ADVANCES IN POLYMER SCIENCE 193

Volume Editors R. Satchi-Fainaro · R. Duncan

Polymer Therapeutics II

Polymers as Drugs, Conjugates and Gene Delivery Systems

Polymer Therapeutics II

Polymers as Drugs, Conjugates and Gene Delivery Systems

Volume Editors: Ronit Satchi-Fainaro · Ruth Duncan

With contributions by

V. Y. Alakhov · S. Allen · C. M. Barnes · E. V. Batrakova
Y. T. A. Chim · M. C. Davies · R. Duncan · J. S. Ellis
J. Fang · K. Greish · A. V. Kabanov · K. Kataoka
H. Maeda · N. Nishiyama · C. J. Roberts
R. Satchi-Fainaro · S. Sherman · S. J. B. Tendler

Springer

The series *Advances in Polymer Science* presents critical reviews of the present and future trends in polymer and biopolymer science including chemistry, physical chemistry, physics and material science. It is adressed to all scientists at universities and in industry who wish to keep abreast of advances in the topics covered.
As a rule, contributions are specially commissioned. The editors and publishers will, however, always be pleased to receive suggestions and supplementary information. Papers are accepted for *Advances in Polymer Science* in English.
In references *Advances in Polymer Science* is abbreviated *Adv Polym Sci* and is cited as a journal.

Springer WWW home page: http://www.springer.com
Visit the APS content at http://www.springerlink.com/

Library of Congress Control Number: 2005933609

ISSN 0065-3195
ISBN-10 3-540-29211-X Springer Berlin Heidelberg New York
ISBN-13 978-3-540-29211-1 Springer Berlin Heidelberg New York
DOI 10.1007/11547785

Springer is a part of Springer Science+Business Media

springer.com

Printed in Germany

Cover design: *Design & Production* GmbH, Heidelberg
Typesetting and Production: LE-TEX Jelonek, Schmidt & Vöckler GbR, Leipzig

Printed on acid-free paper 02/3141 YL – 5 4 3 2 1 0

193
Advances in Polymer Science

Advances in Polymer Science

Recently Published and Forthcoming Volumes

Surface-Initiated Polymerization II
Volume Editor: Jordan, R.
Vol. 198, 2006

Surface-Initiated Polymerization I
Volume Editor: Jordan, R.
Vol. 197, 2006

Conformation-Dependent Design of Sequences in Copolymers II
Volume Editor: Khokhlov, A. R.
Vol. 196, 2006

Conformation-Dependent Design of Sequences in Copolymers I
Volume Editor: Khokhlov, A. R.
Vol. 195, 2006

Enzyme-Catalyzed Synthesis of Polymers
Volume Editors: Kobayashi, S., Ritter, H., Kaplan, D.
Vol. 194, 2006

Polymer Therapeutics II
Polymers as Drugs, Conjugates and Gene Delivery Systems
Volume Editors: Satchi-Fainaro, R., Duncan, R.
Vol. 193, 2006

Polymer Therapeutics I
Polymers as Drugs, Conjugates and Gene Delivery Systems
Volume Editors: Satchi-Fainaro, R., Duncan, R.
Vol. 192, 2006

Interphases and Mesophases in Polymer Crystallization III
Volume Editor: Allegra, G.
Vol. 191, 2005

Block Copolymers II
Volume Editor: Abetz, V.
Vol. 190, 2005

Block Copolymers I
Volume Editor: Abetz, V.
Vol. 189, 2005

Intrinsic Molecular Mobility and Toughness of Polymers II
Volume Editor: Kausch, H.-H.
Vol. 188, 2005

Intrinsic Molecular Mobility and Toughness of Polymers I
Volume Editor: Kausch, H.-H.
Vol. 187, 2005

Polysaccharides I
Structure, Characterization and Use
Volume Editor: Heinze, T.
Vol. 186, 2005

Advanced Computer Simulation Approaches for Soft Matter Sciences II
Volume Editors: Holm, C., Kremer, K.
Vol. 185, 2005

Crosslinking in Materials Science
Vol. 184, 2005

Phase Behavior of Polymer Blends
Volume Editor: Freed, K.
Vol. 183, 2005

Polymer Analysis/Polymer Theory
Vol. 182, 2005

Interphases and Mesophases in Polymer Crystallization II
Volume Editor: Allegra, G.
Vol. 181, 2005

Interphases and Mesophases in Polymer Crystallization I
Volume Editor: Allegra, G.
Vol. 180, 2005

Volume Editors

Dr. Ronit Satchi-Fainaro
Harvard Medical School
and Children's Hospital
Boston Department of Surgery
Vascular Biology Program
1 Blackfan Circle
Boston, MA 02115, USA
ronit.satchi-fainaro@childrens.harvard.edu

Prof. Ruth Duncan
Welsh School of Pharmacy
Cardiff University
Redwood Building
King Edward VII Avenue
Cardiff CF 10 3XF, UK
DuncanR@cf.ac.uk

Editorial Board

Advances in Polymer Science
Also Available Electronically

For all customers who have a standing order to Advances in Polymer Science, we offer the electronic version via SpringerLink free of charge. Please contact your librarian who can receive a password or free access to the full articles by registering at:

springerlink.com

If you do not have a subscription, you can still view the tables of contents of the volumes and the abstract of each article by going to the SpringerLink Homepage, clicking on "Browse by Online Libraries", then "Chemical Sciences", and finally choose Advances in Polymer Science.

You will find information about the

- Editorial Board
- Aims and Scope
- Instructions for Authors
- Sample Contribution

at springeronline.com using the search function.

Contents of Volume 192

Polymer Therapeutics I

Volume Editors: Ronit Satchi-Fainaro, Ruth Duncan
ISBN: 3-540-29210-1

Contents

Adv Polym Sci (2006) 193: 1–65
DOI 10.1007/12_024

Published online: 10 November 2005

Polymer Therapeutics for Cancer: Current Status and Future Challenges

Ronit Satchi-Fainaro[1] (✉) · Ruth Duncan[2] · Carmen M. Barnes[1]

[1]Harvard Medical School, Children's Hospital Boston, Department of Surgery, Vascular Biology Program, Karp Family Research Laboratories 12.007D, 1 Blackfan Circle, Boston, MA 02115, USA
ronit.satchi-fainaro@childrens.harvard.edu

[2]Centre for Polymer Therapeutics Welsh School of Pharmacy, Cardiff University, Redwood Building, King Edward VII Avenue, Cardiff CF10 3XF, UK

Abstract Drug delivery systems for cancer therapeutics have revolutionized medicine. Delivery systems have improved the efficacy and reduced the toxicity of current therapies and resulted in the development of new ones. Today, millions of cancer patients have directly benefited from drug delivery systems, and polymers have been at the frontline of these technological advances. Targeted delivery systems of chemotherapeutics to the tumour compartment can be achieved systemically, either passively or actively. Polymer conjugation radically changes the pharmacokinetics of the bound drug, and conjugates with prolonged circulation times target tumours passively via the enhanced permeability and retention (EPR) effect. Polymer conjugates can also be modified with moieties to directly target the tumour cells or the tumour vasculature. In this chapter, we review the successful clinical application of polymer–protein conjugates, and promising clinical results arising from trials with polymer–anticancer-drug conjugates. Over the last decade more than twelve polymer-drug conjugates have entered Phase I/II clinical trial as intravenously injectable anticancer agents. Only one of the polymer conjugates that has reached clinical trial directly targets tumour cells, while another one targets the tumour vasculature. Conjugation to polymers may save the fate of the many promising drug/peptide chemotherapies that fail each year due to high toxicity or poor pharmacokinetics. Yet, these technologies have not been exploited to their full potential. Only a few combinations of a limited number of chemotherapeutic drugs and polymer delivery systems are being tested in clinical and preclinical trials today. Furthermore, genomics and proteomics research is producing novel peptides, proteins and oligonucleotides that lack effective delivery systems. Thus, the full potential for drug delivery systems based on NCEs (new chemical entities), such as "polymer therapeutics", lies ahead.

Keywords Angiogenesis · Drug targeting · EPR effect · HPMA copolymer · PEG · Polymer therapeutics

Abbreviations

Amino-DAQ	1,5-diazaanthraquinone derivative
ASCO	American Society of Clinical Oncology
ASGP	Asialoglycoprotein
ASGPR	Asialoglycoprotein receptor
ATWLPPR	Alanine-threonine-tryptophan-leucine-proline-proline-arginine
AUC	Area under the curve
BBB	Blood brain barrier
BCNU	1,3-bis(2-chloroethyl)-1-nitrosourea
bFGF	Basic fibroblast growth factor
CM	Carboxymethyl
CPT	Camptothecin
Da	Daltons
DAO	D-amino acid oxidase
DES	Diethylstilboestrol
DLT	Dose limiting toxicity
DMXAA	Dimethyl-xanthenone-4-acetic acid
DOTA	1,4,7,10-tetraazacyclododecane-*N*,*N*′,*N*″,*N*‴-tetraacetic acid
Dox	Doxorubicin
DSPE	Distearoylphosphatidylethanolamine
EC	Endothelial cell
EGF	Epidermal growth factor
en	Ethylenediamine
EPR effect	Enhanced permeability and retention effect
FDA	Food and Drug Administration
FPLC	Fast protein liquid chromatography
HIV/AIDS	Human immunodefficiency virus/Acquired immunodefficiency syndrome
HO	Heme oxygenase
HPLC	High-pressure liquid chromatography
HPMA	*N*-(2-hydroxypropyl)methacrylamide
HuIg	Human immunoglobulin
i.p.	Intraperitonealy
i.v.	Intravenously
IFL	Irinotecan, fluorouracil, and [calcium folinate] leucovorin
IFN-α	Interferon-α
IFN-β	Interferon-β
IgG	Immunoglobulin
IL-6	Interleukin-6
LAK cells	Lymphokine-activated killer cell
LD_{10}	Dose of drug lethal to 10% of animals
MA	Methacryloyil
mAb	Monoclonal antibody
MAG	HPMA: methacryloyl-glycine (MA-Gly)-ONp 95:5 or 90:10
MMP-2	Matrix metalloproteinase-2
MMP-9	Matrix metalloproteinase-2
mPEG	monoPEG

MTD	Maximum tolerated dose
NCE	New chemical entities
NGR	Asparagine-glycine-arginine
NK	Natural killer cells
NSCLC	Non-small-cell lung carcinoma
O_2^-	Superoxide anion
ONp	*p*-Nitrophenyl
PAAm	Polyacrylamide
PCT	Paclitaxel
PDAAm	Polydimethylacrylamide
PDEPT	Polymer directed enzyme prodrug therapy
PEG	Polyethyleneglycol
PEG-G-CSF	PEGylated recombinant methionyl human granulocyte colony stimulating factor
PEI	Poly(ethyleneimine)
PELT	Polymer enzyme liposome therapy
PGA	Poly-*L*-glutamic acid
PLC	Phospholipase C
POG	Pediatric Oncology Group
PS2	Poor performance status 2
PVA	Polyvinyl alcohol
PVP	Polyvinylpyrrolidone
RES	Reticuloendothelial system
RGD	Arginine-glycine-aspartate
ROS	Reactive oxygen species
s.c.	Subcutaneously
ScFv	Single-chain Fv antibody fragment
SCID	Severe combined immunodeficiency disease
SCLC	Small-cell lung cancer
SMANCS	Styrene-co-maleic anhydride-neocarzinostatin
SS-NH-PEG	Succinimidyl ester of PEG
STELLAR	Selective targeting for efficacy in lung cancer, Lower adverse reaction
TBA	Thiobutylamidine
TEM	Tumor endothelial marker
TNFα	Tumor necrosis factor-α
VEGF/VPF	Vascular endothelial growth factor/Vascular permeability factor
VEGFR	Vascular endothelial growth factor receptor
VTA	Vascular targeting agents
XO	Xanthine oxidase
ZnPP	Zinc protoporphyrin

1 Introduction

Chemotherapeutic treatment of neoplastic diseases is often restricted by adverse systemic toxicity, which limits the dose of drug that can be administered, or by the appearance of drug resistance. Lack of selectivity is only one (albeit a major) obstacle hindering the optimisation of drug effective-

ness. Others include inaccessibility of target, premature drug metabolism and allergic reactions [1]. There is a great demand for innovative drug delivery systems that can better target antitumour drugs and that can overcome resistance in its many forms. The question is: how can we meet these challenges?

A great deal of research has concentrated on ways to develop new cancer therapeutics that specifically target tumour cells compared with normal cells, exploiting the differences between neoplastic and normal tissues. These targeted therapies should be more effective and decrease toxicity to normal tissues.

Several systems have been developed in order to restrict the delivery of the chemotherapeutic agent to the tumour site. With the identification of cell-specific receptor/antigens on tumour cells [2] and tumour endothelial cells [3], it has been possible to actively target chemotherapeutic or antiangiogenic agents using ligand- or antibody-bearing delivery systems. Alternatively, the drug can be loaded into high-capacity drug carriers such as liposomes or entrapped in degradable polymers for sustained drug release and localized chemotherapy systems [4]. In the controlled polymer drug delivery systems, the active molecule is released continuously at therapeutic levels by polymer degradation and diffusion through the polymer pores. Clinical approved examples include Zoladex [5, 6], Lupron Depot, and Decapeptyl [7], which are injectable polymer rods or microspheres of luteinizing hormone-releasing hormone (LHRH) analogues for the treatment of advanced prostate cancer [4, 8]. Localized chemotherapy systems have been particularly appealing for the brain, where the presence of the blood–brain barrier limits delivery of therapeutics by blood. Gliadel, an implantable polymer wafer that locally delivers carmustine, has been used successfully for the treatment of malignant gliomas after surgery [9]. Interestingly, we found that HPMA copolymer-TNP-470 (caplostatin) [10] was able to treat orthotopic intracranial U87 human glioblastoma in mice [11], even though it does not cross the blood brain barrier, a fact that eliminated the neurotoxicity associated with the unconjugated TNP-470. This can be attributed to the leakiness of blood vessels in some brain tumours, allowing polymer conjugates to target these tumours by the EPR effect.

Drugs can also be conjugated to polymer carriers, named "polymer therapeutics" [12], that can be either directly conjugated to targeting proteins/peptides or derivatised with adapters conjugated to a targeting moiety. "Polymer therapeutics" [13] is a term used to describe polymeric drugs [14], polymer-drug conjugates [15], polymer-protein conjugates [16], polymeric micelles to which a drug is covalently bound [17], and multicomponent polyplexes that are being developed as nonviral vectors [18] (Fig. 1). All subclasses consist of at least three parts: (a) a specific water-soluble polymer, either as the bioactive itself or as an inert functional part of a multifaceted construct for improved drug, protein or gene delivery; (b) a biodegradable polymer-drug linker, and; (c) the bioactive antitumour drug.

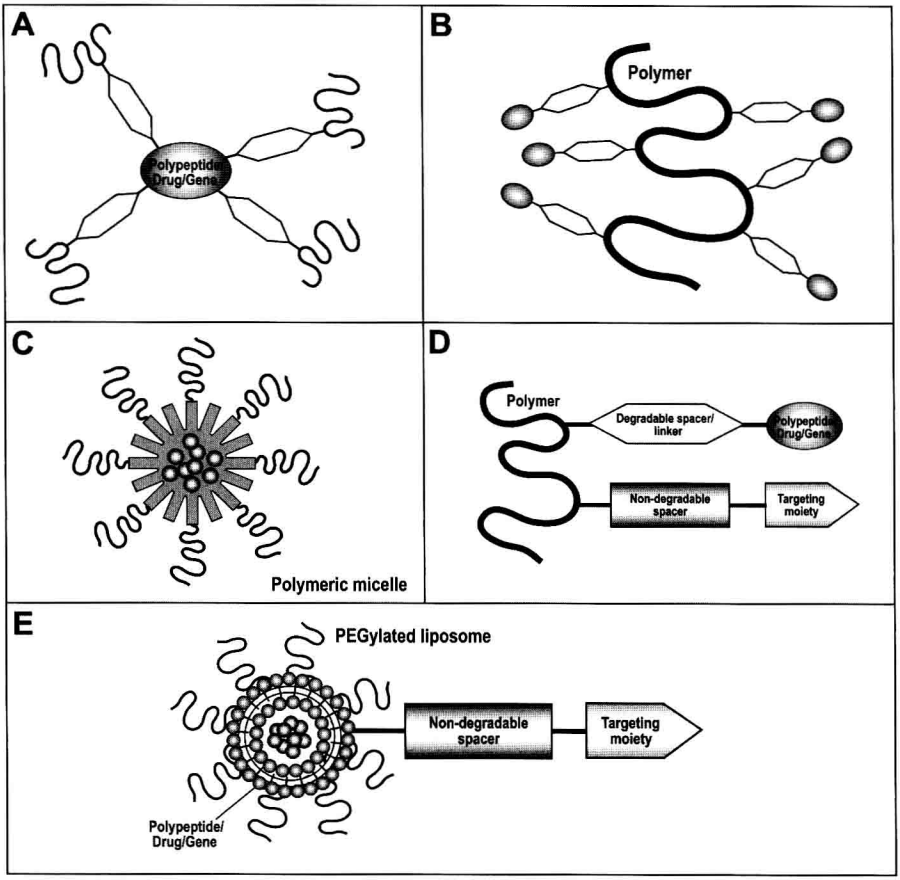

Fig. 1 Schematic diagram of possible combinations of actively targeted conjugates: **A** Soluble polymer-protein conjugate (20 nm) or polyplex: hydrophilic polymers bearing a cationic block-DNA complex (40–60 nm); **B** Soluble polymeric drug (5–15 nm) carrier (polymer therapeutics, modified from [87]); **C** Polymeric micelle (60–100 nm) – amphiphilic block entrapping a drug; **D** Soluble polymeric drug carrier bearing a targeting moiety (5–15 nm); **E** PEGylated stealth liposome carrying the active entity conjugated to a targeting moiety (200–500 nm)

Because in polymer therapeutics the drugs are chemically conjugated, they differ from controlled drug delivery systems in that they are more like new chemical entities (NCE). Not only is their pharmacokinetic profile distinct from that of the parent drug, but the route of cellular uptake may also differ, as the polymer-drug can only enter cells by the endocytic route, leading to lysosomotropic drug delivery. Several conjugates can release drug intracellularly while others release it extracellularly, depending on the polymer-drug linker and the activating moiety. While polymer therapeutics share many

features with other macromolecular drugs and prodrugs (proteins, antibodies, and oligonucleotides, and immunoconjugates), their chemistry makes them amenable to flexible tailoring, for example of their molecular weight, number and types of drugs per polymer, targeting moieties and even bioresponsive elements [12]. Polymer-protein conjugates have made it to the clinic since the early 1990s, with the approval of polyethylene glycol (PEG)-adenosine deaminase, PEG-*L*-asparaginase and styrene maleic anhydride (SMANCS) [19]. During the past two decades, the field of polymer therapeutics has continued to grow due to the advances in both polymer chemistry and biological sciences, and promising results from clinical trials involving polymer-anticancer-drug conjugates [12]. With the emergence of hybrid biotechnologies, which combine the synthesis of innovative polymers with biological macromolecules (proteins, oligonucleotides, antibodies), a number of compounds have been developed that are suitable for clinical development and use (Tables 1, 2, and 3).

It is surprising, however, that with the abundance of novel drugs and targets offered in the post-genomic era and novel sophisticated chemistry available, only four drugs (doxorubicin, camptothecin, paclitaxel and platinate) and four polymers (HPMA copolymer, Poly-*L*-glutamic acid, PEG, and Dextran) are repeatedly used to develop these promising new polymer therapeutics. Therefore, we will examine here future directions and challenges in this field. The purpose of this chapter is to compare different therapeutic targeted delivery systems and strategies for chemotherapeutic and antiangiogenic agents, focusing on those polymer therapeutics that have been approved by the FDA or that are undergoing clinical and preclinical trials. The rationale for the design of preclinical lead compounds is summarised, and the challenges for effective and clinical development of these complex macromolecular prodrugs are discussed.

2
Passive or active targeting?

Targeting can be achieved either actively, by specifically including a recognition moiety into the carrier ("active targeting"), or passively, as a result of some physical or chemical characteristics of the carrier ("passive targeting") [20] (Fig. 2). The active approach relies upon the selective localisation of a ligand at a cell-specific receptor. Passive targeting refers to the exploitation of the natural (passive) distribution pattern of a drug-carrier in vivo. The latter is based upon mechanical entrapment of the carrier by shape or size or uptake by the cells of the reticuloendothelial system (RES). Maeda called the passive targeting phenomenon the "enhanced permeability and retention (EPR) effect" [21], and attributed it to two factors: the disorganised pathology of angiogenic tumour vasculature with its discontinous endothelium,