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Review Article

Replicating viruses for gynecologic cancer therapy

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Summary

Despite advanced therapeutic treatments, gynecologic malignancies such as cervical and ovarian cancers are still the top ten leading cause of cancer death among women in South Korea. Thus a novel and innovative approach is urgently needed. Naturally occurring viruses are live, replication-proficient viruses that specifically infect human cancer cells while sparing normal cell counterparts. Since the serendipitous discovery of the naturally oncotropic virus targeting gynecologic cancer in 1920s, various replicating viruses have shown various degrees of safety and efficacy in preclinical or clinical applications for gynecologic cancer therapy. Cellular oncogenes and tumor suppressor genes, which are frequently dysregulated in gynecologic malignancies, play an important role in determining viral oncotropism. Published articles describing replicating, oncolytic viruses for gynecologic cancers are thoroughly reviewed. This review outlines the discovery of replication-proficient virus strains for targeting gynecologic malignancies, recent progresses elucidating molecular connections between oncogene/tumor suppressor gene abnormalities and viral oncotropism, and the associated preclinical/clinical implications. The authors would also like to propose future directions in the utility of the replicating viruses for gynecologic cancer therapy.

Key words: Replicating virus; Oncolytic virus; Gynecologic cancer; Oncogenes; Tumor suppressor genes.

Introduction

From 2008 to 2012, about 35,000 new cases of gynecologic cancers, such as cervical, endometrial, and ovarian cancers, were diagnosed in South Korea [1]. The most common malignancy of the female genital tract is cervical cancer. Although cervical carcinoma can be reliably detected by both non-invasive methods and HPV typing even in non-invasive stages and incidence rate is slightly reduced recently, it remains the first most common type of malignant tumors among Koreans, and patients with recurrent or metastatic disease often have a poor prognosis and overall survival has not significantly improved [2]. In fact, many deaths also occur from ovarian cancer. Because of its vague symptoms, many cases are diagnosed at late Stages III or IV. Despite advanced surgical techniques and modern chemotherapy, the mortality rate has not changed over the last two decades [2]. Therefore, new innovative therapeutics with a high safety and efficacy are urgently needed.

Oncolytic viruses are live replication-proficient viruses that preferentially infect human cancer cells while sparing normal cell counterparts. Thus replicating oncolytic viruses are highly dependent on host cell physiology for inherent performance as viral cancer-targeting agents. Many naturally occurring viruses have shown a great potential as cancer targeting agents exploiting various oncogene signalling pathways that are established by host cancer cells during tumorigenesis [3-5]. Because carcinogenesis is a multi-step process involving accumulation of not only oncogene abnormalities but also tumor suppressor gene abnormalities, we have recently discovered that cellular tumor suppressor genes such as p53, ataxia telangiectasia mutated (ATM), and retinoblastoma associated (RB) are also important in determining oncolytic viral tropism including reovirus (a RNA virus) and poxvirus (a DNA virus) [6]. Thus, an important mechanism of viral oncolysis can be established by both cellular oncogene and tumor suppressor gene abnormalities.

Using replicating viruses in the treatment of gynecologic malignancies is not a recent idea. In 1912, a Italian clinician, De Pace, described a case history of one of his patients who experienced remission of her enormously large vegetating uterine cervical carcinoma following Pasteur's live attenuated rabies vaccine treatment. The rabies vaccine was developed by Louis Pasteur in order to eradicate rabies not for viral oncolysis. The patient was bitten by a rabid dog and was therefore not treated with the intention for a replicating viral oncolysis. De Pace attributed the regression of the cancer to oncolysis by the virus due to cancer cell death caused by viral cytolysis [7]. Since the use of rabies live vaccine virus, various repli-

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cating viruses have shown various degrees of safety and efficacy in pre-clinical or clinical application for gynecologic cancer therapeutics. The present authors would like review recent progresses in molecular studies as well as clinical aspects of replicating viral oncolysis against gynecologic malignancies.

Mechanism of the replicating viral oncolysis against gynecologic malignancies

Gynecologic malignancies comprise a series of genetic abnormalities involving oncogenes and tumor suppressor genes. Cervical cancer develops from precursor lesions referred to cervical intrepithelial neoplasia. It is now known that specific types of HPV are the principal etiologic agents for cervical cancer. Although the number of known genital HPV types now exceeds 170, only certain highly oncogenic types such as HPV 16 and HPV 18 are clinically important [8]. The highly oncogenic HPVs generate two viral oncoproteins (E6 and E7), which interact with host cellular tumor suppressor proteins (p53 and RB). These interactions between viral oncoproteins and cellular tumor suppressor proteins can lead to cell cycle progression and appear to be critical for the development of cervical cancer [9]. Since oncolytic viruses, such as reovirus and poxvirus, can target tumor suppressor abnormalities [6], these tumor suppressor defective cervical cancers can be a good target for replicating oncolytic viruses. Oncogenes such as c-Myc and Ras are also important for cervical cancer development [10, 11]. Since oncolytic viruses can target oncogene-driven cancers [3-5, 12], not only tumor suppressors but also oncogene-driven cervical cancers are also a good target for replicating oncolytic viruses.

Among gynecological malignancies, ovarian cancer carries the worst prognosis of all gynecological malignancies mainly due to lack of early detection and resistance to conventional therapy. Ovarian cancers may progress along two different pathways: type I cancers are highly differentiated ("low grade") and develop via typical precursor lesions, such as cystadenomas and borderline tumors. Although serous cancers represent the predominant histological subtype of the type I pathway, low-grade mucinous and endometrioid carcinomas, as well as malignant Brenner tumors are also included in this category (group). Type II cancers are poorly differentiated ("high grade") and develop rapidly without known or morphologically visible precursor lesions ("de novo development"). This group includes poorly differentiated serous, endometrioid, clear cell, and transitional cell carcinomas [13]. The morphologic differences of serous type I and type II tumors are reflected by molecular differences: type I tumors are genetically stable; two-thirds of them carry Ras pathway abnormalities. Most of them lack p53 mutations [14, 15]. Low-grade endometrioid cancers carry other mutations such as PTEN and PIK3CA. More than 50% of the low-grade mucinous tumors carry Ras mutations [16]. By contrast, serous high-grade cancers are genetically instable; more than 80 % carry p53 mutations [14, 15]. The majority of hereditary ovarian carcinomas are caused by BRCA1 and/or BRCA2 mutations thereby representing approximately 10% of all ovarian cancer cases [17]. Although ovarian cancers have heterogeneity in genetic abnormalities depending on tumor types, multi-mechanistic replicating viruses could effectively target them.

Since gynecologic malignancies underwent series of genetic abnormalities involving diverse classes of oncogenes and tumor suppressor genes and carcinogenesis is a multistep process involving accumulation both oncogene and tumor suppressor gene abnormality, it would be interesting whether replicating oncolytic viruses could exploit abnormal oncogene/cellular tumor suppressor signaling for their decision of oncolytic specificity and efficacy. Many tumor suppressor genes such as p53, ATM, and RB and oncogenes such as Ras and c-Myc are known to play important roles in genomic fidelity/maintenance. Thus tumor suppressor gene or oncogene abnormalities could affect host genomic integrity and likely disrupt intact antiviral networks due to accumulation of genetic defects resulting in replicating virus susceptibility. In 1998, mechanistic aspects of the oncolytic nature of a replicating virus is intensively studies by Lee's lab in University of Calgary. Strong *et al.* showed that virally resistant NIH 3T3 cells become susceptible to reovirus type 3 Dearing challenge when NIH 3T3 cells transformed with activated Sos or Ras. The Ras oncogene transformed cells showed a compromised antiviral PKR activity compared to untransformed cells [5]. Coffey *et al*. also showed that v-erbB-transformed NIH 3T3 xenografts are highly susceptible to reovirus type 3 Dearing challenge in tumor bearing mouse model [18]. When virally resistant cells are highly expressed by c-Myc oncogene introduction, reovirus type 3 Dearing could preferentially infect them and induce cell death [12, 19]. c-Myc proto-oncogene plays an important role in genomic maintenance and deregulated c-Myc expression generates genomic instability [20-22]. Oncogenes such as Ras and c-Myc were initially to be oncogenic solely because of their role in proliferative control. However, recent studies proposed that chromosome instability caused by combined dysfunctional effect of oncogenes and tumor suppressor genes may be more central to tumorigenesis than previously thought [23]. Thus, not only oncogenes but also tumor suppressor genes may play an important role in determining oncolytic nature of a replicating virus since genomic instability could compromise integrity of normal cellular anti-viral networks. In 2010, the present authors were able to show that p53, ATM, and RB tumor suppressor genes and their abnormal functions are important in determining reovirus oncolytic tropism. Abnormal functions of these tumor suppressors render cells to become susceptible upon replicating oncolytic viral challenges [6]. They proposed that abnormal-

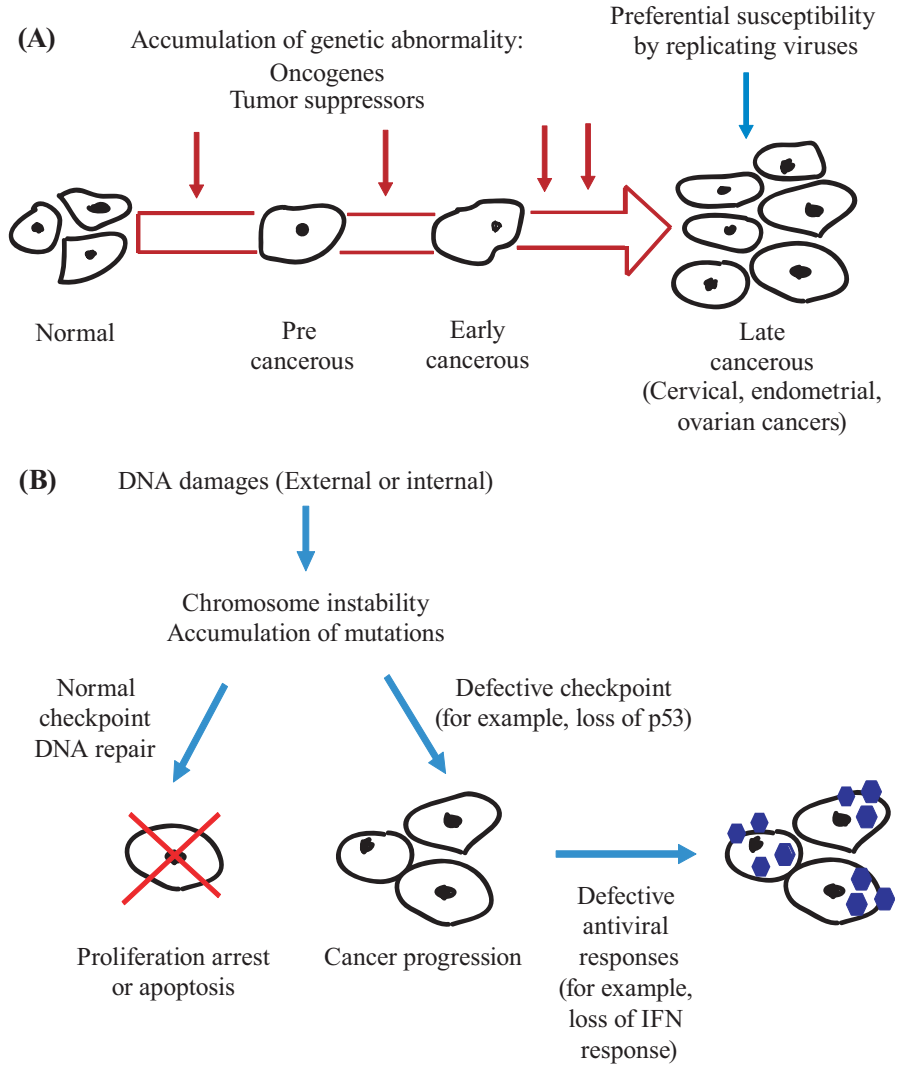

Figure 1. — Mechanistic basis of a replicating viral oncolysis against gynecologic malignancies. (A) Carcinogenesis is a multi-step process involving accumulation of cellular oncogenes and tumor suppressor gene abnormalities. (B) In the case of normal cells, DNA damages (externally or internally) could be repaired by normal checkpoint activities such as cell cycle arrest or apoptosis restricting viral propagation due to preservation of anti-viral integrity. However, tumor suppressor defective cancers could not handle genotoxic challenges, resulting in genomic instability and defective checkpoints/antiviral responses. Taken together, abnormal cells can be preferentially infected by replicating viruses due to loss of intact anti-viral functionality in the host cells.

ity of tumor suppressor genes should increase genomic instability, which would in turn result in the occurrence of new mutations including those affecting antiviral-related host genes. Furthermore, genomic instability appears to be the engine of both tumor progression and stagnation of the normal functions of antiviral-related cellular genes. Thus, mechanistically, replicating viral oncotropism can be established by both oncogenic hyper-activation and/or tumor suppressor abnormalities. Figure 1 shows mechanistic basis of replicating viral oncolysis in regards to cellular oncogenes and tumor suppressor genes.

Oncolytic strains of the replicating viruses for gynecologic cancer therapy

Rhabdoviridae

Using replicating viruses in the treatment of cancers including gynecologic malignancies is not a recent idea. A viral species from *Rhabdoviridae* is perhaps the firstly used oncolytic virus for human gynecological malignancy a century ago. Rabies virus, the prototypic member of the genus Lyssavirus, is an enveloped virus with a negative stranded RNA genome. Rabies is one of the oldest recognized infectious disease. Ancient civilizations were familiar with rabies. An early historical records mentioned the dangers of a rabid dog bites. Pasteur's research on rabies is perhaps the most well-known historical achievement in the vaccine field. Through adaptation of “street” (wild-type) virus to laboratory animals, he was able to change its properties used it as a live vaccine. The wild type virus is naturally attenuated during these passaging events. As mentioned above, in 1912, a Italian clinician, De Pace, reported that one of his patients who experienced remission of her enormously large vegetating uterine cervical carcinoma following Pasteur's live attenuated rabies vaccine treatment [7].

Vesicular stomatitis virus (VSV), the prototypic member of the genus Vesiculovirus, is an enveloped virus with a negative stranded RNA genome. The extensive body of knowledge about the replication of VSV reflects its status as a widely studied prototype for the non-segmented, negative-strand RNA viruses. VSV produces an acute disease in cattle, horses, and pigs characterized by fever and vesicles in the mucosa of the oral cavity and in the skin of the coronary band and teat. Clinically, VS (vesicular stomatitis) pathology is very similar to foot-and-mouth disease (FMB). VSV can also rarely cause an acute febrile disease in humans. Laboratory-adopted strains, however, are rarely pathogenic for humans. Although VS was first reported in the United States in 1916 during an epidemic in cattle and horses, a clinically similar disease was previously described in 1862. In 1925, cattle transported from Kansas City, Missouri, to Richmond, Indiana, initiated an outbreak of VS in the area. The disease was experimentally transmitted to horses and the infectious agent was maintained by serial passages in animals. The strain became the VS-Indiana strain virus [24]. Liu *et al.* reported that a genetically engineered VSV Indiana strain are highly effective in oncolysis of endometrial cancer cells *in vitro* and *in vivo* [25].

Paramyxoviridae

Paramyxomviruses contain non-segmented single-stranded RNA genomes of negative polarity and replicate entirely in the cytoplasm and they are defined by having a protein (F) that causes viral-cell membrane fusion [26]. Mumps virus, a member of the genus Rubulavirus, is the causative agent for mumps, a well-known common childhood disease characterized by swelling of the parotid glands, salivary glands, and other epithelial tissues. Although mumps virus is a human pathogen causing mumps, it is rarely fatal and mostly can be recovered by natural immunity. In 1974, Asada reported that a wild type mumps virus collected from the saliva of a patients with epidemic parotitis showed a significant anti-tumor efficacy against various cancers including gynecologic malignancies. Fourteen terminal cancer patients with uterine, ovarian, and vaginal cancers were administered with mumps virus solution locally and repeatedly, and nine patients showed a very good or good efficacy according to the author's criteria. Most patients well tolerated it and transient high fever was the common adverse effect during the viral therapy [27].

Measles virus, a member of the genus Mobillivirus, is the causative agent for measles. Measles is a highly contagious disease characterized by a prodromal illness of fever, coryza, cough, and conjunctivitis followed by the appearance of a generalized maculopapular rash. In 1954, the virus causing measles was isolated from an 11-year old boy from the United States, David Edmonston, and the virus was adapted and attenuated on chick cells [28, 29]. The Edmonston strain was widely used for eradication of measles worldwide [30]. Galanis *et al.*, reported that a genetically engineered Edmonston strain showed a significant efficacy against conventional therapy-resistant ovarian cancers in a clinical trial [31]. The virus was administrated to 16 patients with ovarian cancers intraperitoneally and repeatedly, and the median overall survival (OS) was 26.5 months, which was significant compared to other conventional therapy OS ranges of six to 12 months. Most patients well tolerated it and there was no significant adverse effects observed during the viral therapy [31].

Adenoviridae

Human adenoviruses belong to the genus of Mastadenovirus. Fifty-six human adenovirus types have been recognized and are classified into seven species (A-G) on the basis of serology and other criteria [32, 33]. Adenoviruses contain a linear, double-stranded DNA genome encapsidated in an icosahedral protein shell. Adenoviruses were firstly isolated and characterized in 1953 by two groups who were searching for the etiologic agents of acute respiratory infections [34, 35]. The two isolated viruses were related and named adenoviruses, after the original tissue (adenoids) in which they were discovered. In humans, besides respiratory disease, adenoviruses cause conjunctivitis and infantile gastroenteritis. In immunocompetent patients, adenoviruses usually cause a mild, self-limiting acute infection. However, in neonates and immunosuppressed patients, adenoviruses can cause fulminant fatal pneumonia, hepatitis, and/or encephalitis [36]. In 1956, Smith *et al.*, reported that wild type adenoviruses showed a significant anti-tumor efficacy against cervical carcinoma patents. Thirty terminal cancer patients with cervical carcinoma were administered with adenoviruses locally, and 11 patients showed a very good or good efficacy according to the author's criteria. [37]. Currently, instead of using wild type viruses, genetically modified versions of human adenoviruses were administered to ovarian and other gynecologic malignancies in clinical trials [38, 39].

Reoviridae

The *Reoviridae* is the largest and most diverse of dsRNA viruses. Mammalian reoviruses belong to the genus of Orthoreovirus. The viral particles display icosahedral symmetry with a diameter of 65 to 80 nm. Due to non-pathogenic nature in general, reoviruses are initially designated as orphan virus coined by Sabin [40]. Although reoviruses have been recovered mostly from children with a variety of mild illnesses such as common cold, diarrhea, etc [41-43], as well as from healthy persons [44], direct inoculation of reoviruses into human volunteers did not cause any significant human diseases. A few volunteers developed mild afebrile illness but these illness could not be definitely attributed to the viruses inoculated [45]. In 2008, phase I clinical trial of reovirus showed a mild illness in a few patients [46]. Taken together, direct inoculation of reovirus into human causes asymptomatic or subclinical illnesses.

Table 1. — *Oncolytic strains of the replicating viruses for gynecologic cancer therapy.*

Replicating viruses			Applications for gynecologic cancer therapy			Comments
Family	Genus/ Species	Backbone Strain	Genomic modification	Pre-clinical	Clinical	
Rhabdoviridae	Lyssavirus / Rabies	Rabies live vaccine (Pasteur's vaccine)	Wild type	ND	Cervical cancer	Wild type in clinical trial
			De Pace. 1912 [7]		De Pace. 1912 [7]	Representative literature
	Vesiculovirus/ Vesicular stomatitis	Indiana	Attenuated, transgene inserted	Endometrial cancer model	ND	
			Balachandran and Barber. 2008 [83]	Liu et al. 2014 [25]		Representative literature
Paramyxoviridae	Rubulavirus/ Mumps	Wild type isolated from a parotitis patient	Wild type	ND	Cervical, ovarian, and other cancers	Wild type in clinical trial
			Asada. 1974 [27]		Asada. 1974 [27]	Representative literature
	Morbillivirus/ Measles virus	Edmonston	Transgene inserted	Ovarian, endometrial cancer model	Ovarian cancer	MV-CEA, MV-NIS in clinical trial
			Peng et al. 2002 [84]; Dingli et al. 2004 [85]	Galanis et al. 2015 [31]; Liu et al. 2014 [25]	Galanis et al. 2015 [31]; Mader et al. 2013 [86]; Pol et al. 2014 [39]	Representative literature
Adenoviridae	Mastadenovirus/ Human adeonviruses B, C, etc	Human adenovirus type 1, 2, 3, 4, 5,6,7,10 group B, etc	Wild type viruses; Chimeric/attenuated	Colon cancer model	Cervical carcinoma; Ovarian cancer	Wild type strains; ONYX-015, ColoAd1, etc in clinical trial
			Smith et al. 1956 [37]; Kuhn et al. 2008 [87]	Kuhn et al., 2008 [87]	Smith et al. 1956 [37]; Vasey et al. 2002 [38]; Pol et al. 2014 [39]	Representative literature
Reoviridae	Orthoreovirus/ Reovirus type1, 2, 3	Reovirus type 3 Dearing, MH	Wild type, Attenuated	Ascites tumor bearing mouse model; Ovarian cancer model	Fallopian tube, ovarian, primary peritoneal carcinoma	Wild type (Reolysin) in clinical trial
			Coffey et al. 1998 [18]; Kim et al. 2010 [6]; Kim et al. 2011 [54]	Bennette. 1960 [47]; Bennette et al. 1967 [49]; Hirasawa et al. 2002 [56]	NCT01199263, NCT00602277	Representative literature/Trial numbers
Poxviridae	Orthopoxvirus/ Vaccinia (VACV)	Lister	Attenuated, transgenes inserted.	Ovarian cancer model	Peritoneal carcinomatosis	GLV-1h68 in clinical trial
			Chen et al. 2001 [91]; Zhang et al. 2007 [88]	Hung et al. 2007 [63]	NCT01443260	Representative literature/Trial number
	Unassigned / Squirrelpox (SQPV)	Kilham	Wild type	Various cancer model including cervical cancer	ND	
			Kim et al. 2014 [71,72]	Kim et al. 2014 [71,72]		Representative literature
Herpesviridae	Simplex virus/ type 1,2	HSV-1 JS-1,McIntyre, HF, HSV-2 HG52, 186	Attenuated, transgene inserted	Ovarian cancer model	ND	
			Chou et al. 1990 [76]; Sivendran et al. 2010[89]	Fu et al. 2007 [80]; Coukos et al. 2000[78]		Representative literature

Abbreviations: YLDV; Yaba-like disease virus, MYXV; Myxoma virus, SQPV; Squirrelpox virus, ND; not done yet.

In 1960s, Bennette isolated an ascite tumor destroying virus during passage of an ascites tumour. It was serially transmissible and seemingly non-pathogenic in host mouse in the absence of neoplastic cells. Filtrates of ascitic fluids and extracts of cells which harbored the unknown virus were highly destructive for the Ehrlich carcinoma and also other ascite tumours [47, 48]. The unknown oncolytic virus, isolated from a mouse ascites tumour, was later identified as a strain of reovirus type 3 MH (Middlesex Hospital) [49]. In 1977, Harshiro *et al.* studied oncolytic nature of reovirus and showed that reovirus type2 D5 Jones strain preferentially induces cytotoxicity against certain transformed cells compared to normal cell counterparts [50]. In 1978, Duncan *et al.* also showed that reovirus type 3 Dearing strain exerted differential sensitivity between normal and SV-40 transformed cells. SV-40 transformed WI-38 cells are highly susceptible to cytotoxic potential of reovirus type 3 Dearing strain challenge [51]. In 1998, Strong *et al.* showed Ras pathway abnormality is important in determining reovirus type 3 Dearing viral oncotropism [5]. In 2010, the present authors were able to show that tumor suppressor abnormities are also important in determining reovirus type 3 Dearing viral oncotropism [6].

Although the naturally occurring reoviruses, which harbor innate oncolytic potential, are generally asymptomatic and sometimes cause only mild symptoms in immuno-

competent humans, immunocompromised animal studies have shown considerable viral diseases such as oily hair effect, myocarditis, black feet syndrome, etc [49, 52, 53]. Because cancer patients can sometimes undergo a severe immunosuppression due to heavy chemo/radiation-treatments or advanced tumor progression, this pathogenic nature of reoviruses could be a hurdle in wild type reovirus-based anticancer therapeutic approaches. In 2007, the present authors isolated a genetically attenuated reovirus variant derived from persistently infected cells and this naturally attenuated reovirus was able to exert potent antitumor activity with a significantly reduced viral pathogenesis in immunocompromised animal models. Importantly, in this instance the attenuated reovirus maintains its oncolytic potential while significantly reducing reoviral pathogenesis in tumor xenograft experiments using SCID mice [53, 54] Furthermore, the attenuated reovirus does not affect murine embryonic stem cells' integrity suggesting that it could be used for pregnant cancer patients [54]. Although efficacy of the attenuated reovirus is not thoroughly compared with wild type reoviruses, it could be used in cases where wild type reoviruses could not be applied due to an unwanted side effect potential of particular cancer patients in a clinical setting. In 2002, Hirasawa *et al.* reported that reovirus type 3 Dearing strain is highly effective in suppressing ovarian tumors in a xenografted animal model [55]. Recently, reovirus type 3 Dearing strain was administrated to ovarian tumor patients in clinical trials as shown in Table 1.

Poxviridae

Poxviruses, which belong to *Poxviridae* family, are ubiquitous, enveloped viruses replicating entirely in the cytoplasm of vertebrate or invertebrate cells. Poxvirus particles (virions) can be externally enveloped (EEV), though the intracellular mature virion (IMV) form of the virus, which contains different envelope, is also infectious. They vary in their shape depending upon the species but are generally brick or oval shaped (similar to a rounded brick) wrapped by the endoplasmic reticulum. The virion is exceptionally large, its size is around 200 nm in diameter and 300 nm in length and carries its genome in a single, linear, double-stranded segment of a DNA molecule comprising 130 to 300 kilobase pairs [56].

Vaccinia virus is a member of the Orthopoxvirus genus of the *Poxviridae* and is the most intensively studied poxvirus. It is famous as the live vaccine virus that was used to eradicate smallpox caused by the variola virus, a feat completed in 1970s that remains the greatest triumph for the World Health Organization to date [57]. Yet despite the effectiveness of vaccinia virus in eradicating the smallpox, its origin and natural history are unknown and remain an enigma of virology [58, 59]. The live vaccinia Lister strain was developed at the Lister Institute in the United Kingdom. From 1968 to 1971, the Lister strain became the most widely used vaccine throughout the world [60]. Oncolytic nature of Lister strain was studies by several groups [61-63]. In 2007, Hung *et al.* showed that a modified version of Lister strain is effective in eradicating ovarian tumors in a xenografted animal model [63]. Currently, a modified version of Lister strain is in clinical trials against peritoneal carcinomatosis as shown in Table 1.

Squirrelpox virus (SQPV) Kilham strain, which belongs to an unassigned genus, isolated from a grey squirrel (*Sciurus carolinensis*) in Maryland, USA [64]. In grey squirrels, SQPV exerts a sub-clinical infection that rarely manifests in disease [65, 66]. However, red squirrel (*Sciurus vulgaris*) infection by SQPV causes ulceration with crusted lesions and scabs around the eyes, lips, feet and genitalia, and an exudative dermatitis which may be similar to myxomatosis pathogenesis caused by the myxoma virus infection in the European rabbit species (*Oryctolagus cuniculus*), and is almost always fatal [67, 68]. Red squirrels have been in decline in Great Britain for the last century due to a combination of habitat loss and the introduction of the North American eastern grey squirrel [69]. The dramatic decline of the native red squirrel in the UK has been attributed to both direct and disease-mediated competition with the grey squirrel, where the competitor acts as a reservoir host of SQPV [70]. SQPV natural infection in humans has not been reported since the decline of the red squirrel population in Great Britain. SQPV seems to have a very narrow and specific host range and exerts its viral pathogenic nature only in a certain squirrel species without affecting other non-squirrel species [70]. Taken together, the human pathogenicity of the SQPV Kilham oncolytic strain seems to be highly unlikely due to the poxviral host range restriction, although further human studies are warranted that include immunocompromised individuals. In 2014, Kim *et al.* showed that SQPV is highly effective in eradicating cervical cancer in a xenografted animal model [71, 72].

Herpesviridae

Herpesviruses contain a linear double-stranded DNA as their genome, an iscosahedral capsid, and an envelope containing viral glycoproteins. Herpes simplex virus-1 (HSV-1) and Herpes simplex virus-2 (HSV-2) belong to Simplexvirus genus and they are well known for causing latent, recurring infections of orolabial herpes and genital herpes. HSV-1 and HSV-2 are now used for replication proficient oncolytic viral therapy [73]. Although the lesions caused by HSV may have been described in the ancient literature millenia ago, a most enlightening description of recurrences of genital lesions was described by Unna in 1883. He wrote that herpes was "so to say a vocational disease", recognized as being "one of the most benign of affections both to the patient and her public" [74]. In 1924, Gruter reported that HSV could be transmitted serially from rabbit to rabbit and the virology com-

Table 2. — *Summary of gynecologic clinical trials with completed and ongoing using the replicating viruses alone or in combination with other therapies.*

Tumor types	Trial location	Phase	Backbone strain/ Highest doses (PFU or $TCID^{50}$)	Safety/ Efficacy	ClinicalTrials.gov identifier	Current status*	Ref
Cervical cancer	Italy	Pilot	Rabies vaccine/ NR	Well tolerated/ Some efficacy		Completed about 1912	De Pace. 1912 [7]
Cervical cancer	US	Pilot	Human adenoviruses/ NR	Well tolerated/ Some efficacy		Completed about 1956	Smith et al. 1956 [37]
Uterine, ovarian, vaginal	Japan	Pilot	Mumps, Wild type/ NR	Well tolerated/ Some efficacy		Completed about 1974	Asada.1974 [27]
Ovarian cancer	UK	1	Human adenovirus/ 10^{11}	Well tolerated/ limited efficacy		Completed about 2002	Vasey et al., 2002 [38]
Ovarian epithelial, primary peritoneal cancer	US	1	Measles, Edmonston/ 10^{9}		NCT00408590	Active, Not recruiting	Galanis et al. 2010 [90]; Galanis et al. 2015 [31]
Ovarian cancer	US	1/2	Measles, Edmonston/ 10^{9}		NCT02068794	Recruiting	Mader et al. 2013 [86]
Peritoneal carcinomatosis	Germany	1/2	Vaccinia, Lister/NR		NCT01443260	Recruiting	
Fallopian tube, ovarian, primary peritoneal carcinoma	US	2	Reovirus type 3 Dearing/10^{10}		NCT01199263	Active, not recruiting	
	US	1	Reovirus type 3 Dearing/10^{10}		NCT00602277	Active, not recruiting	
Ovarian cancer	UK	1/2	Human adenovirus chimeric/NR		NCT02028117	Recruiting	

* As of Jan 2015. Abbreviations: PFU; Plaque forming unit, $TCID^{50}$; 50% Tissue culture infective dose, NR; Not reported.

munity gives him a credit for the isolation of HSV [73, 75]. Importantly, Chou *et al.* reported that the neurovirulence of HSV-1 was mapped to the ICP34.5 gene and the pathogenicity of wild type virus can be significantly attenuated by targeting ICP34.5 gene [76, 77]. Thus oncolytic strain of HSV-1 is genetically engineered for safety and efficacy. In 2000, Coukos *et al.* reported that multi-attenuated HSV-1 mutant G207 exerts cytotoxicity against epithelial ovarian cancer but not normal mesothelium and is suitable for intraperitoneal oncolytic therapy [78]. Mechatabnistically, Farassati *et al.* reported that oncogenic Ras signalling is important in the determination of host-cell permissiveness to HSV-1 [79]. In 2007, Fu *et al.* showed that an oncolytic virus derived from HSV-2 has potent therapeutic effect against metastatic ovarian cancer [80]. Table 1 shows various oncolytic strains of the replicating viruses for gynecologic cancer therapy in preclinical or clinical stages.

Gynecologic clinical trials using the replicating viruses

Since 1910s, replicating viruses has been used for gynecologic cancer therapy. Early clinical studies clearly showed that various replicating viruses are highly effective in eradicating gynecological malignancies with well tolerable safety [7, 27, 37]. Remarkably, Asada reported that 14 terminal cancer patients with uterine, ovarian, and vaginal cancers were administered with wild type mumps virus solution locally and repeatedly, and nine patients showed a very good or good efficacy according to the author's criteria. Most patients well tolerated it and transient high fever was the common adverse effect during the viral therapy [27]. Smith *et al.* also reported that wild type adenoviruses showed a significant efficacy against cervical carcinoma patents. Thirty terminal cancer patients with cervical carcinoma were administered with adenoviruses locally, and 11 patients showed a very good or good efficacy according to the author's criteria [37].

From 2002 to 2014, various replicating viral trials are already conducted or currently in progress. Unlike early trials, the genetically modified version of replicating viruses (except reovirus) are utilized in clinical trials as shown in Table 2. Remarkably, Galanis *et al.* reported that a genetically engineered Edmonston strain showed a significant efficacy against conventional therapy-resistant ovarian cancers in a clinical trial. The virus was administrated to 16 patients with ovarian cancers intraperitoneally and repeatedly, and the median OS was 26.5 months, which was significant compared to other conventional therapy OS ranges of six to 12 months. Most patients well tolerated it and there was no significant adverse effects observed during the viral therapy [31]. Currently, wild type reovirus type 3 Dearing strain, a chimeric form of adenovirus, a genetically modified vaccinia Lister strain, and an engineered measles Edmonston strain are in clinical trials and safety and efficacy of these viruses could be reported in the near future. Table 2 shows summary of gynecologic clinical trials with completed and ongoing using replicating viruses alone or in combination with other therapies.

Future directions

Since the serendipitous use of rabies live vaccine virus in 1912, oncolytic nature of various replicating viruses capable of targeting gynecologic malignancies has been identified. During the past 20 years of molecular research, the involvement of cellular oncogenes and tumor suppressor genes in determining replicating viral oncotropism is now fairly well established. Although replicating viruses are extremely potent in eradicating gynecologic tumors in human, virally resistant tumors can occur in some cases [27, 31]. Thus it would be important to improve the viral efficacy for overcoming viral resistance. Importantly, the present authors were able to show that reovirus-resistant tumors can be eradicated by introducing other oncolytic viruses such as wild type adenovirus [53]. This could be due to the fact that each virus has a unique strategy and low-cross reactivity in replicating inside tumor cells. Thus it would be possible to develop an usage of the replicating oncolytic viruses in a sequential or combinatorial manner to overcome virally resistant tumors. Since combinatorial oncolytic viral regimen using the same or different family of viruses could synergistically enhance the oncolytic viral potency in animal models [53, 81, 82], an optimal combination of replicating DNA or RNA viruses could be clinically applied for an enhanced targeting of the gynecological malignancies in the near future.

Unlike other types of tumors, gynecologic malignancies could occur during pregnancy and immediate treatment should be necessary in some situations. Thus embryonic stem cell sparing oncolytic virus could be useful to target gynecologic pregnant cancer patients. Importantly, the present authors were able to show that the naturally attenuated reovirus could exert oncolytic potency while preserving embryonic stem cells' integrity *in vitro* and *in vivo* [54]. Not only the attenuated reovirus, it is possible that other family of replicating viruses could be identified and utilized for pregnant cancer patients in the future.

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References

[1] Daejeon Statistics Korea: "Korean Statistical Information Service", 2012. Available at: http://kosis.kr.

[2] Fuh K.C., Shin J.Y., Kapp D.S., Brooks R.A., Ueda S., Urban R.R., *et al.*: "Survival differences of Asian and Caucasian epithelial ovarian cancer patients in the United States". *Gynecol. Oncol*., 2014, *16*, pii: S0090-8258(14)01369.

[3] Roberts M.S., Lorence R.M., Groene W.S., Bamat M.K.: "Naturally oncolytic viruses". *Curr. Opin. Mol. Ther.,* 2006, *8*, 314.

[4] Wang G., Barrett J.W., Stanford M., Werden S.J., Johnston J.B., Gao X., *et al.*: "Infection of human cancer cells with myxoma virus requires Akt activation via interaction with a viral ankyrin-repeat host range factor". *Proc. Natl. Acad. Sci. U S A*, 2006, *103*, 4640.

[5] Strong J.E., Coffey M.C., Tang D., Sabinin P., Lee P.W.: "The molecular basis of viral oncolysis: usurpation of the Ras signaling pathway by reovirus". *EMBO J.,* 1998, *17*, 3351.

[6] Kim M., Williamson C.T., Prudhomme J., Bebb D.G., Riabowol K., Lee P.W., *et al.*: "The viral tropism of two distinct oncolytic viruses, reovirus and myxoma virus, is modulated by cellular tumor suppressor gene status". *Oncogene,* 2010, *29,* 3990.

[7] De Pace N.G.: "Sulla scomparsa di un enorme cancro vegetante del collo dell'utero senza cura chirurgica". *Ginecologia*, 1912, *9*, 82.

[8] Bzhalava D., Guan P., Franceschi S., Dillner J., Clifford G.: "A systematic review of the prevalence of mucosal and cutaneous human papillomavirus types". *Virology,* 2013, *445*, 224.

[9] Whang J.D., Lee J.H.: "Molecular genetics of gynecologic cancer". *J. Korean Med. Sci.*, 1997, *12*, 383.

[10] Bourhis J., Le M.G., Barrois M., Gerbaulet A., Jeannel D., Duvillard P., *et al.*: "Prognostic value of c-myc proto-oncogene overexpression in early invasive carcinoma of the cervix". *J. Clin. Oncol.,* 1990, *8*, 1789.

[11] Mammas I.N., Zafiropoulos A., Spandidos D.A.: "Involvement of the ras genes in female genital tract cancer". *Int. J. Oncol*., 2005, *26*, 1241.

[12] Egan C., Kim M., Johnston R.N.: "Increased expression of human c-Myc renders cells sensitive to reovirus oncolysis". AACR Annual General Meeting, Washington, DC, July 2003.

[13] Shih Ie M., Kurman R.J.: "Ovarian tumorigenesis: a proposed model based on morphological and molecular genetic analysis". *Am. J. Pathol*., 2004, *164,* 1511.

[14] Bauerschlag D.O., Schem C., Weigel M.T., Von Kaisenberg C., Strauss A., Bauknecht T., *et al.*: "The role of p53 as a surrogate marker for chemotherapeutical responsiveness in ovarian cancer". *J. Cancer Res. Clin. Oncol*., 2009, *136*, 79.

[15] Hauptmann S., Denkert C., Koch I., Petersen S., Schlüns K., Reles A., *et al.*: "Genetic alterations in epithelial ovarian tumors analyzed by comparative genomic hybridization". *Hum. Pathol.,* 2002, *33*, 632.

[16] Shih I.M., Kurman R.J.: "Molecular pathogenesis of ovarian borderline tumors: new insights and old challenges". *Clin. Cancer Res*., 2005, *11*, 7273.

[17] Meinhold-Heerlein I., Hauptmann S.: "The heterogeneity of ovarian cancer". *Arch. Gynecol. Obstet*., 2014, *289,* 237.

[18] Coffey M.C., Strong J.E., Forsyth P.A., Lee P.W.: "Reovirus therapy of tumors with activated Ras pathway". *Science*, 1998, *282,* 1332.

[19] Kim M., Chung Y.H., Johnston R.N.: "Reovirus and tumor oncolysis". *J Microbiol*. 2007, *45*, 187.

[20] Kuttler F., Mai S.: "c-Myc, Genomic Instability and Disease". *Genome Dyn*., 2006, *1*, 171.

[21] Prochownik E.V., Li Y.: "The ever expanding role for c-Myc in promoting genomic instability". *Cell Cycle*, 2007, *6*, 1024.

[22] Prochownik E.V.: "c-Myc: linking transformation and genomic instability". *Curr. Mol. Med*., 2008, *8*, 446.

[23] Coschi C.H., Dick F.A.: "Chromosome instability and deregulated proliferation: an unavoidable duo". *Cell Mol. Life Sci*., 2012, *69*, 2009.

[24] Douglas S.L., Ivan V.K., Charles E.R.: "Rhabdoviridae." . *In:* Knipe D.M., Howley P.M. (eds). *Fields Virology*, 6th ed. Philadelphia: Lippincott- Raven, 2013, 1298.

[25] Liu Y.P., Steele M.B., Suksanpaisan L., Federspiel M.J., Russell S.J., Peng K.W., *et al.*: "Oncolytic measles and vesicular stomatitis virotherapy for endometrial cancer". *Gynecol. Oncol.,* 2014, *132*, 194.

[26] Robert A.L., Griffith D.P.: "Paramyxoviridae". *In:* Knipe D.M., Howley P.M. (eds). *Fields Virology*, 6th ed. Philadelphia: Lippincott-Raven, 2013, 1407.

[27] Asada, T.: "Treatment of human cancer with mumps virus". *Cancer*, 1974, *34*, 1907.

[28] Enders J.F., Peebles T.C.: "Propagation in tissue cultures of cytopathogenic agents from patients with measles". *Proc. Soc. Exp. Biol. Med*., 1954, *86*, 277.
[29] Enders J.F.: "Measles virus. Historical review, isolation, and behavior in various systems". *Am. J. Dis. Child*., 1962, *103*, 282.
[30] Hornick R.B., Schluederberg A.E., Mccrumb F.R. Jr.: "Vaccination with live attenuated measles virus". *Am. J. Dis. Child*., 1962, *103*, 344.
[31] Galanis E., Atherton P.J., Maurer M.J., Knutson K.L., Dowdy S.C., Cliby W.A., *et al.*: "Oncolytic measles virus expressing the sodium iodide symporter to treat drug-resistant ovarian cancer". *Cancer Res*., 2015, *75*, 22.
[32] Jones M.S. 2nd, Harrach B., Ganac R.D., Gozum M.M., Dela Cruz W.P., Riedel B., *et al.*: "New adenovirus species found in a patient presenting with gastroenteritis". *J. Virol*., 2007, *81*, 5978.
[33] Robinson C.M., Singh G., Henquell C., Walsh M.P., Peigue-Lafeuille H., Seto D., *et al.*: "Computational analysis and identification of an emergent human adenovirus pathogen implicated in a respiratory fatality". *Virology*, 2011, *409*, 141.
[34] Rowe W.P., Huebner R.J., Gilmore L.K., Parrott R.H., Ward T.G.: "Isolation of a cytopathogenic agent from human adenoids undergoing spontaneous degeneration in tissue culture". *Proc. Soc. Exp. Biol. Med*., 1953, *84*, 570.
[35] Hilleman M.R., Werner J.H.: "Recovery of new agent from patients with acute respiratory illness". *Proc. Soc. Exp. Biol. Med*., 1954, *85*, 183.
[36] Arnold J.B.: "Adenoviridae". *In:* Knipe D.M., Howley P.M. (eds). *Fields Virology*, 6th ed. Philadelphia: Lippincott- Raven, 2013, 2613.
[37] Smith R.R., Huebner R.J., Rowe W.P., Schatten W.E., Thomas L.B.: "Studies on the use of viruses in the treatment of carcinoma of the cervix". *Cancer*, 1956, *9*, 1211.
[38] Vasey P.A., Shulman L.N., Campos S., Davis J., Gore M., Johnston S., *et al.*: "Phase I trial of intraperitoneal injection of the E1B-55-kd-gene-deleted adenovirus ONYX-015 (dl1520) given on days 1 through 5 every 3 weeks in patients with recurrent/refractory epithelial ovarian cancer". *J. Clin. Oncol*., 2002, *20*, 1562.
[39] Pol J., Bloy N., Obrist F., Eggermont A., Galon J., Cremer I., *et al.*: "Trial watch: oncolytic viruses for cancer therapy". *Oncoimmunology,* 2014, *3*, e28694.
[40] Sabin A.B.: "Reoviruses. A new group of respiratory and enteric viruses formerly classified as ECHO type 10 is described". *Science*, 1959, *130*, 1387.
[41] Stanley N.F., Dorman D.C., Ponsford J.: "Studies on the pathogenesis of a hitherto undescribed virus (hepato-encephalomyelitis) producing unusual symptoms in suckling mice". *Aust. J. Exp. Biol. Med. Sci*., 1953, *31*, 147.
[42] Ramos-Alvarez M., Sabin A.B.: "Characteristics of poliomyelitis and other enteric viruses recovered in tissue culture from healthy American children". *Proc. Soc. Exp. Biol. Med*., 1954, *87*, 655.
[43] Jackson G.G., Muldoon B.L., Cooper G.S.: "Reovirus type 1 as an etiologic agent of the common cold". *J. Clin. Invest*., 1961, *40*, 1051.
[44] Ramos-Alvarez M., Sabin A.B.: "Intestinal viral flora of healthy children demonstrable by monkey kidney tissue culture". *Am. J. Public Health Nations Health,* 1956, *46*, 295.
[45] Rosen L., Evans H.E., Spickard A.: "Reovirus infections in human volunteers". *Am. J. Hyg.,* 1963, *77*, 29.
[47] Bennette J.G.: "Isolation of a Non-pathogenic Tumour-destroying Virus from Mouse Ascites". *Nature*, 1960, *187*, 72.
[48] Nelson J.B., Tarnowsk G.S.: "An oncolytic virus recovered from Swiss mice during passage of an ascites tumour". *Nature.* 1960, *188,* 866.
[49] Bennette J.G., Bush P.V., Steele R.D.: "Characteristics of a newborn runt disease induced by neonatal infection with an oncolytic strain of reovirus type 3 (REO3MH). I. Pathological investigations in rats and mice". *Br. J. Exp. Pathol*., 1967, *48*, 251.
[50] Hashiro G., Loh P.C., Yau J.T.: "The preferential cytotoxicity of reovirus for certain transformed cell lines". *Arch. Virol*., 1977, *54*, 307.
[51] Duncan M.R., Stanish S.M., Cox D.C.: "Differential sensitivity of normal and transformed human cells to reovirus infection". *J. Virol*., 1978, *28*, 444.
[52] Loken S.D., Norman K., Hirasawa K., Nodwell M., Lester W.M., Demetrick D.J.: "Morbidity in immunosuppressed (SCID/NOD) mice treated with reovirus (dearing 3) as an anticancer biotherapeutics". *Cancer Biol. Ther*., 2004, *3*, 734.
[53] Kim M., Egan C., Alain T., Urbanski S.J., Lee P.W., Forsyth P.A., *et al.*: "Acquired resistance to reoviral oncolysis in Ras-transformed fibrosarcoma cells". *Oncogene*, 2007, *26*, 4124.
[54] Kim M., Garant K.A., zur Nieden N.I., Alain T., Loken S.D., Urbanski S.J., *et al.*: "Attenuated reovirus displays oncolysis with reduced host toxicity". *Br. J. Cancer*, 2011, *104*, 290.
[55] Hirasawa K., Nishikawa S.G., Norman K.L., Alain T., Kossakowska A., Lee P.W.: "Oncolytic reovirus against ovarian and colon cancer". *Cancer Res*., 2002, *62*, 1696.
[56] Moss B.: "Poxviridae". *In:* Fields B.N., Knipe D.M., and Howley P.M. (eds). *Fields Virology*, 6th ed. Philadelphia: Lippincott-Raven, 3292.
[57] Ellner P.D.: "Smallpox: gone but not forgotten". *Infection,* 1998, *26*, 263.
[58] Baxby D.: "The origins of vaccinia virus". *J. Infect. Dis*., 1997, *136*, 453.
[59] Wilkinson L.: "Jenner's smallpox vaccine. The riddle of vaccinia virus and its origin". *Med. Hist*., 1982, *26*, 94.
[60] Rosenthal S.R., Merchlinsky M., Kleppinger C., Goldenthal K.L.: "Developing new smallpox vaccines". *Emerg. Infect. Dis*., 2001, *7*, 920.
[61] Timiryasova T.M., Li J., Chen B., Chong D., Langridge W.H., Gridley D.S., *et al.*: "Antitumor effect of vaccinia virus in glioma model". *Oncol. Res*., 1999, *11*, 133.
[62] Chen B., Timiryasova T.M., Haghighat P., Andres M.L., Kajioka E.H., Dutta-Roy R., *et al.*: "Low-dose vaccinia virus-mediated cytokine gene therapy of glioma". *J. Immunother*., 2001, *24*, 46.
[63] Hung C.F., Tsai Y.C., He L., Coukos G., Fodor I., Qin L., *et al.*: "Vaccinia virus preferentially infects and controls human and murine ovarian tumors in mice". *Gene Ther*., 2007, *14*, 20.
[64] Kilham L., Herman C.M., Fisher E.R.: "Naturally occurring fibromas of grey squirrels related to Shope's rabbit fibroma". *Proc. Soc. Exp. Biol. Med*., 1953, *82*, 298.
[65] Sainsbury A.W., Nettleton P., Gilray J., Gurnell J.: "Grey squirrels have high seroprevalence to a parapoxvirus associated with deaths in red squirrels". *Anim. Conserv*., 2000, *3*, 229.
[66] Tompkins D.M., Sainsbury A.W., Nettleton P., Buxton D., Gurnell J.: "Parapoxvirus causes a deleterious disease in red squirrels associated with UK population declines". *Proc. Biol. Sci*., 2002, *269*, 529.
[67] Duff J.P., Scott A., Keymer I.F.: "Parapox virus infection of the grey squirrel". *Vet. Rec*., 1996, *138,* 527.
[68] Carroll B., Russell P., Gurnell J., Nettleton P., Sainsbury A.W.: "Epidemics of squirrel pox virus disease in red squirrels (Sciurus vulgaris): temporal and serological findings". *Epidemiol. Infect*., 2009, *137*, 257.
[69] Gurnell J.: "The grey squirrel in Britain: problems for management and lessons for Europe". *In:* Mathias M.L., Santos-Reis M., Amori G., Libois R., Mitchell-Jones A., *et al.* (eds). *European Mammals: Proceedings of the European Congress of Mammalogy.* Lisbon: Museu Bocage, 1996, 67.
[70] Collins L.M., Warnock N.D., Tosh D.G., McInnes C., Everest D., *et al.*: "Squirrelpox virus: assessing prevalence, transmission and environmental degradation". *PLoS One*, 2014, *9*, e89521.
[71] Kim M., Ahn J.S., Yun C.O., Kim B.Y.: "Squirrel poxvirus as a novel oncolytic agent". The 40th annual meeting of Korean Cancer Association. Seoul Korea, June 20, 2014.
[72] Kim M., Ahn J.S., Yun C.O., Kim B.Y.: "Therapeutic or preventive pharmacological composition of squirrelpox virus for anticancer therapeutics" (Translated in Korean). Korean patent No.: 10-1370620 Date of Patent grant: Feb 27, 2014.

[73] Roizman B., Knipe D.M., Whitley R.J.. "Herpes Simplex viruses". *In:* Knipe D.M., Howley P.M. (eds), *Fields Virology*, 6th ed. Philadelphia: Lippincott- Raven, 2013, 2803.
[74] Unna P.G.: "On herpes progenitalis, especially in women". *J. Cutan. Vener. Dis*., 1883, *1*, 321.
[75] Gruter W.: "Das herpesvirus, seine atiologische und klinische bedeutung". *Munch. Med. Wschr.,* 1924, *71*, 1058.
[76] Chou J., Kern E.R., Whitley R.J., Roizman B.: "Mapping of herpes simplex virus-1 neurovirulence to gamma 134.5, a gene nonessential for growth in culture". *Science*, 1990, *250*, 1262.
[77] Bolovan C.A., Sawtell N.M., Thompson R.L.: "ICP34.5 mutants of herpes simplex virus type 1 strain 17syn1 are attenuated for neurovirulence in mice and for replication in confluent primary mouse embryo cell cultures". *J. Virol*., 1994, *68*, 48.
[78] Coukos G., Makrigiannakis A., Kang E.H., Rubin S.C,. Albelda S.M., Molnar-Kimber K.L.: "Oncolytic herpes simplex virus-1 lacking ICP34.5 induces p53-independent death and is efficacious against chemotherapy-resistant ovarian cancer". *Clin. Cancer Res*., 2000, *6*, 3342.
[79] Farassati F., Yang A.D., Lee P.W.: "Oncogenes in Ras signalling pathway dictate host-cell permissiveness to herpes simplex virus 1". *Nat. Cell. Biol*., 2001, *3*, 745.
[80] Fu X., Tao L., Zhang X.: "An oncolytic virus derived from type 2 herpes simplex virus has potent therapeutic effect against metastatic ovarian cancer". *Cancer Gene Ther*., 2007, *14*, 480.
[81] Le Boeuf F., Diallo J.S., McCart J.A., Thorne S., Falls T., Stanford M., *et al.*: "Synergistic interaction between oncolytic viruses augments tumor killing". *Mol. Ther*., 2010, *18*, 888.
[82] Alkassar M., Gärtner B., Roemer K., Graesser F., Rommelaere J., Kaestner L., *et al.*: "The combined effects of oncolytic reovirus plus Newcastle disease virus and reovirus plus parvovirus on U87 and U373 cells in vitro and in vivo". *J. Neurooncol*., 2011, *104*, 715.
[83] Balachandran S., Barber G.N.: "Vesicular stomatitis virus (VSV) therapy of tumors". *IUBMB Life*, 2000, *50*, 135.
[84] Peng K.W., Facteau S., Wegman T., O'Kane D., Russell S.J.: "Non-invasive in vivo monitoring of trackable viruses expressing soluble marker peptides". *Nat. Med*., 2002, *8*, 527.
[85] Dingli D., Peng K.W., Harvey M.E., Greipp P.R., O'Connor M.K., Cattaneo R., *et al.*: "Image-guided radiovirotherapy for multiple myeloma using a recombinant measles virus expressing the thyroidal sodium iodide symporter". *Blood*, 2004, *103*, 1641.
[86] Mader E.K., Butler G., Dowdy S.C., Mariani A., Knutson K.L., Federspiel M.J., *et al.*: "Optimizing patient derived mesenchymal stem cells as virus carriers for a phase I clinical trial in ovarian cancer". *J. Transl. Med*., 2013, *11*, 20.
[87] Kuhn I., Harden P., Bauzon M., Chartier C., Nye J., Thorne S., *et al.*: "Directed evolution generates a novel oncolytic virus for the treatment of colon cancer". *PLoS One*, 2008, *3*, e2409.
[88] Zhang Q., Yu Y.A., Wang E., Chen N., Danner R.L., Munson P.J., *et al.*: "Eradication of solid human breast tumors in nude mice with an intravenously injected light-emitting oncolytic vaccinia virus". *Cancer Res*., 2007, *67*, 10038.
[89] Sivendran S., Pan M., Kaufman H.L., Saenger Y.: "Herpes simplex virus oncolytic vaccine therapy in melanoma". *Expert Opin. Biol. Ther*., 2010, *10*, 1145.
[90] Galanis E., Hartmann L.C., Cliby W.A., Long H.J., Peethambaram P.P., Barrette B.A., *et al.*: "Phase I trial of intraperitoneal administration of an oncolytic measles virus strain engineered to express carcinoembryonic antigen for recurrent ovarian cancer". *Cancer Res*., 2010, *70*, 875.
[91] Chen B., Timiryasova T.M., Haghighat P., Andres M.L., Kajioka E.H., Dutta-Roy R., *et al.*: "Low-dose vaccinia virus-mediated cytokine gene therapy of glioma". *J. Immunother*. 2001, *24*, 46.

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