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Progress of non-human primate animal models of cancers

XIA Hou-Jun, CHEN Ce-Shi*

(*Key Laboratory of Animal Models and Human Disease Mechanisms of the Chinese Academy of Sciences & Yunnan Province, Kunming Institute of Zoology, the Chinese Academy of Sciences, Kunming Yunnan* 650223, *China*)

Abstract: Cancer is the second leading disease causing human death. Pre-clinical *in vivo* studies are essential for translating *in vitro* laboratory research results into the clinic. Rodents, including the mouse and rat, have been widely used for pre-clinical studies due to their small size, clear genetic backgrounds, rapid propagation, and mature transgenic technologies. However, because rodents are evolutionarily distinct from humans, many pre-clinical research results using rodent models cannot be reproduced in the clinic. Non-human primates (NHPs) may be better animal models than rodents for human cancer research because NHPs and humans share greater similarity in regards to their genetic evolution, immune system, physiology and metabolism. This article reviews the latest progress of cancer research in NHPs by focusing on the carcinogenesis of different NHPs induced by chemical and biological carcinogens. Finally, future research directions for the use of NHPs in cancer research are discussed.

Key words: Non-human primate; Cancer; Tree shrew

非人灵长类肿瘤模型研究进展

夏厚军, 陈策实*

(中国科学院和云南省动物模型与人类疾病机理重点实验室, 中国科学院昆明动物研究所, 云南 昆明 650223)

摘要：癌症是人类第二大致死的疾病。将体外细胞模型获得的癌症研究结果向临床转化过程中, 动物活体实验是必不可少的一个环节。现在的肿瘤活体实验绝大部分采用啮齿类实验动物如小鼠和大鼠, 这是因为它们具有个体小、繁殖迅速、遗传背景清楚、转基因技术成熟等优势。但是啮齿类和人的亲缘关系比较远, 许多从啮齿类动物模型获得的研究结果不能在人体重现。非人灵长类动物在遗传进化、免疫、生理和代谢等诸多方面与人类高度近似, 理论上更加适合癌症研究。本文对现有的非人灵长类肿瘤研究做一综述, 主要集中介绍用化学和生物致癌剂在不同的非人灵长类动物诱导肿瘤的研究, 为将来用非人灵长类动物研究人类癌症奠定基础。

关键词：非人灵长类; 肿瘤; 树鼩
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Rodents are popular animal models for human cancer research because of their small size, rapid propagation, low cost, the availability of a variety of inbred strains, and transgenic techniques. However, most anti-tumor drugs that cure cancers in rodent models failed in clinical trials because of a lack of efficacy or the presence of toxicity. It is estimated that only 3.8% of patients in phase-I cancer drug trials between 1991 and 2002 achieved an objective clinical response although all drugs worked well against cancer in the mouse (Roberts et al, 2004). This deficiency can be largely attributed to species differences between rodents and humans. Therefore, better animal models are required to increase the success rate for the discovery of anti-cancer drugs and treatments.

Compared to rodents, non-human primates (NHPs) are more similar to humans in regards to their genetic evolution, anatomy, physiology, biochemistry and organ

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* Corresponding author (通信作者), Email: chenc@mail.kiz.ac.cn
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systems. It has been shown that almost all human cancer genes are highly conserved between chimpanzees and humans (Puente et al, 2006). However, the use of NHPs in cancer research is limited because they are rare, expensive, and of a large size. In addition, cancer incidence in NHPs is very low. In this article, we summarize results on spontaneous and experimentally induced tumors in NHPs and discuss future study directions for using NHPs in cancer research.

1 Spontaneous tumors in NHPs

Spontaneous tumors have been observed in NHPs bred in zoos and research institutions all over the world. By 1989, a total of 783 spontaneous tumors in monkeys and apes had been reported (Beniashvili, 1989). From 1989 to 2000, 1 388 cases of benign and malignant tumors (582/806) in NHPs were found (Huang & Cheng, 2002). Most tumors were found in baboons (Cianciolo & Hubbard, 2005) and macaques because these two species were more commonly used for research than other NHP species (Beniashvili, 1989).

The incidence of spontaneous tumors in NHPs depends on the animal species, sex and age. Autopsy records of 373 breeders and normal controls from the National Cancer Institute (NCI), USA showed a very low incidence of spontaneous malignant tumors in cynomolgus (1.5%) and rhesus (2.8%) monkeys, but a considerably higher incidence in African green monkeys (8%) (Thorgeirsson et al, 1994). Most types of spontaneous tumors occurred equally in male and female NHPs. However, spontaneous tumors of the reproductive system were primarily observed in older females and rarely observed in males (Huang & Cheng, 2002). The incidence of spontaneous tumors was approximately 5% in all age groups in NHPs but increased in frequency with age, similar as it does in humans (Beniashvili, 1989; Bunton & Bacmeister, 1989; Yamate et al, 2007; Starost, 2009). For example, the incidence of colon cancer in rhesus monkeys increased with age (Uno et al, 1998). It is worth pointing out that some tumors were observed in infant and young animals (Jayo et al, 1988; Pellegrini et al, 2009).

The spontaneous tumor spectrum in NHPs is very similar to that in humans. For example, nasopharyngeal carcinomas, hepatocellular carcinomas (HCC), renal carcinomas, melanoma, pulmonary neoplasms, and other tumors were reported in chimpanzees (Bailey, 2009). Tumors of the digestive system, reproductive system, and urinary system were more common than those of other organ systems (Beniashvili, 1989). It is clear that while tumors of the respiratory system are rare in NHPs, they are common in humans due to air pollution and smoking (Coggins, 2007). Although the tumor epidemiology of NHPs reflects the natural carcinogenesis, there are some limitations for studying spontaneous tumors. Firstly, only individual cases were described in many studies. Secondly, most tumor cases were found accidentally during an autopsy rather than in live animals. Thus, spontaneous tumors of NHPs provide us with clues for establishing cancer animal models (Huang & Cheng, 2002).

2 Experimental tumors in NHPs

Due to the low incidence of spontaneous tumors, experimental tumor models of NHPs are necessary for human cancer research. It is well known that tumors are initiated by environmental and genetic factors. Approximately 85% of malignant neoplasms are related to chemical, biological and physical environmental factors and chemical, biological and physical carcinogens have been used to induce tumors in NHPs. Several tumor models were established by utilizing combinations of different carcinogens. NHPs can be divided into three groups: anthropoids, prosimians and tree shrews. Carcinogenesis studies have been performed on different species of NHPs from these groups including monkeys (Pfeiffer & Allen, 1948) and tree shrews (Adamson et al, 1970).

2.1 Carcinogenesis studies in anthropoids

Anthropoids, particularly monkeys, have been used to develop cancer models. Monkeys primarily consist of old world monkeys and new world monkeys. Old world monkeys, such as baboons and macaques, inhabit Asia and Africa. New world monkeys are distributed in tropical forests of southern Mexico and Central and South America. Most of them are fairly small in size and are exclusive tree dwellers.

2.1.1 Chemical carcinogens induce different types of cancers

Many different types of human cancers are caused and promoted by chemical carcinogens in food, water and air. Chemical carcinogens can be administrated to NHPs to create a model for human cancer. A long-term study of chemical-induced tumors in macaques began in 1961 by the NCI, USA (Thorgeirsson et al, 1994; Schoeffner & Thorgeirsson, 2000; Takayama et al, 2008).

Three species of NHPs, the rhesus monkey (*Macaca mulatta*), cynomolgus monkey (*Macaca fascicularis*), and African green monkey (*Cercopithecus aethiops*) were tested. A large number of substances, including a variety of food additives, food components, environmental contaminants, N-nitroso compounds, "classical" rodent carcinogens, anti-neoplastic agents, and immunosuppressive agents were evaluated for any long-term carcinogenic effects.

The fungal food contaminants, aflatoxin B1 (AFB1) (Adamson et al, 1973) and sterigmatocystin (SMT) (Thorgeirsson et al, 1994), were found to be potent hepatocarcinogens. Noticeably, AFB1 did not induce the *p53* gene mutation in macaques although *p53* is frequently mutated in human HCC from southern Africa and Qidong in China (Fujimoto et al, 1992). This suggests that the *p53* mutation in humans may be induced by carcinogens other than AFB1. AFB1 also induced adenocarcinomas of the pancreas, osteosarcomas and other tumors (Adamson et al, 1976). Three of four nitrosamines, particularly diethylnitrosamine (DENA), induced HCC (Adamson, 1989; Lapis et al, 1995). Cycasin or its aglycone, methylazoxymethanol (MAM) acetate, induced a variety of tumors, primarily hepatocellular and renal cell carcinomas (Sieber et al, 1980). 2-amino-3-methylimidazo[4,5-f]quinoline (IQ), which is a heterocyclic amines isolated from broiled, fried or barbecued meat and fish, was found to be one of the most potent hepatocarcinogens, inducing malignant liver tumors in 65% of animals over a seven year period of exposure (Adamson et al, 1990; Thorgeirsson et al, 1994). N-Methyl-N-nitrosourea (MNU) was demonstrated to be the only carcinogen persistently inducing tumors in the digestive tract, mostly squamous cell carcinomas of the esophagus (Adamson et al, 1977). Procarbazine, an anti-neoplastic agent, caused acute nonlymphocytic leukemia (Sieber et al, 1978). Several other anti-neoplastic agents, including cyclophosphamide, adriamycin, melphalan and azathioprine also induced a few tumors (Schoeffner & Thorgeirsson, 2000).

Surprisingly, none of the rodent hepatocellular carcinogens tested consistently induced HCC in rhesus and cynomolgus monkeys. This suggests that NHP cancer models may provide different information than rodents models in the study of human cancers. Similarly, dimethylbenzanthracene (DMBA), a strong rodent breast carcinogen, did not induce mammary cancers in eight female *Macaca arctoides* monkeys during a four year observation period (Lillie et al, 2004). Neither cyclamate (Takayama et al, 2000) nor saccharin (Takayama et al, 1998), which had been used as food additives and induced tumors in rats, showed any evidence of carcinogenic effects. The tumorigenic potential of dichlorodiphenyltrichloroethane (DDT), which has been extensively used as a pesticide, was also negligible after dosing for 15–22 years (Takayama et al, 1999).

Other chemicals have been tested in NHPs in addition to those tested in the NCI study. Rhesus macaques treated with DMBA combined with UV light, dodecylbenzene (DDB), or both applied to the skin developed dermal melanosis, papillomas, basal cell tumors and mesodermal sarcomas (Palotay et al, 1976). Ohgaki et al (1986) reported that gastric carcinomas in the pyloric region were developed in five monkeys (*Macaca mulatta* and *Macaca irus*) after adding N-Ethyl-N'-nitro-N-nitrosoguanidine (ENNG) to their drinking water. Similar results were obtained with N-methyl-N-nitro-N-nitrosoguanidine (MNNG) in cynomolgus monkeys (Sharashidze et al, 1989). In conclusion, numerous chemical agents can efficiently induce a variety of cancers in NHPs (Tab. 1).

2.1.2 Hormones and breast cancers

Hormone levels are well known to regulate the occurrence of breast cancer in women, making this an important focus of cancer research using animal models. The mammary epithelial cells of postmenopausal macaques are shown to express estrogen receptors (ERs) (Isaksson et al, 2002) and progesterone receptors (PRs) (Isaksson et al, 2003). Spontaneous breast tumors in macaques showed selective loss of sex steroid receptors and over-expression of the HER2 protein (Wood et al, 2006). Due to these findings, many attempts have been made to establish a breast cancer model with macaques. An early study showed that high doses of estrogen failed to induce breast neoplasm (Geschickter & Hartman, 1959). In the 1960s, a long-term study in adult monkeys sponsored by the FDA, USA assessed the risk of high doses of oral contraceptives in juvenile. A total of 213 treated and 16 control monkeys were used to evaluate the effects of a variety of synthetic oral contraceptive steroids, including synthetic estrogen mestranol, progestogens ethynerone, chloroethyl norgestrel, and anagestone acetate, on the mammary gland over a 10 year period (Drill et al, 1974; Geil & Lamar, 1977; Drill & Golway, 1978; Fitzgerald et al, 1982; Tavassoli

Tab. 1 Summary of experimental cancer models in NHPs

Species	Carcinogens	Cancer types	References
Macaca spp.	AFB1	HCC, osteosarcomas.	Adamson, 1973; Adamson, 1976
	DENA	HCC	Adamson, 1989; Lapis, 1995
	Cycasin or MAM	HCC, renal cell carcinomas	Sieber, 1980
	IQ	Malignant liver tumors	Adamson, 1990; Thorgeirsson, 1994
	MNU	Esophageal squamous cell carcinomas	Adamson, 1977
	Procarbazine	Nonlymphocytic leukemia	Sieber, 1978
	ENNG	Gastric carcinomas	Ohgaki, 1986
	HVS C488	Pleomorphic peripheral T-cell lymphomas	Knappe, 2000
Macaca mulatta	DMBA & UV, (or DDB)	Dermal melanosis, papillomas, basal cell tumors, and mesodermal sarcomas	Palotay, 1976
	Ethynerone, Mestranol, Anagestone acetate, Chloroethynyl norgestrel	Mammary carcinomas	Drill, 1974; Geil, 1977; Drill, 1978; Fitzgerald, 1982; Tavassoli, 1988
	H. pylori & ENNG	Gastric carcinomas	Liu, 2009
	SIV	AIDS-associated lymphomas	Murphey-Corb, 1986
	Radiation	Brain tumors	Wakisaka, 1982
	55-MeV protons	Malignant brain tumors	Wood, 1991
Macaca fascicularis	SIVsm	AIDS-associated lymphomas	Feichtinger, 1990; Putkonen, 1992
	MNNG	Stomach cancers	Sharashidze, 1989
Saguinus oedipus	HVS C488	Acute peripheral T-cell lymphomas	Knappe, 1998;
	HVA	Acute T-cell lymphomas	Hunt, 1972
	EBV	Malignant lymphomas	Shope, 1973a; Cleary, 1985; Epstein, 1985
Callithrix jacchus	HVS C488	Multicentric lymphomas	Duboise, 1998
Galagos spp.	DENA	Nasal mucoepidermoid carcinoma	Thorgeirsson, 1994
Galago crossicaudatus	DMBA & DDB	Papillomas, basosquamous cell epithelioma	Palotay, 1976
Perodicticus potto	DMBA & DDB	Dermal neoplasm	Palotay, 1976
Tupaia belangeri chinenesis	HBV, AFB1	HCC	Yan, 1996; Li, 2004; Li, 2008
Tupaia glis	AFB1	HCC	Reddy, 1976;
	DHPN	Pulmonary adenomas, squamous cell epithelioma	Rao, 1980

et al, 1988). Seventeen mammary carcinomas were identified primarily in the groups treated with high-dose estrogen-progestogen combinations (mestranol+ethynerone, mestranol+anagestone acetate, and mestranol+ chloroethynyl norgestrel) and three were observed in the group treated with ethynerone alone (Tavassoli et al, 1988). Consistently, estradiol and synthetic progestin medroxyprogesterone acetate (MPA) resulted in significantly greater breast epithelial proliferation in ovariectomized adult female cynomolgus macaques (Wood et al, 2007). However, Valerio (1989) summarized data from 17 contraceptive steroid studies in macaques and found that *in situ* ductal carcinomas and invasive breast carcinomas were identified in only 4/264 controls (1.5%) and 13/733 treated animals (1.7%). These results imply that hormones alone are not strong carcinogens in macaques.

2.1.3 Infection-associated cancers

Cancer is not an infectious disease, but some infectious agents can induce cancer. The World Health Organization (WHO) International Agency for Research on Cancer estimated that the total number of infection-associated cancers was approximately 1.9 million in 2002, accounting for 17.8% of human cancers. The principle causes included hepatitis B and C viruses, human papilloma viruses (HPVs), human immunodeficiency virus (HIV), T-lymphotropic viruses, Epstein-Barr virus (EBV), human herpes virus 8, and *Helicobacter pylori* (*H. pylori*) (Parkin, 2006).

2.1.3.1 *Helicobacter pylori* and gastric cancer

Gastric cancer is the fourth most common cancer worldwide (almost 10% of new cancers) and the second leading cause of cancer mortality. *H. pylori* is well known to cause chronic infection in the human gastric

mucosa, to be a major cause of peptic ulcer disease, and a principal risk factor for gastric cancer (Parsonnet et al, 1991). Several rodent models such as the mouse, rat and Mongolian gerbil, have been used extensively to study *H. pylori*-induced diseases. The Mongolian gerbil was found to be the only species in which gastric cancer could be induced by *H. pylori* infection (Ogura et al, 2000). *H. pylori* has been shown to infect several NHPs including chimpanzees (*Pan troglodytes*) (Hazell et al, 1992), rhesus monkeys (Euler et al, 1990), cynomolgus monkeys (Euler et al, 1990) and Japanese monkeys (*Macaca fuscata*) (Shuto et al, 1993). The infection of *H. pylori* caused atrophic gastritis and p53 mutations in the Japanese monkey model (Oda et al, 2002); however, the Japanese monkey model did not develop gastric carcinoma after five years after the first infection. Liu et al reported that gastric intraglandular neoplasia were induced in three rhesus macaques infected with *H. pylori* and that received the ENNG carcinogen, which is similar to the nitrosamines found in pickled vegetables, while no gastric carcinomas were induced by *H. pylori* and ENNG alone (Liu et al, 2009). Thus, *H. pylori* infection is a potential risk factor for inducing gastric neoplasia.

2.1.3.2 Virus-associated cancers

The vast majority of human and animal viruses do not cause cancer after infection. However, some viruses do initiate cancer in a small percentage of cases and are appropriately named tumor viruses. Tumor viruses induce tumors potentially by causing genetic changes in host cells. A number of these viruses have been associated with specific cancers in humans such as HPV and cervical cancer, HBV and liver cancer, EBV and nasopharyngeal cancer and lymphoma, and HIV and lymphoma (Ma & Chan, 2009).

1) Simian herpesvirus and T-cell lymphomas

Both herpesvirus saimiri (HVS) and herpesvirus ateles (HVA) belong to the simian herpesvirus group, and are known to cause no disease or only minor disease in their natural new world monkey hosts squirrel monkeys (*Saimiri sciureus*) and spider monkeys (*Ateles paniscus*) respectively. However, HVS and HVA can induce lymphomas or lymphoblastic leukemias in other monkey species. HVS caused acute peripheral T-cell lymphomas in other new world primates such as tamarins (*Saguinus* spp.), common marmosets (*Callithrix jacchus*), and owl monkeys (*Aotus trivirgatus*) (Meléndez et al, 1969; Fleckenstein et al, 1978). These monkey species are presumably not infected by HVS in the wild. Tamarins are observed to be susceptible to all subgroups (A, B, and C) of HVS. Wild type HVS (type C) caused acute peripheral T-cell lymphomas within only a few weeks in common marmosets and in cotton-top tamarins (*Saguinus oedipus*) (Duboise et al, 1998; Knappe et al, 1998). HVS C488 (a mutant species) even induced pleomorphic peripheral T-cell lymphomas in old world monkeys (Knappe et al, 2000). Similar to HVS, HVA also induced acute T-cell lymphomas in cotton-top tamarins (Hunt et al, 1972).

2) Epstein-Barr virus and lymphomas

EBV is one of the five human herpesviruses, human herpesvirus 4 (HHV-4). EBV is known to cause several lymphoproliferative disorders and cancers, particularly Burkitt's lymphoma, nasopharyngeal carcinoma, and central nervous system lymphomas associated with HIV (Long et al, 2010). The cotton-top tamarin is the only species in which experimental EBV infection is observed to regularly cause malignant lymphomas. Shope et al (1973a) inoculated cotton-top tamarins with EBV and found that one of the four monkeys that received autologous cells transformed *in vitro* by EBV developed lymphoma in mesenteric lymph nodes and three of the four monkeys inoculated with cell-free EBV developed lymphoma. Cleary et al (1985) inoculated cotton-top tamarins with EBV and observed multiple tumors within 14 to 21 days. Additionally, each lymphoma from the same animal induced by EBV arose from different B-cell clones. This resembles EBV-associated human lymphomas that occur with organ transplants. Importantly, tamarins vaccinated with virus-determined membrane antigen gp340 were effectively protected from tumor induction by a massive challenge dose of EBV (Epstein et al, 1985). However, EBV cannot induce tumors in adult squirrel monkeys (Shope et al, 1973b) and gibbons (*Hylobates lar*) (Werner et al, 1972).

3) Simian AIDS-associated lymphomas

The frequency of lymphomas in AIDS patients is approximately 60 to 100 times that of the general population (Spittle, 1998). Despite the fact that Highly Active Anti-Retroviral Therapy (HAART) reduces the incidence of opportunistic infections and prolongs the survival time for HIV-infected patients, the incidence of AIDS-associated lymphomas (AAL) is not reduced. It has been reported that macaques infected with Simian immunodeficiency virus (SIV) developed malignant lymphomas and an AIDS-like syndrome (Feichtinger et al, 1990). Since 1984, complete necropsy examinations

have been performed on more than 1,000 SIV-infected rhesus macaques at the Tulane Regional Primate Research Center and lymphomas have been detected in 38 animals (Murphey-Corb et al, 1986). The low incidence of AAL in SIV-infected rhesus macaques (~4%) is similar to that in AIDS patients. Several approaches, such as using cynomolgus macaques, administering cytokines, using less pathogenic SIV strains, or combining SIV with various herpesviruses have been proposed to increase the incidence of lymphomas in macaques to establish models using less animals (Baskin et al, 2001). The incidence of simian AAL in cynomolgus macaques is eight-fold higher than that in rhesus macaques. Malignant lymphomas were observed in nine of 24 (38%) SIVsm-infected cynomolgus monkeys and all tumors were high-grade B-cell lymphoblastic or pleomorphic lymphomas with extranodal and disseminated growth (Feichtinger et al, 1990). Similar results were observed in another independent study (Putkonen et al, 1992). Thus, SIV-infected macaques (both rhesus and cynomolgus) demonstrated similar features of AAL, including B-cell origin, extranodal anatomic distribution, the absence of SIV in tumor cells and the presence of rhesus lymphocryptovirus (Habis et al, 1999). Interestingly, SIV-infected macaques that had lymphomas with large nuclei survived longer than those that had lymphomas with small nuclei (Habis et al, 1999). This phenomenon was also observed in patients with Burkitt's-like lymphomas. Taken together, SIV-infected monkeys are appropriate animal models to study human AAL.

2.1.4 Radiation-induced cancers

It is well established that radiation can induce cancer in humans. An early study in NHPs showed that radiation induced brain tumors in rhesus monkeys (Wakisaka et al, 1982). Consistently, rhesus monkeys exposed to 55-MeV protons displayed a high incidence of malignant brain tumors with latent periods ranging from 13 months to 20 years (Wood, 1991).

2.2 Carcinogenesis studies in prosimians

Prosimians have few similarities with humans. They are small in size, nocturnal and only found in the old world. To date, only two prosimian species (Galagos and Pottos) have been used in cancer research. When Galagos were administered DENA, a potent hepatocarcinogen in cynomolgus, rhesus, and African green monkeys intraperitoneally the animals developed primarily mucoepidermoid carcinomas of the nasal cavity (Thorgeirsson et al, 1994). In another study, galagos (*Galago crossicaudatus*) were treated with DMBA and DDB biweekly for four years and papillomas, basal cell tumors and basosquamous cell epidermal tumors with satellite lipomas were observed within eight years (Palotay et al, 1976). When DMBA and DDB were applied to the skin of pottos (*Perodicticus potto*) neoplasms were observed in 17 of the 19 primates that lived longer than nine weeks (Palotay et al, 1976).

2.3 Carcinogenesis studies in tree shrews

Tree shrews (*Tupaia* spp.) are non-rodent and primate-like animals native to the tropical forests of Southeast Asia. Previously, tree shrews were grouped with prosimians because of certain internal similarities (brain anatomy for example) but now they have been assigned a separate order, scandentia, based on recent molecular phylogenetic studies. There are 20 species in five genera. All tree shrews share some common characteristics such as a relatively small body mass, generally omnivorous diet, and non-prehensile hands and feet (Martin, 1990). Since 1966 when the first spontaneous case of breast cancer was observed in tree shrews (Elliot et al, 1966) further cases of other spontaneous cancers have been described such as HCC (Hofmann et al, 1981), epidermoid carcinoma, and lymphoma (Brack, 1998). Hence, tree shrews may also be useful experimental animal models for carcinogenesis studies.

2.3.1 Hepatocellular carcinomas

HCC is a major cause of cancer deaths worldwide and accounts for ~6% of all human cancers. The development of HCC is considered to be caused by the HBV infection and chemical carcinogens such as AFB1. During the 1970s, Reddy et al (1976) successfully induced HCC in tree shrews (*Tupaia glis*) by administering AFB1. Yan et al (1996) found that tree shrews (*Tupaia belangeri chinenesis*) could be infected with human HBV. Further investigation revealed that HBV in combination with AFB1 significantly increased the incidence of HCC (52.9%) compared to HBV alone (11.11%) or AFB1 alone (12.50%). Oltipraz treatment protected tree shrews from AFB1-induced liver cancer in a manner similar to that observed in rodents and humans (Li et al, 2000). The tree shrew p53 sequence shares 91.7% (nucleotide) and 93.4% (amino acid) homology with the human p53 sequence (Park et al, 2000). Park et al (2000) detected mutations in the *p53* gene in tree shrews exposed to AFB1 and/or HBV (Park et al, 2000).

In addition to the mutation of p53, AFB1 and HBV up-regulated the expression of the *Ras* gene in tree shrews before HCC development (Su et al, 2004). To understand the mechanism by which AFB1 induced HCC, Li et al (2004) examined gene expression profiles during AFB1-induced hepatocarcinogenesis in tree shrews and identified a set of genes that were up- or down-regulated in HCC. Following that, a group of candidate proteins that were differentially expressed in tree shrew HCC were obtained by proteome analysis (Li et al, 2008). The peroxiredoxin (Prx) II protein was found to be overexpressed in both tree shrew and human HCC. When Prx II expression was inhibited by siRNAs in human Hep3B cells, cell proliferation and survival decreased significantly (Li et al, 2008). These findings suggest that the tree shrew is a suitable animal model for studying the mechanisms of human HCC.

2.3.2 Pulmonary adenomas

Lung cancer is the leading cause of human cancer deaths worldwide. Therefore, the task of finding an effective NHP model to study lung cancer is an important objective in cancer research. In one study with NHPs, the carcinogenic effects of 2, 2'-dihydroxy-di-n-propylnitrosamine (DHPN) on tree shrews (*Tupaia glis*) was examined. DHPN was subcutaneously administered to tree shrews at a dose of 250 mg/kg of body weight once a week for 80 weeks. Eight of the nine males (89%) and 11 of the 14 females (78%) that received DHPN developed pulmonary adenomas between weeks 65 and 102 (Rao & Reddy, 1980). Additionally, 9% of the DHPN-treated tree shrews developed squamous cell carcinomas of the skin and HCC. Taken together, tree shrew may be a suitable animal model for lung cancer research.

3 NHPs for cancer genetics studies, therapies and prevention

NHPs have been used in cancer genetics studies in the past. For instance, the evolution of tumor suppressor genes such as *BRCA1* (Pavlicek et al, 2004; Puente et al, 2006) and *p53* (Contente et al, 2003) in NHPs has been previously reported. To identify potential human cancer-related genes, comparative genomic hybridization (CGH) has been used to generate cytogenetic profiles of malignant gliomas in four monkey tumors (Jaikumar et al, 2007). In 2005, the chimpanzee genome was sequenced (Chimpanzee Sequencing and Analysis Consortium et al, 2005) and two years later, the rhesus macaque genome was sequenced (Rhesus Macaque Genome Sequencing and Analysis Consortium et al, 2007). The sequencing of more NHP genomes will further accelerate comparative cancer genetic studies in NHPs and humans.

In addition to the study of cancer-related genes, NHP models are ideal for studying drugs for cancer therapies and chemoprevention because of their genetic similarity with humans. NHPs have been widely used to evaluate the safety and pharmacokinetics of anti-tumor drugs and vaccines (Parker et al, 1995; Kim et al, 2001; Hassan et al, 2007; Kapetanovic et al 2010; Morera et al, 2010). The rhesus monkey in particular is suggested to be an excellent candidate for a NHP model for ovarian cancer chemoprevention (Brewer et al, 2001a, b). Using cultured rhesus ovarian surface epithelium cells, Wright et al (2005) demonstrated that high-dose estrogen and clinical selective ER modulators induced growth arrest *in vitro*. Rodriguez et al (1998) showed that oral contraceptive progestin induced apoptosis in the ovarian epithelium. Additionally, an agonist of toll-like receptor 5 has been reported to protect rhesus monkeys from lethal irradiation routinely used to treat cancer patients (Burdelya et al, 2008). More recently, caloric restriction has been demonstrated to delay the onset of age-associated diseases, including cancer, in rhesus monkeys (Colman et al, 2009).

4 Conclusions and perspectives

NHPs share numerous similarities with humans and have been widely used to study human diseases such as AIDS, hepatitis and neurodegenerative diseases. However, NHPs have not been widely used for cancer research because of the low incidence of spontaneous tumors, long latency, slow propagation and high costs. Different chemical, biological (including hormones, bacteria, and viruses), and physical carcinogens have been used to induce a variety of tumors in different NHPs, including macaques and tree shrews. These NHP cancer models provide valuable knowledge for human cancer prevention and treatment due to their similarity to human cancers.

More NHP cancer models are required to better understand mechanisms of human carcinogenesis and to design more effective prevention and treatment options for cancer patients. An ideal animal model for cancer research should faithfully reflect various aspects of the human disease such as the etiology, pathology, genetics

and therapy. Additionally, tumors should be expected to develop with a high incidence and a relatively short latency. NHPs have the potential to grow as tools for human cancer research with the development of technology. For example, NHP transgenic techniques have recently been established (Niu et al, 2010). Finally, as primate-like animals, tree shrews which are smaller in size, propagate rapidly and cost comparatively less than larger NHPs and may be a superior model for cancer research and help overcome the disadvantages of using rodents and other NHPs. We are currently trying to establish tree shrew cancer models by using chemical and biological carcinogens. It is our belief that NHPs will play a more significant role in future cancer research.

References:

Adamson RH. 1989. Induction of hepatocellular carcinoma in nonhuman primates by chemical carcinogens [J]. *Cancer Detect Prev*, **14**(2): 215-219.

Adamson RH, Cooper RW, O'Gara RW. 1970. Carcinogen-induced tumors in primitive primates [J]. *J Natl Cancer Inst*, **45**(3): 555-559.

Adamson RH, Correa P, Dalgard DW. 1973. Occurrence of a primary liver carcinoma in a Rhesus monkey fed aflatoxin B1 [J]. *J Natl Cancer Inst*, **50**(2): 549-553.

Adamson RH, Correa P, Sieber SM, McIntire KR, Dalgard DW. 1976. Carcinogenicity of aflatoxin B1 in rhesus monkeys: two additional cases of primary liver cancer [J]. *J Natl Cancer Inst*, **57**(1): 67-78.

Adamson RH, Krollkowski FJ, Correa P, Sieber SM and Dalgard DW. 1977. Carcinogenecity of 1-methyl-1-nitrosourea in nonhuman primates [J]. *J Natl Cancer Inst*, **59**: 415-422.

Adamson RH, Thorgeirsson UP, Snyderwine EG, Thorgeirsson SS, Reeves J, Dalgard DW, Takayama S, Sugimura T. 1990. Carcinogenicity of 2-amino-3-methylimidazo[4,5-f]quinoline in nonhuman primates: induction of tumors in three macaques [J]. *Jpn J Cancer Res*, **81**(1): 10-14.

Bailey J. 2009. An examination of chimpanzee use in human cancer research [J]. *Altern Lab Anim,* **37**(4): 399-416.

Baskin GB, Cremer KJ, Levy LS. 2001. Comparative pathobiology of HIV- and SIV-associated lymphoma [J]. *AIDS Res Hum Retroviruses,* **17**(8): 745-751.

Beniashvili DS. 1989. An overview of the world literature on spontaneous tumors in nonhuman primates [J]. *J Med Primatol*, **18**(6): 423-437.

Brack M. 1998. Spontaneous tumours in tree shrews (Tupaia belangeri): population studies [J]. *J Comp Pathol*, **118**(4): 301-316.

Brewer M, Baze W, Hill L, Utzinger U, Wharton JT, Follen M, Khan-Dawood F, Satterfield W. 2001a. Rhesus macaque model for ovarian cancer chemoprevention [J]. *Comp Med,* **51**(5): 424-429.

Brewer M, Utzinger U, Satterfield W, Hill L, Gershenson D, Bast R, Wharton JT, Richards-Kortum R, Follen M. 2001b. Biomarker modulation in a nonhuman rhesus primate model for ovarian cancer chemoprevention [J]. *Cancer Epidemiol Biomarkers Prev*, **10**(8): 889-893.

Bunton TE, Bacmeister CX. 1989. Diverticulosis and colonic leiomyosarcoma in an aged rhesus macaque [J]. *Vet Pathol*, **26**(4): 351-352.

Burdelya LG, Krivokrysenko VI, Tallant TC, Strom E, Gleiberman AS, Gupta D, Kurnasov OV, Fort FL, Osterman AL, Didonato JA, Feinstein E, Gudkov AV. 2008. An agonist of toll-like receptor 5 has radioprotective activity in mouse and primate models [J]. *Science,* **320**(5873): 226-230.

Chimpanzee Sequencing and Analysis Consortium. 2005. Initial sequence of the chimpanzee genome and comparison with the human genome [J]. *Nature,* **437**(7055): 69-87.

Cianciolo RE, Hubbard GB. 2005. A review of spontaneous neoplasia in baboons (*Papio* spp.) [J]. *J Med Primatol*, **34**(2): 51-66.

Cleary ML, Epstein MA, Finerty S, Dorfman RF, Bornkamm GW, Kirkwood JK, Morgan AJ, Sklar J. 1985. Individual tumors of multifocal EB virus-induced malignant lymphomas in tamarins arise from different B-cell clones [J]. *Science*, **228**(4700): 722-724.

Coggins CR. 2007. An updated review of inhalation studies with cigarette smoke in laboratory animals [J]. *Int J Toxicol*, **26**(4): 331-338.

Colman RJ, Anderson RM, Johnson SC, Kastman EK, Kosmatka KJ, Beasley TM, Allison DB, Cruzen C, Simmons HA, Kemnitz JW, Weindruch R. 2009. Caloric restriction delays disease onset and mortality in rhesus monkeys [J]. *Science,* **325**(5937): 201-204.

Contente A, Zischler H, Einspanier A, Dobbelstein M. 2003. A promoter that acquired p53 responsiveness during primate evolution [J]. *Cancer Res,* **63**(8): 1756-1758.

Drill VA, Golway PL. 1978. Effect of ethynodiol diacetate with ethinyl estradiol on the mammary glands of rhesus monkeys: a preliminary report [J]. *J Natl Cancer Inst*, **60**(5): 1169-1170.

Drill VA, Martin DP, Hart ER, McConnell RG. 1974. Effect of oral contraceptives on the mammary glands of Rhesus monkeys: a preliminary report [J]. *J Natl Cancer Inst*, **52**(5): 1655-1657.

Duboise M, Guo J, Czajak S, Lee H, Veazey R, Desrosiers RC, Jung JU. 1998. A role for herpesvirus saimiri orf14 in transformation and persistent infection [J]. *J Virol*, **72**(8): 6770-6776.

Elliot OS, Elliot MW, Lisco H. 1966. Breast cancer in a tree shrew (Tupaia glis) [J]. *Nature*, **211**(5053): 1105.

Epstein MA, Morgan AJ, Finerty S, Randle BJ, Kirkwood JK. 1985. Protection of cottontop tamarins against Epstein-Barr virus-induced malignant lymphoma by a prototype subunit vaccine [J]. *Nature,* **318**(6043): 287-289.

Euler AR, Zurenko GE, Moe JB, Ulrich RG, Yagi Y. 1990. Evaluation of two monkey species (*Macaca mulatta* and *Macaca fascicularis*) as possible models for human *Helicobacter pylori* disease [J]. *J Clin Microbiol*, **28**(10): 2285-2290.

Feichtinger H, Putkonen P, Parravicini C, Li SL, Kaaya EE, Böttiger D, Biberfeld G, Biberfeld P. 1990. Malignant lymphomas in cynomolgus monkeys infected with simian immunodeficiency virus [J]. *Am J Pathol*, **137**(6): 1311-1315.

Fitzgerald J, de la Iglesia F, Goldenthal EI. 1982. Ten-year oral toxicity study with Norlestrin in rhesus monkeys [J]. *J Toxicol Environ Health*, **10**(6): 879-896.

Fleckenstein B, Daniel MD, Hunt RD, Werner J, Falk LA, Mulder C. 1978. Tumour induction with DNA of oncogenic primate herpesviruses [J]. *Nature*, **274**(5666): 57-59.

Fujimoto Y, Hampton LL, Luo LD, Wirth PJ, Thorgeirsson SS. 1992. Low frequency of p53 gene mutation in tumors induced by aflatoxin B1 in nonhuman primates [J]. *Cancer Res*, **52**(4): 1044-1046.

Geil RG, Lamar JK. 1977. FDA studies of estrogen, progestogens, and estrogen/progestogen combinations in the dog and monkey [J]. *J Toxicol Environ Health*, **3**(1-2): 179-193.

Geschickter CF, Hartman CG. 1959. Mammary response to prolonged estrogenic stimulation in the monkey [J]. *Cancer*, **12**(4): 767-781.

Habis A, Baskin GB, Murphey-Corb M, Levy LS. 1999. Simian AIDS-associated lymphoma in rhesus and cynomolgus monkeys recapitulates the primary pathobiological features of AIDS-associated non-Hodgkin's lymphoma [J]. *AIDS Res Hum Retroviruses*, **15**(15): 1389-1398.

Hassan R, Ebel W, Routhier EL, Patel R, Kline JB, Zhang J, Chao Q, Jacob S, Turchin H, Gibbs L, Phillips MD, Mudali S, Iacobuzio-Donahue C, Jaffee EM, Moreno M, Pastan I, Sass PM, Nicolaides NC, Grasso L. 2007. Preclinical evaluation of MORAb-009, a chimeric antibody targeting tumor-associated mesothelin [J]. *Cancer Immun,* **7**: 20.

Hazell SL, Eichberg JW, Lee DR, Alpert L, Evans DG, Evans DJ Jr, Graham DY. 1992. Selection of the chimpanzee over the baboon as a model for *Helicobacter pylori* infection [J]. *Gastroenterology*, **103**(3): 848-854.

Hofmann W, Möller P, Schwaier A, Flügel RM, Zöller L, Darai G. 1981. Malignant tumours in *Tupaia* (tree shrew). *J Med Primatol*, **10**(2-3): 155-163.

Huang R, Cheng SJ. 2002. Review of spontaneous tumors in non-human primates [J]. *Acta Laboratorium Animalis Scientia Sini,* **10**: 123-128. (in Chinese)

Hunt RD, Meléndez LV, García FG, Trum BF. 1972. Pathologic features of Herpesvirus ateles lymphoma in cotton-topped marmosets (*Saguinus oedipus*) [J]. *J Natl Cancer Inst*, **49**(6): 1631-1639.

Isaksson E, Wang H, Sahlin L, von Schoultz B, Cline JM, von Schoultz E. 2003. Effects of long-term HRT and tamoxifen on the expression of progesterone receptors A and B in breast tissue from surgically postmenopausal cynomolgus macaques [J]. *Breast Cancer Res Treat*, **79**(2): 233-239.

Isaksson E, Wang H, Sahlin L, von Schoultz B, Masironi B, von Schoultz E, Cline JM. 2002. Expression of estrogen receptors (alpha, beta) and insulin-like growth factor-I in breast tissue from surgically postmenopausal cynomolgus macaques after long-term treatment with HRT and tamoxifen [J]. *Breast*, **11**(4): 295-300.

Jaikumar S, Zhuang Z, Mannan P, Vortmeyer AO, Furuta M, Dickerman R, Bedanova J, Lonser RR, Walbridge S, Weil RJ, Lobanenkov VV, Oldfield EH, Pack SD. 2007. Interspecies comparative genomic hybridization (I-CGH): a new twist to study animal tumor models [J]. *Cell Cycle,* **6**(7): 836-842.

Jayo MJ, Jayo JM, Jerome CP, Krugner-Higby L, Reynolds GD. 1988. Maxillo-orbital lymphoma (Burkitt's-type) in an infant *Macaca fascicularis* [J]. *Lab Anim Sci*, **38**(6): 722-726.

Kapetanovic IM, Muzzio M, Hu SC, Crowell JA, Rajewski RA, Haslam JL, Jong L, McCormick DL. 2010. Pharmacokinetics and enhanced bioavailability of candidate cancer preventative agent, SR13668 in dogs and monkeys [J]. *Cancer Chemother Pharmacol,* **65**(6): 1109-1116.

Kim JJ, Yang JS, Dang K, Manson KH, Weiner DB. 2001. Engineering enhancement of immune responses to DNA-based vaccines in a prostate cancer model in rhesus macaques through the use of cytokine gene adjuvants [J]. *Clin Cancer Res,* **7**: 882s-889s.

Knappe A, Feldmann G, Dittmer U, Meinl E, Nisslein T, Wittmann S, Mätz-Rensing K, Kirchner T, Bodemer W, Fickenscher H. 2000. Herpesvirus saimiri-transformed macaque T cells are tolerated and do not cause lymphoma after autologous reinfusion [J]. *Blood,* **95**(10): 3256-3261.

Knappe A, Thurau M, Niphuis H, Hiller C, Wittmann S, Kuhn EM, Rosenwirth B, Fleckenstein B, Heeney J, Fickenscher H. 1998. T-cell lymphoma caused by herpesvirus saimiri C488 independently of ie14/vsag, a viral gene with superantigen homology [J]. *J Virol*, **72**(4): 3469-3471.

Lapis K, Zalatnai A, Timár F, Thorgeirsson UP. 1995. Quantitative evaluation of lysozyme- and CD68-positive Kupffer cells in diethylnitrosamine-induced hepatocellular carcinomas in monkeys [J]. *Carcinogenesis,* **16**(12): 3083-3085.

Li Y, Qin X, Cui J, Dai Z, Kang X, Yue H, Zhang Y, Su J, Cao J, Ou C, Yang C, Duan X, Yue H, Liu Y. 2008. Proteome analysis of aflatoxin B1-induced hepatocarcinogenesis in tree shrew (Tupaia belangeri chinensis) and functional identification of candidate protein peroxiredoxin II [J]. *Proteomics,* **8**(7): 1490-1501.

Li Y, Su J, Qin L, Egner PA, Wang J, Groopman JD, Kensler TW, Roebuck BD. 2000. Reduction of aflatoxin B(1) adduct biomarkers by oltipraz in the tree shrew (*Tupaia belangeri chinensis*) [J]. *Cancer Lett*, **154**(1): 79-83.

Li Y, Wan DF, Su JJ, Cao J, Ou C, Qiu XK, Ban KC, Yang C, Qin LL, Luo D, Yue HF, Zhang LS, Gu JR. 2004. Differential expression of genes during aflatoxin B(1)-induced hepatocarcinogenesis in tree shrews [J]. *World J Gastroenterol,* **10**(4): 497-504.

Lillie MA, Ambrus CM, Pickren JW, Akhter S, Islam A, Ambrus JL. 2004. Breast cancer in intraductal carcinogen-treated non-human primates [J]. *J Med,* **35**(1-6): 271-275.

Liu H, Merrell DS, Semino-Mora C, Goldman M, Rahman A, Mog S, Dubois A. 2009. Diet synergistically affects *Helicobacter pylori*-induced gastric carcinogenesis in nonhuman primates [J]. *Gastroenterology*, **137**(4): 1367-1379.

Long HM, Parsonage G, Fox CP, Lee SP. 2010. Immunotherapy for Epstein-Barr virus-associated malignancies [J]. *Drug News Perspect*, **23**(4): 221-228.

Ma BB, Chan AT. 2009. Infection and cancer: biology, therapeutics and prevention [J]. *Future Oncol*, **5**(2): 149-151.

Martin, R.D. 1990. Primate Origins and Evolution: A Phylogenetic Reconstruction[M]. Princeton University Press: Princeton, N.J.

Meléndez LV, Hunt RD, Daniel MD, García FG, Fraser CE. 1969. Herpesvirus saimiri. II. Experimentally induced malignant lymphoma in primates [J]. *Lab Anim Care*, **19**(3): 378-386.

Morera Y, Bequet-Romero M, Ayala M, Velazco JC, Pérez PP, Alba JS, Ancizar J, Rodríguez M, Cosme K, Gavilondo JV. 2010. Immunogenicity and some safety features of a VEGF-based cancer therapeutic vaccine in rats, rabbits and non-human primates [J]. *Vaccine,* **28**(19): 3453-3461.

Murphey-Corb M, Martin LN, Rangan SR, Baskin GB, Gormus BJ, Wolf RH, Andes WA, West M, Montelaro RC. 1986. Isolation of an HTLV-III-related retrovirus from macaques with simian AIDS and its possible origin in asymptomatic mangabeys [J]. *Nature,* **321**(6068): 435-437.

Niu Y, Yu Y, Bernat A, Yang S, He X, Guo X, Chen D, Chen Y, Ji S, Si W, Lv Y, Tan T, Wei Q, Wang H, Shi L, Guan J, Zhu X, Afanassieff M, Savatier P, Zhang K, Zhou Q, Ji W. 2010. Transgenic rhesus monkeys produced by gene transfer into early-cleavage-stage embryos using a simian immunodeficiency virus-based vector [J]. *Proc Natl Acad Sci U S A*, **107**(41): 17663-17667.

Oda T, Murakami K, Nishizono A, Kodama M, Nasu M, Fujioka T. 2002. Long-term *Helicobacter pylori* infection in Japanese monkeys induces atrophic gastritis and accumulation of mutations in the p53 tumor suppressor gene [J]. *Helicobacter*, **7**(3): 143-151.

Ogura K, Maeda S, Nakao M, Watanabe T, Tada M, Kyutoku T, Yoshida H, Shiratori Y, Omata M. 2000. Virulence factors of

Helicobacter pylori responsible for gastric diseases in Mongolian gerbil [J]. *J Exp Med*, **192**(11): 1601-1610.

Ohgaki H, Hasegawa H, Kusama K, Morino K, Matsukura N, Sato S, Maruyama K, Sugimura T. 1986. Induction of gastric carcinomas in nonhuman primates by N-ethyl-N'-nitro-N-nitrosoguanidine [J]. *J Natl Cancer Inst*, **77**(1): 179-186.

Palotay JL, Adachi K, Dobson RL, Pinto JS. 1976. Carcinogen-induced cutaneous neoplasms in nonhuman primates [J]. *J Natl Cancer Inst*, **57**(6): 1269-1274.

Park US, Su JJ, Ban KC, Qin L, Lee EH, Lee YI. 2000. Mutations in the p53 tumor suppressor gene in tree shrew hepatocellular carcinoma associated with hepatitis B virus infection and intake of aflatoxin B1 [J]. *Gene*, **251**(1): 73-80.

Parker SE, Vahlsing HL, Serfilippi LM, Franklin CL, Doh SG, Gromkowski SH, Lew D, Manthorpe M, Norman J. 1995. Cancer gene therapy using plasmid DNA: safety evaluation in rodents and non-human primates [J]. *Hum Gene Ther,* **6**(5): 575-590.

Parkin DM. 2006. The global health burden of infection-associated cancers in the year 2002 [J]. *Int J Cancer*, **118**(12): 3030-3044.

Parsonnet J, Friedman GD, Vandersteen DP, Chang Y, Vogelman JH, Orentreich N, Sibley RK. 1991. *Helicobacter pylori* infection and the risk of gastric carcinoma [J]. *N Engl J Med*, **325**(16): 1127-1131.

Pavlicek A, Noskov VN, Kouprina N, Barrett JC, Jurka J, Larionov V. 2004. Evolution of the tumor suppressor BRCA1 locus in primates: implications for cancer predisposition [J]. *Hum Mol Genet,* **13**(22): 2737-2751.

Pellegrini G, Bienvenu JG, Meehan JT, Mischler SA, Perry RW, Scott DW, Anderson WI. 2009. Cutaneous melanoma with metastasis in a cynomolgus monkey (*Macaca fascicularis*) [J]. *J Med Primatol*, **38**(6): 444-447.

Pfeiffer CA, Allen E. 1948. Attempts to produce cancer in rhesus monkeys with carcinogenic hydrocarbons and estrogens [J]. *Cancer Res*, **8**(3): 97-127.

Puente XS, Velasco G, Gutiérrez-Fernández A, Bertranpetit J, King MC, López-Otín C. 2006. Comparative analysis of cancer genes in the human and chimpanzee genomes [J]. *BMC Genomics*, **7**: 15.

Putkonen P, Kaaya EE, Böttiger D, Li SL, Nilsson C, Biberfeld P, Biberfeld G. 1992. Clinical features and predictive markers of disease progression in cynomolgus monkeys experimentally infected with simian immunodeficiency virus [J]. *AIDS*, **6**(3): 257-263.

Rao MS, Reddy JK. 1980. Carcinogenicity of 2,2'-dihydroxy-di-n-propylnitrosamine in the tree shrew (*Tupaia glis*): light and electron microscopic features of pulmonary adenomas [J]. *J Natl Cancer Inst*, **65**(4): 835-840.

Reddy JK, Svoboda DJ, Rao MS. 1976. Induction of liver tumors by aflatoxin B1 in the tree shrew (*Tupaia glis*), a nonhuman primate [J]. *Cancer Res*, **36**(1): 151-160.

Rhesus Macaque Genome Sequencing and Analysis Consortium, Gibbs RA, Rogers J, Katze MG, Bumgarner R, Weinstock GM, Mardis ER, Remington KA, Strausberg RL, Venter JC, Wilson RK, Batzer MA, Bustamante CD, Eichler EE, Hahn MW, Hardison RC, Makova KD, Miller W, Milosavljevic A, Palermo RE, Siepel A, Sikela JM, Attaway T, Bell S, Bernard KE, Buhay CJ, Chandrabose MN, Dao M, Davis C, Delehaunty KD, Ding Y, Dinh HH, Dugan-Rocha S, Fulton LA, Gabisi RA, Garner TT, Godfrey J, Hawes AC, Hernandez J, Hines S, Holder M, Hume J, Jhangiani SN, Joshi V, Khan ZM, Kirkness EF, Cree A, Fowler RG, Lee S, Lewis LR, Li Z, Liu YS, Moore SM, Muzny D, Nazareth LV, Ngo DN, Okwuonu GO, Pai G, Parker D, Paul HA, Pfannkoch C, Pohl CS, Rogers YH, Ruiz SJ, Sabo A, Santibanez J, Schneider BW, Smith SM, Sodergren E, Svatek AF, Utterback TR, Vattathil S, Warren W, White CS, Chinwalla AT, Feng Y, Halpern AL, Hillier LW, Huang X, Minx P, Nelson JO, Pepin KH, Qin X, Sutton GG, Venter E, Walenz BP, Wallis JW, Worley KC, Yang SP, Jones SM, Marra MA, Rocchi M, Schein JE, Baertsch R, Clarke L, Csürös M, Glasscock J, Harris RA, Havlak P, Jackson AR, Jiang H, Liu Y, Messina DN, Shen Y, Song HX, Wylie T, Zhang L, Birney E, Han K, Konkel MK, Lee J, Smit AF, Ullmer B, Wang H, Xing J, Burhans R, Cheng Z, Karro JE, Ma J, Raney B, She X, Cox MJ, Demuth JP, Dumas LJ, Han SG, Hopkins J, Karimpour-Fard A, Kim YH, Pollack JR, Vinar T, Addo-Quaye C, Degenhardt J, Denby A, Hubisz MJ, Indap A, Kosiol C, Lahn BT, Lawson HA, Marklein A, Nielsen R, Vallender EJ, Clark AG, Ferguson B, Hernandez RD, Hirani K, Kehrer-Sawatzki H, Kolb J, Patil S, Pu LL, Ren Y, Smith DG, Wheeler DA, Schenck I, Ball EV, Chen R, Cooper DN, Giardine B, Hsu F, Kent WJ, Lesk A, Nelson DL, O'brien WE, Prüfer K, Stenson PD, Wallace JC, Ke H, Liu XM, Wang P, Xiang AP, Yang F, Barber GP, Haussler D, Karolchik D, Kern AD, Kuhn RM, Smith KE, Zwieg AS. 2007. Evolutionary and biomedical insights from the rhesus macaque genome [J]. *Science,* **316**(5822): 222-234.

Roberts TG Jr, Goulart BH, Squitieri L, Stallings SC, Halpern EF, Chabner BA, Gazelle GS, Finkelstein SN, Clark JW. 2004. Trends in the risks and benefits to patients with cancer participating in phase 1 clinical trials [J]. *JAMA*, **292**(17): 2130-2140.

Rodriguez GC, Walmer DK, Cline M, Krigman H, Lessey BA, Whitaker RS, Dodge R, Hughes CL. 1998. Effect of progestin on the ovarian epithelium of macaques: cancer prevention through apoptosis [J]? *J Soc Gynecol Investig,* **5**(5): 271-276.

Schoeffner DJ, Thorgeirsson UP. 2000. Susceptibility of nonhuman primates to carcinogens of human relevance [J]. *In Vivo*, **14**(1): 149-156.

Shope T, Dechairo D, Miller G. 1973a. Malignant lymphoma in cotton-top marmosets after inoculation with Epstein-Barr virus [J]. *Proc Natl Acad Sci U S A,* **70**(9): 2487-2491.

Shope T, Miller G. 1973b. Epstein-Barr virus. Heterophile responses in squirrel monkeys inoculated with virus-transformed autologous leukocytes [J]. *J Exp Med*, **137**(1): 140-147.

Shuto R, Fujioka T, Kubota T, Nasu M. 1993. Experimental gastritis induced by *Helicobacter pylori* in Japanese monkeys [J]. *Infect Immun*, **61**(3): 933-939.

Sieber SM, Correa P, Dalgard DW, Adamson RH. 1978. Carcinogenic and other adverse effects of procarbazine in nonhuman primates [J]. *Cancer Res*, **38**(7): 2125-2134.

Sieber SM, Correa P, Dalgard DW, McIntire KR, Adamson RH. 1980. Carcinogenicity and hepatotoxicity of cycasin and its aglycone methylazoxymethanol acetate in nonhuman primates [J]. *J Natl Cancer Inst*, **65**(1): 177-189.

Sharashidze LK, Beniashvili DSh, Sherenesheva NI, Turkia NG. 1989. Induction of gastric cancer in monkeys by N-methyl-N-nitro-N-nitrosoguanidine (MNNG) [J]. *Neoplasma,* **36**(2): 129-133.

Spittle MF. 1998. Spectrum of AIDS-associated malignant disorders [J]. *Lancet*, **352**(9131): 907.

Su JJ, Ban KC, Li Y, Qin LL, Wang HY, Yang C, Ou C, Duan XX, Lee YL, Yang RQ. 2004. Alteration of p53 and p21 during hepatocarcinogenesis in tree shrews [J]. *World J Gastroenterol*, **10**(24): 3559-3563.

Starost MF. 2009. Adenomyofibroma of the fimbria in a cynomolgus monkey (*Macaca fascicularis*) [J]. *J Vet Diagn Invest*, **21**(6): 892-894.

Takayama S, Renwick AG, Johansson SL, Thorgeirsson UP, Tsutsumi M, Dalgard DW, Sieber SM. 2000. Long-term toxicity and carcinogenicity study of cyclamate in nonhuman primates [J]. *Toxicol Sci*, **53**(1): 33-39.

Takayama S, Sieber SM, Adamson RH, Thorgeirsson UP, Dalgard DW, Arnold LL, Cano M, Eklund S, Cohen SM. 1998. Long-term feeding of sodium saccharin to nonhuman primates: implications for urinary tract cancer [J]. *J Natl Cancer Inst*, **90**(1): 19-25.

Takayama S, Sieber SM, Dalgard DW, Thorgeirsson UP, Adamson RH. 1999. Effects of long-term oral administration of DDT on nonhuman primates [J]. *J Cancer Res Clin Oncol*, **125**(3-4): 219-225.

Takayama S, Thorgeirsson UP, Adamson RH. 2008. Chemical carcinogenesis studies in nonhuman primates [J]. *Proc Jpn Acad Ser B Phys Biol Sci*, **84**(6): 176-188.

Tavassoli FA, Casey HW, Norris HJ. 1988. The morphologic effects of synthetic reproductive steroids on the mammary gland of rhesus monkeys. Mestranol, ethynerone, mestranol-ethynerone, chloroethynyl norgestrel-mestranol, and anagestone acetate-mestranol combinations [J]. *Am J Pathol*, **131**(2): 213-234.

Thorgeirsson UP, Dalgard DW, Reeves J, Adamson RH. 1994. Tumor incidence in a chemical carcinogenesis study of nonhuman primates [J]. *Regul Toxicol Pharmacol,* **19**(2): 130-151.

Yamate J, Tomita A, Kuwamura M, Mitsunaga F, Nakamura S. 2007. Spontaneous peritoneal malignant mesothelioma in a geriatric japanese macaque (*Macaca fuscata*) [J]. *Exp Anim*, **56**(2): 155-159.

Yan RQ, Su JJ, Huang DR, Gan YC, Yang C, Huang GH. 1996. Human hepatitis B virus and hepatocellular carcinoma. II. Experimental induction of hepatocellular carcinoma in tree shrews exposed to hepatitis B virus and aflatoxin B1 [J]. *J Cancer Res Clin Oncol,* **122**(5): 289-295.

Uno H, Alsum P, Zimbric ML, Houser WD, Thomson JA, Kemnitz JW. 1998. Colon cancer in aged captive rhesus monkeys (*Macaca mulatta*) [J]. *Am J Primatol,* **44**(1): 19-27.

Valerio MG. 1989. Comparative aspects of contraceptive steroids: effects observed in the monkey [J]. *Toxicol Pathol*, **17**(2): 401-410.

Wakisaka S, Kemper TL, Nakagaki H, O'Neill RR. 1982. Brain tumors induced by radiation in rhesus monkeys [J]. *Fukuoka Igaku Zasshi,* **73**(10): 585-594.

Werner J, Henle G, Pinto CA, Haff RF, Henle W. 1972. Establishment of continuous lymphoblast cultures from leukocytes of gibbons (*Hylobates lar*) [J]. *Int J Cancer*, **10**(3): 557-567.

Wood CE, Register TC, Lees CJ, Chen H, Kimrey S, Cline JM. 2007. Effects of estradiol with micronized progesterone or medroxyprogesterone acetate on risk markers for breast cancer in postmenopausal monkeys [J]. *Breast Cancer Res Treat,* **101**(2): 125-134.

Wood CE, Usborne AL, Starost MF, Tarara RP, Hill LR, Wilkinson LM, Geisinger KR, Feiste EA, Cline JM. 2006. Hyperplastic and neoplastic lesions of the mammary gland in macaques [J]. *Vet Pathol*, **43**(4): 471-483.

Wood DH. 1991. Long-term mortality and cancer risk in irradiated rhesus monkeys [J]. *Radiat Res,* **126**(2): 132-140.

Wright JW, Stouffer RL, Rodland KD. 2005. High-dose estrogen and clinical selective estrogen receptor modulators induce growth arrest, p21, and p53 in primate ovarian surface epithelial cells [J]. *J Clin Endocrinol Metab,* **90**(6): 3688-3695.