

Advancements in minimal residual disease detection: a practical approach using single-cell droplet PCR for comprehensive monitoring in hematological malignancy

Satoshi Uchiyama*, Kentaro Fukushima*, Seiichiro Katagiri, Junichi Tsuchiya, Tomohiro Kubo, SungGi Chi and Yesuke Minami

Abstract: The identification of chromosomal abnormalities accompanied by copy number alterations is important for understanding tumor characteristics. Testing methodologies for copy number abnormality have limited sensitivity, resulting in their use only for the sample provided at the time of diagnosis or recurrence of malignancy, but not for the monitoring of minimal residual disease (MRD) during and after therapy. We developed the "DimShift" technology which enables to measure the copy number of target gene/chromosome in each cell, which is given by the single cell droplet PCR. Qualitative result of DimShift given by peripheral blood was perfectly concordant with that of bone marrow. These findings and performances are promising to be the new methodology for MRD detection in malignant diseases utilizing bone marrow as well as peripheral blood.

Keywords: chromosomal abnormalities, droplet digital PCR, minimal residual disease, myelodysplastic syndrome

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Chromosomal abnormalities are frequently observed in hematological disorders. For example, myelodysplastic syndrome (MDS) is frequently accompanied by chromosomal aneuploidies, as in chromosomes 5-, 7-, 8+, 17-, or 20-.¹⁻³ These chromosomal aneuploidies are worth identifying and monitoring since they provide information regarding disease status or indicate poor prognosis.⁴⁻⁷ Cytogenetic methods, such as fluorescence *in situ* hybridization or G-banding, are widely used to detect chromosomal copy numbers.⁸ However, because they are low-throughput methods that require numerous steps and may not be welcomed in routine laboratory workflows. Therefore, in this study, we developed a novel high-throughput and sensitive assay for aneuploidy detection using droplet polymerase chain reaction (PCR). A PCR-based method would facilitate more rapid and more frequent monitoring of chromosomal status. Generally speaking, PCR is not suitable for counting just one or two copies, although this technique works as a powerful tool for detecting a novel

genetic marker, such as BCR-ABL,⁹ which is frequently present in cells of patients with chronic myelogenous leukemia and acute lymphoblastic leukemia, but not in cells from healthy patients. In a general real-time PCR, the information of the template molecules is retained during the exponential amplification phase and is reflected as the difference in Ct values. However, the sensitivity is not high enough to determine the ratio of abnormal cells if a sample is a mixture of normal karyotype and aneuploid cells. In digital PCR, where DNA is amplified in a nanoliter-sized compartment, the reaction is efficient enough for the amplification of only one molecule of DNA.¹⁰ At the endpoint, however, the high efficiency makes it impossible to preserve information regarding the number of template molecules. The results only provide information as to whether or not the DNA of interest was amplified, that is, whether the target DNA was present or absent. We speculated that information on the number of template molecules might be retained before the reaction reaches its

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National Library of Medicine Current Catalog National Library of Medicine (U.S.), 1989 *Current Catalog* National Library of Medicine (U.S.),

First multi year cumulation covers six years 1965-70 *Cancer Management in Man: Chemotherapy, Biological Therapy, Hyperthermia and Supporting Measures* Boris Minev, 2011-01-13 This book represents a comprehensive description and evaluation of the most up to date approaches to cancer management. Each chapter prepared by leading basic researchers and clinicians provides an in depth description of a specific method for cancer management. The chemotherapy section of the book is updated to include the newest drugs as well as those currently in development. Organized by drug class this section provides the latest information on most drugs including their mechanisms of action interactions with other agents toxicities side effects and mechanisms of resistance. The biological therapy section of the book provides expanded coverage of the currently used cytokines vaccines and cell based therapies of cancer. Full consideration is also given to other modern treatment approaches such as tyrosine kinase inhibitors inhibitors of tumor angiogenesis and the transcatheter management of cancer. Current advances in hyperthermia in cancer treatment hematologic and nutritional support bone marrow transplantation pain management and care of the terminally ill patients with cancer are also presented. In summary this book provides a comprehensive coverage of the current knowledge on the most innovative systematic and multidisciplinary approaches to the treatment of patients with cancer.

A Multidisciplinary Analysis of Controversies in the Management of Prostate Cancer Donald S. Coffey, 2012-12-06 These proceedings emanate from the Second Prout's Neck Conference on prostate cancer held on October 17-19, 1986 the theme of which was treatment with focus on current issues.

and future research that is needed to answer critical questions related to optimal management of the various stages of prostate cancer. The objective was to reveal the most crucial problems impeding progress and to crystallize the combined multidisciplinary input generated by the conference into focused concepts or recommendations for presentation to the National Cancer Institute NCI with the ultimate intent of targeting research to address the priority issues identified. In organizing the workshop every effort was made to maintain a multidisciplinary balance among nationally renowned authorities on prostate cancer. Thus leading surgeons, radiation and medical oncologists and biostatisticians were in equal presence. While there were spirited exchanges with careful scrutiny and critique of all data presented there was a common belief that the challenge of prostate cancer would be best approached in this multidisciplinary Organ Systems oriented fashion. During the course of the conference it became apparent to all present that major nomenclature and procedural barriers have made it generally difficult and frequently impossible to compare results of clinical research.

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Reference: Oncology Thankamma Ajithkumar, Sarah Jane Jefferies, 2021. This easy to read practical guide distils and compiles all the disparate literature on cancer into one succinct volume. It includes the essential evidence based clinical guidelines needed for the safe and effective management of patients with cancer and has a clear layout to allow for quick reference whilst on the ward. All aspects of cancer and its management are covered including prevention, screening, diagnosis and treatment. The text begins by outlining the clinical approach to suspected cancer and the principles of multidisciplinary prevention and management. It then progresses through site specific cancer management including head and neck, CNS, thoracic, breast, gastrointestinal system, genitourinary system, female genital system, skin, musculoskeletal system, haemopoietic system and endocrine. Later chapters cover oncological emergencies and acute oncology and special situations such as cancer in younger and older people and pregnancy and fertility. The guide also offers information about coping with the lifestyle and social issues that may arise with a diagnosis of cancer such as insurance, travel and support and includes a chapter dedicated to palliative care for the cancer patient. A unique appendix of clinical management flowcharts assists fast appropriate decision making.

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Current Therapy in Genitourinary Surgery Martin I. Resnick, 1992. Features new to this edition include a section on infertility and expanded coverage of impotence therapy. The text emphasizes therapy indications and contra indications and addresses issues of disease severity, patient condition and age as well as potential risks which enter decision making.

Clinical Diagnosis and Management by Laboratory Methods John Bernard Henry, 1996 Provides a scientific information resource in aspects of clinical pathology and laboratory medicine relevant to patient care, health promotion and disease prevention

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