the predefined eligibility criteria.

Main outcome(s) The primary outcomes of interest focus on the quantitative metrics of laboratory-measured cytotoxicity and cell growth inhibition in human nasopharyngeal carcinoma (NPC) cell lines following exposure to bitter compounds or traditional bitter formulations.1. Cell Viability and Proliferation Rates Definition: The percentage or ratio of living/proliferating NPC cells in the intervention group compared to the time-matched untreated or vehicle control group.Measurement: Quantified via standardized colorimetric, fluorometric, or luminescent assays (including, but not limited to, MTT, CCK-8, MTS, XTT, SRB, or Alamar Blue assays).Effect Size Metric: Standardized Mean Difference (SMD) with 95% Confidence Intervals (CIs) will be calculated to pool data across different assay types.2. Half-Maximal Inhibitory Concentration (IC_{50})Definition: The specific concentration of the bitter compound, single-plant extract, or polyherbal formulation required to induce 50% growth inhibition or death of the treated NPC cell lines.Measurement: Reported as numerical concentration values (e.g., micromolar [μ M] or micrograms per milliliter [μ g/mL]) derived from dose-response curves after a specified exposure duration (e.g., 24, 48, or 72 hours).Effect Size Metric: Weighted mean IC_{50} values with corresponding 95% Confidence Intervals (CIs) will be pooled for compounds tested across multiple independent studies.

Additional outcome(s) The secondary outcomes of interest focus on the underlying molecular mechanisms and cellular pathways by which bitter compounds exert cytotoxic effects on human nasopharyngeal carcinoma (NPC) cell lines.These additional parameters include:1. Apoptosis Rates Definition: The quantitative percentage of NPC cells undergoing programmed cell death (early or late apoptosis) following exposure to the

intervention.Measurement: Quantified via standardized flow cytometry assays (e.g., Annexin V/Propitium Iodide [PI] staining) or biochemical assays such as TUNEL staining.2. Apoptotic Biomarkers and Protein Expression Definition: Relative changes or fold-changes in the expression and activation of key regulatory proteins involved in apoptotic pathways.Measurement: Evaluated via Western blotting, Enzyme-Linked Immunosorbent Assay (ELISA), or Quantitative Real-Time PCR (qRT-PCR) for markers including cleaved Caspase-3, Caspase-7, Caspase-9, and the ratio of pro-apoptotic to anti-apoptotic proteins (e.g., Bax/Bcl-2 ratio).3. Intracellular Calcium Concentration (Ca2+) Influx Definition: Changes in the mobilization of cytosolic calcium ions, which is a classic downstream signaling hallmark of bitter taste receptor (TAS2R) activation.Measurement: Measured as relative fluorescence intensity using calcium-sensitive fluorescent indicators (e.g., Fluo-3, Fluo-4, or Fura-2 assays).

Data management To ensure data integrity, reproducibility, and transparency throughout the systematic review and meta-analysis process, data will be managed through the following structured stages:1. Literature Record Management and De-duplication. All literature records retrieved from the electronic database searches (PubMed, Scopus, Web of Science, and Cochrane Library) will be exported into reference management software, specifically EndNote or Zotero. Duplicate records will be automatically identified and removed using the software's de-duplication function, followed by a manual cross-check by the primary reviewer to ensure absolute accuracy.2. Screening and Selection Documentation The unique records will be uploaded to an online systematic review platform (such as Covidence or Rayyan) to facilitate independent screening. Two reviewers will independently screen the titles and abstracts, followed by a full-text assessment against the predefined eligibility criteria. All exclusion reasons at the full-text screening stage will be explicitly documented. Any disagreements will be resolved through consensus or by consulting a third senior reviewer. 3. Data Extraction and Software Tools: A standardized, piloted data extraction sheet will be developed in Microsoft Excel. Two reviewers will independently extract data covering study metadata, cell line characteristics, intervention specifications (including traditional medicine contexts), and quantitative outcomes (sample size, mean, and standard deviation/standard error of IC_{50} values and cell viability percentages).In cases of missing or unclearly reported numerical data, digitized data extraction software (such as WebPlotDigitizer) will be utilized to extract exact numbers from published dose-response curves or bar charts.4. Data Archiving and Accessibility All final extracted datasets, statistical codes, and screening history will be securely stored on a cloud-based repository (such as the Open Science Framework [OSF] or institutional servers). The final dataset will be made open access upon publication of the manuscript to adhere to open science principles.

Quality assessment / Risk of bias analysis To evaluate the methodological quality and internal validity of the included in vitro laboratory studies, two reviewers will independently conduct a risk-of-bias assessment. An adapted version of the National Toxicology Program's Office of Health Assessment and Translation (OHAT) Risk of Bias Tool, specifically modified for in vitro toxicology and cell culture studies, will be utilized. The assessment will evaluate the following core domains of bias: Selection Bias (Allocation and Characterization): whether the human nasopharyngeal carcinoma (NPC) cell lines were properly authenticated (e.g., via STR profiling) and free of mycoplasma contamination. Whether the chemical identity, purity, and concentration of the bitter compounds, single-plant extracts, or polyherbal traditional formulations were clearly characterized and documented.Performance Bias (Experimental Conditions): Whether identical experimental conditions (e.g., incubation time, temperature, culture media volume, and vehicle concentration) were maintained across the intervention and control groups.Whether the researchers were blinded to treatment allocation during assay administration (if applicable).Detection Bias (Outcome Measurement): Whether the cytotoxicity and cell viability outcomes were measured using validated, standardized, and objective quantitative assays (e.g., MTT, CCK-8, or Flow Cytometry).Attrition Bias (Incomplete Outcome Data): Whether all experimental replicates and data points were accounted for, and whether any data exclusions or dropped samples were transparently reported.Reporting Bias (Selective Outcome Reporting): Whether all pre-specified outcomes (such as dose-response curves, all tested time points, and toxicity profiles) were completely reported rather than selectively highlighting only positive findings.Grading and Consensus: Each included study will be graded for each domain as either "Low risk of bias," "Probably low risk of bias," "Probably high risk of bias," or "High risk of bias." Any discrepancies in the quality grading between the two primary reviewers will be resolved through thorough discussion, consensus, or by involving a third senior reviewer.

Strategy of data synthesis The data synthesis will follow a structured approach consisting of both qualitative synthesis and quantitative meta-analysis, in accordance with the PRISMA guidelines.
1. Qualitative Synthesis (Narrative Synthesis)A narrative synthesis will be conducted for all included studies to outline the general landscape of the research. Data will be summarized in tabular format, describing the specific human nasopharyngeal carcinoma (NPC) cell lines, types and traditional classifications of bitter compounds/polyherbal formulations, experimental parameters (dosage and exposure duration) and mechanistic findings (such as apoptosis markers or calcium influx).
2. Quantitative Synthesis (Meta-Analysis)If the included studies exhibit sufficient homogeneity in terms of population (NPC cell lines) and outcome measures, a quantitative meta-analysis will be performed using statistical software (such as Review Manager [RevMan] or R with the meta and metafor packages).Effect Size Calculation: For continuous outcomes (e.g., cell viability and proliferation rates measured via MTT/CCK-8 assays), the Standardized Mean Difference (SMD) with corresponding 95% Confidence Intervals (CIs) will be calculated to account for variations in laboratory assay techniques. For half-maximal inhibitory concentration (IC50) values, the weighted mean with 95% CIs will be pooled across studies. Assessment of Heterogeneity: Statistical heterogeneity among the studies will be evaluated using Cochran's Q test ($p < 0.10$) and quantified with the I2 statistic. An I2 value greater than 50% will be considered indicative of substantial heterogeneity.Choice of Meta-Analysis Model: A Random-effects model (DerSimonian and Laird method) will be preferentially used to pool effect sizes, as it accounts for variations inherent in different laboratory environments, cell line passages, and the chemical profiles of traditional remedies. A fixed-effect model will only be used if statistical heterogeneity is negligible (\(I^2 < 25\% \)). Subgroup and Sensitivity Analyses: To explore potential sources of heterogeneity, predefined subgroup analyses will be performed by chemical classification or by traditional medicine type of the bitter entity (e.g., purified compounds vs. polyherbal formulations).Specific NPC cell lines and their Epstein-Barr Virus (EBV) status. Exposure durations (e.g., 24 h vs. 48 h vs. 72 h). Sensitivity analysis will be conducted by systematically omitting one study at a time to assess the stability and robustness of the pooled estimates. Publication Bias Assessment: If a meta-analysis includes 10 or more studies, potential publication bias will be visually assessed using a Funnel plot and statistically evaluated using Egger's linear regression test.

Subgroup analysis To explore potential sources of statistical heterogeneity and investigate how specific biological, chemical, and methodological characteristics influence the cytotoxic efficacy of bitter taste receptor agonists, predefined subgroup analyses will be performed when sufficient data are available (at least 3 studies per subgroup).
Subgroup analyses are planned based on the following specific criteria:
1. By Classification and Source of the Bitter Entity: Purified single molecules/compounds (e.g., isolated terpenoids, alkaloids, flavonoids, or synthetic TAS2R agonists).Crude mono-herbal extracts (extracts derived from a single bitter medicinal plant).Traditional polyherbal formulations (multi-ingredient recipes classified under traditional medicine frameworks such as Traditional Thai Medicine, Traditional Chinese Medicine, or Ayurveda).
2. By Nasopharyngeal Carcinoma (NPC) Cell Line Characteristics: Epstein-Barr Virus (EBV) status: EBV-positive cell lines (e.g., C666-1) versus EBV-negative cell lines (e.g., HONE-1, SUNE-1, CNE-2).Differentiation and cell line origin (e.g., highly differentiated versus undifferentiated NPC laboratory models).
3. By Methodological and Experimental Parameters: Exposure duration: Short-term exposure (24 hours) versus intermediate exposure (48 hours) versus long-term exposure (72 hours or more). Type of cell viability/cytotoxicity assay utilized: Metabolic-based assays (e.g., MTT, MTS, XTT, CCK-8) versus protein/DNA-binding assays (e.g., SRB assay, crystal violet) versus fluorescent cell staining.

Sensitivity analysis To evaluate the robustness, stability, and reliability of the pooled cytotoxic effect sizes (Standardized Mean Differences [SMD] and pooled IC_{50} values), sensitivity analyses will be systematically performed using the following approaches:
1. Leave-One-Out Sensitivity Analysis: A "leave-one-out" validation method will be conducted by sequentially removing one individual study at a time from the meta-analysis and re-calculating the pooled summary effect size and the heterogeneity statistic (I^2). This process helps determine whether the overall statistical conclusions are disproportionately driven by a single dominant study or a specific extreme outlier.
2. Methodological Quality (Risk of Bias) Filter: A sensitivity analysis will be performed by restricting the meta-analysis exclusively to high-quality studies. Studies classified as having a "High risk of

bias" or "Probably high risk of bias" during the quality assessment phase will be temporarily excluded to observe if their removal substantially reduces statistical heterogeneity or alters the estimated cytotoxic efficacy.
3. Statistical Model Comparison: The robustness of the data will be further tested by comparing the pooled results generated by the primary Random-effects model against those generated by a fixed-effects model. If both models yield statistically consistent results with overlapping 95% confidence intervals, the meta-analytic findings will be considered highly robust against model assumptions.

Language restriction No language restrictions will be applied during the initial electronic database search to ensure comprehensive retrieval of all potentially relevant studies, particularly those documented within localized traditional medicine contexts.However, for.

Country(ies) involved Primary Country: Thailand Affiliation of Authors: Faculty of Medicine , Thammasat University, Thailand.

Other relevant information Ethical Approval: Since this study is a systematic review and meta-analysis based entirely on previously published in vitro laboratory data, it does not involve direct contact with human subjects, animal experimentation, or the collection of fresh clinical tissue. Therefore, formal institutional review board (IRB) or ethical approval is not required.
Conflict of Interest: The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this protocol.
Dissemination Plan: The final findings of this systematic review and meta-analysis will be submitted to a peer-reviewed, international scientific journal (preferable within the fields of Oncology, Ethnopharmacology, or Chemosensory Research).
Amendments: Any post-registration deviations, modifications, or updates to this protocol during the course of the study will be transparently documented, dated, and updated within this registry platform to maintain absolute research transparency.

Keywords Nasopharyngeal carcinoma; Bitter compounds; Cytotoxicity; In vitro; Meta-analysis; Traditional medicine; TAS2Rs.
Dissemination plans The findings of this systematic review and meta-analysis will be broadly disseminated to the scientific and medical community through the following channels:
-Peer-Reviewed Journal Publication: The primary dissemination route will be the submission and publication of a full-length original research manuscript in a high-impact, international peer-reviewed journal. Target journals will be selected from those indexed in major citation databases (such as Scopus Q1/Q2 or Web of Science) that specialize in Oncology, Ethnopharmacology, Chemosensory Research, or Phytomedicine.
-Conference Presentations: Key findings and statistically pooled outcomes from this study will be prepared for abstract submission and presented as either oral or poster presentations at relevant national and international biomedical sciences and cancer research conferences.
-Open Access Archiving: To ensure maximum visibility and adherence to open science principles, a pre-print or the post-print version of the manuscript, along with its complete extracted datasets and statistical R/RevMan codes, will be securely archived and made accessible on a public repository (such as Open Science Framework [OSF]).

Contributions of each author
Author 1 - Nanatphong Paje - Principal Investigator will be responsible for the following core research activities: Conceptualization and Design: Formulating the initial research question, designing the PICO framework, and establishing the overall study protocol. Search Strategy Development: Developing and refining the comprehensive electronic search strings across the targeted databases.
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Author 2 - Arunporn Itharat - Conceptualization and Design: Formulating the initial research question, designing the PICO framework, and establishing the overall study protocol. Search Strategy Development: Developing and refining the comprehensive electronic search strings across the targeted databases.
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