

Chapter 8.2

Cryopreservation of Lymphocytes in Bulk

DOUGLAS M. STRONG

Introduction

Since the early discovery of the cryoprotective properties of glycerol, a great deal of investigation has gone into the determination of the mechanisms of freezing injury (Polge et al., 1949). Very briefly, cells that are cooled too slowly, to below freezing temperatures, are damaged by the resulting increase in salt concentration and cellular shrinkage which occurs as water is removed during the formation of extracellular ice (Lovelock, 1953). Conversely, if cooling is too rapid, a new mechanism is invoked in which shrinkage no longer occurs but the cell is damaged by the formation of intracellular ice, either during freezing or upon thawing (Mazur et al., 1969). Cryoprotectants, such as Me₂SO, reduce the amount of ice present during freezing and reduce solute concentration thus reducing ionic stress (Farrant et al., 1974; Lovelock, 1953; Mazur et al., 1969; Polge et al., 1949). However, these compounds can themselves cause osmotic injury since they are hypertonic and can cause damage during their addition or removal (Strong, 1976). Optimum cooling rates vary from cell type to cell type depending on differences in membrane permeability and intracellular water, which is removed during the dehydration phase of slow cooling and extracellular ice formation. In general, the larger the cell volume, the slower the rate of cooling to allow equilibration of intra- and extra-cellular water during freezing.

Although the primary use of cryopreserved lymphocytes in the histocompatibility laboratory may be as reagents in lymphocytotoxicity assays, where the preservation requirements are less rigid, the use of frozen thawed cells in functional assays or in assays for determination of other cell surface markers, may require closer attention to protocol. Several laboratories have reported that, under certain conditions, selection of lymphoid subsets may occur following freezing and thawing (Farrant et al., 1972; Knight et al., 1972). Furthermore, lymphoid clones and B- or T-LCL have different optimum cooling rates (Strong et al., 1985). Although sophisticated controlled rate freezing devices are not absolutely required for lymphocyte cryopreservation, such equipment usually increases reproducibility and improves recovery (Dubey et al., 1986; Ichino and Ishiiwa, 1988). In addition, the handling of cells prior to and following freezing and thawing is at least as important as the freezing itself.

Abbreviations

Me₂SO: Dimethylsulfoxide
DNase: Deoxyribonuclease

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ASHI Laboratory Procedure Manual American Society for Histocompatibility and Immunogenetics,Amy B. Hahn,Geoffrey A. Land,Rosemarie M. Strothman,2000 *Manual of Molecular and Clinical Laboratory Immunology* John L. Schmitz,Barbara Detrick,Maurice R. O'Gorman,2024-12-24 THE authoritative guide for clinical laboratory immunology For nearly 50 years the Manual of Molecular and Clinical Laboratory Immunology has been the premier resource for laboratories students and professionals involved in the clinical and technical details of diagnostic immunology testing The 9th Edition continues its tradition of providing comprehensive clinical and technical information on the latest technologies used in medical and diagnostic immunology Led by a world renowned group of authors and editors this new edition reflects substantial changes aimed at improving and updating the Manual's utility while reflecting the significant transformations that have occurred since the last edition including the revolution of gene editing and the widespread adoption of molecularly engineered cellular therapies Topical highlights include Laboratory Management three new chapters cover essential aspects of quality assurance quality improvement and quality management aligning with the increasingly stringent and demanding regulatory environment Inborn Errors of Immunity the primary immunodeficiency section has been completely updated to align with the latest International Union of Immunological Societies classifications of inborn errors of immunity Functional Cellular Assays expanded content includes detailed discussions on various functional assays critical for modern immunologic testing Autoimmune Diseases expanded chapters on systemic and organ specific autoimmune disorders including new chapters on Sjgren's syndrome and deficiency of ADA2 as well as significant updates on organ specific autoimmune diseases Transplantation Immunology updated chapters detail the assessment of immune reconstitution and ABO testing reflecting latest practices The 9th Edition of the Manual of Molecular and Clinical Laboratory Immunology serves as an invaluable resource for laboratory directors clinicians laboratory managers technologists and students It provides critical insights into the selection application and interpretation of immunologic tests offering practical guidance on troubleshooting clinical application and an understanding of test limitations This comprehensive and up to date manual remains an essential tool for anyone involved in the diagnosis evaluation and management of immune mediated and immune system related disorders

Handbook of Human Immunology Mary S. Leffell,Albert D. Donnenberg,Noel R. Rose,1997-04-10 Combining basic explanations of laboratory tests with 115 tables full of reference data and applications the Handbook of Human Immunology provides practicing clinicians with a current complete guide to molecular immunology Introductory chapters overview the molecular basis of immune responses and immunological disorders focusing on the role of cell receptors accessory molecules and cytokines in these processes Emphasis is placed on immunological parameters that are clinically useful The basic principles underlying assays of the immune system are discussed and the book stresses the application and interpretation of immune tests Comprehensive coverage is given to immunoglobulins and their age dependent concentration Cellular

immunology is discussed from the perspectives of lymphocyte functional parameters as well as through immunophenotyping of lymphocytes and other leukocytes Both serological and molecular diagnosis of infectious diseases are reviewed The Handbook of Human Immunology contains up to date information on exciting developments in immunogenetics covering the application of T cell receptor genes and the HLA alleles in disease associations and transplantation

Manual of Molecular and Clinical Lab Immunology Barbara Detrick, Robert G. Hamilton, James D. Folds, 2006-03-10 Introduces new material that reflects the significant advances and developments in the field of clinical laboratory immunology Provides a comprehensive and practical approach to the procedures underlying clinical immunology testing Emphasizes molecular techniques used in the field of laboratory immunology Updates existing chapters and adds significant new material detailing molecular techniques used in the field Presents guidelines for selecting the best procedures for specific situations and discusses alternative procedures Covers aspects of immunology related disciplines such as allergy autoimmune diseases

cancers and transplantation immunology

Molecular Biology of Hematopoiesis 5 Nader G. Abraham, Shigetaka Asano, G. Brittinger, G.J.M. Maestroni, R.K. Shadduck, 2012-12-06 This volume of Molecular Biology of Hematopoiesis is dedicated to John W Adamson M D Tadamitsu Kishimoto M D Robert C Gallo M D Arthur W Nienhuis M D and Franco Mandelli M D for their contributions in developing an overall view of the state of the art knowledge in the field of hematopoiesis Richard Champlin among other renowned clinicians presented updated information on stem cells and T cell depletion for bone marrow transplant A clinical update on thrombopoietin was presented by Pamela Hunt of Amgen and by Kenneth Kaushansky Arthur Nienhuis and Katherine Turner's contributions to our current knowledge and advances in the fields of growth factors and gene transfer were also recognized during the 9th Symposium on Molecular Biology of Hematopoiesis in Genoa The chapters cover such diverse areas as preclinical and clinical updates on growth factors and positive and negative regulatory molecules Advances in Leukemia Mechanism and Treatment by Interferon was presented by Professor Sante Tura Readers will find presentation of exciting advances that have occurred in the area of hematopoiesis The elucidation of gene structures of key growth factor proteins such as IL 12 and IL II will lead to new insights and new approaches in understanding the regulation of hematopoiesis as well as application of new growth factors

Histocompatibility Testing Jeffrey L Bidwell, Cristina Navarrete, 2000-03-08 This invaluable book provides comprehensive coverage of contemporary serological cellular and molecular methodologies in histocompatibility testing and their application to human organ transplantation and transfusion The contributors are internationally respected authorities in histocompatibility and immunogenetics and are closely involved in the development or application of state of the art technologies The first three sections of the book are primarily intended for use as a bench manual for histocompatibility testers immunologists and immunogeneticists the fourth and fifth sections on selection of donors and statistical methods will further assist medical practitioners involved in clinical transplantation and its outcome The final section of the book reviews

the genetics and clinical relevance of minor histocompatibility antigens **The SNU Manual of Surgical Care 5 Edition**
Department of Surgery, Seoul National University College of Medicine, 2024-03-13 This book serves as a concise surgical manual that quickly references the latest information essential for determining the policies of surgical patient care and pre and post operative treatment It is a necessary guide not only for surgical residents and medical students but also for practicing surgeons Features of this book 1 Provides information on surgical care through a systematic approach to surgery related topics 2 Reflects the latest research findings and recommendations on the rapid development and evolution of the surgical field 3 Covers a wide range of topics and areas in surgical surgery including patient management before and after surgery basic techniques and procedures management of complications fluids and nutrition 4 Offers practical guidance that can help in real surgical situations and practice 5 Available in a pocket size that can be easily referred to at any time 6 An English edition that can be referenced by medical professionals both in the Korea and abroad Table of contents Contributors Preface to the 5th edition of SNU Manual of Surgical Preface to the 1st edition Preface to the 2nd edition Preface to the 3rd edition Preface to the 4th edition I Basic Considerations 01 Surgical Infections and Antibiotics 02 Fluids and Nutrition 03 Critical Care Medicine 04 Surgical Complications 05 Preoperative Evaluation and Management for Medical Disease 06 Basic Surgical Skills 07 Symptomatic Surgical Management II Specific Considerations 01 Liver 02 Bile Duct and Pancreas 03 Gastrointestinal Tract 04 Small Intestines 05 Colon Rectum and Anus 06 Breast 07 Endocrine 08 Vascular 09 Kidney and Pancreas Transplantation 10 Pediatric Surgery 11 Trauma 12 Liver Transplantation Index

Clinical Laboratory Medicine Kenneth D. McClatchey, 2002 This thoroughly updated Second Edition of Clinical Laboratory Medicine provides the most complete current and clinically oriented information in the field The text features over 70 chapters seven new to this edition including medical laboratory ethics point of care testing bone marrow transplantation and specimen testing providing comprehensive coverage of contemporary laboratory medicine Sections on molecular diagnostics cytogenetics and laboratory management plus the emphasis on interpretation and clinical significance of laboratory tests why a test or series of tests is being done and what the results mean for the patient make this a valuable resource for practicing pathologists residents fellows and laboratorians Includes over 800 illustrations 353 in full color and 270 new to this edition Includes a Self Assessment and Review book **Immunohematology: Principles and Practice** Eva D Quinley, 2020-06-15 Immunohematology Principles and Practice Third Edition an ideal text for anyone who wants to master the theory and practices of today s blood banking **Manual of Molecular and Clinical Laboratory Immunology** Barbara Detrick, John L. Schmitz, Robert G. Hamilton, 2020-07-16 THE authoritative guide for clinical laboratory immunology For over 40 years the Manual of Molecular and Clinical Laboratory Immunology has served as the premier guide for the clinical immunology laboratory From basic serology testing to the present wide range of molecular analyses the Manual has reflected the exponential growth in the field of immunology over the past decades This eighth edition reflects the latest advances and developments in the diagnosis and

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The editor has incorporated scientific contributions from a diverse group of leading researchers in the field of hematology and related blood cell research This book aims to provide an overview of current knowledge pertaining to our understanding of hematology The main subject areas will include blood cell morphology and function the pathophysiology and genetics of hematological disorders and malignancies blood testing and typing and the processes governing hematopoiesis Blood cell physiology biochemistry and blood flow are covered in this book This text is designed for hematologists pathologists and laboratory staff in training and in practice The work presented in this book will be of benefit to medical students and to researchers of hematology and blood flow in the microcirculation This book is written primarily for those who have some knowledge of chemistry biochemistry and general hematology The authors of each section bring a strong clinical emphasis to the book **Клінічна імунологія та алергологія. Англ. мовою** Бажора Ю.І., 2013 III IV *Manual of Clinical*

Microbiology, 4 Volume Set Karen C. Carroll, Michael A. Pfaller, 2024-11-19 Revised by a collaborative international interdisciplinary team of editors and authors this edition of the Manual of Clinical Microbiology includes the latest applications of genomics and proteomics and is filled with current findings regarding infectious agents leading edge diagnostic methods laboratory practices and safety guidelines This edition also features four new chapters Diagnostic Stewardship in Clinical Microbiology Salmonella Escherichia and Shigella and Morganellaceae Erwiniaceae Hafniaceae and Selected Enterobacterales This seminal reference of microbiology continues to set the standard for state of the science laboratory practice as the most authoritative reference in the field of microbiology If you are looking for online access to the latest from this reference or site access for your lab please visit www.wiley.com/learn/clinmicronow

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Technical Manual American Association of Blood Banks, 1996 **Flow Cytometry** Zbigniew Darzynkiewicz, J. Paul Robinson, Harry A.

Crissman, 1994 **Bone Marrow and Stem Cell Transplantation** Meral Beksaç, 2007-05-03 This volume is a compendium of cutting edge molecular methods for the successful transplantation of hematopoietic stem cells The contributors are world renown leaders in the field They describe promising tools for stem cell transplant research models such as in vivo bioluminescence imaging They discuss HLA typing PCR SSP typing and HLA antigens This volume is an invaluable source for biochemists molecular biologists and clinicians

The HLA Complex in Biology and Medicine Narinder K Mehra, 2010-11-26 A comprehensive guide to the HLA Human Leukocyte Antigen system for immunologists and clinicians this book contains up to date information on the MHC Major Histocompatibility Complex and its role in the immune response and in various diseases The book explores the biological significance and role of the HLA system in organ and haematopoietic stem cell transplantation management This volume is an invaluable guide to the full spectrum of HLA related science while also serving as a conceptual and technical resource for those involved in HLA related research and in clinical or surgical practice In addition it will be a primary point of contact for individuals working in other areas who suddenly find that their research is drawing them into the complexities of HLA genetics

Accreditation Requirements Manual of the American Association of Blood Banks American Association of Blood Banks, Robert E. Klein, 1987

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