Advanced Drug Delivery Reviews 130 (2018) 3–11

Contents lists available at ScienceDirect

Advanced Drug Delivery Reviews

journal homepage: www.elsevier.com/locate/addr

Perspectives on the past, present, and future of cancer nanomedicine☆

Yu Seok Youn [a], You Han Bae [b,*]

[a] School of Pharmacy, Sungkyunkwan University, Suwon, Gyeonggi-do, Republic of Korea
[b] Department of Pharmaceutics and Pharmaceutical Chemistry, University of Utah, Salt Lake City, UT, USA

ARTICLE INFO

Article history:
Received 18 March 2018
Received in revised form 27 April 2018
Accepted 15 May 2018
Available online 18 May 2018

ABSTRACT

The justification of cancer nanomedicine relies on enhanced permeation (EP) and retention (R) effect and the capability of intracellular targeting due primarily to size after internalization (endocytosis) into the individual target cells. The EPR effect implies improved efficacy. Affinity targeting for solid tumors only occur after delivery to individual cells, which help internalization and/or retention. The design principles have been supported by animal results in numerous publications, but hardly translated. The natures of EP and R, such as frequency of large openings in tumor vasculature and their dynamics, are not understood, in particular, in clinical settings. Although various attempts to address the issues related to EP and delivery, by modifying design factors and manipulating tumor microenvironment, are being reported, they are still verified in artificial rodent tumors which do not mimic the nature of human tumor physiology/pathology in terms of transport and delivery. The clinical trials of experimental nanomedicine have experienced unexpected adverse effects with modest improvement in efficacy when compared to current frontline therapy. Future nanomedicine may require new design principles without consideration of EP and affinity targeting. A possible direction is to set new approaches to intentionally minimize adverse effects, rather than aiming at better efficacy, which can widen the therapeutic window of an anticancer drug of interest. Broadening indications and administration routes of developed therapeutic nanotechnology would benefit patients.

Contents

1. Introduction

The word “nano” has become extremely popular not only in the scientific community but also among the general public. The use of “nano” suggests objective way and subjective feeling of renewing existing things more innovative and/or opening up new possibilities. Academia and industries have achieved remarkable progress through nanotechnology; in pharmaceutics and pharmacology, the generic property suggested by “nano”, innovation, have provoked significant anticipation in nanomedicine for cancer treatment. Looking back on the past 30 years, the overall clinical outcomes of cancer nanomedicine are, however, suboptimal, thus raising questions about reasons for their underperformance and true prescriptions for better cancer nanomedicine. In this short review, a concise summary of cancer nanomedicine from both retrospective and prospective views is provided. Herein, the prototypes of nanomedicine described are limited to intravenously administered therapeutic (not diagnostic) nanoparticles, such as polymer-drug

☆ This review is part of the *Advanced Drug Delivery Reviews* theme issue on “Perspectives and review articles on nanomedicine from NanoDDS 2017”.

* Corresponding author.
E-mail address: you.bae@utah.edu (Y.H. Bae).

https://doi.org/10.1016/j.addr.2018.05.008
0169-409X/

conjugates (hereafter referred to as nanoconjugates), liposomes, polymeric micelles, and protein-based nanoparticles, because of the breadth of preclinical and clinical cases available to guide potential future directions.

2. Therapeutic cancer nanomedicine

Colloid (or colloidal particle) was discovered long before the modern nanomedicine era that started in the 1990s. Since then, the scientific term "colloidal" was replaced by "nano." Liposomes were discovered and described in the early 1960's [1,2], and their size range (except for giant or large multilamellar vesicles) are largely identical to the size range of colloids: small unilamellar vesicles are in a range of 20–100 nm and large ones100–400 nm [3,4]. There is no doubt that the unilamellar liposomes fit for the category of modern nanomedicine. Researchers have often used the term 'nano-liposomes' until the early 2000s, adding unnecessary "nano" to liposomes, although both showed little disparity in terms of colloidal size range and physical aspects in most studies [4,5]. Conventional liposomal formulations have become a unique category of therapeutic nanomedicine that shows the highest potential for Food and Drug Administration (FDA) approval [6,7].

Since the approval of Doxil® by FDA in 1995, we have noticed no nanomedicine products with significant clinical outcomes, i.e., improving efficacy, alleviating toxicity, and thereby extending the lifespan of cancer patients. Despite hundreds of thousands of scientific papers published and trillions of research dollars invested in cancer nanomedicine, the prototype formulations available on the market are all liposomes [DaunoXome® (daunorubicin, 1996), DepoCyt® (cytarabine, 1996), Marqibo® (vincristine, 2012), and Onivyde® (irinotecan, 2012)] [6,8,9], except for the protein-based nanoparticle [Abraxane® (paclitaxel, 2005)] [8]. The polymeric micellar formulation of Genexol-PM® (approved in South Korea, 2007) is yet under clinical phase II/III investigation in the USA. Supposedly, Abraxane® and Genexol-PM® are closer to non-toxic solubilizing formulations for paclitaxel because they quickly dissociate, breaking into smaller fragments (~10 nm) in the blood [10–15], despite their original sizes claimed to be 130 and 20–50 nm, respectively [9,16–18]. The clinical approval of both Doxil® and Abraxane® was attributed to reduced or altered toxicity profiles rather than improved efficacy [19]. On the other hand, nanoconjugates, which have been popular in industry for decades, have faded away without a single pharmaceutical product getting beyond clinical trials. The clinical status of polymeric micelles is similar to or little better than that of nanoconjugates, with Genexol-PM® being the exception, to date.

It was one of prime importance to focus on intracellular targeting of nanomedicine after internalization. Modern functional nanomedicines have emerged due primarily to their typical size range of 10–300 nm, which is two orders smaller than the average size of ordinary cancer cells. It was hypothesized that the designed/planned functions are expected to occur at targeted subcellular organelles in a controlled manner. This belief started with anticancer drugs and expanded to the delivery of genetic materials. However, prior to this, nanoparticles must be delivered to individual target cells, taking a long journey in harsh in vivo environment [20,21], and a large quantity of internalized nanoparticles are prone to be sequestered and degraded in endosomes or lysosomes [22–25], not going to cytoplasm or nucleus [26,27]. Unless nanoparticles are transported into the right site of action in cancer cells in a timely and accurate manner, there may be less pharmacological manifestations, despite seemingly sufficient levels of drugs and nanoparticles accumulated in the peripheral tumor region [19].

3. Design principles

Since the enhanced permeability (EP) and retention (R) effect was first coined and described by Dr. Hiroshi Maeda based on his observation of the Lipiodol/SMANCS [an anticancer protein neocarzinostatin (NCS) conjugated with a styrene-maleic acid (SMA) copolymer] system [13,28–30], this widespread formalized concept has been a catalyst to facilitate the use of nanotechnology in pharmaceutical field. However, the EPR effect has also been an advocate to justify indiscriminate clinical trials of cancer nanomedicine regardless of efficacy validation. For this reason, almost all articles dealing with therapeutic cancer nanoparticles include the term EPR, making it one of the most cited terms in the field of drug delivery and formulations [13,31].

The design basis of cancer nanomedicine, in particular for solid tumors, are the (i) EPR effect and (ii) cancer cell-specific affinity targeting, with the aim of broadening the therapeutic window by enhancing efficacy and reducing toxicity [32,33]. More accumulation in target sites and thereby less exposure to other organs are expected by the EPR effect. In this regard, long circulation is thought to contribute accumulation by EP [34]. Such a conception based on "passive" and "active" targeting has been emphasized for a long time in describing the in vivo fate of nanoparticles [20,32].

Much of the optimistic anticipation surrounding the clinical potential of modern cancer nanomedicine has been due to this EPR effect [35,36]: in brief, nanoparticles are highly permeable through tumor vasculatures due to large pores (sometimes reaching several hundred nm to 2 μm) [37] and, in turn, retain long by dysfunctional and immature drainage in lymphatic vessels, vs. normal vasculatures. However, this idealized concept is partially correct in animal cancer models where it goes well but not optimally applied in human cancer physiology [38–40]. Recently, many articles have spoken up the voice that the EPR effect is controversial and not a universal solution to cancer nanomedicine, on the basis of many conflicting clinical studies [38,41]. Doxil® showed only limited therapeutic success in soft tumors, such as Kaposi's sarcomas or multiple myeloma, where the EPR is applied well due to less transport barriers to cancer cells [42–44], whereas it did not show superior efficacy over free doxorubicin formulations in most solid tumors, such as, ovarian and breast cancers [45,46].

The EPR effect and targeting rely on a few assumptions: (i) mass transport occurs toward cancer cells regardless of transport resistance, concentration gradient (can be reversed over time), and interstitial fluid pressure (IFP); (ii) IFP applies to collapsed lymphatic vessels selectively, not blood vessels; (iii) long-range strong interactions attract the circulating nanoparticles in the blood to the target cells in tumors. However, such assumptions have been proven inappropriate by many studies based on the following evidence.

First, nanoparticles do not accumulate easily in and penetrate into solid tumors by the EPR due to reverse pressure gradient and transport resistance by tumor physiology [19]. In normal tissues, the forces that determine the net filtration pressure, determined by hydrostatic and colloid osmotic pressures of blood vessels and interstitum, is slightly positive toward interstitum (from blood vessels) [47]. However, in tumor tissues, the forces become reverse, mainly due to elevated IFP that is derived from complex physiological reasons, including poorly organized and leaky vasculature, dysfunctional lymphatic vessels, or dense and highly organized extracellular matrix (ECM) [48,49]. The elevated IFP, described as an interstitial "hypertension" in solid tumors, exacerbates bulk fluid flow to interstitum from blood vessels, which in turn limits the nanoparticle extravasation/penetration [50–53]. Nanoparticles are impelled to pass through the openings by diffusion rather than bulk flow and sometimes pushed back to the blood vessels [54]. Recent studies show that normalization of tumor blood vessels reduces IFP and allows improved delivery of smaller nanoparticles, describing that the first assumption of the EPR is not universal [50,55].

Second, through a series of studies on a Lipiodol/SMANCS formulation, Dr. Maeda reached a conclusion that extensive accumulation of macromolecules in tumors was due to enhanced vascular permeability and lack of the lymphatic recovery [30,31]. This phenomenon is an analogy to a traffic jam resulting from a wide entrance and a narrow exit, similar with a bottleneck. In reality, elevated IFP is responsible not

only for lack of lymphatic drainage but also collapsed tumor vasculature [51], and has been well reported in clinical human tumors [54,56,57]. Aside from exit of lymphatic drainage, entrance into tumor tissue is not so amenable to nanoparticles because of poor blood supply and IFP [56,58]. The dynamics of openings between endothelial cells of the tumor blood vessels could critically influence on accumulation and retention of nanoparticles [59], but is not counted at all in conventional EPR concept.

Third, "active targeting" is based on the idea that targeted drugs are able to exert specific selectivity only on target cells. This classical concept goes back to the hypothetical "magic bullet," in which a targeted system can solely attack cancer cells without damage to surrounding healthy tissues, suggested by Paul Ehrlich [21,60]. However, clinical active targeting has been hardly observed because human tumor system and cancer biology is much more complicated and unfavorable to targeted nanomedicine. In addition, current targeted nanomedicine is unable to automatically guide themselves to solely cancer cells, even despite the use of external trigger systems [20,61,62]. More accurately, active targeting refers to specific interactions between carrier nanoparticles and target cells, depending upon ligand-receptor interactions, which take place in close proximity (within a few nanometers) [63]. Therefore, attraction forces between ligands and receptors become weak beyond the distance range, despite high specific affinity (K_d) between the two.

We must be aware that nanomedicine requires systemic targeting based on blood circulation and extravasation in vivo prior to close ligand-receptor interactions. Clinically, the in vivo environment is dynamic and chaotic, unlike static in vitro experiments. Blood flow is so fast that nanoparticles are forced to circulate the whole body approximately once per minute because cardiac output is >5 L/min in human adults having a ~5-L total body fluid volume [64–66]. The velocity of blood flow is approximately 1.5–33 cm/s (minimum at capillaries and venules) [67]. In addition, the total length of blood vessels in the average human adult is ~100,000 km [68]. Although this is so exorbitantly huge to match our mathematical calculation, we can easily assume that drugs/nanoparticles are placed at injection sites truly far from tumor sites (presumably > several tens to hundreds of meters). Consequently, nanomedicine (if ~100 nm; 10^{-5} cm) faces a very long journey at a high speed (> several cm/s), after which it is apt to bypass tumors (at most a few centimeters in size) in sub seconds. The point is that ligands and receptors (even if exist at endothelial surface) must rendezvous within a few seconds at a distance of a few nanometers while transiting via rapid blood flow. It is like the rendezvous docking usually done in a few tens centimeter distance between spacecraft and space station, both of which orbit around tremendously fast in space. Meanwhile, nanomedicines must be designed to avoid their accumulation in normal healthy tissue. It is very much unlikely. As such, in reality, the net contribution of so-called active targeting has not been noticeable in clinical settings. On this basis, long-lasting nanomedicines may have significantly more opportunity to target tumors, in either passive or active mode, but unfortunately have an equal chance of targeting non-tumor cells.

Only a limited fraction of blood volume, depending on tumor size (typically smaller than 5 cm in diameter in human patients) and degree of blood vessel density in the tumors, circulates into tumor vasculature. Nanomedicine having ligands penetrated into the tumor interstitial space may encounter more favorable conditions for receptor interactions because interstitial fluid flow is much slower (< tens of μm/s) than in blood vessels [69]. Nevertheless, there are many factors that interfere with the docking of ligands and receptors. Again, ligand-receptor interactions take place within a few nanometers of proximity under amenable static conditions [63] via non-covalent bonding, such as, Van der Waals forces, hydrophobic, π-, ionic- and electrostatic interactions [70]. Tumor interstitial space is chaotic due to elevated IFP and dense ECM, pushing nanoparticles back into the blood vessels and preventing access to individual tumor cells having relevant receptors [19]. Large regions that are poorly vascularized in rapidly growing tumors also provide a considerable distance for the diffusion of nanoparticles toward cancer cell receptors [71]. Some nanoparticles require hours or even days to diffuse only 200 μm, failing to reach tumor core [72,73].

Cancer cell heterogeneity is one of the most critical factors in the nullification of active targeting. Cancer cells are not homogenous, consist of multiple cell types and in a dynamic state caused by genetic instability and epigenetic diversity. Cancer cells respond differently to chemotherapeutics and show diverse survival rates [19,74]. In this regard, the targeted delivery of nanomedicine is only able to kill a fraction of cancer cells having specific receptors located in the accessible region of bulk tumors [32]. Many articles have highlighted the potential of active targeting based on receptor overexpression in tumors, but in most human tumors, unlike in rodents, the expression percentage of a specific receptor is insufficient to achieve successful tumor control via nanomedicine. It is well-documented that if only 10% of the cells in breast cancer samples are strongly positive to HER-2/*neu*, such patients should undergo Herceptin® therapy [20,75]. This clinical example indicates the real difficulty of targeting using ligand-receptor interactions in bulk tumors on account of tumor heterogeneity. Saying, a large fraction of nanomedicine is abandoned with little therapeutic use even though 100% of them are perfectly delivered to relevant bulk tumors. Also, the fact that many normal tissues express specific receptors for certain relevant ligands, such as, transferrin [76–78], folic acid [79,80], biotin [81,82], and RGD [83], among others, is a continuing concern. Nanomedicines that do not reach tumor cells can affect normal healthy cells. Moreover, the expression of specific receptors on tumors is heterogeneous and the molecular characteristics of the relevant tumors may change with time [84]. As such, active targeting is much far from our design and desire.

4. Retrospective view

The history of modern nanomedicine is short but has a firm basis in rational design compared with conventional screening of oncology drugs. The overall clinical success rate of oncology drugs is ~5% from phase I to FDA approval, but that of targeted drugs (including therapeutic antibodies) has reached ~10% [85]. By far, the net contribution of nanomedicine to success rate has not met initial expectations. Focusing on prototypes of "therapeutic" nanomedicines encapsulating anticancer drugs, since the approval of Doxil®, only five nanomedicines have appeared on the global market (DaunoXome®, DepoCyt®, Abraxane®, Marqibo® and Onivyde®) [6,8,9]. Among these, Abraxane® is the only unique prototype, albumin-bound nanoparticles, that is not a liposome. Although over twenty years have passed since the release of Doxil®, liposomal formulations are still a major category. As mentioned above, polymeric micellar formulations (including Genexol-PM®) are still under clinical trials in the USA. Doxil® significantly reduced cardiac toxicity but caused notable skin toxicity in hands and feet due to its extended blood circulation time [41]. However, it failed to prove improved antitumor efficacy over existing doxorubicin formulations, which indicates that the EPR effect could not be practically applied to human tumor physiology. The clinical success of nanomedicines approved to date relies heavily on altered toxicity profile rather than enhanced anticancer efficacy [86].

One important note is that nanomedicine review papers always list drug candidates in clinical trials and approved products. Table 1 shows the clinical trial status of nanomedicine candidates from 1993 to 2013, most of which belong to nanoconjugates, polymeric micelles, and liposomes. Where are these many nanomedicine candidates now? In fact, many of them, if not all, have been fading away from the clinical trials in silence. On account of the proprietary nature of clinical results, the final status or cause of failure cannot be disclosed, only assumed. However, with the high cost and urgency of clinical trials, most are assumed to be terminated by failure, and some of them to be suspended

Table 1
Cancer nanomedicines in silence in clinical trials (1993–2013).

Category	Trade name	Formulations	Company	Start year	Clinical status	Current descriptions/indications (start/completion year)	ClinicalTrials.gov Identifier or [References]
Nano-conjugates	AD-70 (DOX-OXD)	Dextran-doxorubicin	–	1993	Phase I	• Unidentified @ ClinicalTrials.gov • No progress	[118]
	PK1 (FCE28068)	HPMA-doxorubicin	–	1997	Phase II	• Advanced breast cancer (1997/~) • Unknown status	NCT00003165
	PK2 (FCE28069)	HPMA-doxorubicin-galactosamine	–	1999	Phase I/II	• Unidentified @ ClinicalTrials.gov • Hepatocellular carcinoma	[116]
	PNU166945	HPMA-paclitaxel	–	2001	Phase I	• Unidentified @ ClinicalTrials.gov • Refractory solid tumors	[116]
	CT-2103 (Xyotax; Opaxio™)	Polyglutamate-paclitaxel (Paclitaxel polyglumex)	Cell Therapeutics	2002	Phase II	• Recurrent ovarian or primary peritoneal cancer (2002/2006) • Metastatic breast cancer (2004/2007) • Advanced prostate cancer (2005/2008) • Glioblastoma - orphan drug status (FDA 2012) • Other studies: • in combination with premetrexed, gemcitabine, cetuximab, caboplastin, capecitabine, estradiol, temozolomide or radiation	NCT00045682 NCT00148707 NCT00446836 [87]
	CT-2106	Polyglutamate-camptothecin	Cell Therapeutics	2003	Phase I/II	• Advanced malignancy (2003/2005) (phase I) • Advanced/metastatic ovarian cancer (2004/2007) • Metastatic colorectal cancer (2004/2007) (+ folinic acid & 5-FU)	NCT00059917 NCT00291837 NCT00291785
	Prothecan (Pegamotecan)	PEG-camptothecin	Enzon	2003	Phase I/II	• Stomach cancer (2003/~) • Advanced/metastatic sarcoma (2003/~) • Suspended status until 2012 (last posted)	NCT00080002 NCT00079950
	AP5280	HPMA-platinate (carboplatin derivative)	Access Pharmaceuticals	2004	Phase I	• Unidentified @ ClinicalTrials.gov • No progress, unknown status since	[119]
	AP5346 (ProLindac™)	HPMA-DACH (diaminocyclohexane)-platinate (oxaplatin derivative)	Access Pharmaceuticals	2006	Phase II	• Recurrent/unresectable carcinoma (head & neck) (2006/~) • No progress, unknown for recruitment	NCT00415298
	EZN-2208	Multi-arm mPEG-SN38	Enzon	2009	Phase II	• Metastatic breast cancer (2007/~) • Metastatic breast cancer (2009/2013) (estimated) • Other studies: • in combination with cetuximab/-irinotecan, bevacizumab	NCT00520637 NCT01036113
	XMT-1001	Polyacetal-camptothecin	Mersana Therapeutics	2011	Phase I	• Advanced solid tumors (2011/2011) • No progress	NCT00455052 & [117]
	CRLX-101 (IT-101)	Poly-β-cyclodextrin-PEG-camptothecin	Cerulean Pharma	2011	Phase II	• Advanced non-small cell lung cancer (2011/2014) • Other studies: • in combination with olaprib, capecitabine, bevacizumab, topotecan hydrochloride, campthothecin, ciclesonide	NCT01380769
Polymeric micelles	NC-6004 (Nanoplatin™)	mPEG-*b*-Poly(glutamic acid) cisplatin	NanoCarrier	2009	Phase I/II	• Advanced/metastatic pancreatic cancer (2009/2013) (+ gemcitabine) • All other studies: • in combination with gemcitabine or cetuximab/5-FU	NCT00910741
	NK012	mPEG-*b*-Poly(glutamic acid) SN38	Nippon Kayaku	2009	Phase II	• Small cell lung cancer (2009/2012) (primary completion) • Advanced/metastatic breast cancer (2009/2015) • Refractory solid tumors (2009/2011) • Other studies: • in combinations with carboplatin, 5-FU	NCT00951613 NCT00951045 NCT00542958
	SP1049-C	Pluronic-*b*-copolymer doxorubicin (Pgp targeting)	Supratek Pharma	2010	Phase II	• Advanced adenocarcinoma of the esophagus • No progress	[120]
	NK911	mPEG-Poly(aspartic acid) doxorubicin	Nippon Kayaku	2011	Phase I	• Unidentified @ ClinicalTrials.gov • Solid tumors in Japan (2011/~)	[117]
	BIND-014 (Accurins™)	mPEG-PL(G)A docetaxel (targeting prostate-specific membrane antigen (PSMA))	BIND Therapeutics	2011	Phase I/II	• Advanced/metastatic cancer (2011/2016) (phase I) • Metastatic prostate cancer (2013/2016) • Squamous non-small cell lung cancer (2013/2016)	NCT01300533 NCT01812746 NCT01792479
	NK105	mPEG-*b*-Poly(aspartic acid) paclitaxel	Nippon Kayaku	2012	Phase III	• Metastatic/recurrent breast cancer (2012/2017)	NCT01644890

Table 1 (*continued*)

Category	Trade name	Formulations	Company	Start year	Clinical status	Current descriptions/indications (start/completion year)	ClinicalTrials.gov Identifier or [References]
	NC-4016	PEG-*b*-Poly(glutamic acid) oxaliplatin	NanoCarrier	2013	Phase I	• Advanced solid tumors or lymphoma (2013/2017)	NCT03168035
	NC-6300	mPEG-*b*-Poly(aspartate-hydrazone) epirubicin	NanoCarrier	2013	Phase I	• Advanced solid tumors or soft tissue sarcoma (2013/2020) (recruiting/estimate completion)	NCT03168061
Liposomes	L-Annamycin	Liposomal annamycin	Callisto	1998	Phase I/II	• Anthracycline-resistant breast cancer (1998/2001) • Acute lymphocytic leukemia (2005/2008) • Acute lymphocytic leukemia (2007/2009) • Unknown status	NCT00012129 NCT00271063 NCT00430443
	OSI-211	Liposomal lurtotecan	OSI Pharmaceuticals	1999	Phase I/II	• Advanced solid tumors (1999/2009) (phase I) • Advanced solid tumors (2000/2009) (+ cisplatin) (phase I) • Recurrent carcinoma (head & neck) (2001/2002) • Recurrent small cell lung cancer (2002/2003) • Relapsed ovarian cancer (2002/2003) • No progress	NCT00003891 NCT00006036 NCT00022594 NCT00046787 NCT00046800
	IHL-305	Liposomal irinotecan hydrochloride	Yacult Honsha	2006	Phase I	• Advanced solid tumors (2006/~) • Unknown status	NCT00364143
	CPX-1	Liposomal irinotecan hydrochloride plus floxuridine	Celator Pharmaceutical	2006	Phase II	• Advanced colorectal carcinoma (2006/2008)	NCT00361842
	ThermoDox™	Thermosensitive liposomal doxorubicin	Celsion	2006	Phase I/II	• Recurrent breast cancer (2006/2011) (phase I) (+ hyperthermia) • Primary metastatic liver cancer (2007/2009) (+ radiofrequency ablation, phase I) • Painful bone metastases (2012/2013) (+ MRI-guided high intensity focused ultrasound) • Recurrent breast cancer (2013/2016) (+ microwave hyperthermia)	NCT00346229 NCT00441376 NCT01640847 NCT00826085
	MBP-426	Transferrin-conjugated liposomal oxaplatin	Mebiopharm	2009	Phase I/II	• Advanced/metastatic solid tumors (2006/2009) (phase I) • Gastric/esophageal adenocarcinoma (2009/2015) (+ Leucovorin/5-FU) • Unknown status, no progress	NCT00355888 NCT00964080
Other platforms	AI-850	Sponge-like sugar particles (< 2 μm)	Acusphere	2007	Phase I	• Unidentified @ ClinicalTrials.gov • No progress	[121]
	CALAA-01	Transferrin-β-cyclodextrin-based nanoparticles containing siRNA targeting RRM2	Calando Pharmaceuticals	2008	Phase I	• Solid tumor cancers (2008/2012) • No progress	NCT00689065

by "potential" failures. In fact, many nanomedicines have not made it beyond clinical trials mainly due to lack of significant improved clinical efficacy [86,87]. Comprehensively, the development status of nanomedicine has been poor.

The first category of disappearing nanomedicine is nanoconjugates. Herein, PEGylated protein drugs are excluded because they are much closer to modified protein drugs rather than nanomedicines. Since the introduction of SMANCS in 1994 (Zinostatin stimalamer in Japan) [9,88], nanoconjugates had become a popular template for nanomedicine and their development was rapid up until ~2010. Up to 2011, twelve nanoconjugates were evaluated in clinical phase I/II trials, mostly based on existing chemotherapeutics, such as, doxorubicin, paclitaxel, camptothecin and platinates. All of the nanomedicines listed in many review papers and on ClinicalTrials.gov are out of date or expired, regardless of the polymer types used, such as dextran or HPMA (N-(2-hydroxypropyl)methyacrylamide) polymers, polyglutamate or polycyclodextrin, and polyethylene glycol (Table 1). In most cases, the average required time for the go/no-go decision has elapsed since initiation, and most clinical studies seem to have made no progress and have been officially (or presumably) terminated, indicating inactive state or overall failure. In addition, some nanoconjugates (Xyotax® and XMT-1001) were evaluated in combination with one or more chemotherapeutics. Many of them seem like a dilemmatic choice due to unsatisfied clinical results from the monotherapy of original nanomedicines.

The second category is polymeric micelles, which entered clinical trials in the early or middle 2000s and some of them are still on going. Some candidates that entered clinical trials around 2010 may show significant efficacy. Nevertheless, it is probable that a similar conclusion will be reached for polymeric micelles because the performance of other nanomedicines based on EPR have shown repetitive patterns without true success, i.e., little improvement in efficacy and continuing toxicity concerns, which cannot be overcome at once without full understanding of reasons for poor outcomes. That is why there are still many questions concerning the clinical effectiveness as therapeutic cancer nanomedicine.

The third category is liposomes that produced approved drug products. Due to the advantage of established formulations, many liposomal candidates have been intensively applied in clinical trials since the pioneering era of middle 1990s. Despite no critical technological advance, we have seen several more liposomal formulations, which were approved by the European Medicines Agency (EMA) (Caelyx® and Myocet®) or belonged to other therapeutic indications (AmBisome®, Visudyne®, Abelcet®) [9]. Nonetheless, similar with other categories,

multiple clinical studies listed in Table 1 seem inactive and suspended. For example, the Lyso-thermosensitive liposomal doxorubicin (ThermoDox®), in conjunction with microwave-assisted hyperthermia, MRI-guided ultrasound or radiofrequency ablation, has attracted great interest by a series of clinical trials since 2006 but has not heralded the clinical success to date [89]. Also, the transferrin-conjugated liposomal oxaplatin (MBP-426) formulated by the active targeting approach has been under clinical trials since 2009 but not approved until now [90].

The cause of relatively poor clinical success rate of oncology drug candidates cannot be simply explained because there are many factors to be considered, i.e., therapeutic efficacy, toxicity, cost-effectiveness, patent issues, patient compliance/acceptance, emergence of competitive products, and so on. In spite of enormous advantages coming from nanomedicine that have been claimed by academia, such as EPR and active targeting, it remains to prove that the relevant outcomes and achievements are superior to oncology drug discovery rates. Why do such unsuccessful results seem to be repeated in clinical trials? This may be because we continue to use similar approaches with different designs without knowing what went wrong in prior examples. It is time for us to reflect on these failures in retrospective views and understand why such candidate nanomedicines were not successfully translated. The discrepancy between preclinical and clinical outcomes needs to inform better design, but no theories are available to explain such deviations. Recent news increased our disappointment, especially those for BIND-014, ThermoDOX®, NK105, CRLX101, CALLA-01, and others from research labs and industry. BIND Therapeutics filed Chapter 11 after BIND-014 clinical trials [91]. Some might survive or be resurrected and reach the market, but real advances in clinical efficacy seem beyond our reach. The reality is that we have not seen any of approved nanomedicine, which satisfies technological or clinical requirements, based on the theory of modern cancer nanomedicine. The theory of EPR, which has been the basis for developing nanomedicine for a long time, becomes merely one of many possible pathological observations, if it exists at all, in human clinical tumors, but may not be so solid. Nanomedicine should be regarded as an option, a solubilizing hydrophobic drug, and an alternative to existing formulation strategies, and should be discussed as a controversial issue, rather than a belief.

5. Preclinical cancer models

The decision on clinical trials of cancer nanomedicine heavily relies on preclinical outcomes from rodent models. Almost all scientific papers dealing with preclinical results of cancer nanomedicine have routinely shown excellent tumor suppression data: tumors completely disappeared from ectopic or orthotopic implant sites in rodents in some cases. Most studies have been carried out using similar tumor xenograft protocols as well as dosing schedules. In many cases, tumors grow abnormally fast because of a great number of cancer cells xenografted [92] and their weight reached almost 10% of the total body weight of the rodents [13,93], whereas real human tumors range from a few millimeters to a few centimeters [19,94]. The dose of nanoparticles was aggressively determined (almost highest dose desired for maximum efficacy) without careful consideration of the health of the rodents. All of such preclinical settings are like a stereotyped soap drama acted by preordained ending scenario. Thus, nanomedicines are ready to exert their full antitumor efficacy on highly EPR-responsive murine tumors prepared to provide the best conditions for treatment.

In active targeting, nanomedicines with highly surface-modified specific ligands, without physiological or toxicological consideration, tends to suppress tumors comprised of relatively homogenous cancer cells with well-expressed receptors. A series of similar animal study protocols, which do not reflect human cancer biology, must be one of reasons for the repetitive failure of nanomedicine in clinical trials. We just have to use such animal models to compare the basic efficacy versus a naïve formulation or free drugs in early development stage, and we should not exaggerate these results.

Typical cancer nanomedicines to treat solid tumors seek to alter biodistribution of anticancer drugs and rely on delivery to individual target cells. A range of factors in physiology, pathology, histology, and nanomedicine are associated to biodistribution. Designed nanoparticles circulate for long periods in the blood stream but are not beneficial to diffusional permeation into dense tumor tissues in humans when compared to the diffusivity of small molecules. Furthermore, the relevance of physiology, tumor pathology, cancer biology, and immunology of the given models to human patients will vary depending on target disease, drug candidate, and nanomedicine because a single model does not represent all aspects of the target diseases in human. Therefore, it may come to a fallacious conclusion to hastily give a meaning to preclinical results obtained from a single cancer model at a single therapy using a single drug formulation. The biological diversity and heterogeneity in human cancer cells may depreciate the value of a single cancer model.

It is time for scientists in related fields to make sincere efforts in developing an enhanced platform of preclinical models that closely reflect human cancer biology. Nanomedicine scientists have acquired all design parameters from rodent models instead of from human patients and thus are only able to treat artificial rodent disease models. Disease models are employed for prediction of the clinical translation of rationally designed nanomedicines and we are still waiting for a better success rate to appear than that of randomly screened anticancer drugs. Rodent models do not represent a miniature human cancer model and lack predictive power for nanomedicine. Alternative in vitro and in vivo models that can yield improved predictive power for efficacy as well as adverse effects are needed.

6. Prospective view

A superficial reason for the discrepancy between preclinical and clinical outcomes is that nanomedicines are unable to behave in the human body as they do in rodents. Continuous hope and promise for better cancer nanomedicine persist by (i) fine-tuning nanoparticles responsive to tumor environment or (ii) altering tumor biology favorable to nanoparticle delivery. Some factors of fine-tuning, for examples, physical aspects (size and shape) and targeting functionality in response to internal (tumor pH or enzymes etc.) [24,95–98] or external (light, temperature, magnetic field or ultrasound etc.) [99–101] stimuli, can contribute to the delivery of cancer nanomedicines. These strategies would hardly lead to quantum improvements unless they overcome the practical problems related to physiology and pathology in human. Normalization of elevated tumor IFP could be an effective strategy allowing for interstitial permeation of nanomedicine. This can be achieved by (i) tumor vascular normalization using anti-angiogenesis agents [102–104] and (ii) remodeling by artificial degradation of ECM using collagenase or hyaluronidase. These approaches have also showed improved performance of nanoparticles in preclinical studies, but their clinical usefulness has not been validated yet. In particular, early studies have shown the possibility of intratumoral or intravenous delivery of collagenase for this purpose [49,105], but systemically administered enzymes that destroy ECM may present general toxicity concern in humans [53,106,107]. In reality, a series of design changes and approaches seem insufficient to overcome all barriers of unfavorable tumor environment in human. We may not be able to take even one small step forward to the actual solution unless the issues associated with EPR and targeting are fundamentally understood. Nevertheless, it is time to think of a post-EPR world [13]. We need to seek out the practical values of nanomedicine from both technological and clinical viewpoints, although some may not appear to be major breakthroughs.

First, clinical translation of cancer nanomedicine is often concluded by toxicity profiles that are a major concern of FDA, despite enormous efforts seeking better efficacy by formulation designs and manufacturing. For many decades, we have paid much attention to improving efficacy rather

than alleviating toxicity in the development of cancer nanomedicine. When the toxicity of anticancer treatments is manipulated through the use of nanomedicines (i.e., a focus on decreasing toxicity without compromising efficacy), this may guide a new direction for cancer nanomedicine, providing better opportunities for translation. Surely, this kind of paradigm shift is not a fundamental solution but a compromise, and is disappointing from the angle of efficacy improvement. Nevertheless, the approval of liposomal doxorubicin formulations (Doxil® and others) and Abraxane®, which bring in almost one billion US dollars in sales per year (estimated in 2022 and 2019, respectively) [86,108,109], was based on altered toxicity profiles and tolerated dose increase [19]. However, predicting toxicity in human patients from what we have learned from animal models is significantly more challenging. For instance, the observed toxicity in human while translating nanomedicine is hardly observed or monitored in animal cancer models.

Second, broadening the potential applications of nanomedicine beyond cancer to other diseases, which have less resistance to transport of nano-sized materials, would expand the uses of the technologies developed. This issue does not fit well with the current topic of cancer nanomedicine, but changing indications can be an alternative means of expanding the clinical usage of nanomedicines. In this respect, there are already many therapeutic nanomedicines for diseases other than cancers, for example, Ambisome® (liposomal amphotericin B) for fungal treatment, Visudyne® (liposomal verteporfin) for macular degeneration treatment, Diprivan® (liposomal propofol) for sedation or anesthesia induction, and a series of iron-replacement nanoparticles (CosmoFer®, DexFerrum®, Venofer®, Feraheme®, Injectafter®, Monofer®, and Diafer®) for iron-deficient anemia treatment as a naïve colloidal form [6]. In these fields, nanoparticles seem to play a rather primitive role in storing and releasing drugs.

Third, therapeutic nanomedicine is mostly assumed to be injectable, despite that several other routes, such as, nasal [110,111], pulmonary [112,113], and oral/peroral [114,115] administration, are used for specific applications. Recent studies on the delivery of therapeutic nanomedicine via such routes have displayed meaningful (antitumor) efficacy, despite shortcomings at the preclinical levels. It seems highly probable that nanomedicine administered locally via such routes could cure tumors present at local sites. In terms of systemic efficacy, these routes might have potential in the treatment of other indications, but not cancers, because these nanomedicines will face the same in vivo hardships in tumor regions and systemic circulation as those injected intravenously. Also, the topical application of liposomes is already popular in both academia and industry; for example, the topical products of a liposomal amphotericin B (Ambisome®) and a polymeric micellar estradiol (Estrasorb®) are already on commercial market [116], and the inhaled liposomal amikacin (Arikace™) has been applied in clinical phase III for cystic fibrosis treatment [117]. All things, such as, pros and cons, and disappointment and compromise, can be raised, but practical and non-invasive administration of nanoparticles would contribute to the future of therapeutic nanomedicine.

Developments in nanomedicine require a team effort with a range of expertise in various areas relevant to target diseases, including physiology, pathology, oncology, material science, and mass transport, among others. Unfortunately, cancer nanomedicine has attracted material scientists, chemists, and engineers who are not truly specialized in such areas. When all goes fine with preclinical and clinical outputs, the story becomes simple. However, anything goes wrong it is extremely challenging to identifying any clues for reasoning. A better understanding of target diseases, previous examples that were translated, the strengths and limitations of the disease models we employ, and differences in the biology and physiology between species are all important components of future technology development. We need to accept the heterogeneity of human diseases and that no theory is a universal concept that will apply to every cancer, and to pay closer attention to preclinical outcomes and their relevance to future clinical studies. The current development of cancer nanomedicines seems to be a repetitive beginning of a new game that cannot be concluded, seeking for a game changer.

Acknowledgements

This research was supported by the Basic Science Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Science and ICT (2017R1A2B2004955).

References

[1] A. Bangham, M.M. Standish, J.C. Watkins, Diffusion of univalent ions across the lamellae of swollen phospholipids, J. Mol. Biol. 13 (1965) 238–252.
[2] T. Allen, A. Chonn, Large unilamellar liposomes with low uptake into the reticuloendothelial system, FEBS Lett. 223 (1987) 42–46.
[3] V.P. Torchilin, Recent advances with liposomes as pharmaceutical carriers, Nat. Rev. Drug Discov. 4 (2005) 145–160.
[4] K. Khosravi-Darani, M. Mozafari, Nanoliposome potentials in nanotherapy: a concise overview, Int. J. Nanosci. and Nanotechnol. 6 (2010) 3–13.
[5] O. Felfoul, M. Mohammadi, S. Taherkhani, D. De Lanauze, Y.Z. Xu, D. Loghin, S. Essa, S. Jancik, D. Houle, M. Lafleur, Magneto-aerotactic bacteria deliver drug-containing nanoliposomes to tumour hypoxic regions, Nat. Nanotechnol. 11 (2016) 941–947.
[6] A.C. Anselmo, S. Mitragotri, Nanoparticles in the clinic, Bioeng. Transl. Med. 1 (2016) 10–29.
[7] U. Bulbake, S. Doppalapudi, N. Kommineni, W. Khan, Liposomal formulations in clinical use: an updated review, Pharmaceutics 9 (2017) 12.
[8] D. Bobo, K.J. Robinson, J. Islam, K.J. Thurecht, S.R. Corrie, Nanoparticle-based medicines: a review of FDA-approved materials and clinical trials to date, Pharm. Res. 33 (2016) 2373–2387.
[9] S. Tran, P.-J. DeGiovanni, B. Piel, P. Rai, Cancer nanomedicine: a review of recent success in drug delivery, Clinical and translational medicine 6 (2017) 44.
[10] V. Narayanan, C. Weekes, Nanoparticle albumin-bound (nab)-paclitaxel for the treatment of pancreas ductal adenocarcinoma, Gastrointest. Cancer. 5 (2015) 11–19.
[11] M. Zhao, C. Lei, Y. Yang, X. Bu, H. Ma, H. Gong, J. Liu, X. Fang, Z. Hu, Q. Fang, Abraxane, the nanoparticle formulation of paclitaxel can induce drug resistance by up-regulation of P-gp, PLoS One 10 (2015), e0131429. .
[12] N. Chen, C. Brachmann, X. Liu, D.W. Pierce, J. Dey, W.S. Kerwin, Y. Li, S. Zhou, S. Hou, M. Carleton, Albumin-bound nanoparticle (nab) paclitaxel exhibits enhanced paclitaxel tissue distribution and tumor penetration, Cancer Chemother. Pharmacol. 76 (2015) 699–712.
[13] J.W. Nichols, Y.H. Bae, EPR: evidence and fallacy, J. Control. Release 190 (2014) 451–464.
[14] R. Savić, T. Azzam, A. Eisenberg, D. Maysinger, Assessment of the integrity of poly (caprolactone)-b-poly (ethylene oxide) micelles under biological conditions: a fluorogenic-based approach, Langmuir 22 (2006) 3570–3578.
[15] H.M. Burt, X. Zhang, P. Toleikis, L. Embree, W.L. Hunter, Development of copolymers of poly (D, L-lactide) and methoxypolyethylene glycol as micellar carriers of paclitaxel, Colloids Surf. B: Biointerfaces 16 (1999) 161–171.
[16] E.S. Lee, Y.S. Youn, Albumin-based potential drugs: focus on half-life extension and nanoparticle preparation, J. Pharm. Investig. 46 (2016) 305–315.
[17] T.-Y. Kim, D.-W. Kim, J.-Y. Chung, S.G. Shin, S.-C. Kim, D.S. Heo, N.K. Kim, Y.-J. Bang, Phase I and pharmacokinetic study of Genexol-PM, a cremophor-free, polymeric micelle-formulated paclitaxel, in patients with advanced malignancies, Clin. Cancer Res. 10 (2004) 3708–3716.
[18] M. Green, G. Manikhas, S. Orlov, B. Afanasyev, A. Makhson, P. Bhar, M. Hawkins, Abraxane®, a novel Cremophor®-free, albumin-bound particle form of paclitaxel for the treatment of advanced non-small-cell lung cancer, Ann. Oncol. 17 (2006) 1263–1268.
[19] J.W. Nichols, Y.H. Bae, Odyssey of a cancer nanoparticle: from injection site to site of action, Nano Today 7 (2012) 606–618.
[20] Y.H. Bae, K. Park, Targeted drug delivery to tumors: myths, reality and possibility, J. Control. Release 153 (2011) 198.
[21] P. Ehrlich, Aus theorie und praxis der chemotherapie, W. Klinkhardt, 1911.
[22] S. Mukherjee, R.N. Ghosh, F.R. Maxfield, Endocytosis, Physiol. Rev. 77 (1997) 759–803.
[23] J. Panyam, W.-Z. Zhou, S. Prabha, S.K. Sahoo, V. Labhasetwar, Rapid endo-lysosomal escape of poly (DL-lactide-co-glycolide) nanoparticles: implications for drug and gene delivery, FASEB J. 16 (2002) 1217–1226.
[24] R.A. Petros, J.M. DeSimone, Strategies in the design of nanoparticles for therapeutic applications, Nat. Rev. Drug Discov. 9 (2010) 615–627.
[25] E.-L. Eskelinen, Y. Tanaka, P. Saftig, At the acidic edge: emerging functions for lysosomal membrane proteins, Trends Cell Biol. 13 (2003) 137–145.
[26] T.-G. Iversen, T. Skotland, K. Sandvig, Endocytosis and intracellular transport of nanoparticles: present knowledge and need for future studies, Nano Today 6 (2011) 176–185.
[27] W. Fan, D. Xia, Q. Zhu, L. Hu, Y. Gan, Intracellular transport of nanocarriers across the intestinal epithelium, Drug Discov. Today 21 (2016) 856–863.
[28] H. Maeda, M. Ueda, T. Morinaga, T. Matsumoto, Conjugation of poly (styrene-co-maleic acid) derivatives to the antitumor protein neocarzinostatin: pronounced improvements in pharmacological properties, J. Med. Chem. 28 (1985) 455–461.

[29] T. Konno, H. Maeda, K. Iwai, S. Maki, S. Tashiro, M. Uchida, Y. Miyauchi, Selective targeting of anti-cancer drug and simultaneous image enhancement in solid tumors by arterially administered lipid contrast medium, Cancer 54 (1984) 2367–2374.
[30] Y. Matsumura, H. Maeda, A new concept for macromolecular therapeutics in cancer chemotherapy: mechanism of tumoritropic accumulation of proteins and the antitumor agent smancs, Cancer Res. 46 (1986) 6387–6392.
[31] Y. Matsumura, T. Oda, H. Maeda, General mechanism of intratumor accumulation of macromolecules: advantage of macromolecular therapeutics, Gan to kagaku ryoho. Cancer & Chemother. 14 (1987) 821–829.
[32] Y.H. Bae, Drug targeting and tumor heterogeneity, J. Control. Release 133 (2009) 2–3.
[33] H. Maeda, J. Wu, T. Sawa, Y. Matsumura, K. Hori, Tumor vascular permeability and the EPR effect in macromolecular therapeutics: a review, J. Control. Release 65 (2000) 271–284.
[34] A. Gabizon, D. Papahadjopoulos, Liposome formulations with prolonged circulation time in blood and enhanced uptake by tumors, Proc. Natl. Acad. Sci. U. S. A. 85 (1988) 6949–6953.
[35] J. Fang, H. Nakamura, H. Maeda, The EPR effect: unique features of tumor blood vessels for drug delivery, factors involved, and limitations and augmentation of the effect, Adv. Drug Deliv. Rev. 63 (2011) 136–151.
[36] E.A. Azzopardi, E.L. Ferguson, D.W. Thomas, The enhanced permeability retention effect: a new paradigm for drug targeting in infection, J. Antimicrob. Chemother. 68 (2012) 257–274.
[37] J.D. Byrne, T. Betancourt, L. Brannon-Peppas, Active targeting schemes for nanoparticle systems in cancer therapeutics, Adv. Drug Deliv. Rev. 60 (2008) 1615–1626.
[38] R.K. Jain, Vascular and interstitial barriers to delivery of therapeutic agents in tumors, Cancer Metastasis Rev. 9 (1990) 253–266.
[39] R. Weiner, The mechanism of 67Ga localization in malignant disease, Nucl. Med. Biol. 23 (1996) 745–751.
[40] K.J. Harrington, S. Mohammadtaghi, P.S. Uster, D. Glass, A.M. Peters, R.G. Vile, J.S.W. Stewart, Effective targeting of solid tumors in patients with locally advanced cancers by radiolabeled pegylated liposomes, Clin. Cancer Res. 7 (2001) 243–254.
[41] F. Danhier, To exploit the tumor microenvironment: since the EPR effect fails in the clinic, what is the future of nanomedicine? J. Control. Release 244 (2016) 108–121.
[42] D.W. Northfelt, B.J. Dezube, J.A. Thommes, B.J. Miller, M.A. Fischl, A. Friedman-Kien, L.D. Kaplan, C. Du Mond, R.D. Mamelok, D.H. Henry, Pegylated-liposomal doxorubicin versus doxorubicin, bleomycin, and vincristine in the treatment of AIDS-related Kaposi's sarcoma: results of a randomized phase III clinical trial, J. Clin. Oncol. 16 (1998) 2445–2451.
[43] S. Stewart, H. Jablonowski, F.D. Goebel, K. Arasteh, M. Spittle, A. Rios, D. Aboulafia, J. Galleshaw, B.J. Dezube, Randomized comparative trial of pegylated liposomal doxorubicin versus bleomycin and vincristine in the treatment of AIDS-related Kaposi's sarcoma. International Pegylated Liposomal Doxorubicin Study Group, J. Clin. Oncol. 16 (1998) 683–691.
[44] F. Kiessling, M.E. Mertens, J. Grimm, T. Lammers, Nanoparticles for imaging: top or flop? Radiology 273 (2014) 10–28.
[45] M. O'brien, N. Wigler, M. Inbar, R. Rosso, E. Grischke, A. Santoro, R. Catane, D. Kieback, P. Tomczak, S. Ackland, Reduced cardiotoxicity and comparable efficacy in a phase III trial of pegylated liposomal doxorubicin HCl (CAELYX™/Doxil®) versus conventional doxorubicin for first-line treatment of metastatic breast cancer, Ann. Oncol. 15 (2004) 440–449.
[46] A. Gabizon, H. Shmeeda, Y. Barenholz, Pharmacokinetics of pegylated liposomal doxorubicin, Clin. Pharmacokinet. 42 (2003) 419–436.
[47] C.-H. Heldin, K. Rubin, K. Pietras, A. Östman, High interstitial fluid pressure—an obstacle in cancer therapy, Nat. Rev. Cancer 4 (2004) 806–813.
[48] P.A. Netti, D.A. Berk, M.A. Swartz, A.J. Grodzinsky, R.K. Jain, Role of extracellular matrix assembly in interstitial transport in solid tumors, Cancer Res. 60 (2000) 2497–2503.
[49] M. Kato, Y. Hattori, M. Kubo, Y. Maitani, Collagenase-1 injection improved tumor distribution and gene expression of cationic lipoplex, Int. J. Pharm. 423 (2012) 428–434.
[50] R.K. Jain, R.T. Tong, L.L. Munn, Effect of vascular normalization by antiangiogenic therapy on interstitial hypertension, peritumor edema, and lymphatic metastasis: insights from a mathematical model, Cancer Res. 67 (2007) 2729–2735.
[51] X. Hu, R. Adamson, B. Liu, F. Curry, S. Weinbaum, Starling forces that oppose filtration after tissue oncotic pressure is increased, Am. J. Physiol. Heart Circ. Physiol. 279 (2000) H1724–H1736.
[52] Y. Boucher, R.K. Jain, Microvascular pressure is the principal driving force for interstitial hypertension in solid tumors: implications for vascular collapse, Cancer Res. 52 (1992) 5110–5114.
[53] P.A. Netti, L.T. Baxter, Y. Boucher, R. Skalak, R.K. Jain, Time-dependent behavior of interstitial fluid pressure in solid tumors: implications for drug delivery, Cancer Res. 55 (1995) 5451–5458.
[54] Y. Boucher, H. Salehi, B. Witwer, G. Harsh IV, R. Jain, Interstitial fluid pressure in intracranial tumours in patients and in rodents, Br. J. Cancer 75 (1997) 829.
[55] V.P. Chauhan, T. Stylianopoulos, J.D. Martin, Z. Popović, O. Chen, W.S. Kamoun, M.G. Bawendi, D. Fukumura, R.K. Jain, Normalization of tumour blood vessels improves the delivery of nanomedicines in a size-dependent manner, Nat. Nanotechnol. 7 (2012) 383–388.
[56] R. Gutmann, M. Leunig, J. Feyh, A.E. Goetz, K. Messmer, E. Kastenbauer, R.K. Jain, Interstitial hypertension in head and neck tumors in patients: correlation with tumor size, Cancer Res. 52 (1992) 1993–1995.
[57] Y. Boucher, L.T. Baxter, R.K. Jain, Interstitial pressure gradients in tissue-isolated and subcutaneous tumors: implications for therapy, Cancer Res. 50 (1990) 4478–4484.
[58] Y. Boucher, M. Leunig, R.K. Jain, Tumor angiogenesis and interstitial hypertension, Cancer Res. 56 (1996) 4264–4266.
[59] Y. Matsumoto, J.W. Nichols, K. Toh, T. Nomoto, H. Cabral, Y. Miura, R.J. Christie, N. Yamada, T. Ogura, M.R. Kano, Vascular bursts enhance permeability of tumour blood vessels and improve nanoparticle delivery, Nat. Nanotechnol. 11 (2016) 533–538.
[60] K. Strebhardt, A. Ullrich, Paul Ehrlich's magic bullet concept: 100 years of progress, Nat. Rev. Cancer 8 (2008) 473–480.
[61] K.F. Pirollo, E.H. Chang, Does a targeting ligand influence nanoparticle tumor localization or uptake? Trends Biotechnol. 26 (2008) 552–558.
[62] D.B. Kirpotin, D.C. Drummond, Y. Shao, M.R. Shalaby, K. Hong, U.B. Nielsen, J.D. Marks, C.C. Benz, J.W. Park, Antibody targeting of long-circulating lipidic nanoparticles does not increase tumor localization but does increase internalization in animal models, Cancer Res. 66 (2006) 6732–6740.
[63] J.Y. Wong, T.L. Kuhl, J.N. Israelachvili, N. Mullah, S. Zalipsky, Direct measurement of a tethered ligand-receptor interaction potential, Science 275 (1997) 820–822.
[64] D.B. Young, Chapt. 3. Cardiac Function, in: Control of Cardiac Output, Morgan & Claypool Life Sciences, 2010.
[65] C. Starr, R. Taggart, C. Evers, L. Starr, Biology: The Unity and Diversity of Life, Nelson Education, 2015.
[66] L. Shagel, A.B.C. Yu, Chapter 11. Physiologic drug distribution and protein binding, Applied Biopharmaceutics & Pharmacokinetics, McGraw-Hill Global Education Holdings, LLC, 2012.
[67] S. Bae, K. Ma, T.H. Kim, E.S. Lee, K.T. Oh, E.-S. Park, K.C. Lee, Y.S. Youn, Doxorubicin-loaded human serum albumin nanoparticles surface-modified with TNF-related apoptosis-inducing ligand and transferrin for targeting multiple tumor types, Biomaterials 33 (2012) 1536–1546.
[68] R.T. Jones, Blood flow, AnRFM 1 (1969) 223–244.
[69] J.M. Munson, A.C. Shieh, Interstitial fluid flow in cancer: implications for disease progression and treatment, Cancer Manag. Res. 6 (2014) 317–328.
[70] I. Guryanov, S. Fiorucci, T. Tennikova, Receptor-ligand interactions: advanced biomedical applications, Mater. Sci. Eng. C 68 (2016) 890–903.
[71] J.R. Less, T.C. Skalak, E.M. Sevick, R.K. Jain, Microvascular architecture in a mammary carcinoma: branching patterns and vessel dimensions, Cancer Res. 51 (1991) 265–273.
[72] F. Yuan, M. Leunig, S.K. Huang, D.A. Berk, D. Papahadjopoulos, R.K. Jain, Mirovascular permeability and interstitial penetration of sterically stabilized (stealth) liposomes in a human tumor xenograft, Cancer Res. 54 (1994) 3352–3356.
[73] S. Ramanujan, A. Pluen, T.D. McKee, E.B. Brown, Y. Boucher, R.K. Jain, Diffusion and convection in collagen gels: implications for transport in the tumor interstitium, Biophys. J. 83 (2002) 1650–1660.
[74] L.D. Greller, F.L. Tobin, G. Poste, Tumor heterogeneity and progression: conceptual foundations for modeling, Invasion Metastasis 16 (1996) 177–208.
[75] P. Birner, G. Oberhuber, J. Stani, C. Reithofer, H. Samonigg, H. Hausmaninger, E. Kubista, W. Kwasny, D. Kandioler-Eckersberger, M. Gnant, Evaluation of the United States Food and Drug Administration-approved scoring and test system of HER-2 protein expression in breast cancer, Clin. Cancer Res. 7 (2001) 1669–1675.
[76] L. Recht, C.O. Torres, T.W. Smith, V. Raso, T.W. Griffin, Transferrin receptor in normal and neoplastic brain tissue: implications for brain-tumor immunotherapy, J. Neurosurg. 72 (1990) 941–945.
[77] Y. Kohgo, T. Nishisato, H. Kondo, N. Tsushima, Y. Niitsu, I. Urushizaki, Circulating transferrin receptor in human serum, Br. J. Haematol. 64 (1986) 277–281.
[78] Z.M. Qian, H. Li, H. Sun, K. Ho, Targeted drug delivery via the transferrin receptor-mediated endocytosis pathway, Pharmacol. Rev. 54 (2002) 561–587.
[79] J.F. Ross, P.K. Chaudhuri, M. Ratnam, Differential regulation of folate receptor isoforms in normal and malignant tissues in vivo and in established cell lines. Physiologic and clinical implications, Cancer 73 (1994) 2432–2443.
[80] J. Sudimack, R.J. Lee, Targeted drug delivery via the folate receptor, Adv. Drug Deliv. Rev. 41 (2000) 147–162.
[81] D.L. Vesely, S.F. Kemp, M.J. Elders, Isolation of a biotin receptor from hepatic plasma membranes, Biochem. Biophys. Res. Commun. 143 (1987) 913–916.
[82] W. YoungáKim, J. SeungáKim, Biotin-guided anticancer drug delivery with acidity-triggered drug release, Chem. Commun. 51 (2015) 9343–9345.
[83] E. Ruoslahti, Integrins, J. Clin. Invest. 87 (1991) 1–5.
[84] J. Ledermann, S. Canevari, T. Thigpen, Targeting the folate receptor: diagnostic and therapeutic approaches to personalize cancer treatments, Ann. Oncol. 26 (2015) 2034–2043.
[85] A. Mullard, Parsing clinical success rates, Nature 15 (2016) 447.
[86] C.L. Ventola, Progress in nanomedicine: approved and investigational nanodrugs, P T 42 (2017) 742–755.
[87] J.M. Caster, A.N. Patel, T. Zhang, A. Wang, Investigational nanomedicines in 2016: a review of nanotherapeutics currently undergoing clinical trials, Wiley Interdisciplinary Reviews: Nanomed. Nanobiotechnol. 9 (2017) 1–8.
[88] E. Masuda, H. Maeda, Antitumor resistance induced by zinostatin stimalamer (ZSS), a polymer-conjugated neocarzinostatin (NCS) derivative, Cancer Immunol. Immunother. 40 (1995) 329–338.
[89] B. Wood, R. Poon, Z. Neeman, M. Eugeni, J. Locklin, S. Dromi, S. Kachala, R. Probhakar, W. Hahne, S. Libutti, Phase I Study of Thermally Sensitive Liposomes Containing Doxorubicin (ThermoDox) Given during Radiofrequency Ablation (RFA) in Patients with Unresectable Hepatic Malignancies, Am. Soc. Clin. Oncol. Gastrointest, Cancers Symposium, 2007.
[90] K. Sankhala, A. Mita, R. Adinin, L. Wood, M. Beeram, S. Bullock, N. Yamagata, K. Matsuno, T. Fujisawa, A. Phan, A phase I pharmacokinetic (PK) study of MBP-426, a novel liposome encapsulated oxaliplatin, J. Clin. Oncol. 27 (2009) 2535.
[91] H. Ledford, Bankruptcy of nanomedicine firm worries drug developers, Nature 533 (2016) 304–305.

[92] J. Jonkers, A. Berns, Conditional mouse models of sporadic cancer, Nat. Rev. Cancer 2 (2002) 251–265.
[93] J. Ozao-Choy, G. Ma, J. Kao, G.X. Wang, M. Meseck, M. Sung, M. Schwartz, C.M. Divino, P.-Y. Pan, S.-H. Chen, The novel role of tyrosine kinase inhibitor in the reversal of immune suppression and modulation of tumor microenvironment for immune-based cancer therapies, Cancer Res. 69 (2009) 2514–2522.
[94] C.L. Carter, C. Allen, D.E. Henson, Relation of tumor size, lymph node status, and survival in 24,740 breast cancer cases, Cancer 63 (1989) 181–187.
[95] E.S. Lee, K. Na, Y.H. Bae, Super pH-sensitive multifunctional polymeric micelle, Nano Lett. 5 (2005) 325–329.
[96] E.S. Lee, K.T. Oh, D. Kim, Y.S. Youn, Y.H. Bae, Tumor pH-responsive flower-like micelles of poly (L-lactic acid)-b-poly (ethylene glycol)-b-poly (L-histidine), J. Control. Release 123 (2007) 19–26.
[97] R. De La Rica, D. Aili, M.M. Stevens, Enzyme-responsive nanoparticles for drug release and diagnostics, Adv. Drug Deliv. Rev. 64 (2012) 967–978.
[98] C. Zhang, D. Pan, K. Luo, N. Li, C. Guo, X. Zheng, Z. Gu, Dendrimer–doxorubicin conjugate as enzyme-sensitive and polymeric nanoscale drug delivery vehicle for ovarian cancer therapy, Polym. Chem. 5 (2014) 5227–5235.
[99] C. Lee, H.S. Hwang, S. Lee, B. Kim, J.O. Kim, K.T. Oh, E.S. Lee, H.G. Choi, Y.S. Youn, Rabies virus-inspired silica-coated gold Nanorods as a Photothermal therapeutic platform for treating brain tumors, Adv. Mater. 29 (2017) 1605563.
[100] N. Lee, D. Yoo, D. Ling, M.H. Cho, T. Hyeon, J. Cheon, Iron oxide based nanoparticles for multimodal imaging and magnetoresponsive therapy, Chem. Rev. 115 (2015) 10637–10689.
[101] L. Cheng, C. Wang, L. Feng, K. Yang, Z. Liu, Functional nanomaterials for phototherapies of cancer, Chem. Rev. 114 (2014) 10869–10939.
[102] C.J. Cheng, W.M. Saltzman, Nanomedicine: downsizing tumour therapeutics, Nat. Nanotechnol. 7 (2012) 346–347.
[103] R.K. Jain, Normalizing tumor vasculature with anti-angiogenic therapy: a new paradigm for combination therapy, Nat. Med. 7 (2001) 987–989.
[104] R.K. Jain, Normalization of tumor vasculature: an emerging concept in antiangiogenic therapy, Science 307 (2005) 58–62.
[105] X. Zheng, B.A. Goins, I.L. Cameron, C. Santoyo, A. Bao, V.C. Frohlich, G.D. Fullerton, Ultrasound-guided intratumoral administration of collagenase-2 improved liposome drug accumulation in solid tumor xenografts, Cancer Chemother. Pharmacol. 67 (2011) 173–182.
[106] L. Eikenes, Ø.S. Bruland, C. Brekken, C. de Lange Davies, Collagenase increases the transcapillary pressure gradient and improves the uptake and distribution of monoclonal antibodies in human osteosarcoma xenografts, Cancer Res. 64 (2004) 4768–4773.
[107] T.T. Goodman, P.L. Olive, S.H. Pun, Increased nanoparticle penetration in collagenase-treated multicellular spheroids, Int. J. Nanomedicine 2 (2007) 265–274.
[108] H.A. Havel, Where are the nanodrugs? An industry perspective on development of drug products containing nanomaterials, AAPS J. 18 (2016) 1351–1353.
[109] Liposomal Doxorubicin Market Size, Share, Industry Report, 2013–2024 (2016) Available from : https://www.grandviewresearch.com/industry-analysis/liposomal-doxorubicin-market.
[110] R. Fernández-Urrusuno, P. Calvo, C. Remuñán-López, J.L. Vila-Jato, M.J. Alonso, Enhancement of nasal absorption of insulin using chitosan nanoparticles, Pharm. Res. 16 (1999) 1576–1581.
[111] A. Vila, A. Sánchez, K. Janes, I. Behrens, T. Kissel, J.L.V. Jato, M.a.J. Alonso, Low molecular weight chitosan nanoparticles as new carriers for nasal vaccine delivery in mice, Eur. J. Pharm. Biopharm. 57 (2004) 123–131.
[112] S.H. Choi, H.J. Byeon, J.S. Choi, L. Thao, I. Kim, E.S. Lee, B.S. Shin, K.C. Lee, Y.S. Youn, Inhalable self-assembled albumin nanoparticles for treating drug-resistant lung cancer, J. Control. Release 197 (2015) 199–207.
[113] W.H. Roa, S. Azarmi, M.K. Al-Hallak, W.H. Finlay, A.M. Magliocco, R. Löbenberg, Inhalable nanoparticles, a non-invasive approach to treat lung cancer in a mouse model, J. Control. Release 150 (2011) 49–55.
[114] A.A. Date, J. Hanes, L.M. Ensign, Nanoparticles for oral delivery: design, evaluation and state-of-the art, J. Control. Release 240 (2016) 504–526.
[115] Y. Yun, Y.W. Cho, K. Park, Nanoparticles for oral delivery: targeted nanoparticles with peptidic ligands for oral protein delivery, Adv. Drug Deliv. Rev. 65 (2013) 822–832.
[116] L. Zhang, F. Gu, J. Chan, A. Wang, R. Langer, O. Farokhzad, Nanoparticles in medicine: therapeutic applications and developments, Clin. Pharmacol. Ther. 83 (2008) 761–769.
[117] S. Svenson, Clinical translation of nanomedicines, Curr. Opin. Solid State Mater. Sci. 16 (2012) 287–294.
[118] S. Danhauser-Riedl, E. Hausmann, H.-D. Schick, R. Bender, H. Dietzfelbinger, J. Rastetter, A.-R. Hanauske, Phase I clinical and pharmacokinetic trial of dextran conjugated doxorubicin (AD-70, DOX-OXD), Investig. New Drugs 11 (1993) 187–195.
[119] J.M. Rademaker-Lakhai, C. Terret, S.B. Howell, C.M. Baud, R.F. de Boer, D. Pluim, J.H. Beijnen, J.H. Schellens, J.-P. Droz, A phase I and pharmacological study of the platinum polymer AP5280 given as an intravenous infusion once every 3 weeks in patients with solid tumors, Clin. Cancer Res. 10 (2004) 3386–3395.
[120] J.W. Valle, A. Armstrong, C. Newman, V. Alakhov, G. Pietrzynski, J. Brewer, S. Campbell, P. Corrie, E.K. Rowinsky, M. Ranson, A phase 2 study of SP1049C, doxorubicin in P-glycoprotein-targeting pluronics, in patients with advanced adenocarcinoma of the esophagus and gastroesophageal junction, Investig. New Drugs 29 (2011) 1029–1037.
[121] A.C. Mita, A.J. Olszanski, R.C. Walovitch, R.P. Perez, K. MacKay, D.P. Tuck, C. Simmons, S. Hammond, M.M. Mita, M. Beeram, Phase I and pharmacokinetic study of AI-850, a novel microparticle hydrophobic drug delivery system for paclitaxel, Clin. Cancer Res. 13 (2007) 3293–3301.