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Review

Towards targeted cancer therapy: Aptamer or oncolytic virus?

Kei X. Tan [a], Michael K. Danquah [a,*], Amandeep Sidhu [b,c], Clarence M. Ongkudon [d], Sie Yon Lau [a]

[a] Department of Chemical Engineering, Curtin University, Sarawak 98009, Malaysia
[b] Curtin Sarawak Research Institute, Curtin University, Sarawak 98009, Malaysia
[c] Faculty of Health Sciences, Curtin University, Perth 6102, Australia
[d] Biotechnology Research Institute, Universiti Malaysia Sabah, Kota Kinabalu, Sabah 88400, Malaysia

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ABSTRACT

Cancer is a leading cause of global mortality. Whilst anticancer awareness programs have increased significantly over the years, scientific research into the development of efficient and specific drugs to target cancerous cells for enhanced therapeutic effects has not received much clinical success. Chemotherapeutic agents are incapable of acting specifically on cancerous cells, thus causing low therapeutic effects accompanied by toxicity to surrounding normal tissues. The search for smart, highly specific and efficient cancer treatments and delivery systems continues to be a significant research endeavor.

Targeted cancer therapy is an evolving treatment approach with great promise in enhancing the efficacy of cancer therapies via the delivery of therapeutic agents specifically to and into desired tumor cells using viral or non-viral targeting elements. Viral oncotherapy is an advanced cancer therapy based on the use of oncolytic viruses (OV) as elements to specifically target, replicate and kill malignant cancer cells selectively without affecting surrounding healthy cells. Aptamers, on the other hand, are non-viral targeting elements that are single-stranded nucleic acids with high specificity, selectivity and binding affinity towards their cognate targets. Aptamers have emerged as a new class of bioaffinity targeting elements can be generated and molecularly engineered to selectively bind to diverse targets including proteins, cells and tissues. This article discusses, comparatively, the potentials and impacts of both viral and aptamer-mediated targeted cancer therapies in advancing conventional drug delivery systems through enhanced target specificity, therapeutic payload, bioavailability of the therapeutic agents at the target sites whilst minimizing systemic cytotoxicity. This article emphasizes on effective site-directed targeting mechanisms and efficacy issues that impact on clinical applications.

Contents

* Corresponding author at: Department of Chemical Engineering, Curtin University, Malaysia.
E-mail address: mkdanquah@curtin.edu.my (M.K. Danquah).

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1. Introduction

Cancer continues to be one of the primary causes of global mortality. It is estimated that approximately 1.6 million new cancer cases with around 580 thousands deaths were recorded in the United States in 2015 (Siegel et al., 2015). Cancer originates from uncontrolled cell division with potential metastasis into other normal neighboring tissues. It is caused by internal factors (such as extreme hormonal changes, inherited genetic disorders, immunity, and family history), external factors (such as poor diet, unhealthy lifestyles, tobacco, and alcohol), and environmental factors (such as exposure to chemicals, ultraviolet light, radiation, pollution, and infectious organisms) (Ferlay et al., 2015; American Cancer Society, 2015), and it is often diagnosed only after years of exposure to these factors. Cancerous cells differ from other normal cells by unlimited replication and proliferation, persistent angiogenesis, metastasis, evading apoptosis, and invading tissues (Li et al., 2014a, 2014b; Singh et al., 2012). These cancerous/tumor cells can survive even with redundant signaling pathways (Li et al., 2014a, 2014b).

Today, there are various cancer treatments available. These include surgery, chemotherapy, radiotherapy, immune therapy, and targeted therapy. Although recent advances in medicine have resulted in increased patient survival rates, cancer recurrence after recovery still persists, and this is a key hindrance to long-term survival (Smith et al., 2013). Targeted therapy is the most advanced form of cancer therapies aimed at delivering treatment to specific and targeted cancerous cells with minimal cytotoxic effects (Shaikh, 2012). It makes use of targeting agents which are either viral or non-viral elements with specific functional features (Kim et al., 2011b).

Viruses have been demonstrated as effective vaccination vectors and are now developed as novel antitumor agents with the capability of activating lytic activity and antitumor immune responses (Woller et al., 2014). They can kill cancerous cells via diverse mechanisms including apoptosis, autophagy, direct lysis, necrosis, toxic protein expression and immune response stimulation (Wong et al., 2010). Viral vectors are widely used as effective therapeutic delivery vehicles for both in vitro and in vivo gene expression for cancer treatment (Giacca and Zacchigna, 2012). Examples include the activation of the immune system by delivering genes that encode for co-stimulatory proteins into tumor cells; and the inhibition of tumor cell proliferation by hindering the regulatory proteins of the cell cycle (Giacca and Zacchigna, 2012). Over the past decades, viral oncotherapy has been used widely in cancer treatments for specific tumor targeting and inactivation due to the inherent anticancer properties of oncolytic viruses (OV) that enable them to replicate, spread and kill tumor cells without damaging surrounding normal non-cancerous cells (Singh et al., 2012; Russell and Peng, 2007; Lu et al., 2012; Chiocca and Rabkin, 2014). OVs are made of DNA and RNA viruses that are either tumor selective by nature or genetically-engineered (Chiocca and Rabkin, 2014). OVs employed in cancer treatments are often non-pathogenic naturally occurring viruses of either the wild-type that are only cytotoxic to malignant cells (Cripe et al., 2009), or naturally-existing mutants that are attenuated (Eager and Nemunaitis, 2011). The first commercialized OV anticancer drug is the H101 type 5 adenovirus with E1B-55KD and partial deletions of E3 gene. This OV anticancer drug was granted in 2005 by Chinese regulators as a result of its safety and superior anti-tumor performance in treating head and neck cancer when combined with chemotherapy (Russell and Peng, 2007; Vähä-Koskela et al., 2007).

Non-viral targeting elements such as aptamers show significant therapeutic potential due to their favorable biophysical and biochemical characteristics such as low immunogenicity, high productivity, biodegradability and biocompatibility (Kim et al., 2011b). Aptamers are oligonucleotides which can either be single-stranded deoxyribonucleic acid (ssDNA) or ribonucleic acid (ssRNA) molecules (Sun et al., 2014). They are widely used as advanced cell targeting elements in clinical diagnostics and targeted therapeutic delivery due to their high selectivity, specificity and binding affinity to their targets (McKeague and DeRosa, 2012). Aptamers fold into specific 3-D structures (Song et al., 2012; Baird, 2010) to bind to their targets via hydrogen bonding, Van der Waal interactions, electrostatic interaction and/or hydrophobic interactions (McKeague and DeRosa, 2012; Upadhyay et al., 2013; Witt et al., 2015). The conformation of the interactions between an aptamer and its target is based on the 3-D structure of the aptamer. Systematic Evolution of Ligands by Exponential enrichment (SELEX) technology is an in vitro iterative selection process used to generate aptamers specific for a desired target with high binding affinity. The SELEX methodology comprises of repetitive selection, amplification and enrichment schemes until the library is enriched with a specific target clone to derive the aptameric sequence (Radom et al., 2013; Orava et al., 2010; Alibolandi et al., 2015). The emergence of several SELEX modifications allows the generation of aptamers with high specificity towards a wide range of targets including proteins, cells and tissues (Ye et al., 2012; Tan et al., 2016; Santosh and Yadava, 2014). Hence, they are often experimented and used as drug carriers for targeted pharmaceutical delivery and as biological drugs for therapeutic treatments. The approval of pegatanib aptamer by US Food and Drug Administration (FDA) in 2004 for the treatment of vascular age-related macular degeneration has become the landmark in the clinical applications of aptamers (Song et al., 2012). There are a number of therapeutic aptamers in clinical trials. For example, the nucleolin-specific AS1411 aptamer for treating acute myeloid leukemia and renal cell carcinoma (Sun et al., 2014), and the NU172 aptamer for targeting thrombin molecules to treat anticoagulation in heart disease (Song et al., 2012). This article discusses the prospects of aptamer-mediated targeted cancer therapies for enhanced cancer treatment in juxtaposition with viral oncotherapy. It focuses on the targeting mechanisms, challenges faced and milestones achieved by both OVs and aptamers as targeting agents for effective pharmaceutical delivery for cancer treatment.

2. Global cancer scenario

Cancer is a major global health problem causing about one in every seven deaths worldwide (American Cancer Society, 2015). The high mortality rate of cancer persists regardless of the significant developments in cancer therapies over the past few decades (Singh et al., 2012). It has been reported that over 60% of cancer deaths happen in low-medium resource economies due to poverty, ignorance, environmental pollution, and poor medical and health systems (Siegel et al., 2015). Notwithstanding the advancements in modern medicine, the global cancer 'epidemic' is increasing significantly. The American Cancer Society predicts that there will be approximately 21.7 million of new cancer cases with 13 million deaths in 2030 (American Cancer Society, 2015). According to GLOBOCAN, about 15 million new cancer cases and 8.8 million cancer mortality cases, excluding non-melanoma skin cancer, are expected globally in 2015 (Ferlay et al., 2013). It has been reported that about one-third of cancer cases in developed nations are due to unhealthy behaviors including poor nutrition, obesity, and physical inactivity (American Cancer Society, 2015). Cancers that are common in men are lung and bronchus, colorectal, prostate, pancreas and liver cancers whilst women are mostly diagnosed with breast, colorectum, pancreas, ovary, and lung and bronchus cancers (Siegel et al., 2015; Ferlay et al., 2015). According to American Cancer Society, the top five cancers in both men and women are breast, prostate, cervix uteri, lung and colorectal cancers (Siegel et al., 2015; Saranath and Khanna, 2014). Globally, most of the cancer related deaths are caused by lung, stomach and liver cancers (Sharma et al., 2014) (See Table 1.).

Cancer cases are expected to increase, and the challenge is to develop effective medical interventions to address this top public health concern. Cancer therapies such as chemotherapy and radiotherapy provide immediate pathways to treat cancerous cells, and have been partially or completely successful in many cases. However, these therapies are challenged with low therapeutic index due to their infectiveness to

Table 1
A summary of different types of cancers with causes, mortality, region effects and treatment.

Cancer type	Common causes	Estimated number of cancer deaths in 2015	Region	Treatment	References
Lung	Tobacco intake and consumption, infectious agents such as hepatitis B virus (HBV), radon gas exposure, asbestos, secondhand smoke, some metals and organic chemicals, air pollution, radiation, tuberculosis (medical history), and genetic susceptibility	1,732,185	Central and Eastern Europe, Eastern Asia, Northern America, Northern Europe, and South East Asia	Cessation or tobacco avoidance, appropriate vaccination, surgery, chemotherapy, targeted therapies, and radiation therapy	(Ferlay et al., 2015; American Cancer Society, 2015)
Breast	First birth at older age, high usage of menopausal hormone therapy, poor medical treatment, obesity, long-term heavy smoking, genetic susceptibility (family history), and oral contraceptives consumption	560,407	Northern America, Northern and Southern Europe, Australia, New Zealand, Europe, Oceania, and the Americas	Surgical removal of cancerous cells, mastectomy, radiation therapy, hormone therapy, and chemotherapy,	(Ferlay et al., 2015; American Cancer Society, 2015)
Liver	Poor prognosis, tobacco consumption, alcohol consumption, infectious agents such as HBV and/or hepatitis C virus (HCV), alcoholic liver disease, diabetes, and overweight	806,873	Eastern and South-Eastern Asia, Southern Europe, Northern America, and Western Africa	Alcohol avoidance, vaccination, surgical removal of part of the liver, liver transplantation, embolization, ablation, and targeted drugs	(Siegel et al., 2015; Ferlay et al., 2015; American Cancer Society, 2015)
Stomach	Poor hygiene, high salt intake, poor food preservation methods, poor nutrition, infectious agents, and high prevalence of *Helicobacter pylori*	785,558	Eastern Asia, Central and Eastern Europe, Central and South America	Surgery (gastrectomy), chemotherapy, radiation therapy, and targeted therapy	(Siegel et al., 2015; Ferlay et al., 2015)
Colorectal	Poor prognosis, obesity, moderate to heavy alcohol intake, high intake of red meat, long-term smoking, poor nutrition, low calcium consumption, chronic inflammatory bowel disease, and family history	752,731	Australia, New Zealand, Central and Eastern Europe	Surgery, colostomy, chemotherapy, radiation therapy, and targeted therapy	(Ferlay et al., 2015; American Cancer Society, 2015)
Prostate	Family history, age, inherited genetic susceptibility, obesity, and high consumption of dairy foods and/or processed meats	335,643	Australia, New Zealand, Northern America, South America, Southern Africa, Caribbean, Western and Northern Europe, and Oceania	Active surveillance, surgery, brachy therapy, external beam radiation, hormonal therapy, radiation therapy, and vaccination (sipuleucel-T)	(Ferlay et al., 2015; American Cancer Society, 2015)
Cervical	Infectious agents such as human papillomavirus (HPV), sexual activity at early age, multiple sexual partners, suppressed human immune system, consumption of oral contraceptives, and tobacco intake.	284,923	Eastern Africa, Melanesia, Southern and Middle Africa	Prophylactic vaccination against HPV16/18, surgery, radiation therapy, chemotherapy, loop electrosurgical excision procedure, laser ablation, cryotherapy, and targeted drug therapy	(Siegel et al., 2015; Ferlay et al., 2015; American Cancer Society, 2015; Saranath and Khanna, 2014)
Pancreatic	Tobacco intake, family history, poor prognosis, heavy alcohol consumption, overweight, diabetes, and genetic syndromes	359,354	Asia, Eastern Asia, Europe	Surgery, chemotherapy, radiation therapy, and targeted drugs	(Ferlay et al., 2015; American Cancer Society, 2015)
Leukemia	Poor prognosis, genetic susceptibility (family history), ionizing radiation exposure (e.g.: medical radiation), down syndrome, genetic abnormalities, obesity, parental smoking, and chemical exposure (e.g.: benzene, formaldehyde)	283,373	Northern America, Australia, and New Zealand	Chemotherapy, and stem cell transplantation	(Siegel et al., 2015; Ferlay et al., 2015; American Cancer Society, 2015)
Ovarian	Family history of ovarian or breast cancer, pelvic inflammatory disease, Lynch syndrome, menopausal hormone therapy, obesity, and tobacco intake	163,765	Asia, Europe, and the Americas	Surgery (salpingo-oophorectomy, hysterectomy, omentum), and chemotherapy	(Ferlay et al., 2015; American Cancer Society, 2015)

completely block cancer growth and metastasis; significant adverse effects to normal cells; and the potential evolution of cancer cells with higher drug resistance (Singh et al., 2012). Unfortunately, for many recovered cancer patients after treatment with chemotherapy, surgery, immunotherapy, and targeted agents, the malignant cells recur after months or years later (Cripe et al., 2009). There are several possible factors responsible for such recurrence. These include the low targeting efficacy of chemotherapy and radiotherapy to desired tumor cells, the difficulty in developing tailored treatment regimen for specific cancer scenarios, and the resistance to therapeutic agents by some cancerous cells (Cripe et al., 2009). Multifunctional targeted therapy presents the opportunity to design new targeting agents with high specificity and minimal or no systemic cytotoxicity, directing high drug dosage to desired tumor site, and thus creating enhanced therapeutic index (Singh et al., 2012). Today, there are several molecular targeting agents for cancer available for clinical applications whilst many more are being developed with the aim of revolutionizing the range and efficacy of cancer therapies (Shaikh, 2012).

3. Oncolytic viruses: conventional cancer targeting agents

Chemotherapy targets and kills both normal and tumor cells that divide rapidly. In contrast, targeted cancer therapies are intended to target desired tumor cells without affecting the growth of other normal cells (Aravind et al., 2012). It employs targeting agents to interact with specific targeted molecules in order to inhibit the growth and spread of desired cancer cells (Alibolandi et al., 2015; Aravind et al., 2012; Balashanmugam et al., 2014). With advanced knowledge and capabilities in medical bioscience, biotechnology and molecular biology, it has become possible to molecularly engineer viruses as targeting agents

with enhanced selectivity and oncolysis for cancer treatments and gene therapy (Singh et al., 2012; Cripe et al., 2009). Oncolytic virotherapy is a new cancer treatment that enhances the specificity of conventional tumor cell inactivation mechanisms by employing tumor-selective OVs that infect, replicate and lyse tumor cells specifically within cancerous tissues without affecting normal cells and tissue counterparts, thus providing an improved therapeutic index with minimal effects on normal cells (Singh et al., 2012; Cripe et al., 2009; Bartlett et al., 2013; Guo et al., 2008). Also, cancer cells lack many natural protection mechanisms against viruses and thus, are susceptible to virus infection (Svyatchenko et al., 2012). OVs possess various targeting mechanisms including pro-apoptotic, transductional, transcriptional, and translational (Russell and Peng, 2007). OVs work by infecting tumor cells and replicating within them. After replication, cytolysis occurs to trigger cancer cell death and release their progeny for further infection to surrounding tumor cells (Smith et al., 2013; Guo et al., 2008) (See Fig. 1.). Other specific anti-tumoral mechanisms include direct cytotoxicity, transgene expression, triggering anticancer immunity, and sensitization to radiotherapy and chemotherapy (Vähä-Koskela et al., 2007). Examples of OVs are adenovirus, adeno-associated virus, gammaretrovirus, parvovirus, lentivirus, herpes simplex virus, newcastle disease virus, measles virus, reovirus, and chicken anaemia virus (Singh et al., 2012; Smith et al., 2013; Smith et al., 2013; Cripe et al., 2009; Guo et al., 2008). An ideal viral vector is capable of substituting most of its viral genome with the desired therapeutic gene for expression at the tumor site (Giacca and Zacchigna, 2012). OVs possess desirable characteristics when they are the wild-type with non-pathogenic animal viral components, and cause cytotoxicity to tumor cells without infecting other normal cells (Chiocca and Rabkin, 2014). OVs are convenient and easy to genetically manipulate (Svyatchenko et al., 2012) to form attenuated viral mutants with deleted or mutated genes that are essential for replication (Cripe et al., 2009). OVs are promising agents to target cancer stem cells (CSCs) which are responsible for initiating tumor sites, self-renewing and differentiating for tumor growth as well as inducing metastasis (Smith et al., 2013). Extensive research works have been carried out on the application of OVs to target the cell surface markers of CSCs such as prominin-1 and CD133, in order to inhibit the spread of cancer cells. The evolution of cancer is seeded from a small number of active CSCs. In comparison with normal cancer cells, CSCs are more resistant to conventional chemo/radio-therapy, and can lead to the recurrence of a more resilient and metastatic cancer. Hence, OVs provides the opportunity to not only inactivate normal cancer cells but also targeting CSCs to improve survival rate and reduce the possibility of cancer recurrence (Smith et al., 2013).

Numerous clinical studies have demonstrated the efficacy of OVs in killing cancer cells that are resistant to radiotherapy or chemotherapy (Eager and Nemunaitis, 2011). One research study reported that oncolytic adenovirus conjugated with actively targeting Arg-Gly-Asp(RGD)-poly(cystaminebisacrylamide-diaminohexane) (poly(CBA-DAH)) biopolymer can be delivered safely to specific cancer cells for inducing apoptosis and suppressing both IL-8 and VEGF expression by expressing the short hairpin RNA. The observed therapeutic effects included anti-angiogenesis, inhibition of tumor migration, invasion and growth, high gene transfection efficiency, and low cytotoxicity (Kim et al., 2011a). Adenoviruses can selectively and actively kill tumor cells via cell lysis with amplified transgene expression, conditional replication and progeny production, and diffusion into neighbored cancer cells. The concerns associated with potential metathesis of oncolytic viruses is dependent on the transformations in the viral surface protein in terms of structural reorganisation and epitope mapping which are critical for specific and targeted tumor attack. Hence since intracapsular nucleic acid replication and translations during metathesis could result in surface transformations that can potentially affect the nature of the recognition proteins of progenies then the specificity of the signaling pathway would be affected. For instance, Ayala-Breton et al. (2014) genetically engineered the vesicular stomatitis virus by replacing its G glycoprotein with fusion (F) and hemagglutinin (H) envelope glycoproteins. This resulted in a higher recognition of virus towards CD46-overexpressing cancer cells, rapid viral replication and stronger cytopathic effect. The viral surface proteins of some virus have been modified to target surface receptors of specific cancer cells with high affinity. This modification is usually inherent to the viral genome and it is inherited upon subsequent duplications (Verheije and Rottier, 2012).

Another study by Kim et al. (2011a, 2011b) demonstrated the hepatoma-specific adenovirus (YKL-1001) conjugated with arginine-grafted bioreducible polymer (ABP) as an effective tumor targeting and therapeutic agent to inactivate liver cancer cells such as HepG2 and Huh 7 cells via systemic administration. This bioconjugate is more resistant to neutralizing antibodies, resulting in longer circulatory half-life, increased gene transduction efficiency and lytic potency, and limited cytotoxic effects for a more efficient and safe therapeutic delivery (Kim et al., 2011b). The result also illustrated the efficacy of adenovirus-ABP complex in killing coxsackie-adenovirus receptor (CAR)-deficient tumor cells via CAR-independent routes (Kim et al., 2011b). Additionally, a gibbon ape leukemia virus (GALV)-based replicating retrovirus vector (RRV) armed with a yeast cytosine deaminase (CD) suicide gene was indicated to provide high cancer cell inactivation

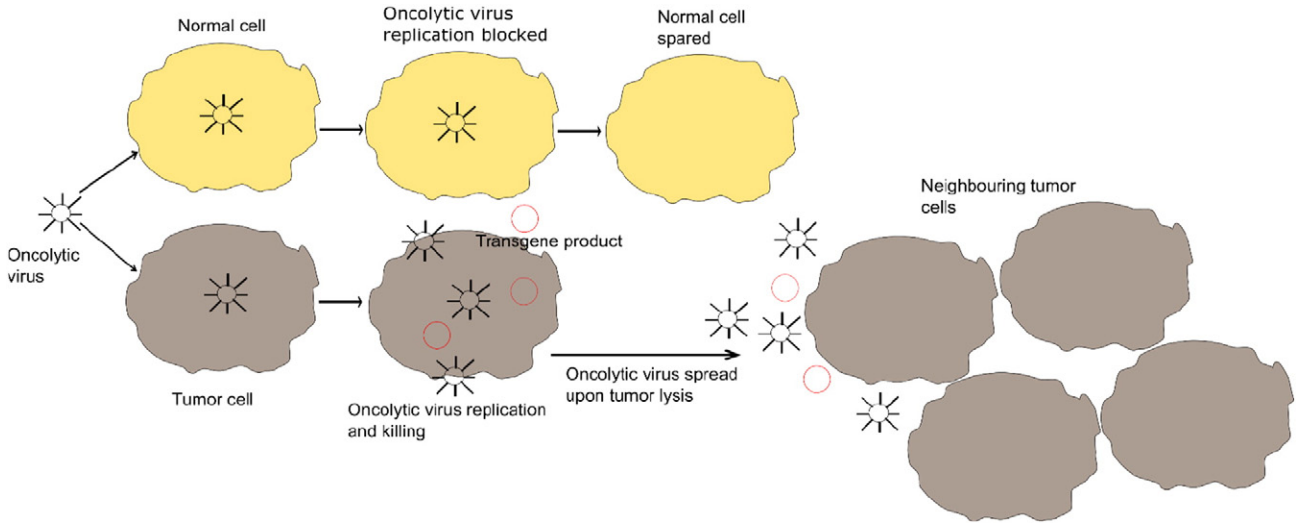

Fig. 1. Mechanism of OVs in killing tumor cells.

and inhibit in vivo cancer growth to treat hepatocellular carcinoma with enhanced gene delivery and transduction efficiency (Lu et al., 2012). This study showed significantly suppressed cancer growth due to the production of high suicide gene toxicity by GALV-RRV with no cytotoxicity to surrounding normal tissues (Lu et al., 2012). The suicide genes encoded enzymes that intracellularly converted the non-toxic prodrug into toxic metabolites, leading to cancer cell deaths via suicide gene therapy (Lu et al., 2012). Zhang et al., 2007 engineered a recombinant adeno-associated virus (rAAV) vector to deliver thrombospondin-1 type 1 repeats (3TSR) and endostatin for anti-angiogenic gene therapy with significant anti-angiogenic and anticancer effects in vivo (Zhang et al., 2007). The experimental results illustrated the transgene expression of 3TSR and endostatin that caused efficient suppression of the vascular endothelial growth factor (VEGF)-induced angiogenesis at both local and distant sites in mice with pancreatic cancer cells (Zhang et al., 2007). Bhutia et al., 2013 investigated the efficacy of adenovirus in delivering melanoma differentiation-associated gene-7 (*mda-7*) to targeted tumor cells for blocking the proliferation of breast cancer stem cells via the suppression of Wnt/β-catenin signaling pathway, endoplasmic reticulum stress and apoptosis without harming normal stem cells (Bhutia et al., 2013). The result also showed the enhancement of chemotherapy and antibody-elicited killing due to the expression of *mda-7*/interleukin-24 within the breast tumor cells after the adenovirus.*mda*-7 infection (Bhutia et al., 2013) (See Table 2.).

OVs possess positive traits that make them effective as targeting and delivery agents. For instance, adenovirus provides large foreign DNA capacity; efficient intracellular delivery of therapeutic genes; cancer-selective killing; low mutagenesis rate and genotoxicity; non-integration into genome or chromosomes of host; and high titers concentration (Kim et al., 2011b; Kim et al., 2011a). One of the favorable characteristics of OVs is their viral persistence whereby infected cancer cells turn into a source of constant viral production to help in spreading the viral vector to metastatic tumor cells (Lu et al., 2012; Chiocca and Rabkin, 2014). Most genes that encode for viral proteins are knocked off or removed from the viral genome hence, OVs do not infect human whilst killing cancer cells. Also, the microenvironment of cancer cells is optimal for viral replication (Smith et al., 2013). Many research studies have demonstrated the effectiveness of OVs in combating a wide range of human cancer cells in both preclinical and clinical trials (Cripe et al., 2009). Safety concerns such as sensitivity towards immune system and drug can be armed in Ovs (Chiocca and Rabkin, 2014). Likewise, larger OVs can be equipped to express immune-stimulating transgenes to enhance their therapeutic effects by attracting more immune effector cells (Smith et al., 2013). Therefore, OVs are able to undergo genetic manipulations to provide enhanced gene therapies that target specific receptors as well as various activated cellular pathways (Smith et al., 2013; Kim et al., 2011a). Additionally, OVs with various tropisms are capable of combating multiple tumor types. They are capable on acting on distant metastasis besides primary tumors (Chiocca and Rabkin, 2014).

4. Challenges of OVs as targeting elements

Undoubtedly, more research efforts are needed to fully exploit the therapeutic potential of oncolytic virotherapy. In 1999, a healthy teenager was killed as a result of toxic shock in a clinical trial involving the use of adenovirus to treat inherited Ornithine Transcarbamylase Deficiency (Svyatchenko et al., 2012). Another incident happened in 2002 when 2 volunteers developed leukemia after receiving a trial gene therapy to treat X-linked severe combined immunodeficiency (Svyatchenko et al., 2012). These cases, amongst other recently reported serious side-effects and cancer resurgence cases, demonstrate the limitations of OVs in gene therapy particularly in relation to the triggering of immune responses against the viral vectors and transgenes, and also inducing mutations as a result of inappropriate insertion of transgenes (Svyatchenko et al., 2012). OVs possess several other shortcomings which limit their

Table 2
Summary of some oncolytic viruses for targeted delivery.

Oncolytic virus (OV)	Target	Cancer	Positive effects	Reference
Adenovirus, reovirus, herpes simplex virus-1, vaccinia virus, measles virus	Prominin-1 and CD133 of CSCs	Cancer stem cells (CSCs)	Inhibited the spread of tumor cells; minimized cancer relapse; and increased survival rate	(Smith et al., 2013)
Adenovirus conjugated with Arg-Gly-Asp(RGD)-poly(cystaminebisacrylamide-diaminohexane) (poly(CBA-DAH)) biopolymer	Interleukin-8 and vascular endothelial growth factor	Human cancer cells (A549 lung carcinoma, MCF7 breast adenocarcinoma, HT1080 fibrosarcoma)	Induced apoptosis; suppressed expression of IL-8 and VEGF; and anti-angiogenesis	(Kim et al., 2011a)
Adenovirus (YKL-1001) conjugated with arginine-grafted bio-reducible polymer (ABP)	A-fetoprotein expressing HepG2 and Huh 7 liver cancer cells, and CAR-deficient tumor cells	Hepatocellular carcinoma	Increased the gene transduction efficiency; enhanced the lytic potency, prolonged the circulatory half-life; and minimized cytotoxicity	(Kim et al., 2011b)
Gibbon ape leukemia virus (GALV)-based replicating retrovirus vector (RRV)	Prodrug 5-fluorocytosine (5-FC)	Hepatocellular carcinoma	Enhanced gene delivery; improved transduction efficiency; and increased cancer killing effect	(Lu et al., 2012)
Recombinant adeno-associated virus (rAAV) vector	Vascular endothelial growth factor	Pancreatic cancer cells	Successfully delivery of thrombospondin-1 type 1 repeats (3TSR) and endostatin; and anti-angiogenesis	(Zhang et al., 2007)
rAAV serotypes 6	NEU antigen in NEU-expressing breast cancer cells	Breast cancer	Expressed neu oncogene; and triggered cell-mediated and humoral immune responses	(Steel et al., 2013)
Adenovirus. melanoma differentiation-associated gene-7 (*mda-7*)	Wnt/β-catenin signaling pathway	Breast cancer stem cells	Expression of *mda-7*; suppression of Wnt/β-catenin signaling pathway; endoplasmic reticulum stress; and apoptosis	(Bhutia et al., 2013)

therapeutic applications. These shortcomings include high immunogenicity, low capacity for gene insertion, short-term gene expression, limited viral vector delivery as a result of poor viral transduction, and the propensity of eliciting insertional mutagenesis and tumorigenicity (Luo et al., 2015). Some OVs can only be therapeutically effective to cancer cells that contain specific oncogenic profiles such as the activated Ras pathway in the case of reovirus (Smith et al., 2013). Therapeutic efficiency of certain OVs including adenovirus and vaccinia virus is limited by pre-existing immunity. In such cases, OVs are eradicated by phagocytic cells and neutralizing antibodies before they kill the tumor cells (Smith et al., 2013; Russell and Peng, 2007; Kim et al., 2011a; Ferguson et al., 2012). OVs such as adenovirus tend to accumulate in the liver, causing liver toxicity, short half-life and adverse side effects within the host, and be excreted rapidly by neutralizing antibodies in vivo (Kim et al., 2011b; Kim et al., 2011a). OVs also face challenges such as non-specific uptake and clearance by the liver, lung and spleen to hinder their systemic availability (Ferguson et al., 2012).

OVs administered intravenously are removed from blood circulation rapidly and the increasing antiviral immunity speeds up this elimination process for subsequent exposures (Russell and Peng, 2007). This limits the systemic administration and delivery of therapeutic drugs which are essential for targeting both primary and metastatic tumors (Kim et al., 2011b). It has been reported that interactions between adenovirus and red blood cells and platelets result in toxicity, reduced therapeutic availability, and undesirable side effects (Kim et al., 2011b; Ferguson et al., 2012). In addition, the expression of coxsackie-adenovirus receptor (CAR) was observed to be low on cancer cells, leading to low CAR-mediated viral transduction efficiency (Kim et al., 2011b). Viral vectors like retrovirus have various disadvantages in term of flexibility, storage, production and safety issues including activating latent disease (Nie et al., 2011). Gammaretrovirus, which used to be the most utilized gene vehicle, has been reported to be inefficient in transducing non-replicating cells and possesses the propensity to induce insertional mutagenesis (Giacca and Zacchigna, 2012). Also, there is a possibility that OVs can evolve into a pathogen and evade the host immune system, and might result in person-to-person transmission of the original or pathogenic derivative (Russell and Peng, 2007). Although adenoviruses are effective and safe to be involved in cancer therapy based on various clinical trials (Phase I and II), they are incapable of being used as a monotherapy due to their low efficacy in tumor-killing (Svyatchenko et al., 2012).

5. Aptamers as a new class of targeting agents for cancer therapy

Aptamers have gained significant interests in biomedical science and pharmaceutical delivery as ideal targeting agents for drug delivery. Aptamers have the potential to recognize and bind specifically to targeted cells in vivo for successful therapeutic outcomes with minimal cytotoxic effects to surrounding normal cells (Balashanmugam et al., 2014). Aptamers fold into specific 3-D structures to interact with their targets under affinity binding conditions with dissociation constants ranging from pico- to nano-molar (Sun et al., 2014). The strong and specific binding characteristics of aptamers make them effective targeting elements. Aptamers are chemically and thermally stable due to their strong phosphodiester bonds, and those with small molecular weights are able to diffuse through blood circulation more rapidly with high cellular penetration. Aptamers are mostly non-immunogenic, and their production technique is scalable and inexpensive (Sun et al., 2014; Tan et al., 2016; Gedi and Kim, 2014). With great targeting mechanisms, aptamers are able to specifically recognize minor structural differences within the target molecule of interest. For example, aptamers can identify and bind to hidden epitopes, a characteristic feature which is not easily achieved with conventional targeting ligands (Sun et al., 2014; Radom et al., 2013).

Aptamer can be used either alone or as the aptamer-drug conjugate whereby it is used alone when it acts as a medical drug. For example, pegatanib aptamer is a FDA-approved aptamer-based drug to target, bind and antagonize vascular endothelial growth factor (VEGF) with high affinity via its unique 3-D conformation. This inhibits the binding of VEGF to its receptors in order to prevent angiogenesis and reduce vascular permeability (Song et al., 2012). Vavalle and Cohen (2012) demonstrated that REG1 aptamer (clinical trial Phase II) is capable of treating acute coronary syndrome by selectively inhibiting factor IXa in the REG1 anticoagulation system. Another research study by Gao et al. (2014) demonstrated the efficiency of NS2-specific aptamer as an inhibitor that impedes the binding of NS2 towards NS5A protein to prevent viral RNA replication, which then leads to anti-hepatitis C virus infection. Aptamers can also be used as ideal drug carriers to act as a navigator and guide the medical drug to targeted cancerous site without causing cytotoxic effect to surrounding healthy cells. This is predominantly due to the high affinity and selectivity of aptamers as targeting element towards extracellular receptors or surface biomarkers of cancer cells as shown in Table 3. Aptamer-mediated targeted drug delivery system is highly specific and significantly increases the dosage of delivered drug to targeted tumor cells as well as improving the therapeutic index (Radom et al., 2013; Zhu et al., 2014). There are many research conducted on the efficiency of different type of aptamer-drug conjugates and the details are well documented in Section 9 (page 19) of this review. It has been scientifically proven that the use of aptamers as targeting agents in targeted pharmaceutical delivery can enhance the therapeutic action of antitumor agents on a variety of cancers including brain cancer (Guo et al., 2011), breast cancer (Alibolandi et al., 2015; Aravind et al., 2012), pancreatic cancer (Sun et al., 2014), colorectal cancer (Li et al., 2014b), lymphoblastic leukemia (Aravind et al., 2012; Huang et al., 2009), hepatic cancer (Alibolandi et al., 2015) and prostate cancer (Min et al., 2011). (See Tables 4 and 5.)

Aptamers act as navigators to direct therapeutic payloads to desired targeted sites while minimizing systemic cytotoxicity to other normal cells and effectively triggering receptor-mediated internalization, called endocytosis, to increase the cellular uptake of therapeutic molecules into targeted tumor cells (Radom et al., 2013; Huang et al., 2009; Zhu et al., 2014). In order to enhance interaction with their desired targets, aptamers spontaneously re-conform their molecular binding structures to maximum binding at their active sites via hydrogen bonding, and electrostatic, hydrophobic and Van der Waal interactions (Upadhyay et al., 2013; Witt et al., 2015; Acquah et al., 2015). Aptamer-mediated targeted drug delivery is a promising approach for cancer therapy as aptamers can be generated and engineered to possess high binding affinities towards specific internalized cell surface receptors or biomarkers such as nucleolin for breast cancer cells, protein tyrosine kinase (PTK7) for acute lymphoblastic leukemia, and prostate-specific membrane antigen (PSMA) for prostate cancer cells as shown in Table 3 (Orava et al., 2010). The mechanism of aptamer targeting and cellular uptake comprises of three steps including targeting, endocytosis and cytotoxic effects (Orava et al., 2010) (See Fig. 2.). The targeting mechanism of aptamers begins when the aptamer binds to its desired biomarker to trigger a receptor-mediated internalization. The aptamer-receptor complex is then internalized via either clathrin-

Table 3
A list of surface biomarkers expressed onto different tumor cells.

Surface biomarker	Surface biomarker-expressing tumor cells
Nucleolin	Melanomas, gastric, breast, leukemia, lung tumor cells (Aravind et al., 2012).
Protein tyrosine kinase 7 (PTK 7)	Lung, gastric, colon tumor cells (Min et al., 2011; Li et al., 2014a, 2014b).
Epithelial cell adhesion molecule (EpCAM)	Bladder, ovarian, breast, pancreas, hepatocellular tumor cells (Alibolandi et al., 2015).
Prostate specific membrane antigen (PSMA)	Prostate, kidney tumor cells (Sun et al., 2014).

Table 4
Comparison between OVs and aptamers as targeting elements.

Parameters	OVs	Aptamer	Reference
Size	Nano-sized, and bigger than aptamers	8–25 kDa	(Giacca and Zacchigna, 2012; Sun et al., 2014; Lakhin et al., 2013)
Charge	Neutral, negative or positive depending on the total charge of its genes and coated protein	Negatively charged due to phosphate backbone	(Radom et al., 2013; Ahmad et al., 2014)
Therapeutic effect	Can be engineered for improved high therapeutic efficiency	Can be engineered for improved high therapeutic efficiency	(Smith et al., 2013; Giacca and Zacchigna, 2012; Russell and Peng, 2007; Alibolandi et al., 2015; Balashanmugam et al., 2014; Guo et al., 2011)
Mechanism of action	Transcriptional, translational, pro-apoptotic, transductional	Receptor-dependent internalization via hydrogen bonding, electrostatic and Van der Waals interaction	(Russell and Peng, 2007; Huang et al., 2009)
Versatility of application	Vaccination, targeted cancer therapy, and gene therapy	Biosensing, diagnostic and therapeutic applications, in vivo imaging, targeted therapy, food inspection, targeted drug delivery, new drug and biomarker discovery	(Kim et al., 2011b; Woller et al., 2014; Song et al., 2012)
Stability	Thermally stable	Thermally and more chemically stable	(Sun et al., 2014; Song et al., 2012)
Ease of formulation	Can be engineered to improve formulation	Easily formulated	(Russell and Peng, 2007; Baird, 2010; Santosh and Yadava, 2014)

independent or clathrin-dependent vesiculation into the endosome of the targeted tumor cells (Erdmann et al., 2014). This internalization process leads to increased drug uptake into the target cells (Tuerk and Gold, 1990; Radom et al., 2013; Alibolandi et al., 2015). Huang et al., 2009 conjugated doxorubicin (Dox) anticancer drug to sgc8c aptamer via a hydrazone linker or covalent bond to form the Dox-aptamer complex with no effect on the bioactivity of Dox and aptameric binding properties (Radom et al., 2013; Huang et al., 2009). After internalization, the acidic environment of the endosome triggers hydrolytic bonding cleavage that hydrolyzes the covalent bond between the conjugated chemotherapeutic drug and the aptamer. Consequently, the released drug passively diffuses through the endosomal membrane into the cytosol and finally enters the nucleus and intercalate into the gDNA of the targeted cancer cells to induce therapeutic effects (Radom et al., 2013; Huang et al., 2009). The aptamer is eliminated later during the fusion of endosome and lysosomes containing endonucleases that degrade the aptamers. Briefly, the endosomic acidic environment induces hydrolysis of acid-susceptible linkers whereas the acidic environment of the lysosome degrades linkers that are labile to proteolytic, hydrolytic or digestive enzymes (Vilar et al., 2012). The effectiveness of aptamer-mediated targeted therapeutic delivery can be affected by several factors such as the size, charge and nature of the conjugated chemotherapeutic drugs, amplitude of target cell surface biomarkers, and the endocytic nature of the targeted cell (Orava et al., 2010).

6. Biophysical limitations of aptamer-mediated targeted delivery

Although numerous reported pre-clinical targeted delivery studies using aptamers have reported promising results, there are only several commercialized aptamer-based drugs on the market as most aptamer-mediated targeted delivery technologies fail at various phases of clinical trials (Zhu et al., 2014; Liechty et al., 2010) For example, AS1411 DNA aptamer clinical study failed at phase II clinical trial for the treatment of renal cell carcinoma notwithstanding the numerous pre-clinical validations (Zhu et al., 2014). Also, BAX499 aptamer clinical study for the treatment of hemophilia was terminated at phase I due to low therapeutic index (Peyvandi et al., 2013). This demonstrates that major biochemical and biophysical challenges that impede effective aptamer-

Table 5
A list of FDA-approved oncolytic viruses and aptamers and those undergoing clinical trials.

	Developmental stage	Medical application
Oncolytic virus		
Imlygic (oncolytic herpes simplex virus type 1)	FDA-approved	A genetically modified virus to cure melanoma skin cancer found in lymph glands or on skin. It is administered by injections targeting melanoma lesions (FDA, 2015). Retrieved from http://www.fda.gov/NewsEvents/Newsroom/PressAnnouncements/ucm469571.htm on 1st August 2016.
MG1MA3 maraba virus and AdMA3 adenovirus	Clinical trial Phase I	To target MAGE-A3 protein-expressing cancer cells in order to trigger immune responses (Ottawa Hospital Research Institute, 2015). Retrieved from http://www.ohri.ca/newsroom/newsstory.asp?ID=649 on 1st August 2016.
LOAd703 adenovirus	Clinical trial Phase I/IIa	To treat pancreatic cancer by stimulate the immune system in addition to the killing effect of adenovirus towards cancer cells (U.S. National Institutes of Health, 2016). Retrieved from https://clinicaltrials.gov/ct2/show/NCT02705196?term=oncolytic+virus&rank=3 on 1st August 2016.
CG0070 adenovirus	Clinical trial Phase III	To cure bladder cancer using granulocyte macrophage-colony stimulating factor (GM-CSF)-encoded adenovirus to trigger systemic immune response (U.S. National Institutes of Health, 2016). Retrieved from https://clinicaltrials.gov/ct2/show/NCT02705196?term=oncolytic+virus&rank=3 on 1st August 2016.
Aptamer		
Pegatanib aptamer (Macugen)	FDA-approved	To inhibit angiogenesis and medicate neovascular age-related macular degeneration (Song et al., 2012).
Nuclein-specific AS 1411	Clinical trial Phase II completed	To cure acute myeloid leukemia (Sun et al., 2014).
REG1 aptamer-based anticoagulant	Clinical trial Phase III recruiting	To inhibit factor IXa in treating acute coronary syndrome (Vavalle and Cohen, 2012).
E10030 DNA aptamer	Clinical trial Phase III recruiting	To target platelet-derived growth factor in treating age-related macular degeneration (Ni et al., 2011).

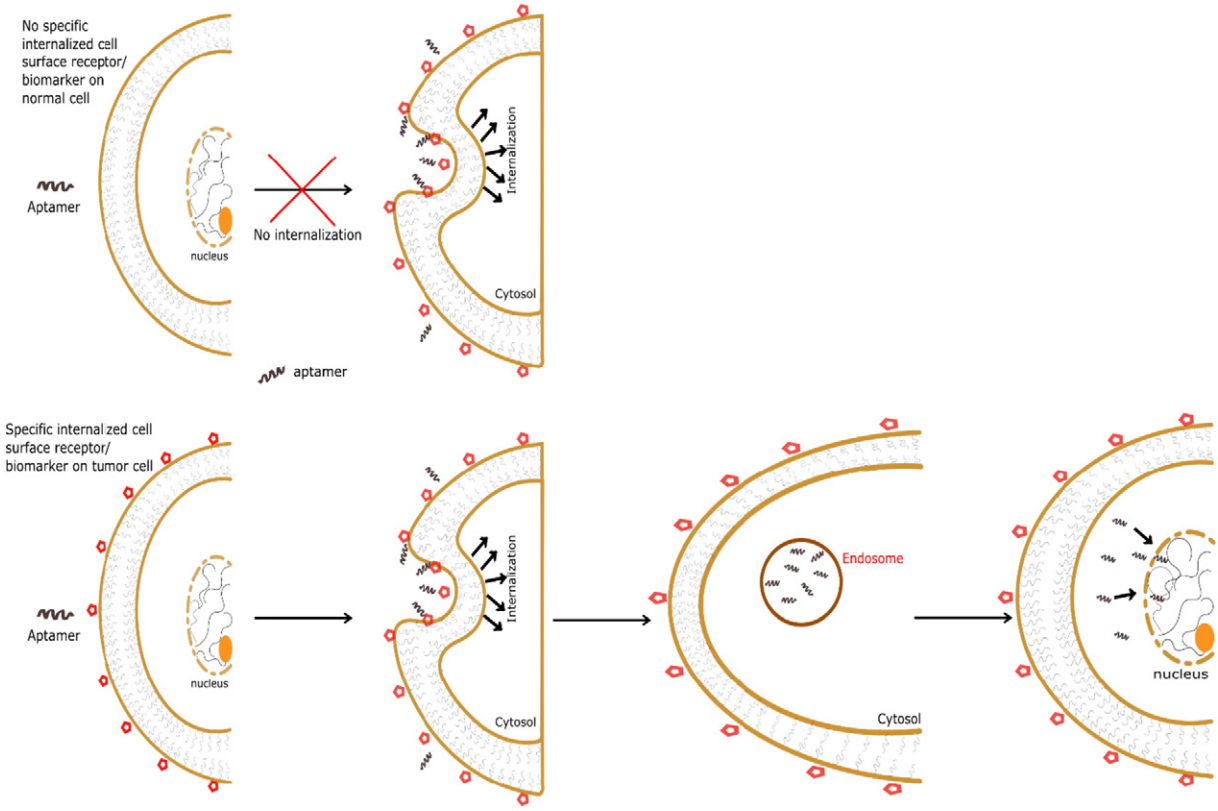

Fig. 2. The killing mechanism of aptamer-mediated formulations towards tumor cells.

mediated in vivo targeted delivery still remain to be addressed. Aptameric targets are often internalized surface protein receptors or biomarkers of the target cells residing in interstitial fluids or blood plasma. It is therefore essential that the aptamers are biochemically stable in these extracellular fluid compartments for a period of time. However, the stability of aptamers in these compartments is challenged by negatively charged cell membrane electrostatic repulsion, endonuclease degradation, rapid renal clearance, and rapid bio-distribution of aptamers from the blood plasma into tissues (Sundaram et al., 2013; Lakhin et al., 2013; Wengerter et al., 2014; Wengerter et al., 2014). Consequently, aptamers have a short circulating half-life and this affects their performance as biological drugs, drug carriers and/or targeting agents (Sun et al., 2014; Orava et al., 2010; Liechty et al., 2010).

Aptamers are oligonucleotides with a low molecular weight of 8-25 kDa, making them labile to rapid renal excretion. The kidney filters and removes molecules from the bloodstream within the molecular weight range of 30–50 kDa (Sundaram et al., 2013). The delivery of sufficient drug dosage to targeted sites is therefore significantly impeded by rapid renal excretion, leading to ineffective in vivo therapies (Lakhin et al., 2013; Wengerter et al., 2014). Additionally, nuclease-mediated degradation is one of the major drawbacks limiting the application of aptamers for effective pre-clinical and clinical therapeutic delivery. Aptamers are generally degraded in the bloodstream within minutes based on their conformation and concentration. Consequently, the amount of aptamers remaining in the bloodstream is inadequate for an effective therapeutic event to take place (Sundaram et al., 2013; Lakhin et al., 2013). Also, aptamers are hydrophilic negatively-charged molecules that are impermeable to biological barriers such as the cell membrane. Electrostatic repulsive forces exist between the aptamer and the target cell membrane surface, and this significantly obstructs effective aptamer-receptor binding, cell membrane permeation, receptor-mediated internalization, intracellular target-specific binding, and the desired cytotoxic effect (Orava et al., 2010; Wu et al., 2011). Furthermore, the simple structures of aptamers result in a small drug loading capacity, and this consequently decreases the drug dosage delivered to the targeted site (Sun et al., 2014). Aptameric binding mechanism can also affect the effective application of aptamers as targeting elements. Aptamers target cell surface protein receptors and this can potentially alter the drug bisorption rate to eventually affect the pharmacokinetic and pharmacodynamic properties. There is the possibility of aptamers fusing into the human genome to express foreign proteins which can lead to the evolution of non-specific normal cells into cancerous cells (Du et al., 2006).

7. Molecular mechanism of action: OVs vs aptamers

OVs are highly selective to cancerous site and this allows them to only duplicate and spread within the cancerous cell zone without killing surrounding healthy cells. For instance, OVs such as measles virus and coxsackie virus have been reported to possess a natural preference towards cancer cells due to the presence of unique surface protein markers that induce specific binding affinities to corresponding cancer cells (Thorne et al., 2007). As a result of the protein attachment, OVs latch onto the cancer cells for penetration followed by replication inside the cancer cells using the replication machinery of the host cells. OV particles then leave the host cells via cell lysis or budding after completing viral replication in order to target other surrounding cancer cells. Other OVs, including adenovirus, can be genetically engineered to knock off specific enzymes that are available only in tumor cells, and this leads to competition between the OV and tumor cells for the same mechanism of survival. The tumor antigens produced by tumor cells are usually undetected by the human immune system for destruction. However, in the presence of OVs that engage in tumor cells destruction, intracellular tumor antigens are released to activate the immune system. Thus, there is a synergetic effect between OVs and the body immune system (Ledford, 2015).

Aptamers can be engineered to kill cancer cells by interacting specifically with a wide range of surface biomarkers expressed onto various cancer cells as shown in Table 3. The mode of action begins when the aptamer selectively binds onto the surface receptors or internalized surface biomarkers of targeted cancer cells to stimulate endocytosis (Alibolandi et al., 2015; Huang et al., 2009; Radom et al., 2013). The mechanism of action for aptamer can be found in page 13 as follows:

> The mechanism of aptamer targeting and cellular uptake comprises of three steps including targeting, endocytosis and cytotoxic effects (Orava et al., 2010). The targeting mechanism of aptamers begins when the aptamer binds to its desired biomarker to trigger a receptor-mediated internalization. The aptamer-receptor complex is then internalized via either clathrin-independent or clathrin-dependent vesiculation into the endosome of the targeted tumor cells (Erdmann et al., 2014). This internalization process leads to increased drug uptake into the target cells (Tuerk and Gold, 1990; Radom et al., 2013; Alibolandi et al., 2015). Huang et al., 2009 conjugated doxorubicin (Dox) anticancer drug to sgc8c aptamer via a hydrazone linker or covalent bond to form the Dox-aptamer complex with no effect on the bioactivity of Dox and aptameric binding properties (Radom et al., 2013; Huang et al., 2009). After internalization, the acidic environment of the endosome triggers hydrolytic bonding cleavage that hydrolyzes the covalent bond between the conjugated chemotherapeutic drug and the aptamer. Consequently, the released drug passively diffuses through the endosomal membrane into the cytosol and finally enters the nucleus and intercalate into the gDNA of the targeted cancer cells to induce therapeutic effects (Radom et al., 2013; Huang et al., 2009). The aptamer is eliminated later during the fusion of endosome and lysosomes containing endonucleases degrade the aptamers.

8. Aptamer-conjugated polymeric particulates as targeted delivery systems

Aptamers serve as great cell-targeting elements with much potential in advancing conventional targeted drug delivery systems with increased therapeutic index and minimal systemic cytotoxicity. Notwithstanding, unresolved challenges associated with aptamer-mediated in vivo delivery have highly impacted practical applications of aptamers for effective clinical therapies. These challenges have triggered significant interests into the use of synthetic delivery systems as drug carriers. Biomedical research into effective synthetic therapeutic delivery carriers to overcome the drawbacks of aptamers has become a significant research endeavor. The emergence of biocompatible and biodegradable polymers that can be molecularly engineered to tune their biophysical and biochemical properties such as particulate size and distribution, surface area and morphology, chemical compositions, and toxicity has created opportunities to optimize the capacity of aptamers as targeting elements for sustained and controlled release of drug at effective dosages.

Polymeric particles have gained recognition as efficient and safe delivery vehicles in pharmaceutical drug delivery and in vivo therapeutic treatment as compared to other delivery systems such as bacterial and viral delivery systems. This is due to their unique properties including biocompatibility, low or non-immunogenicity, biodegradability, and bioavailability in addition to their tunable physicochemical features to generate a controlled drug release profile (Saranya and Radha, 2014). The use of polymeric particles as synthetic delivery carriers for aptamers conjugated with drug to target desired cells in pharmaceutical delivery is a promising approach to improve the specificity, selectivity and therapeutic efficacy whilst enhancing transfection efficiency (Alibolandi et al., 2015). There are various reported studies on aptamer-conjugated polymeric micro/nano-particles as therapeutic delivery carriers. These aptamer-polymer formulations have demonstrated promising results to improve the specificity of drug delivery systems by targeting various biomarkers including epithelial cell adhesion protein molecule (EpCAM) on breast and colon cancerous cell surfaces (Alibolandi et al., 2015; Subramanian et al., 2015), nucleolin on acute myeloid leukemia, gliomas and renal tumor cells (Aravind et al., 2012; Guo et al., 2011), PSMA on prostate tumor cell surfaces (Baird, 2010), and mucin-1 (MUC-1) on lung epithelial cancer cells (Lu et al., 2012; Zhang et al., 2007). An ideal polymeric carrier is capable of protecting the encapsulated drug and aptamer molecules from the physiological environment without compromising their bioactivity and biophysical properties whilst providing a controlled release of the active agents in optimal dosages with the appropriate release kinetics (Tan and Danquah, 2012). Consequently, a reproducible and predictable drug release profile for a sustained period of time can be achieved; the therapeutic effects of drugs with short half-lives can be prolonged for enhanced therapy; side effects, drug waste and frequent drug dosing can be significantly reduced for a better patient compliance (Vilar et al., 2012). There are many reported efficient aptamer-polymeric formulations using polymers such as poly(lactic-*co*-glycolic acid) (PLGA), poly(ethylene imine) (PEI), polyethylene glycol (PEG), poly(β-amino ester) (PBAE), chitosan, and poly (ortho esters) (POE). PLGA is one of the widely used biopolymer for pharmaceutical delivery. It is an approved synthetic biopolymer by the Food and Drug Administration (FDA) and World Health Organization (WHO) as a safe delivery carrier of active therapeutic molecules (Vilar et al., 2012; Stevanovic and Uskokovic, 2009). In addition, PLGA has positive biochemical and biophysical properties for enhanced drug delivery. These include high DNA cargo capacity (Balashanmugam et al., 2014), ease of formulation into different shapes and sizes (Makadia and Siegel, 2011), provision of prolonged and controlled release profile, and modifiable temperature and pH sensitive degradation rates (Vilar et al., 2012; Aravind et al., 2013).

9. Current research on aptamer-mediated polymeric formulations for targeted cancer therapy

Aptamer mediated-targeted delivery using tunable polymeric systems is undoubtedly a promising development to improve conventional pharmaceutical delivery. There are various research studies reporting drug-aptamer conjugated polymeric formulations with positive outcomes. Min et al. (2011) developed a dual PSMA aptamer conjugated with the anticancer drug, Dox, to target and treat both PSMA (+) and PSMA (−) highly expressed prostate tumor cells. The study demonstrated efficient delivery of Dox to targeted prostate tumor cells with enhanced selective cell uptake and successful induction of apoptosis in desired target cells. Guo et al. (2011) reported the therapeutic efficacy of nucleolin specific AS1411 DNA aptamer-functionalized PEG-PLGA formulation in targeted delivery of paclitaxel, a drug to treat gliomas and brain tumor cells, by targeting internalized cell surface receptors called nucleolin. Blood circulation and retention of paclitaxel at the targeted site was prolonged. As a result, the drug dosage administration and cytotoxicity on the targeted tumors was improved significantly with prolonged survival of experimental animals. Aravind et al., 2012 designed an aptamer-labeled paclitaxel-conjugated PLGA polymeric system with accurate tumor cell targeting, and it induced significant endocytosis and intracellular accumulation of the drug-conjugated polymeric particles to trigger apoptosis (Liechty et al., 2010). Another clinical research conducted by Aravind et al., 2012 demonstrated that drug-encapsulated AS1411 aptamer conjugated PLGA-lecithin-PEG polymeric particles are effective in improving specific cell targeting with greater tumor killing effects for chronic leukemia and MCF-7 breast cancer cells as well as sustained drug release at the target site

for enhanced therapeutic performance. A clinical study reported that targeted drug delivery using aptamer conjugated PEG-PLGA particles can offer a better therapeutic index as compared to non-targeted polymeric formulations. The study demonstrated a greater drug encapsulation efficiency using polymeric formulations and enhanced drug cellular uptake with stronger killing effects on human breast adenocarcinoma cells and minimal systemic cytotoxicity to surrounding normal cells using epithelial cell adhesion molecule (EpCAM) targeted RNA aptamer (Alibolandi et al., 2015). The work of Li et al. (2014b) further supported that EpCAM aptamer functionalized polymeric particles result in efficient targeted drug delivery. Their work showed that EpCAM aptamer-PLGA-lecithin-curcumin-PEG formulation can improve in vivo therapy significantly with enhanced drug bioavailability, increased binding to targeted colon tumor cells, greater cellular uptake as well as high cytotoxicity specifically to colorectal cancer cells.

PEG biopolymer is an FDA approved polymer for nasal, injectable and rectal formulations. It is one of the biopolymers of interest for pharmaceutical delivery due to its favorable characteristics such as enhancing the dissolution rate of partially soluble drugs (Dhar et al., 2008); hydrophilic sheltering proteins and peptides from the immune system; prolonging the circulation time of encapsulated molecules to reduce systemic removal (Guo et al., 2011; Li et al., 2014b); and enhancing the formulation stability to protect encapsulated bioactives from the physiological environment (Makadia and Siegel, 2011). Numerous studies have been reported on the use of PEI biopolymer as a powerful non-viral transfection agent to boost aptamer-mediated delivery for high transfection efficiency, improved release rate, and enhanced cargo capability. A study using mucin 1(MUC1) aptamer-labeled pDNA conjugated PEI complex in treating human lung tumor cells has demonstrated an efficient targeted gene delivery with improved gene expression, greater transfection efficiency and controlled release rate (Kurosaki et al., 2012). A recent study by Subramanian et al., 2015 also illustrated the use of PEI nanocomplex in targeted delivery of siRNA using EpCAM aptamer to selectively inhibit tumor cell proliferation (Subramanian et al., 2015). Chitosan is a modified natural polysaccharide, and has been widely used as a therapeutic delivery carrier. Sayari et al., 2014 have demonstrated the use of chitosan polymeric particles for specific delivery of the anticancer drug, SN38, to cure colon cancer using MUC1 DNA aptamer to increase the targeting ability and cellular uptake of targeted tumors (Sayari et al., 2014). Another research study also reported the use of chitosan particles in encapsulating S58 aptamer that targets human Tenon's capsule fibroblasts to treat myofibroblast trans-differentiation by antagonizing TGF-β receptor II. The results showed chitosan as a potential drug carrier with good encapsulation efficiency, low systemic cytotoxic effect, and sustained release profile (Chen et al., 2013). Poly(beta-amino ester) (PBAE) is a novel pH sensitive biopolymer, and has been utilized for endosomal delivery of various drugs and genes. For instance, Zhang and his team designed a dual-functional pH sensitive nucleolin-specific AS1411 aptamer conjugated D-α-tocopheryl polyethylene glycol 1000-*block*-PBAE copolymer (Apt-TPGS-*b*-PBAE) with improved synergistic effect of target recognition and pH-sensitive release of drug in the endosomal acidic environment. As a result, the retention of drug at the targeted site was prolonged with significant inhibition of tumor proliferation due to enhanced cytotoxicity as compared to individual drug therapies (Zhang et al., 2014). pH-responsive PBAE has also been conjugated with PLGA as a single formulation to deliver drugs in the research study of Vlerken et al. (2008). This study demonstrated an extended circulation time and a high drug accumulation at the target site to enhance breast cancer treatment.

10. Future outlook

The diversity of cancer cells and intra-tumor genetic heterogeneity continue to be the most significant challenge to the development of effective cancer therapies. It is improbable that a single therapy will be effective to completely destroy tumors cells in all cancer patients. More research efforts are essential to develop robust and tunable therapies capable of providing specific and tailored treatment to different cancer scenarios. This can be achieved by developing multimeric formulated aptamers with high specificity, binding affinity and sensitivity to boost the discovery of malignant cells as well as providing a sustained therapeutic effect. Multimodal tumor treatments that combine several therapies such as chemotherapy and gene therapy along with multiple agents including aptamers and OVs for specific therapeutic activity, will be a great promise with huge potency in killing tumor cells and increasing the survival rate of patients.

11. Conclusion

This article discusses the characteristics and significance of viral and aptamer-based targeted cancer therapies, alongside the research advancement guiding the choice of OVs and aptamers as targeting elements for cancer treatment. The use of targeting elements is a promising strategy for targeted cancer therapies to address the limitations of therapeutic agents such as low specificity towards targeted tumor cells; cytotoxicity to surrounding normal cells; poor pharmacokinetics; and insufficient effective therapeutic dosages at the targeted sites. Both OVs and aptamers have their unique advantages and limitations. OVs have the ability to kill cancer cells using mechanisms different from conventional chemotherapies to eradicate chemotherapy-resistant tumors cells and reduce cancer relapse for better survival rates. Aptamers are powerful targeting agents due to their high binding affinity and specificity towards their cognate targets as well as other advantageous such as low immunogenicity, low molecular weight, and chemical and thermal stability. Aptamers, however present minimal safety concerns post-transfection compared to OVs. Biopolymeric particles have demonstrated to improve the efficacy of drugs in relation to enhanced solubilization, bioavailability and biocompatibility as well as boosting the full potential of aptamers as targeting agents via sustained and controlled release profile. Both OVs and aptamers have demonstrated to be effective in targeted cancer therapies either as standalone or in combination with other targeting agents. However, more research efforts are required to minimize their limitations for longstanding cancer treatment with minimal side effects.

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