Radioimmunoassay Laboratory at Radiation Medicine Centre

Implementation and Impact of Liquid Handling System for Thyroid Disorder Diagnostics

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Abstract

Radiation Medicine Centre (RMC) is one of the largest centers for thyroid research and is also one of the leading referral bases for managing thyroid disorders, with a robust patient registry and an established infrastructure for diagnostic innovations. With approximately 15,000 thyroid cancer patients and over 5,000 follow-up cases annually, the center is at the forefront of thyroid disorder research, particularly thyroid cancer. A significant part of the research involves the development and validation of *in-vitro* diagnostic assays, particularly radioisotopic assays such as radioimmunoassays (RIA) and immunoradiometric assays (IRMA) which have proven instrumental in diagnosing and monitoring thyroid conditions. However, large-scale production of these assays at laboratory level has been constrained by labor-intensive manual biomolecule immobilization processes. To address this, an automated liquid handling system (ASSIST PLUS-INTEGRA) has been implemented to automate the immobilization process, thus increasing efficiency, reducing contamination, improving reproducibility, and enhancing data quality. The implementation of this system has notably advanced the production of in-house thyroglobulin (Tg) IRMA kits for routine thyroid cancer monitoring, marking a significant stride in both research and routine clinical diagnostics. This article provides an overview of the integration of this advanced liquid handling system into the RIA laboratory and discusses the benefits and impacts of its implementation.

Introduction

Thyroid disorders, including thyroid cancer, hypothyroidism, and hyperthyroidism, are among the most common endocrine disorders globally. The accurate diagnosis and monitoring of these conditions require the use of specialized diagnostic tools, particularly immunoassays [1]. Immunodiagnostic radioisotopic techniques such as RIA and IRMA play a critical role in the detection and quantification of thyroid-specific biomarkers like total T4 (TT4), Free T4 (FT4), Tg and anti-thyroid peroxidase antibodies (anti-TPOAb). These biomarkers are essential for monitoring thyroid cancer progression and recurrence, and they also aid in the management of other thyroid-related disorders [2].

The manual process of immobilizing biomolecules, such as antibodies and analytes, onto solid-phase surfaces like polystyrene tubes or plates is labor-intensive, requiring skilled personnel and a significant amount of time. In a high-throughput laboratory setting, where large-scale production of assays is necessary, this process becomes even more challenging. The need for precision, reproducibility, and cost efficiency is paramount. At RMC, TT4, FT4, anti-TPOAb, Tg and anti-Tg autoantibody (TgAb) assays have been developed and validated and some of them also found applications, as an import substitute; for routine *in-vitro* patient services [3]. In this article, we discuss how the implementation of an advanced liquid handling system has addressed these challenges and improved the efficiency of antibody immobilization in the production of Tg IRMA kits for the management of routine thyroid cancer patients.

Materials and Methods

Patient Population and Sample Collection

RMC manages a registry of approximately 15,000 thyroid cancer patients, with over 5,000 follow-up cases annually. Clinical samples, are collected from these patients for biomarker analysis using RIA and IRMA techniques. The biomarker assays developed in-house are being used for diagnostic and monitoring purposes for routine clinical care.

Radioimmunoassay Development

RIA and IRMA are essential diagnostic tools for quantifying specific biomarkers related to thyroid function and cancer. These assays rely on the binding of radiolabeled antibodies to their respective antigens (thyroid hormones, antibodies, etc.) on solid-phase surfaces. In the conventional assay production process, polyclonal antibodies are manually coated onto polystyrene tubes using manual pipetting followed by indigenously developed and fabricated equipment for aspiration and washing (Fig. 1-6). This is typically done by adding a solution containing the biomolecule to each tube and allowing it to adsorb. This step demands highly skilled personnel and is time-intensive, with the entire process taking up to a week. Until now, the immobilization of the analytes has been done manually, making the task cumbersome.

Integration of the Liquid Handling System (ASSIST PLUS-INTEGRA)

A customized (for the in-house racks) liquid handling system (ASSIST PLUS-INTEGRA) was procured to automate the biomolecule immobilization process (Fig.7). This device incorporates independent electronic pipettes into a platform to transform manual work into an automated workflow and provides precision pipetting and liquid handling capabilities, enabling the rapid and reproducible coating of biomolecules on solid-phase surfaces. The system is capable of performing a wide range of liquid handling tasks, including dilution, transfer and biomolecule immobilization; and can process large volumes in a high-throughput setting.

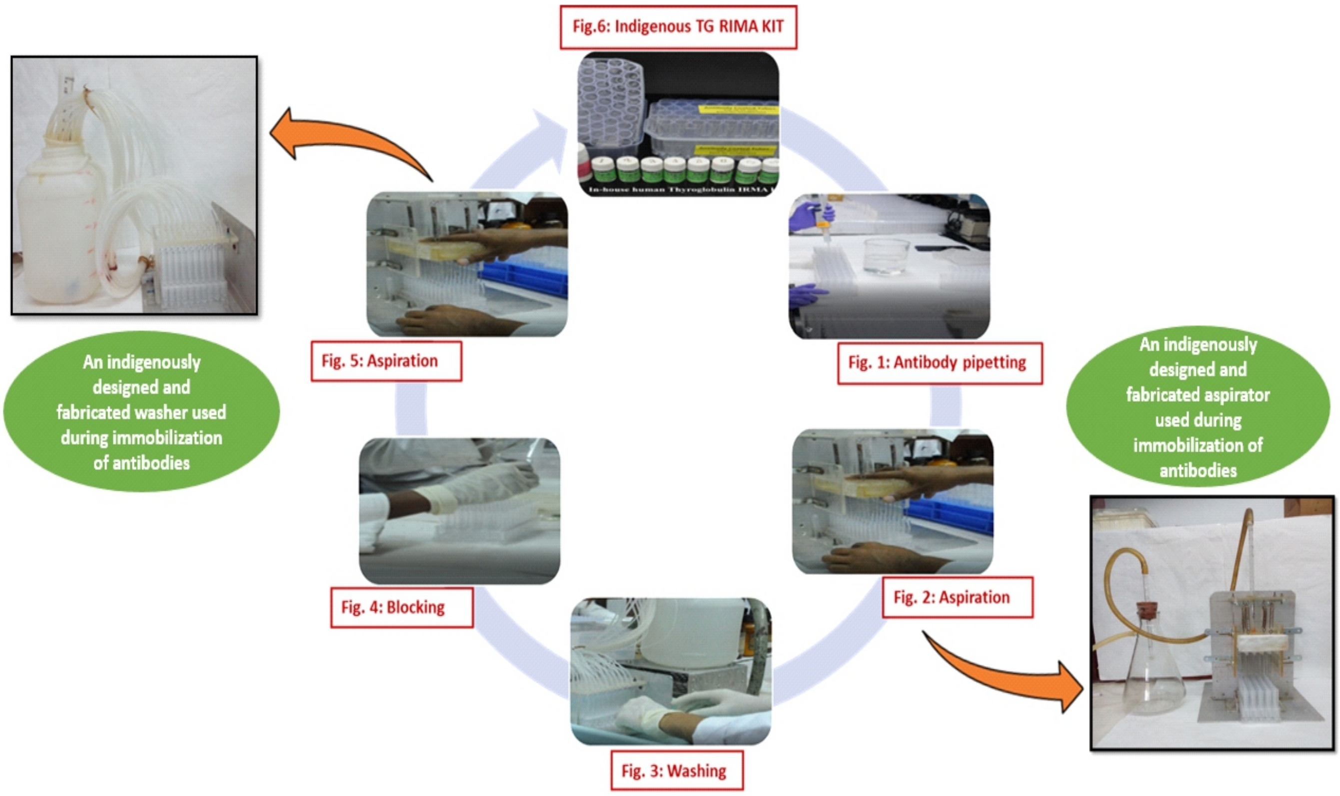

Fig.1-6: Various steps involved in the manual immobilization of antibodies at RMC for in-house Tg IRMA kit production.

In one specific instance, the liquid handling system was used to coat 10,700 tubes with polyclonal anti-thyroglobulin antibodies, which were then used for the production of in-house Tg IRMA kits. These kits are used to estimate Tg in serum for monitoring thyroid cancer patients for the detection of residual or recurrent disease.

Assay Validation

The developed Tg IRMA kits (liquid handling system used for preparing antibody coated tubes) underwent validation to ensure their performance in clinical settings. Additionally, assays for other hormones are also being developed in small batches for research and development purpose in our laboratory.

Results and Discussion

Improved Efficiency and Throughput

The integration of the liquid handling system significantly improved the efficiency of the biomolecule (antibody) immobilization process. In a single batch, the liquid handling system was able to coat 10,700 tubes, a task that would have otherwise taken a significant amount of manual labor. This customized automation not only saved time but also allowed for greater throughput, enabling the laboratory to scale its production of diagnostic kits for routine purpose without sacrificing quality [4].

Enhanced Precision and Reproducibility

One of the primary challenges in manual biomolecule immobilization is ensuring consistency across batches. Variability in antibody coating, volume dispensation, and even slight inconsistencies in technique can lead to variations in assay performance. By automating this process, the liquid handling system significantly reduced these sources of error. The system's precision pipetting capabilities ensured that each tube was coated with a desired amount of antibody, resulting in more reproducible assay performance. Sensitivity, specificity, and reproducibility were assessed using patient samples, and the performance was found to be satisfactory [5].

Reduced Contamination and Labor Costs

Manual coating processes are prone to contamination, particularly when handling large numbers of samples. The automated system minimizes the risk of cross-contamination, as the pipetting steps are performed in a controlled, enclosed environment. Additionally, by reducing the reliance on manual labor, the system has resulted in cost savings, both in terms of personnel time and the potential for errors that could lead to wasted materials or rework. Due to customization, the spare racks could be utilized for assembling the polystyrene tubes for immobilization process which further reduced the cost.

Improved Data Quality and Flexibility

The liquid handling system's ability to control the volume and precision of reagents added to each tube enhances the data quality of the final assays. Moreover, the system is highly flexible, allowing for the easy scaling up or down of assay production depending on research needs. This flexibility has been invaluable in developing assays for additional thyroid hormones and other biomarkers, as well as in customizing assays for research purposes.

Cost Efficiency and Increased Productivity

With the high throughput and reduced labor costs, the integration of the liquid handling system has made the production of radioimmunoassay more cost-effective. These improvements in productivity and cost efficiency have contributed to the sustainability of in-house assay development and production. The system can be used not only for coating the polystyrene tubes but also plates.

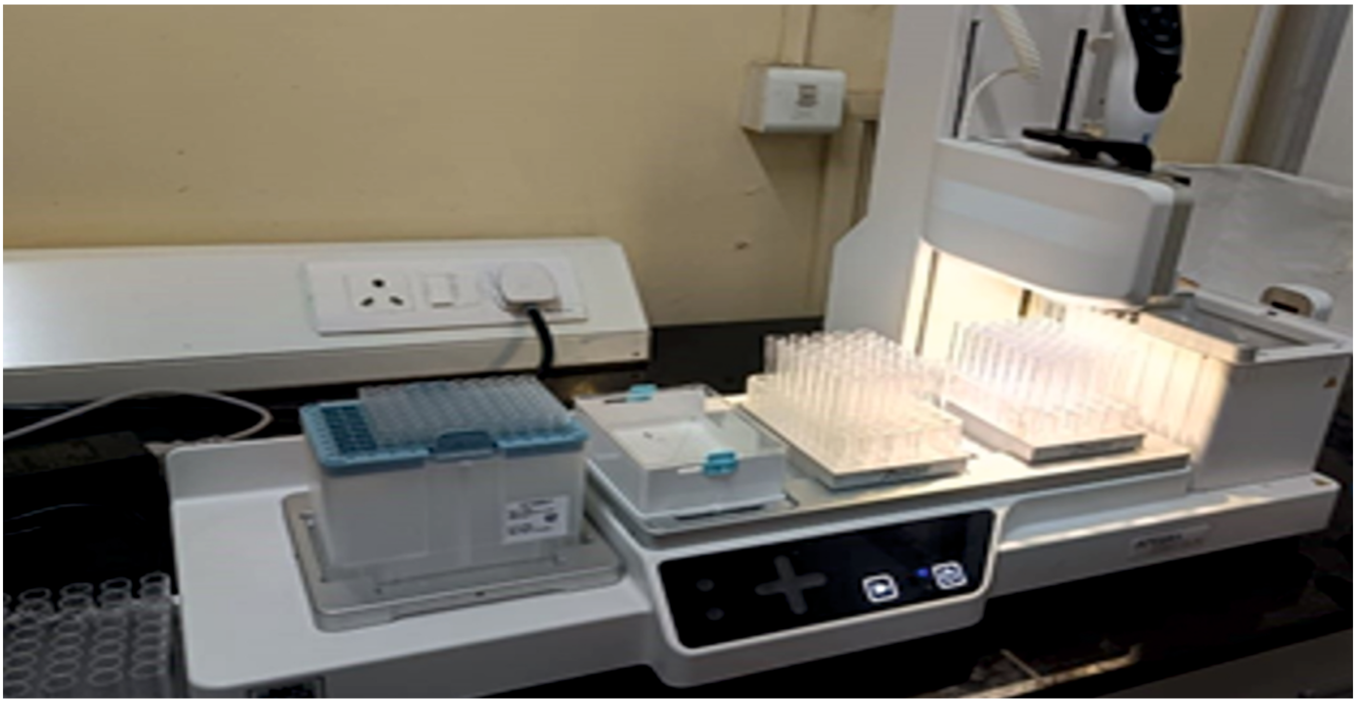

Fig.7: Customized liquid handling system at RMC: Enabling precise immobilization of biomolecules for immunodiagnostic applications.

Conclusion

The integration of the automated liquid handling system has significantly enhanced the capabilities of the radioimmunoassay laboratory in terms of efficiency, reproducibility, and productivity. By automating the biomolecule immobilization process, the system has not only streamlined the production of thyroid cancer diagnostic kits but also paved the way for future innovations in immunodiagnostic assays at the laboratory level. The benefits of this integration extend beyond increased throughput and reduced costs, improving overall laboratory performance and contributing to better patient outcomes. As the system continues to support research and development in the field of thyroid disorder diagnostics, it underscores the importance of adopting advanced technologies in the laboratory to meet the growing demands of modern healthcare.

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