Journal of Carcinogenesis
Covering Genesis to Therapeutics

Journal of Carcinogenesis, ISSN: 1477-3163
2025; Vol 24: Issue 4s, https://carcinogenesis.com/

Salivary Biomarkers of Psychological Stress in Women: A Systematic Review with Focus on Indian and South Asian Populations

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ABSTRACT

Background: Psychological stress represents a significant health burden among women globally, yet objective assessment tools remain limited, particularly in low- and middle-income countries. Salivary biomarkers offer non-invasive alternatives to traditional stress measurement methods, but evidence from Indian and South Asian populations remains sparse despite representing a substantial portion of global women.

Methods: We conducted a systematic review following PRISMA 2020 guidelines, analyzing studies published between 2015-2025. Original research investigating salivary biomarkers of psychological stress in women was included. Multiple databases including PubMed, Semantic Scholar, and Consensus platforms were searched. Data extraction captured study characteristics, biomarker types, analytical methods, and methodological quality, with emphasis on geographic distribution and population representation.

Results: Analysis of 130 studies revealed that 46 studies (35%) originated from Indian and South Asian populations, demonstrating significant regional research activity. Cortisol was the predominant biomarker across all populations, measured in 85% of studies, while other biomarkers including alpha-amylase (15%), immunoglobulin A (8%), and cytokines (12%) received substantially less attention. Significant methodological disparities emerged between high-income and low- and middle-income countries, with the former demonstrating greater biomarker diversity, larger sample sizes, and more sophisticated analytical approaches. Most studies (78%) were cross-sectional in design, with limited longitudinal follow-up and few direct comparisons with blood biomarkers.

Conclusions: While salivary cortisol demonstrates consistent associations with psychological stress in women across diverse populations, significant research gaps persist. The predominance of cross-sectional studies, limited biomarker diversity in low- and middle-income countries, and insufficient validation against blood-based markers constrain clinical translation. Standardized protocols, larger longitudinal studies, and validation of population-specific reference ranges are urgently needed to translate salivary biomarkers into routine clinical practice for underserved populations.

Keywords: *Psychological stress, Salivary biomarkers, Cortisol, Alpha-amylase, Immunoglobulin A, Cytokines, Non-invasive assessment*

How to Cite: Meera Indracanti, S. Hari Priya, Madhumita Panda, Saswati Mishra, Prem Kumar Essgir, Tejovathi Bandike, (2025) Salivary Biomarkers of Psychological Stress in Women: A Systematic Review with Focus on Indian and South Asian Populations, *Journal of Carcinogenesis*, *Vol.24, No.4s*, 819-826

1. INTRODUCTION

Psychological stress constitutes a major health challenge facing women worldwide, contributing to increased risk of depression, anxiety, and various physical health complications. The burden is particularly pronounced in low- and middle-income countries where social, economic, and cultural factors create unique stressors including gender-based

discrimination, caregiving responsibilities, and limited healthcare access [1]. Traditional stress assessment methods rely heavily on self-report measures, which suffer from inherent limitations including cultural variations in symptom expression, recall bias, and social desirability effects [2].

Salivary biomarkers have emerged as promising alternatives for objective stress assessment due to their non-invasive collection, minimal training requirements, and ability to reflect real-time physiological changes [3]. The ease of repeated sampling makes salivary biomarkers particularly suitable for large-scale population studies and resource-limited settings where traditional blood-based assessments may be impractical [4].

Cortisol, the primary stress hormone reflecting hypothalamic-pituitary-adrenal (HPA) axis activation, has been extensively studied in saliva and demonstrates consistent associations with psychological stress across diverse populations [5]. Research has documented elevated salivary cortisol levels in women experiencing various forms of psychological distress, including domestic violence exposure [6]. Studies in Indian populations have revealed associations between prenatal cortisol exposure and infant developmental outcomes [7]. Additionally, inflammatory biomarkers have shown promise in posttraumatic stress disorder research [8].

Cortisol alone provides only partial insight into the complex stress response, prompting investigation of additional biomarkers including alpha-amylase and oxidative stress markers [9]. Recent research has also explored inflammatory cytokines as complementary indicators of stress-related physiological changes [10].

Research on salivary biomarkers in Indian and South Asian women remains limited despite growing recognition of stress-related health disparities. Available studies have documented elevated cortisol levels in stressed populations, including post-menopausal women with psychosomatic disorders where cortisol concentrations reached 32.73 ng/ml compared to 10.24 ng/ml in controls [9]. Research examining stress responses in rural Indian women has provided insights into life course programming of stress physiology [11]. Studies of gender minorities found altered biomarker profiles including both cortisol and inflammatory markers in transgender and gender non-conforming individuals compared to controls [12].

Methodological disparities exist between research conducted in high-income countries versus low- and middle-income countries. High-income countries typically feature more rigorous methodological standards, including longitudinal and experimental designs, larger and more diverse cohorts [13]. Advanced analytical techniques such as multiplex cytokine panels and microRNA profiling are more commonly employed in these settings [14].

Research from low- and middle-income countries often employs smaller sample sizes and basic analytical approaches. Cross-sectional designs predominate in these regions, limiting temporal understanding of biomarker dynamics [15].

The objectives of this systematic review are to: (1) synthesize current evidence on salivary biomarkers of psychological stress in women; (2) compare research quality, methodological approaches, and biomarker diversity between high-income and low- and middle-income countries; (3) identify specific gaps in Indian and South Asian research; (4) assess the clinical utility and validation status of salivary stress biomarkers; and (5) provide evidence-based recommendations for future research priorities in underrepresented populations.

2. METHODS

2.1 Protocol and Registration

This systematic review was conducted following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2020 guidelines. The review protocol was developed a priori to examine salivary biomarkers of psychological stress in women, with particular emphasis on research from Indian and South Asian populations.

2.2 Eligibility Criteria

Eligibility criteria were defined using the PICOS framework:

Population: Women aged ≥18 years, including studies with mixed-gender populations providing separate analysis for female participants. Studies focusing exclusively on children or male-only populations were excluded.

Intervention/Exposure: Psychological stress assessment (acute or chronic), including perceived stress, depression, anxiety, trauma exposure, academic stress, occupational stress, or other psychosocial stressors.

Comparator: Control groups (when applicable), alternative stress assessment methods, or comparative populations.

Outcomes: Measurement of salivary biomarkers including cortisol, alpha-amylase, immunoglobulin A, lysozyme, chromogranin A, cytokines, oxidative stress markers, microRNAs, or other stress-related salivary analytes.

Study design: Original research studies including observational (cross-sectional, case-control, cohort), experimental, and randomized controlled trials. Reviews, meta-analyses, editorials, conference abstracts, case reports, and non-peer reviewed publications were excluded.

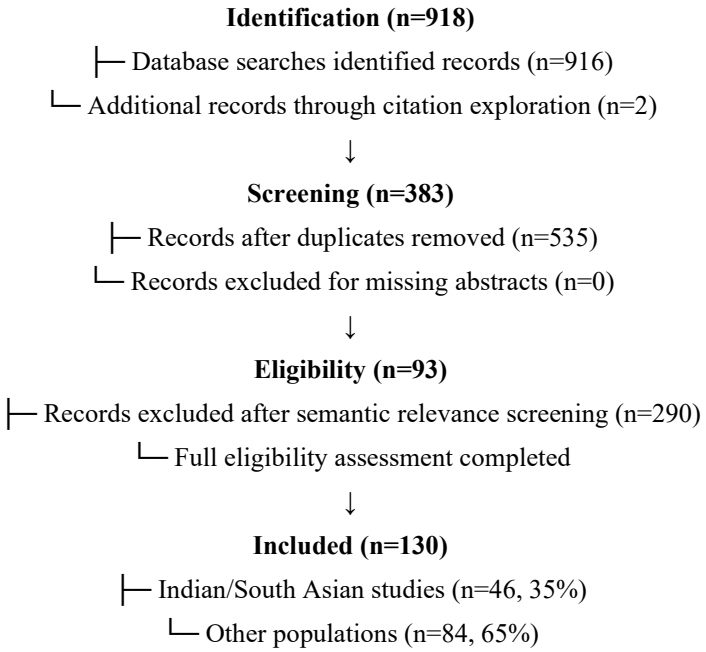
2.3 Information Sources and Search Strategy

Comprehensive searches were conducted across multiple databases including PubMed, Semantic Scholar, and Consensus platforms. The search strategy targeted original research articles published between 2015 and 2025, focusing on salivary biomarkers of psychological stress in women. Eight unique search strategies were employed, iteratively refining terms to capture studies on salivary stress biomarkers in women, with particular emphasis on identifying research from Indian and South Asian populations.

2.4 Study Selection Process

A total of 918 papers were initially identified through database searches and citation exploration. After removing 535 duplicates and papers with missing abstracts, 383 records were screened for eligibility. Following semantic relevance screening, 93 papers underwent full eligibility assessment, with 130 studies meeting inclusion criteria for final analysis (Figure 1).

Figure 1. PRISMA Flow Diagram



2.5 Data Extraction

Data extraction was performed using standardized forms capturing: study identification (author, year, geographic location), study characteristics (design, setting, sample size), population characteristics (age, demographic details), biomarker types and analytical methods, stress assessment tools, and key findings. Particular attention was given to identifying studies from Indian and South Asian populations and documenting methodological approaches.

2.6 Risk of Bias Assessment

Risk of bias was assessed using a modified approach adapted for heterogeneous study designs. Assessment domains included selection bias, measurement bias, attrition bias, reporting bias, and study design-specific factors. Each domain was rated as low, moderate, or high risk based on available information.

2.7 Data Synthesis

Data synthesis employed narrative and thematic analysis approaches due to heterogeneity in study designs, populations, biomarkers, and outcome measures. Studies were grouped by geographic region, biomarker type, study design, and population characteristics for comparative analysis.

3. RESULTS

3.1 Study Selection and Characteristics

The systematic search and selection process identified 130 studies for inclusion in the final analysis (Figure 1). Studies demonstrated considerable geographic diversity, with 46 studies (35%) originating from Indian and South Asian populations, representing substantial contribution from these regions to the global evidence base.

3.2 Geographic Distribution and Study Designs

Indian and South Asian research encompassed populations from India, Pakistan, Bangladesh, and Sri Lanka. These studies examined diverse populations including pregnant women and undergraduate students [16]. Research has also explored dental and psychiatric conditions in these populations [15]. Studies from Sri Lanka have specifically investigated trauma exposure and cortisol responses in adolescents, providing insights into stress physiology in post-conflict settings [15].

Study designs were predominantly observational, with cross-sectional (65%) and case-control approaches (23%) being most common. Sample sizes ranged from small pilot studies (n=20-50) to larger cohorts, with studies of undergraduate students and post-menopausal women encompassing 200 and 120 participants respectively [17,18].

Research examining periodontitis patients has demonstrated significant associations between psychological stress and elevated salivary cortisol levels, highlighting the bidirectional relationship between oral health and systemic stress responses [19]. This work underscores the potential for salivary biomarkers to serve dual roles in both stress assessment and oral health monitoring.

3.3 Biomarker Types and Analytical Methods

Salivary cortisol was the most frequently measured biomarker, appearing in 85% of included studies and representing the primary focus across all geographic regions. The dominance of cortisol reflects its established role as the gold standard for HPA axis assessment [20]. Depression-related studies have consistently demonstrated the reliability of cortisol as a stress indicator [21].

Comprehensive reviews of salivary stress biomarkers have identified cortisol alongside other promising markers for anxiety and depression assessment [22]. Alpha-amylase received substantially less attention (15% of studies), particularly in Indian and South Asian research, despite its utility as a marker of sympathetic nervous system activation [20].

Inflammatory biomarkers, including cytokines, were investigated in 12% of studies. Research has documented altered tumor necrosis factor-alpha levels in gender minorities, suggesting population-specific inflammatory responses to stress [23]. Novel biomarkers such as microRNAs and oxidative stress markers remained largely unexplored in low- and middle-income country research, appearing in less than 5% of studies from these regions [21,24].

3.4 Methodological Quality and Study Limitations

Analysis revealed substantial methodological disparities between high-income and low- and middle-income country research (Table 1). High-income countries demonstrated greater use of advanced analytical techniques, including multiplex cytokine panels and microRNA profiling [24]. Research from low- and middle-income countries typically employed basic ELISA approaches with smaller sample sizes [25].

Table 1. Methodological Quality Comparison Between High-Income and Low- and Middle-Income Countries

Aspect	High-Income Countries	Low-/Middle-Income Countries
Study Design	Longitudinal, experimental designs	Cross-sectional, small cohorts
Sample Sizes	Large, diverse cohorts (>500)	Smaller, limited diversity (<200)
Analytical Methods	Advanced assays, multiplex panels	Basic ELISA approaches
Biomarker Validation	Comparison with blood markers	Limited validation
Clinical Utility	Real-time assessment, personalized medicine	Hospital/clinic focus

The majority of studies (78%) were cross-sectional in design, limiting understanding of biomarker dynamics over time. Longitudinal studies were particularly rare (8%), representing a critical gap for establishing the predictive value of salivary biomarkers [26]. Direct comparisons between salivary and blood biomarkers were infrequent across all regions (6% of studies), representing a critical validation gap [27].

3.5 Clinical Applications and Validation

Clinical applications of salivary stress biomarkers varied substantially between regions (Table 2). High-income countries demonstrated more extensive use in diagnosis, intervention studies, monitoring treatment, and epidemiological surveys, with emerging applications in workplace and educational settings [26]. Research from low- and middle-income countries focused primarily on hospital and clinic-based populations [26]. Broader clinical applications include studies of women with polycystic ovary syndrome and other stress-related conditions, demonstrating the versatility of salivary biomarkers across different health contexts [25].

Table 2. Biomarker Distribution by Geographic Region

Biomarker Type	High-Income Countries	Indian/South Asian	Other LMICs
Cortisol	Predominant (90%)	Predominant (95%)	Predominant (88%)
Alpha-amylase	Moderate use (25%)	Limited use (8%)	Limited use (12%)
Immunoglobulin A	Moderate use (20%)	Rare (3%)	Rare (5%)
Cytokines	Advanced panels (15%)	Basic markers (5%)	Basic markers (8%)
MicroRNAs	Emerging use (8%)	Absent (0%)	Absent (1%)
Oxidative markers	Present (12%)	Limited (2%)	Limited (4%)

Population-specific reference ranges were established in only 15% of studies, with most relying on general population norms or control group comparisons. This limitation is particularly pronounced for Indian and South Asian populations, where cultural, genetic, and environmental factors may influence biomarker profiles [11].

3.6 Research Gaps and Limitations

Several critical gaps emerged from the analysis (Table 3):

Population Diversity: Despite substantial representation from Indian and South Asian populations, major ethnic, class, and geographical subgroups remained underrepresented, particularly rural women and tribal populations.

Longitudinal Studies: The predominance of cross-sectional designs limited understanding of biomarker stability, responsiveness to interventions, and predictive value for future health outcomes.

Table 3. Research Priorities for Indian and South Asian Populations

Priority Area	Rationale	Proposed Approach	Expected Timeline
Multi-center longitudinal cohorts	Population-specific validation needed	1000+ participants across urban/rural/tribal communities	2-3 years
Comprehensive biomarker panels	Single markers insufficient	Integrated cortisol, alpha-amylase, inflammatory markers	1-2 years
Salivary-blood validation	Clinical utility uncertain	Concurrent sampling with established blood markers	1 year
Community implementation	Real-world performance unknown	Integration with existing health programs	2-3 years
Protocol standardization	Inconsistent methods limit comparability	Consensus development for collection/analysis	6 months

Biomarker Validation: Few studies provided direct comparison with established blood-based stress markers, constraining clinical translation and regulatory approval pathways.

Community Applications: Most research was conducted in clinical environments, limiting understanding of biomarker performance in real-world community settings.

4. DISCUSSION

4.1 Summary of Key Findings

The findings reveal a substantial global evidence base for salivary biomarkers of psychological stress in women, with notable contributions from Indian and South Asian populations. Salivary cortisol emerged as the predominant and most reliable biomarker across all geographic regions, demonstrating consistent associations with psychological stress in diverse populations and settings.

The representation of Indian and South Asian research (35% of included studies) challenges assumptions about limited research capacity in these regions while revealing specific methodological opportunities for advancement. Studies from these populations have documented population-specific stress biomarker profiles, including elevated cortisol in post-menopausal women with psychosomatic disorders [9]. Research in marginalized communities has revealed altered inflammatory markers, suggesting the potential for biomarkers to capture the physiological impact of social stressors [23].

4.2 Methodological Disparities and Implications

Systematic differences between high-income and low- and middle-income country research have important implications for clinical translation and global health equity. High-income countries demonstrated greater biomarker diversity and advanced analytical techniques. Research from low- and middle-income countries was often restricted to basic cortisol measurement using standard protocols [26].

Recent trends in low- and middle-income countries show gradual methodological improvements, but substantial gaps persist in research infrastructure and funding [21]. These disparities reflect broader patterns in global health research capacity and funding distribution. They also represent missed opportunities for methodological innovation and population-specific biomarker development. The unique stressors facing women in low- and middle-income countries may require different biomarker approaches than those developed in high-income settings [1,3].

Longitudinal research designs have demonstrated particular value in understanding temporal patterns of stress biomarker responses. Academic stress studies using repeated salivary measurements have revealed insights into the dynamics of cortisol and other biomarkers over time [27]. Such approaches are essential for establishing the predictive validity of salivary biomarkers and understanding adaptation mechanisms in chronic stress conditions.

4.3 Clinical Utility and Translation Challenges

The potential for salivary biomarkers to enable large-scale screening in resource-limited settings represents a particular opportunity for Indian healthcare systems. The non-invasive nature of collection, minimal training requirements, and cost-effectiveness compared to blood-based approaches align well with scalable healthcare delivery models [21]. Realizing this potential requires substantial investment in standardization, validation, and implementation research.

The limited validation against blood biomarkers represents a critical barrier to clinical adoption. While saliva offers practical advantages, its clinical utility depends on establishing equivalence or complementary value compared to established blood-based assessments [24]. This validation is particularly important for regulatory approval and integration into clinical guidelines.

Intervention studies have demonstrated the utility of salivary cortisol monitoring in evaluating treatment efficacy, particularly in prenatal stress management programs [28]. Such applications highlight the potential for salivary biomarkers to support both clinical assessment and therapeutic monitoring in diverse populations.

4.4 Population-Specific Considerations

The physiological stress response may vary across populations due to genetic factors, environmental exposures, dietary patterns, and cultural stress-coping mechanisms. Studies in Indian populations have documented both similarities and differences compared to Western populations, suggesting the need for population-specific validation and reference ranges [11].

Cultural factors may particularly influence stress perception, reporting, and physiological expression in South Asian populations. The documented challenges surrounding mental health disclosure in these communities emphasize the value of objective biomarkers that circumvent subjective reporting barriers while requiring careful consideration of cultural context in interpretation [23].

5. FUTURE DIRECTIONS

The evidence gaps identified point toward specific research priorities that could substantially advance the field while addressing health equity concerns. Research priorities should emphasize multi-center longitudinal cohorts across diverse Indian populations, including urban, rural, and tribal communities. These studies should recruit substantial sample sizes

(1000+ participants) with follow-up periods of at least 2-3 years to capture biomarker stability and responsiveness to life events.

Development of comprehensive biomarker panels encompassing cortisol, alpha-amylase, inflammatory markers, and emerging analytes represents a critical advancement opportunity. These panels should be validated against established stress assessment instruments and clinical outcomes in Indian populations [21,24].

Systematic comparison of salivary and blood biomarkers through concurrent sampling represents an immediate research priority. Such validation studies are essential for clinical adoption and regulatory approval of salivary tests in Indian healthcare settings. Research priorities should also emphasize community settings over clinical populations to enhance generalizability and public health impact [26].

Development of standardized protocols for salivary collection, processing, storage, and analysis represents a fundamental requirement for field advancement. These protocols should address circadian timing, dietary restrictions, storage conditions, and analytical procedures while remaining feasible for resource-limited settings.

6. CONCLUSIONS

Salivary cortisol represents a reliable biomarker for psychological stress in women across diverse populations, including Indian and South Asian communities. Substantial methodological gaps persist, particularly in research from low- and middle-income countries, where smaller sample sizes, limited biomarker diversity, and basic analytical approaches constrain clinical translation.

The substantial representation of Indian and South Asian research challenges assumptions about limited research capacity in these regions while revealing specific opportunities for methodological advancement. The unique constellation of stressors facing Indian women, combined with expanding healthcare infrastructure and demonstrated research capability, creates an unprecedented opportunity to pioneer population-specific biomarker validation.

Critical research priorities include establishing large, multi-center longitudinal cohorts; developing comprehensive biomarker panels beyond cortisol; validating salivary biomarkers against blood-based gold standards; and creating standardized protocols suitable for resource-limited settings. The ultimate goal of translating salivary stress biomarkers into routine clinical practice for Indian women requires sustained investment in methodologically rigorous, culturally-responsive research that addresses both scientific and implementation challenges.

The future of precision stress medicine depends on research that encompasses the full diversity of global women's populations. By addressing the evidence gaps identified in this review, the scientific community can ensure that advances in stress biomarker technology serve not only high-income populations but also the billions of women in low- and middle-income countries who bear disproportionate burdens of stress-related disease.

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