

REASSESSING RIBONUCLEIC ACID ISOLATION FROM HUMAN MONONUCLEAR CELL CULTURE WITH MAGNETIC BEADS PRE-ENRICHMENT FOR MOLECULAR ANALYSIS

Sengul Ilker^{1, 2, 3} Sengul Demet⁴

¹ Giresun University Faculty of Medicine, Thyroidology Unit, TR-28100 Giresun, Turkey

² Giresun University Faculty of Medicine, Division of Endocrine Surgery, TR-28100 Giresun, Turkey

³ Giresun University Faculty of Medicine, Department of General Surgery, TR-28100 Giresun, Turkey

⁴ Giresun University Faculty of Medicine, Department of Pathology, TR-28100 Giresun, Turkey

Primljen/Received: 18. 11. 2025.

Prihvaćen/Accepted: 13. 12. 2025.

Online First: December 2025.

Dear Editor,

It is with considerable interest that I have perused the recently published contribution by Bhatia, entitled “*Ribonucleic Acid Isolation from Human Mononuclear Cell Culture with Magnetic Beads Pre-enrichment for Molecular Analysis*,” set forth within *Sanamed*, volume 19(1). The author addresses a conundrum of pronounced import for those engaged in molecular analyses and the development of immunotherapies. As such, the established difficulty of extracting ribonucleic acid (RNA), particularly from mononuclear cell (MNC) cultures exceeding a few months in age, is well documented within the author’s own laboratory experience, having persisted, as stated, for over two years of consistent failure.

The developed methodology, centered upon the pre-enrichment of the cultured MNCs utilizing Cluster of Differentiation 45 (CD45)-specific magnetic beads antecedent to the customary mini column isolation, proves efficacious where previous attempts faltered. The successful isolation of RNA from cultures exceeding six months in duration—confirmed through spectrophotometric yield measurements and subsequent conventional and real-time Polymerase Chain Reaction (PCR) assays for the beta-actin housekeeping gene—is indeed a notable technical advancement. The assertion that this study constitutes the inaugural demonstration of isolating RNA from aged human MNC cultures via specific magnetic beads is, moreover, a claim that warrants careful consideration.

Notwithstanding the demonstrated success of this technique, one might cast an inquiring gaze upon certain aspects of the execution and presentation. Firstly,

the affiliation of the esteemed author with Genekam Biotechnology AG, which entity serves as the sole source for the crucial magnetic beads, the mini column isolation kit, the PCR kits, and the specialized magnetic rack, invites circumspection. Whilst proprietary methods often feature in novel protocols, the near-total reliance upon reagents and apparatus supplied by the author’s own commercial interest renders the protocol less immediately accessible or generalizable for laboratories not possessed of the aforementioned instruments and supplies. The efficacy of the method, therefore, appears for the present to be closely associated with this particular commercial supply chain.

Secondly, whilst the author reports that the initial method failed consistently over two years, and that isolation without magnetic beads was not achieved, the results presented lack detailed quantitative metrics comparing the failed isolations to the successes. The presentation of “failure” in Table 1 illustrates the necessity of the pre-enrichment step, yet does not afford the readership an optimal means to gauge the precise extent of nucleic acid degradation or inhibition encountered previously—data which would further illuminate the magnitude of the technical challenge overcome.

Finally, the discussion alludes to further consequential applications of the isolated RNA—namely, its conversion to complementary DNA (cDNA) and subsequent use for other purposes—yet these vital data are deferred for “future publications.” Herewith, a more comprehensive elucidation of the robustness of the isolated material might have been advantageously reserved for this singular publication, ensuring that the full scope of the methodology’s utility is presented forthwith (1).

In essence, whilst the developed magnetic bead pre-enrichment methodology provides a much-needed solution for obtaining RNA from refractory MNC cultures, it is sincerely hoped that future work will include more extensive comparative data and endeavor to ascertain the protocol's viability utilizing reagents sourced from diverse suppliers, thereby further affirming its widespread applicability in molecular diagnostics and therapeutic development. This issue merits further investigation. We thank Bhatia et al. (1) for their valuable study on RNA isolation from human MNC culture with molecular analysis.

Abbreviations

MNC – Mononuclear Cells

CD45 – Cluster of Differentiation 45

PCR – Polymerase Chain Reaction

RNA – Ribonucleic Acid

cDNA – Complementary DNA

Funding: No funding was available.

REFERENCES

1. Bhatia S. Ribonucleic Acid Isolation from Human Mononuclear Cell Culture with Magnetic Beads Pre-enrichment for Molecular Analysis. *Sanamed*. 2025;19(3):275–9. doi: 110.5937/sanamed0-50158.

*Accepted papers are articles in press that have gone through due peer review process and have been accepted for publication by the Editorial Board of *Sanamed*. The final text of the article may be changed before the final publication. Accepted papers can already be cited using the year of online publication and the DOI, as follows: the author's last name and initial of the first name, article title, journal title, online first publication month and year, and the DOI. When the final article is assigned to volumes/issues of the journal, the Article in Press version will be removed and the final version will appear in the associated published volumes/issues of the journal. The date the article was made available online first will be carried over.

How to cite this article: Sengul I, Sengul D. Reassessing Ribonucleic Acid Isolation from Human Mononuclear Cell Culture with Magnetic Beads Pre-enrichment for Molecular Analysis. *Sanamed*. Online First, December 2025. doi: 10.5937/sanamed0-62870.

Correspondence to/Autor za korespondenciju

Prof. Demet Sengul, M.D.

Editor-in-Chief „*Sanamed*” journal, Serbia

Founder & Chair, Department of Pathology

Founder, Scientific and Research Laboratories

Giresun University Faculty of Medicine

TR-28100, Giresun, Turkey

email: demet.sengul.52@gmail.com

WoS: A-3035-2019; Scopus: 22938589200

ORCID numbers

Sengul Demet 0000-0002-0416-0621

Sengul Ilker 0000-0001-5217-0755

Conflicts of Interest: None declared.

Author Contribution & Responsibilities: The authors take full responsibility for the accuracy and integrity of the content, as well as the validity of institutional affiliations. The publisher remains neutral regarding jurisdictional claims in institutional affiliations. All authors have read and agreed to the published version of the manuscript. **Authors' contributions:** **IS:** Conceptualization, Formal analysis, Investigation, Methodology, Project administration, Resources, Software, Validation, Visualization, Writing – original draft, Writing – review & editing; **DS:** Conceptualization, Formal analysis, Investigation, Methodology, Project administration, Resources, Software, Supervision, Validation, Visualization, Writing – original draft, Writing – review & editing.

Note: Artificial intelligence was not utilized as a tool in this study.

Licensing: This work is licensed under a Creative Commons Attribution 4.0 International (CC BY 4.0) License.