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Long-term fate of terminally differentiated skeletal muscle cells following E1A-initiated cell cycle reactivation

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Abstract

We have previously shown that E1A reactivates the cell cycle in 'irreversibly' growth arrested, terminally differentiated (TD) cells. The molecular events following E1A-mediated reactivation of TD skeletal muscle cells have been extensively investigated. However, the long-term fate of the reactivated cells has not been directly determined. In this paper, E1A is used to reactivate TD myotubes derived from established cell lines or primary myoblasts. We show that the reactivated muscle cells continue proliferating beyond the end of the first cell cycle and progress through at least a second one. Experiments performed with an inducible E1A/estrogen receptor chimera indicate that the reactivated cell cycle is self-sustained, since E1A is exclusively necessary to reactivate TD cells, but is dispensable for both the continuation of the first cycle and the progression into the following one. Finally, we report that E1A-mediated reactivation of muscle cells results in apoptotic cell death that can be delayed by the antiapoptotic, adenoviral E1B 55 kDa oncogene. *Cell Death and Differentiation* (2000) **7**, 145–154.

Keywords: terminal differentiation; mitosis; cytokinesis; time-lapse videomicroscopy

Abbreviations: TD, terminally differentiated; MSC, mouse satellite cells; Ara-C, β-D-arabinofuranoside; BrdUrd, 5-bromo-2'-deoxyuridine; MHC, myosin heavy chain; PI, post infection; TUNEL, terminal deoxyribonucleotide transferase-mediated dUTP nick-end labeling; SiF, serum free

Introduction

Terminally differentiated cells are defined by specialized properties, which are the result of tissue-specific gene expression, associated with a physiologically irreversible growth arrest. Terminal differentiation characterizes the majority of the cells in an adult mammal. The inability of TD cells to replicate poses strategic problems to the organism. These cells must live as long as the organism itself, which requires extraordinary measures to ensure their survival. In addition, organs and systems devoid of stem cell compartments and whose parenchyma is composed exclusively of TD cells cannot renew their tissues by means of cell proliferation. This makes them especially vulnerable to irreplaceable cell loss caused by diseases or injuries. The ability to induce sustained proliferation of TD cells such as neurons, myocardiocytes, or endocrine cells would potentially have a major impact on the therapy of diseases and traumas of such organs.

One example of terminal differentiation is provided by skeletal muscle cells.[1] The differentiation process of these cells can be recapitulated *in vitro*. Primary as well as established myoblasts can be cultured extensively. Mitogen withdrawal starts the differentiation program, which begins with irreversible exit from the cell cycle (commitment). Postmitotic myoblasts then express muscle-specific genes, turning into mononucleated myocytes (biochemical differentiation). The final stage of skeletal muscle differentiation *in vitro* involves fusion of myocytes into multinucleated structures called myotubes (morphological differentiation).[1]

It has been shown that a number of DNA tumor virus oncogenes are able to reactivate the cell cycle in TD cells.[2–6] Adenoviruses bear the E1A oncogene, which shares this ability.[6] Adenoviruses are particularly well suited for cell cycle reactivation of TD cells, since they can infect almost all cell types[7,8] across a wide range of species,[6] irrespective of the proliferation status of the cells. In fact, we and others have shown that adenovirus infection and the consequent E1A expression can reactivate a variety of TD cells, including myotubes, adipocytes,[6,9] myocardiocytes,[9,10] and neurons (M Crescenzi, unpublished). E1A exerts such activity by activating genes normally expressed at the G1/S boundary.[11] By this mechanism, E1A in effect bypasses the cell cycle block that prevents TD myotubes from proceeding beyond mid-G1 phase upon mitogenic stimulation.[12]

We have shown that E1A-reactivated myotubes undergo DNA replication, mitosis, and cytokinesis.[6,9] However, the long-term fate of these cells has not been investigated in depth. Open questions include whether further cell cycles follow the first one, whether any fraction of the reactivated cells can survive indefinitely, and whether the activity of E1A is required to sustain the cell cycle or it is only needed to initiate it. All of these issues are relevant in view of potential therapeutic applications.

In this paper, we present evidence that TD skeletal muscle cells reactivated by E1A undergo more that one cell cycle. We show that the reactivated cells eventually die by apoptosis. Such outcome can be at least partially opposed by the antiapoptotic adenovirus E1B oncogene, which allows extended survival. Finally, making use of a

Reactivation Of The Cell Cycle In Terminally Differentiated Cells

Lingjun Ying

Reactivation Of The Cell Cycle In Terminally Differentiated Cells:

Reactivation of the Cell Cycle in Terminally Differentiated Cells Marco Crescenzi,2002 This volume deals with the most advanced areas of reactivation of the cell cycle in terminally differentiated cells Terminally differentiated cells have long been regarded as irreversibly unable to proliferate However this view is being overturned with great implications for both biological knowledge and potential therapeutic applications The basic science is presented in detail and the potentialities for exploitation in cell replacement therapy and tissue repair are highlighted For the first time large parts of this research field are covered in a single resource contributed by scientists who have given the most to its advancement in recent years This volume will be valuable for young scientists wishing to enter this field and will serve as an authoritative reference for those already working in it Reactivation of the Cell Cycle in Terminally Differentiated Cells Marco Crescenzi,2002 The DNA Damage Response: Implications on Cancer Formation and Treatment Kum Kum Khanna,Yosef Shiloh,2009-09-18 The eld of cellular responses to DNA damage has attained widespread recognition and interest in recent years commensurate with its fundamental role in the ma tenance of genomic stability These responses which are essential to preventing cellular death or malignant transformation are organized into a sophisticated s tem designated the DNA damage response This system operates in all living organisms to maintain genomic stability in the face of constant attacks on the DNA from a variety of endogenous by products of normal metabolism as well as exogenous agents such as radiation and toxic chemicals in the environment The response repairs DNA damage via an intricate cellular signal transduction network that coordinates with various processes such as regulation of DNA replication tr scriptional responses and temporary cell cycle arrest to allow the repair to take place Defects in this system result in severe genetic disorders involving tissue degeneration sensitivity to speci c damaging agents immunode ciency genomic instability cancer predisposition and premature aging The nding that many of the crucial players involved in DNA damage response are structurally and functionally conserved in different species spurred discoveries of new players through similar analyses in yeast and mammals We now understand the chain of events that leads to instantaneous activation of the massive cellular responses to DNA lesions This book summarizes several new concepts in this rapidly evolving eld and the advances in our understanding of the complex network of processes that respond to DNA damage **The CDK-Activating Kinase (CAK)** Philipp Kaldis,2003-01-31 This volume aims to extract and summarize all information about CAK by pointing out commonly accepted facts and unresolved issues It takes the reader from yeast to mammals and describes all areas that CAK is thought to be involved in This volume is designed to serve newcomers to the field as well as specialists any person interested in cell growth signal transduction and cancer will find this a useful tool to own *Ceramide Signaling* Anthony H. Futerman,2002 The volume assembles current information on the role of ceramide as a signalling molecule in 16 chapters written by leading workers in this area Specific attention is given to mechanisms of analysis of ceramide and its biophysical properties on enzymes of ceramide metabolism and down stream

targets of ceramide on the cross talk of ceramide signalling with other signalling pathways and on the role of ceramide in neuronal signalling Finally the book closes with a section on the therapeutic implications of ceramide action in the areas of cannabinoid action chemotherapy and atherosclerosis and illustrates the potential medical significance of delineating the roles of ceramide in cell signalling This is the first volume specifically devoted to ceramide signalling and will act as an invaluable resource for basic and medical researchers and graduate students wishing to get a state of the art overview of this rapidly moving field Handbook of Neurochemistry and Molecular Neurobiology Abel Lajtha,Regino Perez-Polo,Steffen Roßner,2008-06-06 The nervous system is highly fragile especially during aging illness and trauma This book addresses a small sampling of major constituents of neural function at the cellular and molecular level that play crucial roles in development and aging DNA Repair Inna Kruman,2011-11-07 The book consists of 31 chapters divided into six parts Each chapter is written by one or several experts in the corresponding area The scope of the book varies from the DNA damage response and DNA repair mechanisms to evolutionary aspects of DNA repair providing a snapshot of current understanding of the DNA repair processes A collection of articles presented by active and laboratory based investigators provides a clear understanding of the recent advances in the field of DNA repair **Stem Cells and Regenerative Medicine** Philippe Taupin,2008 The subject of this book is stem cell research and regenerative medicine Stem cells are undifferentiated cells that have the ability to differentiate into different lineages of the body Stem cells carry tremendous potential for the treatment of a broad range of disease and injuries Stem cells exist in embryonic fetal and adult tissues including the adult central nervous system This book aims at in depth the recent developments in stem cell research and regenerative medicine Though this book encompasses all the fields of stem cell research and regenerative medicine it emphasises adult neurogenesis and neural stem cell research and therapy **Biological Nitrogen Fixation, Sustainable Agriculture and the Environment** Yi-Ping Wang,Min Lin,Zhe-Xian Tian,Claudine Elmerich,William E. Newton,2006-01-30 The 14th International Nitrogen Fixation Congress was held in Beijing China from October 27th through November 1st 2004 This volume constitutes the proceedings of the Congress and represents a compilation of the presentations by scientists from more than 30 countries around the World who came to Beijing to discuss the progress made since the last Congress and to exchange ideas and information This year marked the 30th anniversary of the first Congress held in Pullman Washington USA in 1974 Since then this series of Congresses has met five times in North America three in the United States and once each in Canada and Mexico once in South America Brazil four times in Western Europe once each in Spain The Netherlands Germany and France once in Eastern Europe Russia and once in Australia and now for the first time in Asia China was a most appropriate choice because China is a big country with the largest population in the World about 1 3 billion people which is about 22% of the World s population It is traditionally an agricultural country even though China has only 7% of the available farming land This situation explains why agriculture and its productivity are major issues for the Chinese people its

government and the scientists in the field *Cumulated Index Medicus* ,1995 Cell-Cycle Mechanisms and Neuronal Cell Death Agata Copani,Ferdinando Nicoletti,2007-03-06 Cell Cycle Mechanisms and Neuronal Cell Death examines the role of cell cycle activation in the molecular mechanisms leading to neuronal degeneration Leading Authors discuss this topic in relation to the major neurological disorders including Alzheimer s disease stroke and epilepsy This book serves to gain new insights into the molecular determinants of neuronal death and to establish new targets for therapeutic intervention

Post-Translational Modifications and Compartmentalized Protein Quality Control in Cardiac Muscle and Disease Huabo Su,Aldrin V. Gomes,Mark J. Ranek,2021-10-18 Medical Aspects of Disability, Fourth Edition Herb Zaretsky,Herbert H. Zaretsky, PhD,Steven Flanagan,Alex Moroz,2010-07-20 Like its predecessor the book remains one of the best sources of rehabilitation practice preparation for students and the most enlightened guide for rehabilitation professionals Nan Zhang Hampton PhD CRC Department of Counseling and School PsychologyUniversity of Massachusetts Medical Aspects of Disability has continually been the go to resource for health care professionals educators and students Now in its fourth edition this landmark volume has been substantially revised updated and expanded comprehensively describing aspects of disability pertaining to medical conditions commonly encountered in rehabilitation settings This edition discusses important topics that have come to the forefront of medical rehabilitation and disability covering disabling conditions and disorders not only from clinical but also functional and psychological perspectives Chapter authors among the most widely respected authorities in their respective fields provide comprehensive guides on what to expect and how to manage each medical issue discussing the causative agents classification pain management psychological factors and much more Emerging Topics Discussed Social work in physical medicine The information revolution disability and assistive technology Complementary and alternative medicine Trends in medical rehabilitation delivery and payment systems Legislation and rehabilitation professionals Telerehabilitation solutions to distant and international care Disabling Conditions and Disorders Included Geriatric rehabilitation Limb deficiency Organ transplantation Cardiovascular disorders Traumatic brain injury Diabetes mellitus Epilepsy Visual impairments Peripheral vascular disorders Rehabilitation in cancer patients AIDS and HIV Rehabilitation in burns Speech language hearing and swallowing disorders *New Insights into Mechanisms of Epigenetic Modifiers in Plant Growth and Development* Ming Luo,Gabino Ríos,Tomasz Jacek Sarnowski,Shoudong Zhang,Marc Libault,Nitin Mantri,Jean-Benoit Charron,2020-02-12 Structure-Function Relationships of Human Pathogenic Viruses Andreas Holzenburg,Elke Bogner,2007-05-08 Structure Function Relationships of Human Pathogenic Viruses provides information on the mechanisms by which viruses enter the cell replicate package their DNA into capsids and mature into new virions The relation between structural features and the pathogenicity and oncogenicity of some of the most relevant human viral pathogens are demonstrated and the acquisition of defense mechanisms through virus host interactions are presented In contrast to textbooks this volume combines timely research data to provide a holistic view of viral pathogenesis

Furthermore Structure Function Relationships of Human Pathogenic Viruses illustrates in a single volume the fundamental processes involved in viral life cycles using up to date information from research laboratories around the world Knowledge of these processes is crucial to develop rationales for the design of future drugs The timeliness of the data and the comprehensive yet concise approach this book takes in order to present the world of viral pathogens should make it a frontrunner in higher education and R D **Alzheimer's and Parkinson's Diseases** Abraham Fisher,Israel Hanin,Werner Poewe,Manfred Windisch,2007 This issue is a dedicated supplement published in addition to the regular issues of Neurodegenerative Diseases containing congress abstracts Neurodegenerative Diseases is a well respected international peer reviewed journal in Neurology Supplement issues are included in the subscription *Abeta Peptide and Alzheimer's Disease* Colin J. Barrow,David H. Small,2006-12-22 Recent advances in genetics and brain biochemistry point to the Abeta peptide as the major culprit in causing neurodegeneration in Alzheimer s Disease AD Abeta Peptide and Alzheimer s Disease is specifically targeted at summarizing current knowledge of the Abeta peptide and its role in AD Written by leaders in the industrial and academic world specializing in this rapidly moving area the book covers fundamental biochemical studies on this peptide the genetic impact on Abeta expression and processing and various AD therapeutic strategies that target Abeta Although specifically focusing on the Abeta peptide and AD there is also some discussion on the similarity and differences of this peptide and AD with other amyloidogenic diseases such as prion disease Parkinsons and Huntingtons This book will primarily be of interest to AD researchers and clinicians but also to researchers of other amyloid diseases **Rb and Tumorigenesis** Maurizio Fanciulli,2007-02-26 Rb and Tumorigenesis examines how recent advances have demonstrated the interaction of Rb with chromatin remodeling enzymes This new title explores the potential roles of these interactions in Rb functions and provides some evidence that distinct Rb co repressor may target different genes in different phases of the cell cycle *The Cell Cycle in the Central Nervous System* Damir Janigro,2008-01-23 Cell Cycle in the Central Nervous System overviews the changes in cell cycle as they relate to prenatal and post natal brain development progression to neurological disease or tumor formation Topics covered range from the cell cycle during the prenatal development of the mammalian central nervous system to future directions in postnatal neurogenesis through gene transfer electrical stimulation and stem cell introduction Additional chapters examine the postnatal development of neurons and glia the regulation of cell cycle in glia and how that regulation may fail in pretumor conditions or following a nonneoplastic CNS response to injury Highlights include treatments of the effects of deep brain stimulation on brain development and repair the connection between the electrophysiological properties of neuroglia cell cycle and tumor progression and the varied immunological responses and their regulation by cell cycle **Journal of Cell Science** ,2001

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