INPLASY

Effectiveness of miRNA's in the follow up of testicular tumors: a systematic review

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ADMINISTRATIVE INFORMATION

Support - Self founded.

Review Stage at time of this submission - Completed but not published.

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 11 December 2025 and was last updated on 11 December 2025.

INTRODUCTION

Review question / Objective OBJECTIVES AND HYPOTHESES

Nuclear or Central Target
To describe the usefulness of miRNAs as biomarkers in the clinical follow-up of patients with testicular cancer through a systematic review.

Peripheral or Secondary Objectives
1. Identify the most studied miRNAs in testicular cancer.
2. To analyse the available evidence on the use of miRNA in the monitoring of patients with testicular cancer.

Rationale Testicular cancer, particularly germ cell tumours (GCTs), represents a significant challenge in modern oncology. Despite the high cure rates achieved with conventional therapies, significant challenges remain. The high incidence underscores the need for advances in diagnostic and therapeutic strategies to improve clinical outcomes for affected patients.
Despite advances in current treatments, significant limitations persist in the early identification of relapses, prediction of treatment response, and reduction of long-term side effects. Conventional tumour markers have demonstrated limited sensitivity and specificity, being insufficient for accurate patient monitoring, especially in subtypes such as seminomas, where their elevation is minimal or absent.
In this context, miRNAs are emerging as promising tools in precision oncology due to their stability in bodily fluids, their ability to reflect real-time tumour dynamics, and their specificity in key processes of tumour progression, such as cell proliferation, invasion, and chemotherapy resistance. Some miRNA clusters have demonstrated high sensitivity and specificity, overcoming the limitations of traditional biomarkers and offering new perspectives for risk stratification, clinical monitoring, and personalized treatments in testicular cancer.

Conducting a systematic review to evaluate the usefulness of miRNAs in the follow-up of patients with testicular cancer is crucial for consolidating existing evidence, identifying knowledge gaps, and proposing practical recommendations to optimize the management of this disease. Furthermore, this study could lay the groundwork for future research, facilitating the implementation of miRNAs as standard biomarkers in clinical practice and contributing to improving patients' quality of life through more accurate diagnoses and more effective therapies. This approach not only benefits patients but also has the potential to reduce costs associated with unnecessary treatments and related complications.

Condition being studied Testicular cancer is the most common malignancy in young men between the ages of 15 and 44. Although current treatments, such as cisplatin-based chemotherapy, have significantly improved cure rates, challenges related to treatment resistance, relapse, and long-term adverse effects persist. In this context, microRNAs (miRNAs) have emerged as promising biomarkers due to their fundamental role in regulating cellular processes involved in carcinogenesis, tumour progression, and therapeutic response in testicular germ cell tumours (TGCTs).

The purpose of this project is to conduct a systematic review of the literature to evaluate the usefulness of miRNAs in the clinical follow-up of patients with testicular cancer. The aim is to analyse their potential as prognostic and disease-monitoring biomarkers, as well as their potential to predict treatment response and detect relapse in early stages. The review will integrate evidence on the regulatory mechanisms of miRNAs in testicular cancer, including their interaction with key signalling pathways.

Based on the growing body of literature supporting the role of miRNAs in tumour biology and precision medicine, this project aims to consolidate current knowledge to propose recommendations for their implementation in clinical practice. The results could contribute to optimizing the management of patients with testicular cancer, reducing complications resulting from conventional treatments, and improving the quality of life of survivors.

METHODS

Participant or population Protocol and reporting. We followed PRISMA 2020 and PRISMA-DTA guidance for diagnostic test accuracy reviews. A prespecified (unregistered) protocol defined the clinical question, eligibility criteria, outcomes, and analysis plan.

Review question and target condition. Population: individuals evaluated for testicular germ cell tumors (TGCT). Index test: circulating miR-371a-3p measured in serum or plasma (qPCR, targeted amplification assays, or digital PCR). Comparators: alpha-fetoprotein (AFP), beta-human chorionic gonadotropin (β-hCG), and lactate dehydrogenase (LDH). Target condition: viable TGCT.

Eligibility criteria. We included observational cohorts (prospective or retrospective) and cross-sectional studies reporting per-patient diagnostic accuracy for the index test and/or comparators with sufficient data to derive 2×2 tables (TP, FP, FN, TN) in one of the following scenarios: (a) primary diagnosis prior to orchiectomy, (b) surveillance after curative treatment, (c) assessment of treatment response, or (d) evaluation of residual masses. We excluded case reports, editorials, narrative reviews, duplicate cohorts, tissue-only miRNA studies, non-TGCT tumors, and studies lacking per-patient accuracy data.

Information sources and search strategy. We searched PubMed/MEDLINE, ScienceDirect, Wiley Online Library, the Virtual Health Library (BVS/VHL), and SciELO from January 2015 to the most recent update November 2025. Searches combined terms for “testicular cancer”/TGCT and “microRNA/miRNA” with “biomarker”, “follow-up”, “surveillance”, and “diagnosis” in English and Spanish. We also screened reference lists of relevant articles and prior reviews. Full search strings for each database are provided in the Supplement.

Study selection. Two reviewers independently screened titles/abstracts and full texts against eligibility criteria (RC, OV); disagreements were resolved by consensus. Selection is summarized in a PRISMA flow diagram with counts at each stage.

Data extraction. Using a standardized form, we recorded: study design; setting/country; clinical scenario; specimen (serum/plasma); assay platform/normalization; cut-off definition (pre-specified vs optimized); histology/stage spectrum; denominators for cases/controls; index and comparator 2×2 data (or sensitivity/specificity with denominators to reconstruct counts); AUC when available; and pre-analytical conditions (e.g., hemolysis control, processing time, freeze–thaw). When 95% CIs were not reported, we calculated Wilson CIs using study denominators.

Risk of bias and applicability. We used QUADAS-2 (patient selection, index test, reference standard, flow/timing) with signaling questions tailored to biomarker studies; we also assessed applicability concerns per domain. Case–control designs and

post-hoc threshold optimization were considered higher risk.

Outcomes. The primary outcomes were sensitivity and specificity of circulating miR-371a-3p for the primary-diagnosis setting. Secondary outcomes included AUC, accuracy in surveillance/recurrence and treatment-response settings, and comparative performance of AFP, β-hCG, and LDH.

Statistical analysis. We modeled study-level sensitivity and specificity using a bivariate random-effects model (Reitsma) on the logit scale. Within-study variances were derived from binomial assumptions; a 0.5 continuity correction was pre-specified for zero cells, if present. Between-study variance components (τ^2 for sensitivity and specificity) and the between-study correlation (ρ) were estimated by maximum likelihood (L-BFGS-B). We plotted a hierarchical SROC (HSROC) and, where pooling was infeasible, a descriptive Moses–Littenberg SROC with Q*. Primary analysis was restricted to pre-orchiectomy diagnosis. Other scenarios (surveillance, treatment response, residual masses) were summarized narratively or by single operating points. Planned sensitivity analyses included restriction to prospective cohorts and to pre-specified thresholds. Analyses used Python 3.11 (NumPy/SciPy/matplotlib); Wilson 95% CIs were computed for study-level proportions.

Software and reproducibility. Analyses were performed in Python 3.11 (NumPy/SciPy) with matplotlib for figures; Wilson CIs were computed for proportions when needed. All figure-generation scripts are available on request and summary figures are supplied as supplements.

Intervention Determination of miRNA.

Comparator Usual monitoring (laboratory, DHL, AFP, h-GCH).

Study designs to be included We included observational cohorts (prospective or retrospective) and cross-sectional studies reporting per-patient diagnostic accuracy for the index test and/or comparators with sufficient data to derive 2×2 tables (TP, FP, FN, TN) in one of the following scenarios.

Eligibility criteria We included observational cohorts (prospective or retrospective) and cross-sectional studies reporting per-patient diagnostic accuracy for the index test and/or comparators with sufficient data to derive 2×2 tables (TP, FP, FN, TN) in one of the following scenarios: (a) primary diagnosis prior to orchiectomy, (b) surveillance after curative treatment, (c) assessment of treatment response, or (d) evaluation of residual masses. We excluded case reports, editorials, narrative reviews, duplicate cohorts, tissue-only miRNA studies, non-TGCT tumors, and studies lacking per-patient accuracy data.

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Information sources Information sources and search strategy. We searched PubMed/MEDLINE, ScienceDirect, Wiley Online Library, the Virtual Health Library (BVS/VHL), and SciELO from January 2015 to the most recent update November 2025. Searches combined terms for “testicular cancer”/TGCT and “microRNA/miRNA” with “biomarker”, “follow-up”, “surveillance”, and “diagnosis” in English and Spanish. We also screened reference lists of relevant articles and prior reviews. Full search strings for each database are provided in the Supplement.

Main outcome(s) Outcomes. The primary outcomes were sensitivity and specificity of circulating miR-371a-3p for the primary-diagnosis setting. Secondary outcomes included AUC, accuracy in surveillance/recurrence and treatment-response settings, and comparative performance of AFP, β-hCG, and LDH.

Data management Statistical analysis. We modeled study-level sensitivity and specificity using a bivariate random-effects model (Reitsma) on the logit scale. Within-study variances were derived from binomial assumptions; a 0.5 continuity correction was pre-specified for zero cells, if present. Between-study variance components (τ^2 for sensitivity and specificity) and the between-study correlation (ρ) were estimated by maximum likelihood (L-BFGS-B). We plotted a hierarchical SROC (HSROC) and, where pooling was infeasible, a descriptive Moses–Littenberg SROC with Q*. Primary analysis was restricted to pre-orchiectomy diagnosis. Other scenarios (surveillance, treatment response, residual masses) were summarized narratively or by single operating points. Planned sensitivity analyses included restriction to prospective cohorts and to pre-specified thresholds. Analyses used Python 3.11

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Software and reproducibility. Analyses were performed in Python 3.11 (NumPy/SciPy) with matplotlib for figures; Wilson CIs were computed for proportions when needed. All figure-generation scripts are available on request and summary figures are supplied as supplements.

Quality assessment / Risk of bias analysis Statistical analysis. We modeled study-level sensitivity and specificity using a bivariate random-effects model (Reitsma) on the logit scale. Within-study variances were derived from binomial assumptions; a 0.5 continuity correction was pre-specified for zero cells, if present. Between-study variance components (τ^2 for sensitivity and specificity) and the between-study correlation (ρ) were estimated by maximum likelihood (L-BFGS-B). We plotted a hierarchical SROC (HSROC) and, where pooling was infeasible, a descriptive Moses–Littenberg SROC with Q*. Primary analysis was restricted to pre-orchiectomy diagnosis. Other scenarios (surveillance, treatment response, residual masses) were summarized narratively or by single operating points. Planned sensitivity analyses included restriction to prospective cohorts and to pre-specified thresholds. Analyses used Python 3.11 (NumPy/SciPy/matplotlib); Wilson 95% CIs were computed for study-level proportions.
Software and reproducibility. Analyses were performed in Python 3.11 (NumPy/SciPy) with matplotlib for figures; Wilson CIs were computed for proportions when needed. All figure-generation scripts are available on request and summary figures are supplied as supplements.

Strategy of data synthesis Statistical analysis. We modeled study-level sensitivity and specificity using a bivariate random-effects model (Reitsma) on the logit scale. Within-study variances were derived from binomial assumptions; a 0.5 continuity correction was pre-specified for zero cells, if present. Between-study variance components (τ^2 for sensitivity and specificity) and the between-study correlation (ρ) were estimated by maximum likelihood (L-BFGS-B). We plotted a hierarchical SROC (HSROC) and, where pooling was infeasible, a descriptive Moses–Littenberg SROC with Q*. Primary analysis was restricted to pre-orchiectomy diagnosis. Other scenarios (surveillance, treatment response, residual masses) were summarized narratively or by single operating points. Planned sensitivity analyses included restriction to prospective cohorts and to pre-specified thresholds. Analyses used Python 3.11 (NumPy/SciPy/matplotlib); Wilson 95% CIs were computed for study-level proportions.
Software and reproducibility. Analyses were performed in Python 3.11 (NumPy/SciPy) with matplotlib for figures; Wilson CIs were computed for proportions when needed. All figure-generation scripts are available on request and summary figures are supplied as supplements.

Subgroup analysis Fourteen studies were selected, the characteristics of which are presented in Table 1. We constructed another table for the per-study diagnostic performance in which 4 studies were selected (MiR-371a-3p). Table 2. And the analysis per QUADAS-2 is presented in Table 3.

Sensitivity analysis Across four primary-diagnosis studies, the bivariate model estimated summary sensitivity 0.89 and specificity 0.93. Between-study heterogeneity was notable (report τ^2(Se), τ^2(Sp), ρ), consistent with threshold variation. Study-level estimates (Wilson 95% CIs) appear in Figure 2. The HSROC (Figure 3) shows high discrimination; the Q* from descriptive SROC (~0.90) aligns with the bivariate summary. The prospective multicenter cohort (2019) contributed a single operating point (Sensitivity 0.901, Specificity 0.940), displayed in Supplementary Figure S1. The hierarchical SROC (Figure 3) demonstrates high discriminative ability for circulating miR-371a-3p in the primary-diagnosis setting. In a complementary descriptive analysis using Moses–Littenberg, the Q* point (sensitivity = specificity) was ~0.90, aligning with the bivariate summary and supporting robustness of the overall signal.
In the strictly prospective multicenter cohort (2019), the operating point for primary diagnosis was Sensitivity 0.901 and Specificity 0.940. Because only one prospective study reported both metrics, a bivariate meta-analysis was not performed; the prospective SROC therefore corresponds to that single operating point (Supplementary Figure S1). The location of this point relative to the hierarchical SROC underscores the consistency of performance in real-world, multicenter settings.
Figure 4 compares SROCs for miR-371a-3p vs AFP, β-hCG, and LDH using available operating points across scenarios/thresholds. Curves are descriptive; interpretation should emphasize study-level points. miR-371a-3p showed higher sensitivity than AFP/LDH with preserved high specificity; β-hCG retained excellent specificity but moderate sensitivity.

Language restriction English.

Country(ies) involved México.

Keywords germ cell; miRNA; precision medicine.

Contributions of each author

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