Published in final edited form as:
Curr Pharm Biotechnol. 2011 February 1; 12(2): 182–188.

Infection, Stem Cells and Cancer Signals

S. Sell*
Wadsworth Center and Ordway Research Institute Empire State Plaza, Albany, NY 12201, USA

Abstract

The association of cancer with preceding parasitic infections has been observed for over 200 years. Some such cancers arise from infection of tissue stem cells by viruses with insertion of viral oncogenes into the host DNA (mouse polyoma virus, mouse mammary tumor virus). In other cases the virus does not insert its DNA into the host cells, but rather commandeers the metabolism of the infected cells, so that the cells continue to proliferate and do not differentiate (human papilloma virus and cervical cancer). Cytoplasmic Epstein Barr virus infection is associated with a specific gene translocation (Ig/c-myc) that activates proliferation of affected cells (Burkitt lymphoma). In chronic osteomyelitis an inflammatory reaction to the infection appears to act through production of inflammatory cytokines and oxygen radical formation to induce epithelial cancers. Infection with Helicobacter pylori leads to epigenetic changes in methylation and infection by a parasite. Clonorchis sinensis also acts as a promoter of cancer of the bile ducts of the liver (cholaniocarcinoma). The common thread among these diverse pathways is that the infections act to alter tissue stem cell signaling with continued proliferation of tumor transit amplifying cells.

Keywords

Cancer stem cells; infection; cancer etiology; cancer signaling

INTRODUCTION

The goal of this chapter is to explore how infections cause cancer via effects on stem cell signaling. The idea that parasitic infections cause cancer is one of the oldest theories of the origin of cancer. Here I will use specific examples to show the relationship of infections to the concept that cancer arises from stem cells as a result of affecting stem cell signaling. It is now well known that many cancers are caused by infections, mainly of viral origin. Viral infections can cause genetic changes by integrating into host DNA (mouse polyoma and mammary tumor viruses); they can cause gene translocations, resulting in constitutive activation of cell signaling pathways for proliferation (Epstein-Barr Virus); they can produce products that take over the signaling pathways of tissue stem cells and allow continued proliferation of transit amplifying cells derived from the stem cells (human papilloma virus), or they can produce tissue injury, resulting in an enhancing or promoting effect on initiated cells (viral hepatitis). In addition, examples are presented in which bacterial infection leads to epigenetic changes associated with development of cancer (*Helicobacter pylori*) or acts as a promoting agent (chronic osteomyelitis). Infection by a parasite, *Clonorchis sinensis* also acts as a promoter of cancer of the bile ducts of the liver (cholaniocarcinoma). For each of

*Address correspondence to this author at the Wadsworth Center and Ordway Research Institute Empire State Plaza, Albany, NY 12201, USA; Tel: 518-474-0547; Fax: 518-473-2900; ssell@wadsworth.org.

these infectious pathways, evidence is presented that tissue determined stem cells are the target of the infection.

INFECTION AND THE STEM CELL ORIGIN OF CANCER

The relationship between infection and the stem cell origin of cancer has been the subject of repeated controversy for several hundred years. In the last 40-50 years, appreciation of the role of stem cells in cancer has resulted in a change in how we think about cancer [1-5]. Prior to the 19th century cancer was believed to arise from an excess of black bile. A view consistent with the humoral theory of disease as espoused by such luminaries as Hippocrates, Celsus, Galen [6], and St. Thomas Aquinas. In the first half of the 19th century, Joseph Claude Anselme Recamier [7] and Robert Remak [8] observed that cancer tissues looked more like embryonic tissues than like adult tissues. That observation led to the embryonal rest theory of cancer proposed by Franco Durante [9] and Julius Cohnheim [10]. The embryonal theory of cancer was a forerunner of the stem cell theory of cancer that would be adumbrated later [1-3]. However, during the latter half of the 19th century, the embryonal rest theory of cancer fell out of favor, to be supplemented by the infectious origin of cancer; cancer was thought to derive from de-differentiation of mature tissues as a result of infection.

The de-differentiation theory of cancer posits that cancers arise from changes in mature differentiated cells, so that the de-differentiated cancer cells do not die and continue to proliferate. A number of observations led to general acceptance of the de-differentiation theory. As late as 1853, Sir James Paget wrote that cancers came from morbid material in the blood, essentially a variation of the black bile theory [11]. Even Rudolf Virchow, the “Father of Pathology” [12], who described embryonic tissues in teratocarcinomas, concluded that cancers arose from adult connective tissue during chronic inflammation. The earlier observation that chimney sweeps developed cancer of the scrotum [13,14], was interpreted to mean that cancers were induced in mature epithelial tissues by chemicals. Others considered cancer to arise from a disequilibrium between connective tissue and epithelium [15], a changed “habit of growth” of normal cells [16,17], or the loss of a restraining environment of the body on displaced tissue cells, leading to de-differentiation of mature cells [18]. Amedee Borrel [19] and a number of other scientists believed that infection by parasites caused cancers; by 1911, Peyton Rous had identified the Rous sarcoma virus as a cause of cancer in poultry [20]. Finally, Theodore Boveri [21] discovered that cancer cells have abnormal chromosome content. Each of these observations was considered to support de-differentiation as the main mechanism of development of cancer. By 1914, Bainbridge, in his authoritative book “The Cancer Problem” [22], concluded “The congenital or embryonic theory of the origin of cancer has received no support whatever from the experimental and comparative investigations of recent times”. De-differentiation remained the dominant theory of the origin of cancer into the 1980s. However, the role of stem cells in cancer was restored by studies on teratocarcinoma and leukemia [1,3], as well as other cancers, that convincingly showed that most cancers, if not all, must arise from stem cells and retained cells with stem-cell properties. For many years, infections agents, such as parasites, were thought to cause cancer, but definitive proof was lacking [19,22].

VIRUSES AND CANCER

The ability to transfer cancers with bacterial and cell free filtrates of tumor material in chickens led to the discovery of Rous sarcoma virus in 1910 [20,23], the first convincing evidence that viruses can cause cancer. However, for over 40 years, that remained an isolated finding. Then, in 1951, Ludwig Gross discovered that mouse leukemias [24], mammary carcinomas [25], and salivary gland carcinomas [26] were also transplantable

using cell free filtrates of fluids from cancerous tissues. A few years later, Eddy and collaborators were able to culture from mice a tumor virus that would cause cancer when injected back into normal mice [27]. This cultured “virus” induced tumors in various organs when transmitted to mice, so these workers named it mouse polyoma virus (MPyV,28).

MOUSE POLYOMA VIRUS

Studies on MPyV and simian virus 40 (SV40) led the way to a new understanding of the genes and gene products responsible for initiation of cancer [29]. Both MPyV and SV40 are small DNA tumor viruses. MPyV rarely causes any adverse effects in the wild, and persists as an endemic harmless infection. Tumor induction was only noted when large amounts of the virus were injected into newborn mice, or into adult mice that were immune deficient [29]. Cells infected *in vitro* with MPyV or SV40 can undergo either lytic infection (the infected cells die) or viral transformation (the infected cells become immortal). In a lytic or permissive infection, the virus replicates in the infected cells, and fully formed infectious viral particles are released when the cells lyse [30]. During transformation infection, the infected cells are stimulated to proliferate by the virus, but the cells are not lysed, and few, if any, viral particles are produced. Thus, transformation can be considered a “failed” infection that stimulates proliferation of the infected cells without formation of complete competent viral particles.

During lytic infection, the virus attaches to the cell, and is endocytosed and transported to the nucleus; there it is uncoated, and the viral DNA integrates into the host genome and is then transcribed [31]. When sera from infected animals were used to identify antigens in the MPyV virus, three viral antigens and a host antigen were identified. The viral antigens were named large T (LT), middle T (MT) and small T (ST); the host antigen was p53, now widely known as a tumor suppressor, but found in mutated form after MPyV infection [32]. Viral transformation of infected cells is largely determined by both the LT and MT antigens, but the effects of the two antigens are quite different [33]. LT converts primary mouse cells into immortal cell lines that are not tumorigenic; whereas MT converts cultured cells into a fully tumorigenic cell line [34]. Thus, the oncogenic function of MPyV is related to the effects of the polyoma MT antigen, PyMT [35]. The transforming activity of PyMT is associated with tyrosine phosphorylation of the product of *c-src* [36], a cellular oncogene. The interaction of PyMT with other proteins in the cell-activation signaling cascade that leads to malignant transformation has been reviewed by Cheung [37]. Compelling experimental models of breast cancer can be are generated if the PyMT oncogene is linked to the mouse mammary tumor (MMTV) virus promoter. Before discussion of the MMTV-PyMT is model, let us consider MMTV and the cellular origin of breast cancer in the mouse.

MOUSE MAMMARY TUMOR VIRUS

MMTV specifically infects mouse mammary cells and produces random interruptions in the somatic DNA via insertion of pro-viral DNA copies into the DNA of the infected cells. In infected mammary glands, hyperplastic lesions containing MMTV-induced mutations are precursors to invasive lesions [38]. The MMTV-induced mutations occur in single long-lived epithelial cells present in normal mammary glands that possess the properties of stem cells, i.e., self-renewal, and the ability to produce divergent epithelial progeny [39]. Genes families commonly affected by MMTV insertion in multiple individual tumors include Wnt, FGF, RSpo, as well as eIF3e and Notch 4 that are known to play essential roles in stem-cell maintenance and behavior [39]. Thus, MMTV acts at the level of mammary stem cells to induce proliferation and immortality.

MMTV PROMOTER-DRIVEN ONCOGENE TRANSGENIC MICE

The MMTV long terminal repeat promoter has been used to create very informative transgenic mouse models for breast cancer [40]. Carcinogenic transgenes are manufactured by linking the MMTV promoter to a selected oncogene. Then, when the MMTV promoter is activated in the developing mammary glands of transgenic mice, the oncogene products will be produced. MMTV-driven oncogenes producing mammary cancers in mice include PyMT, neu/ErbB2, cyclin D1, Cyclin E, Ras, Myc, Int-1, and c-rel [41,42]. Transgenic mice with these genes develop mammary tumors with variable ages of onset, histology, and invasiveness, related to the oncogenic pathways activated by the oncogenes [40,41]. We will now consider the MMTV-PyMT transgenic mouse.

MMTV-PyMT

In the MMTV-PyMT transgenic mouse the mouse mammary tumor virus promoter is linked to the polyoma middle T-antigen oncogene [42]. These mice are available from NIH. The MMTV promoter is activated early during development of the mouse mammary gland epithelium leading to expression of PyMT. Hemizygous female MMTV-PyMT mice develop palpable BCAs within 5 weeks of age and large tumors by 10 weeks of age. Males first develop BCA at about 16 weeks and must be euthanized at about 7 months of age. The BCAs in these mice are similar to ductal or basal breast cell breast carcinomas of humans. They grow very rapidly and have a varied histological appearance including medullary, glandular and ductal morphology. Early multifocal preneoplastic lesions involve the ends of growing ducts [42] where putative breast stem cells are located [1]. Thus, in this, and other mouse models of mammary cancer, the evidence is that activation of the oncogene occurs in the self-renewing mammary stem cell and that proliferation of this cell leads to accumulation of immortal breast cancer cells. Recent evidence indicates that bone marrow stem cells from MMTV-PyMT mice may be able to migrate to the breast and transdifferentiate into breast epithelial cells, which then become cancerous [43]. Thus, the transgenic oncogene is present in all the cells of the transgenic mice, but only becomes activated to produce cancers in breast tissue. Cell lines derived from the early ductal carcinoma in situ lesions of MMTV-PyMT mice are able to be transplanted into mammary fat pads cleared of normal breast tissue of syngeneic recipients and “progress” to invasive cancer with bi-potential for formation of myoepithelial and luminal cells [44]. Invasive cancer in this model most likely derives from a pre-cancer stem cell that is capable of self-renewal and multi-lineage differentiation, without further mutation. What is the evidence, if any, that MMTV is associated with human breast cancer?

MMTV AND HUMAN BREAST CANCER

Circumstantial evidence suggests that viruses such as MMTV, Epstein-Barr virus (EBV) and human papilloma virus (HPV) are involved in causation of human breast cancer [41,45]. Homologous sequences of the env gene of MMTV are found in approximately 40% of human breast cancers, but not in normal breast tissue or other types of cancer [46]. In addition, MMTV-specific RNA sequences have been found in human breast cancer specimens [47]. The poor prognosis of gestational associated breast cancer arises from stimulation of expression of MMTV-like sequences present in the human genome by pregnancy hormones [48]. The MMTV provirus is associated with progesterone receptor-positive advanced breast cancer with p53 mutations, and MMTV has hormone-responsive elements that correlate with enhanced replication and poor prognosis [49]. MMTV infection rapidly expands in cultured human breast cells [50], but productive infection of human cells *in vivo* has not been reported. However, viral particles with 95% homology to MMTV have been isolated from primary cultures of human breast cancer cells; the isolated virus has been

named human mammary tumor virus [HMTV; 51]. Thus, although there is highly suggestive evidence that MMTV is involved in the carcinogenic process for at least some human breast cancers, the evidence remains circumstantial [41,45,49]. We can speculate that integration of the MMTV envelope (env) sequence into mammary stem cells, especially during gestation, is the first step in development of at least some human breast cancers. HPV and EBV have also been indicted as suspects in the causation of human breast cancer, but not convicted [45].

HUMAN PAPILLOMA VIRUS (HPV)

In 1983, Harald zur Hausen and his co-workers in 1983 found that certain types of HPV were associated with cervical cancer [52]. Most of the normal cervical epithelium is made up of terminally differentiated cells containing large amounts of glycogen. HPV infects the proliferation-competent basal stem cells of the cervix. The virus hijacks the metabolism of these cells and directs the infected cells to produce viral proteins. Viral E6 and E7 proteins inhibit tumor suppressor genes, E6 inhibits p53, while E7 inhibits p53, p21, and RB [53]. Thus, the viral products reprogram the cervical stem cells for the purposes of their own proliferation and survival. The infected cells no longer differentiate and fill with glycogen, but rather they proliferate and replace the normal cervical epithelium with immature proliferating cells (cervical intraepithelial neoplasia, or CIN). The cervical cells continue to divide by symmetric rather than asymmetric division and eventually replace the normal cervical epithelium. If the lesion is not treated, the malignant cervical cells invade the basement membrane of the cervix. Fortunately, invasive cancer of the cervix can often be prevented. The development of CIN can be detected through periodic examination of the exfoliated cells of the cervix [Papanicolaou smear, 54]. If immature cells are found, the lesions in the cervix can be identified by failure of the poorly differentiated cells lacking glycogen to be colored blue by iodine. The unstained epithelial tissue can then be removed by superficial surgery, and the future growth of cancer prevented the future this approach may become less necessary with the use of HPV vaccines to immunize against infection; a process that prevents development of cervical cancer. Recently, evidence has been accumulating that a genus of HPV is associated with squamous cell carcinoma of the skin [55].

HEPATITIS VIRUS-ASSOCIATED LIVER CANCER

Other that exposure to aflatoxin, the major risk factor for human liver cancer is hepatitis virus infection [56]. In experimental models designed for the study of these risk factors, hepatitis-associated liver injury acts as a promoter of chemically initiated liver stem cells [57]. The situation with viral hepatitis and human liver cancer is much more complicated. Many lines of epidemiologic evidence connect hepatitis B virus (HBV) and hepatitis C virus (HCV) to human liver cancers, but exactly how these viruses cause cancer in the absence of exposure to a chemical carcinogen remains controversial. There appear to be two pathways for each virus [58,59]. In HBV-associated liver cancer, the HBV viral genome integrates into host DNA potentially resulting in loss of tumor suppression or activation of oncogenes [59]. Alternatively, production of the HBV X-protein causes disruption of cell cycle regulation, signaling, cell adhesion, and apoptosis [60]. Which of these disruptions is actually causative of cancer is not clear. For HCV, the viral DNA does not integrate into the host DNA, but the core HCV protein acts on host-cell mitochrondria to produce oxidative stress, which induces aberrations in cell growth-associated genes [60,61]. Modulation of cellular gene expression also occurs, leading to up-regulation of mitogen-activated protein kinases and activating factor 1 (AP-1), resulting in stimulation of cell proliferation [60,61]. At least four HCV gene products, namely HCV core, NS3, NS4B, and NS5A, can induce

transformation in tissue culture [61]; HCV core and NS5A activate the Wnt-β-catenin cell activation pathway.

The relevant question for the cellular origin of human liver cancer is, at what level of the liver cell hierarchy (stem cell, ductal progenitor cell or hepatocyte) do these combinations of genetic and epigenetic events act? HBV and HCV clearly infect mature liver cells, activate proliferation of otherwise quiescent hepatocytes, inhibit apoptotic pathways, and produce hepatocellular carcinoma (HCC). The fact that mature liver cells can give rise to cancer could be argued to be an example of de-differentiation. However, mature hepatocytes respond to loss [62,63], or injury [64,65] of liver cells by proliferation. Since mature liver cells can proliferate, we argue that virus-induced HCCs represent examples of late maturation arrest of cells in the liver cell lineage [1], as is hypothesized for experimental HCC induced chemically by diethynitrosamine [66,67].

MUTATIONS

Mutations were first proposed as a cause of cancer by Theodor Boveri in 1914, following his identification of abnormal chromosomes in sea urchin embryos that resulted in morphologically distorted mitoses [68]. The direct association with cancer was tenuous, and van Hanesmann later argued that if abnormal chromosomes were present in cancer, they were more likely to be the result of cancer than the cause of it [69]. Abnormal mitoses were seen in many cancers, but not until the identification of specific mutations in leukemia could mutations be definitively linked to the induction of cancer. It would be over 50 years later that the mutation theory of cancer was proved for human leukemia.

LEUKEMIA

Peter Nowell in the 1970s [70] identified a specific gene translocation in two chromosomes of chronic myeloid leukemia (CML) cells, i.e., the Philadelphia chromosome. In the Philadelphia chromosome, the break point cluster region (bcr) on chromosome 22 is repositioned next to a known oncogene (abl) on chromosome 9. The translocation produces a transgene product (bcr-abl) that increases proliferation, blocks apoptosis, and liberates the leukemia cells from their tissue *niche*. Since that discovery, a large number of gene translocations have been identified in leukemia and lymphoma [71,72]. The role played by infections in the translocation process remains unclear for most leukemia. However, in Burkitt lymphoma a translocation appears to be introduced as a result of a virus infection.

BURKITT LYMPHOMA

Burkitt lymphoma was discovered to be strongly associated with infection by the EBV in 1964 [73]. The continued proliferation of cells in Burkitt lymphoma is due to translocation of the immunoglobulin promoter (Ig) next to the powerful oncogene c-myc, with resultant proliferation of B-cells. It is not clear how the viral infection results in this translocation, but an experimental model of it illustrates how activation of the Ig-myc transgene is related to the stage of maturation arrest of the lymphoma.

The experimental model is a transgenic mouse that has the Ig promoter linked to c-myc and bcl2 [74,75]. When the immunoglobulin promoter is activated, there is expression of one gene that increases proliferation (c-myc) and another that blocks apoptosis (bcl2), numbers of lymphoma cells thus increase synergistically. The key point here is the linkage to the Ig promoter. The characteristic property of B-cells is production of immunoglobulin, i.e., activation of the Ig promoter. Although the transgene is present in all of the cells of the transgenic mouse it is only activated in B-cells. Thus, the inserted transgenes produce maturation arrest and proliferation at the B-cell level, i.e., a B-cell lymphoma, even though

the same translocation is present in precursor cells in the B-cell lineage. Although other translocations are found in various myeloid leukemias, viral involvement has not been demonstrated, and the causes such of translocations are unknown [76].

In humans, Burkitt lymphoma is associated with infection of cells in the B-cell lineage with EBV. Infected cells may spontaneously lose EBV plasmids during proliferation and then do not become transformed [77]. EBV is a member of the herpes-virus family and infects more that 90% of human during their lifetimes [78]; 55% of seropositive individuals actively shed virus in the saliva at any given time [79]. EBV DNA does not incorporate into host DNA, but the in the infected cells the virus produces its own DNA polymerase and replicates by usurping host cell mechanisms during mitosis. The activation of the proto-oncogene *c-myc* appears to be the critical genetic mechanism causing Burkitt lymphoma in humans. However, the product of *c-myc* activates both proliferation and apoptosis. Since increased apoptosis offsets c-MYC induced proliferation, infection with EBV is generally symptomless [80]; so another event must produce lymphoma-genesis. How infection with EB virus actually produces such an inhibition of apoptosis remains controversial. Mutations resulting in loss of function of pro-apoptotic proteins such as p53, Rb, or Bim (Bcl-2 family) could be responsible in some cases [81]. Viral EBNA-1 or EBNA3A could also play a role, or EBNAec could prevent apoptosis through repression of BIM [81]. In addition, microRNA clusters, such as EBR1 and EBR2, which are highly expressed in EBV associated tumors, could block apoptosis [82]. Other infections, such as malaria and HIV, can induce polyclonal B-cell activation and could cooperate with EBV to produce lymphoma in some cases [83]. Thus, EBV infection is necessary, but not sufficient, to produce lymphoma. In any case, if the lymphoma is to be maintained, EBV infection must occur at the level of a precursor cell in the B-cell lineage, a cell in which it establishes a lifelong latent infection [84]; then a second event must occur that results in activation and maturation arrest of memory B-cells.

BACTERIAL INFECTIONS

The association of cancer with bacterial infections has been noted many times in the past, but a direct relationship between cancer and bacterial infections has been difficult to delineate.

CHRONIC OSTEOMYELITIS

Chronic osteomylelitis is caused by an infection, usually staphylococcal, in the bone, where host defense mechanisms and antibiotics are unable to reach the established organisms. Approximately 1 in 100 patients with chronic osteomyelitis develops squamous cell carcinoma [85] of the skin where the infection drains. The chronic drainage of purulent exudate leads to degeneration and metaplasia of the epithelialized lining of drainage tracts that extend from the infected site in the bone to the skin surface [86]. The stimulation of the epithelium by inflammatory cytokines over many years appears to produce transformation of the basal stem cells of the skin. This process likely represents an example of “cancerization” whereby a chronic infection produces a field of epithelium with altered expression of tumor suppressor genes, such as p53; such fields have increased probability for occurrence of a second mutation in an ocogene, such as *c-myc* [87-89]. Recently, an epigenetic mechanism has been demonstrated for another cancer associated with bacterial infection, gastric cancer.

DNA METHYLATION AND FIELD CANCERIZATION IN GASTRIC CANCER ASSOCIATED WITH *HELICOBACTER PYLORI* INFECTION

H. pylori infection of the stomach is a major risk factor for development of gastric cancer. Areas of hypermethylation are seen in the gastric mucosa of patients with *H. pylori* infection, and it is in these areas that gastric cancers arise [90-92]. *H. pylori* infection leads to increased methyltransferase activity, with resulting increased methylation of genes in gastric stem cells. It is the infection-associated inflammatory response, rather than *H. pylori* itself, that appears to be responsible for inducing DNA methylation [93]. Using mitochondrial DNA (mtDNA) mutations as a marker of clonal expansion, McDonald [94], showed that mtDNA mutations establish themselves in gastric stem cells and are passed on to all of their differentiated progeny. Thus, by clonal expansion, the mutation is spread to form patches of mutated cells in the mucosa. In this way, the increased methylation caused by *H. pylori* infection is inherited in stem cells and transit amplifying cells, and causes repression of the p53 gene with resultant increased proliferation and loss of apoptosis [91,92]. The identification of both stem cells and transitamplifying cells as targets was based on the observed response to therapy. If the *H. pylori* infection is treated, some of the areas of hypermethylation disappear, whereas others do not [90]. This finding is interpreted as follows. The areas of hypermethylation that disappear represent hypermethylation of transit-amplifying cells. Since these cells turn over rapidly, they are replaced with cells that do not have the increased methyltransferase activity. Thus, active infection is required to maintain hypermethylation. When the *H. pylori* infection cleared by antibiotic treatment, newly formed transit-amplifying cells from non-methylated stem cells will not be hypermethlyated. On the other hand, in areas where stem cells are affected, the self-renewing stem cells continue to give rise to hypermethylated transit-amplifying cells [91,92].

PARASITES

Liver Flukes

Cholangiocarcinoma of the liver occurs in association with infection with liver flukes in endemic areas of infection in Southeast Asia [95,96]. During infection with *Clonorchis sinensis* or *Opisthorchis viverrini*, the organisms live in the bile ducts and stimulate proliferation of bile duct cells [97,98]. Release of cytokines by inflammatory cells during the infection or the presence of growth factors in the secretory-excretory products of the flatworms is believed to stimulate proliferation of biliary progenitor cells [99,100]. In experimental models of this infection in Syrian golden hamsters previously exposed to an hepatocarcinogen, there is rapid development of atypical ductal proliferation, cholangofibrosis, cholantiofibromas and cholangiocarcinomas [97,98]. This response involves activation of small intraportal or ductal “oval” cells [101], believed to be liver stem cells [102,103]. It is proposed that the infection with *C. sinensis* acts as a promoter, i.e., it activates proliferation of ductal stem cells that have been “initiated” by exposure to dimethylnitrosamine [DMN, 98,99]. Without such promotion, exposed animals would be expected to develop hepatocellular carcinomas [104]. Thus this model serves as the prototype whereby infections act as promoters for cancer.

CONCLUSIONS

It is no longer questioned that infections can cause cancer. Some such cancers arise from infection of tissue stem cells by viruses with insertion of viral oncogenes into the host DNA (mouse polyoma virus, mouse mammary tumor virus). In other cancers. the virus does not insert its DNA into the host cells, but rather commandeers the metabolism of the infected cells, so that the cells continue to proliferate and do not differentiate (cervical cancer).

Cytoplasmic EBV virus infection results in a gene translocation (Ig/c-myc) that activates proliferation of infected cells; however, in other cancers an inflammatory reaction to the infection, or the occurrence of additional mutations may be necessary if the infected cells are to acquire the characteristics of cancer (Burkitt lymphoma). Still other cancers are associated with bacterial and parasitic infections. In these cancers the infection acts to promote presumptively initiated cells (stimulate proliferation of mutated cells). The common thread among these diverse pathways is that the infections act to alter tissue stem cell signaling.

Acknowledgments

Supported by NIH R01 grant CA112481

REFERENCES

[1]. Sell S, Pierce GB. Biology of Disease, Maturation arrest of stem cell differentiation is a common pathway for the cellular origin of teratocarcinomas and epithelial cancers. Lab. Invest. 1994; 70:6–21. [PubMed: 8302019]

[2]. Reya T, Morrison SJ, Clarke MF, Weissman IL. Stem cells, cancer, and cancer stem cells. Nature. 2001; 414:105–111. [PubMed: 11689955]

[3]. Sell S. Stem cell origin of cancer and differentiation therapy. CR Oncol. Hematol. 2004; 51:1–28.

[4]. Sell S. Potential gene therapy for cancer stem cells. Curr. Gene Ther. 2006; 6:579–591. [PubMed: 17073603]

[5]. Wicha MS, Liu S, Dontu G. Cancer stem cells, an old idea-a paradigm shift. Cancer Res. 2006; 66:1883–1890. [PubMed: 16488983]

[6]. Shimkin, M. Contrary to *n*ature. NIH, USDOH; 1977.

[7]. Recamier JCA. Recherches sur the traitement du cancer, par la compression méthodique simple ou combinée, et sur l'histoire général de la même maladie. Paris Gabon. 1829; 2

[8]. Remak R. Ein Beitrag zur Entwickelungsgeschichte der Krebshaften geschwulste. Deut. Klin. 1854; 6:70–174.

[9]. Durante F. Nesso fisio-patologico tra la struttura dei nei materni e la genesi di alcuni tumori maligni. Arch. Memorie Osservazioni Chirugia Practica. 1874; 11:217–226.

[10]. Cohnheim J. Congenitales, Quergestreiftes Muskelsarkon der Nireren. Virchows Arch. 1875; 65:64.

[11]. Paget, J. Lectures on surgical Pathology. London: 1853.

[12]. Virchow R. Der Krankhoften Geschwulste. Berlin. Onkologie. 1863; II(Pt 1)

[13]. Pott, P. Chirurgical Observations Relative to the Cataract, the Polypus of the Nose, the Cancer of the Scrotum, the Different Kinds of Ruptures, and the Mortification of the Toes and Feet. Clarke and Collins; London, Hawes: 1775.

[14]. Potter M. Percivall Pott's contribution to cancer research. NCI Monograph. 1963; 10:1–13.

[15]. Thiersch, C. Lepiz. 1865. Der Epithelialkrebs namenlich der Haut. Eine anatomisch kliniche Untersuchung. Mit einen Atlas microscopischer Abbildungen.

[16]. Benecke. Zeigler's Beitruge zur Path. Anat. 1892-1893

[17]. Adami, GA. The principles of pathology. London: 1909.

[18]. Rippert, H. Geschwulstelehre fur Aerzte und Studierende. Bonn: 1904.

[19]. Borrel A. Le problème du cancer. Bull Inst. Pasteur. 1907; 5:497–512. 545, 593-608, 642-662.

[20]. Rous P. A sarcoma of the fowl transmissible by an agent separable from the tumor cells. J. Exp. Med. 1911; 13:397–411. [PubMed: 19867421]

[21]. Boveri, R. Zur Frage der Entstehung maligner Tumoren. Fischer; Jena: 1914.

[22]. Bainbridge, WS. The Cancer Problem. The Macmillan Co; New York: 1914.

[23]. Rous PA. Transmissible avian neoplasm (sarcoma of the common fowl). J. Exp. Med. 1910; 12:696–705. [PubMed: 19867354]

[24]. Gross L. “Spontaneous” leukemia developing in C3H mice following inoculation in infancy, with AK-leukemic extracts, or AK-embryos. Proc. Soc. Exp. Biol. Med. 1951; 76:27–32. [PubMed: 14816382]

[25]. Gross L. The “vertical” transmission of mouse mammary carcinoma and chicken leukemia; its possible implications for human pathology. Cancer. 1951; 3:626–633. [PubMed: 14839617]

[26]. Gross L. A filterable agent, recovered from Ak leukemia extracts, causing salivary gland carcinomas in C3H mice. Proc. Soc. Exp. Biol. Med. 1953; 83:414–421. [PubMed: 13064287]

[27]. Eddy BE, Sell S, Berkeley W. Cytopathogenicity in tissue culture by a tumor virus from mice. Proc. Soc. Exp. Biol. Med. 1958; 98:848–851. [PubMed: 13591363]

[28]. Sell S, Eddy BE. Neoplasms in mice inoculated with a tumor agent carried in tissue culture. J. Nat. Cancer Inst. 1958; 20:1223–1243. [PubMed: 13549981]

[29]. Atkin SJL, Griffin BE, Dilworth SM. Polyoma virus and simian virus 40 as cancer models, history and perspectives. Semin. Cancer Biol. 2009; 19:211–217. [PubMed: 19505648]

[30]. Hurwitz J, Cean FB, Kwong AD, Lee SH. The *in vitro* replication of DNA containing the SV40 origin. J. Biol. Chem. 1990; 265:18043–18046. [PubMed: 1976634]

[31]. Bullock PA. The initiation of simian virus 40 DNA replication *in vitro*. Crit. Rev. Biochem. Mole. Biol. 1997; 32:503–568.

[32]. Ito Y, Brocklehurst JR, Dulbecco R. Virus-specific proteins in the plasma membrane of cells lytically infected or transformed by polyoma virus. Proc. Nat. Acad. Sci. USA. 1977; 74:4666–4670. [PubMed: 73183]

[33]. Rassoulzadegan M, Cowie A, Carr A, Glaichenhaus N, Kamen R, Cuzon F. The roles of individual polyoma virus early proteins in oncogenic transformation. Nature. 1982; 300:713–718. [PubMed: 6294529]

[34]. Land H, Parada LF, Weinberg RA. Tumorigenic conversion of primary embryo fibroblasts requires at least two cooperating oncogenes. Nature. 1983; 304:596–602. [PubMed: 6308472]

[35]. Dilworth SM. Polyoma virus middle T antigen and its role in identifying cancer-related molecules. Nat. Rev. 2002; 2:951–956.

[36]. Eckhart W, Hutchison MA, Hunter T. An activity phosphorylating tyrosine in polyoma T antigen immunoprecipitates. Cell. 1979; 18:925–933. [PubMed: 229973]

[37]. Cheng J, DeCaprio JA, Fluck MM, Schaffhausen BS. Cellular transformation by Simian Virus 40 and murine polyoma virus T antigens. Semin. Cancer Biol. 2009; 19:218–228. [PubMed: 19505649]

[38]. Medina D. Premalignant and malignant mammary lesions induced by MMTV and chemical carcinogens. J. Mammary Gland Biol. Neoplasia. 2008; 13:271–277. [PubMed: 18663563]

[39]. Callahan R, Smith GH. Common integration sites for MMTV in viral induced mouse mammary tumors. J. Mammary Gland Biol. Neoplasia. 2008; 13:309–321. [PubMed: 18709449]

[40]. Cardiff RD, Kenney N. Mouse mammary tumor biology, a short history. Adv. Cancer Res. 2007; 98:53–116. [PubMed: 17433908]

[41]. Taneja P, Frazier DP, Kending RD, Maglic D, Sugiyama T, Kai R, Taneja NK, Inoe K. MMTV mouse models and the diagnostic values of MMTV-like sequences in human breast cancer. Expert Rev. Mol. Diagn. 2009; 9:423–440. [PubMed: 19580428]

[42]. Guy CR, Cardiff RD, Muller WL. Induction of mammary tumors by expression of polyomavirus middle T oncogene, a transgenic mouse model for metastatic disease. Mol. Cell Biol. 1992; 12:954–961. [PubMed: 1312220]

[43]. Guest I, Ilic Z, Ma J, Glinsky G, Sell S. Direct and indirect contribution of bone marrow derived cells to cancer. Int. J. Cancer. 2010; 126(10):2308–2318. [PubMed: 19816927]

[44]. Damone P, Hodgson JG, Chen JQ, Young LJT, Cardiff RD, Borowsky. Breast Cancer Res. 2008; 10:R50. [PubMed: 18522749]

[45]. Amarante MK, Watanabe MAE. The possible involvement of virus in breast cancer. J. Cancer Res. Clin. Oncol. 2009; 135:329–337. [PubMed: 19009309]

[46]. Wang Y, Holland JF, Bleiweiss IJ. Expression of mouse mammary tumor virus-like env gene-like sequences in human breast cancer. Cancer Res. 1995; 55:5173–5179. [PubMed: 7585568]

[47]. Wang Y, Go V, Holland JF. Expression of mouse mammary tumor virus-like env gene sequences in human breast cancer. Clin. Cancer. Res. 1998; 4:2565–2568. [PubMed: 9796992]

[48]. Wang Y, Melana SM, Baker B. High prevalence of MMTV-like env gene sequences in gestational breast cancer. Med. Oncol. 2003; 20:233–236. [PubMed: 14514972]

[49]. Lawson JS, Gunzburg WH, Whitaker NJ. Viruses and human breast cancer. Future Microbiol. 2006; 1:33–51. [PubMed: 17661684]

[50]. Indik S, Gunzburg WE, Salmons B. Rapid spread of mouse mammary tumor virus in cultured human breast cells. Retrovirology. 2007; 4:73. [PubMed: 17931409]

[51]. Melana SM, Nepomnaschy I, Sakalian M. Characterization of viral particles isolated from primary cultures of human breast from the same individuals. Cancer Res. 2007; 67:8960–8965. [PubMed: 17875739]

[52]. Durst M, Gissmann L, Ikenberg H, zur Hausen H. A papillomavirus DNA from a cervical carcinoma and its prevalence in cancer biopsy samples from different geographic regions. Proc. Natl. Acad. Sci. USA. 1983; 80:3812–3815. [PubMed: 6304740]

[53]. Deligeoroglou E, Christopoulos P, Aravantinos L, Papadias K. Human papilloma virus molecular profile and mechanisms of cancerogenesis, a review. Eur. J. Gynaecol. Oncol. 2009; 30:128–132. [PubMed: 19480239]

[54]. Papanicolaou GN. Historical development of cytology as a tool in clinical medicine and in cancer research. Acta Unio Int. Contra Cancrum. 1958; 14:249–254. [PubMed: 13594631]

[55]. Plasmeijer EI, Neale RE, de Koning MNC, Quint WGV, McBride P, Feltkamp MCW, Green AC. Persistence of Betapapillomavirus infections as a risk factor for actinic keratoses, precursor to cutaneous squamous cell carcinoma. Cancer Res. 2009; 69:8926–8931. [PubMed: 19903846]

[56]. Gomaa AI, Khan SA, Toledano MB, Waked I, Taylor-Robinson SD. Hepatocellular carcinoma, epidemiology risk factors and pathogenesis. World J. Gastroenterol. 2008; 14:4300–4308. [PubMed: 18666317]

[57]. Sell S. Mouse models to study the interaction of risk factors for human liver cancer. Cancer Res. 2003; 63:7553–7562. [PubMed: 14633666]

[58]. Lee AT, Lee CG. Oncogenesis and transforming viruses, the hepatitis B virus and hepatocellular carcinoma—the etiopathogenic link. Front. Biosci. 2007; 12:234–245. [PubMed: 17127296]

[59]. Azam F, Koulaouzidis A. Hepatitis B virus and hepatocarcinogenesis. Ann. Hepatol. 2008; 7:125–129. [PubMed: 18626429]

[60]. Koike K. Hepatitis C virus contributes to hepatocarcinogenesis by modulating metabolic and intracellular signaling pathways. J. Gastroenterol. Hepatol. 2007; 22(Suppl.1):S108–111. [PubMed: 17567457]

[61]. Levrero M. Viral hepatitis and liver cancer, the case of hepatitis C. Oncogene. 2006; 27:3834–3847. [PubMed: 16799625]

[62]. Higgins GM, Anderson RM. Experimental pathology of the liver. Restoration of the liver of the white rat following partial surgical removal. AMA Arch. Pathol. 1931; 12:186–202.

[63]. Bucher, NR.; Malt, RA. Regeneration of the liver and kidney. Little Brown and Co; Boston: 1971. p. 1-278.

[64]. Stowell RE, Less CW. Histochemical studies of mouse liver after single feeding of carbon tetrachloride. Arch. Pathol. 1972; 50:519–537.

[65]. Wahl PN, Tandon HD, Bharadwaj TP. Acute carbon tetrachloride hepatic injury. Composite histological, histochemical and biochemical study, Part I. Histological and histochemical studies. Acta Pathol. Microbiol. Scand. 1955; 37A:305–314.

[66]. Sell S. Alpha-fetoprotein, stem cells, and cancer. The Abbot Award Lecture. Tumor Biol. 2008; 2008; 29:161–180.

[67]. Sell S. Stem cells in hepatocarcinogenesis. Cell Sci. Rev. 2006:1742–8130. ISN NO. *3*.

[68]. Boveri, R. Zur Frage der Entstehung maligner Tumoren. Fischer; Jena: 1914. AN AACR SCIENTIFIC LANDMARK

[69]. von Hansemannn D. Das Problem der Krebsmalignität. Zeitschr. Krebsforsch. 1920; 17:172–191.

[70]. Nowell PC. Diagnostic and prognostic value of chromosome studies in cancer. Ann. Clin. Lab. Sci. 1974; 4:234–240. [PubMed: 4533399]

[71]. Rowley JD. Nonrandom chromosomal abnormalities in hematologic disorders of man. Proc. Natl. Acad. Sci. USA. 1975; 72:152–156. [PubMed: 1054491]

[72]. Chalandon Y, Schwaller J. Targeting mutated protein tyrosine kinases and their signaling pathways in hematologic malignancies. Haematologica. 2005; 90:949–968. [PubMed: 15996933]

[73]. Epstein MA, Achong BG, Barr YM. Virus particles in cultured lymphoblasts from Burkitt's lymphoma. Lancet. 1964; 1:702–703. [PubMed: 14107961]

[74]. Cory S, Vaux DL, Harris AW, Adams JM. Insights from Bcl-2 and Myc, malignancy involves abrogation of apoptosis as well as sustained proliferation. Cancer Res. 1999; 597:1681s–1692s. Suppl.

[75]. Park SS, Kim JS, Tessarollo L, Owens JD, Peng L, Han SS, Tae Chung S, Torrey TA, Cheung WC, Plakiewica RD, McNeil N, Ried T, Buchinski JF, Morse HC 3rd, Janz S. Insertion of c-Myc into Igh induces B-cell and plasma-cell neoplasms in mice. Cancer Res. 2005; 65:1306–1315. [PubMed: 15735016]

[76]. Sell S. Leukemia, stem cells, maturation arrest and differentiation therapy. Stem Cell Rev. 2005; 1:197–205. [PubMed: 17142856]

[77]. Cohen JL. Epstein-barr virus infection. N. Engl. J. Med. 2000; 343:481–492. [PubMed: 10944566]

[78]. Yao QY, Rickinson AB, Epstein MA. A re-examination f the Epstein-Barr virus carrier state in healthy seropositive individuals. Int. J. Cancer. 1985; 35:35–42. [PubMed: 2981780]

[79]. Vereide D, Sugden B. Proof of EBV's sustaining role in Burkitt's lymphoma. Semin. Cancer Biol. 2009; 19:389–393. [PubMed: 19628040]

[80]. Klein E, Klein G. Burkitt lymphoma, Editorial. Semin. Cancer Biol. 2009; 19:345–346. [PubMed: 19962087]

[81]. Klein G. Burkitt lymphoma-a stalking horse for cancer research? Semin. Cancer Biol. 2009; 19:347–350. [PubMed: 19607918]

[82]. Allday MJ. How does Epstein-Barr virus (EBV) complement the activation of Myc in the pathogenesis of Burkitt's lymphoma? Semin. Cancer Biol. 2009; 19:366–376. [PubMed: 19635566]

[83]. Bornkamm GW. Epstein-barr virus and its role in the pathogenesis of burkitt's lymphoma. Semin. Cancer Biol. 2009; 19:351–365. [PubMed: 19619654]

[84]. Hochberg D, Middeldorp JM, Catalina M, Sullivan J, Luzuriaga K, Thorley-Lawson DA. Demonstration of the Burkitt's lymphoma Epstein-Barr virus phenotype in dividing latently infected memory cells *in vivo*. Proc. Natl. Acad. Sci. USA. 2004; 101:239–244. [PubMed: 14688409]

[85]. Sedlin ED, Fleming JL. Epidermoid carcinoma arising in chronic osteomyelitis foci. J. Bone Joint Surg. AM. 1963; 45:827–838.

[86]. Sankaran-Kutty M, Corea JR, Ali MS, Kutty MK. Squamous cell carcinoma in chronic osteomyelitis. Clin. Orthop. 1982; 198:264–267. [PubMed: 4028559]

[87]. Slaughter DP, Southwick HW, Smejkal W. Field "cancerization" in oral stratified squamous epithelium. Clinical implications of multicentric origin. Cancer. 1953; 6:963–968. [PubMed: 13094644]

[88]. Carlson JA, Scott D, Wharton J, Sell S. Incidental histopathologic patterns, possible evidence of "Field Cancerization" surrounding skin tumors. Am. J. Dermatopathol. 2001; 223:494–497. [PubMed: 11801788]

[89]. Braakhuis JMB, Tabor MP, Kummer JA, Leemans CR, Brakenhoff RH. A Genetic Explanation of slaughter's concept of field cancerization, evidence and clinical implications. Cancer Res. 2003; 63:1727–1730. [PubMed: 12702551]

[90]. Ushijima T, Nakajima T, Maekita T. DNA methylation as a marker for the past and future. J. Gastroenterol. 2006; 41:401–407. [PubMed: 16799880]

[91]. Ushijima T. Epigenetic field for cancerization. J. Biochem. Mol.Biol. 2007; 40:142–150. [PubMed: 17394762]

[92]. Ushijima T. Detection and interpretation of altered methylation patterns in cancer cells. Nat. Rev. Cancer. 2005; 5:223–231. [PubMed: 15719030]

[93]. Niwa T, Tsukamoto T, Toyoda T, Mori A, Tanaka H, MAekita T, Ichinose M, Tatematsu M, Ushimjima T. Inflammatory processes triggered by Helicobacter pylori infection cause aberrant DNA methylation in gastric epithelial cells. Cancer Res. 2010; 70:1430–1440. [PubMed: 20124475]

[94]. McDonald SA, Greaves LC, Gutierrez-Gonzalez L, Rodriquez-Justo M, Deheragoda M, Leedham SJ, Taylor RW, Preston SL, Lovell M, Hunt T, Wlia G, OUfrif D, Harrison R, Novelli MR, Mitchell I, Stoker DL, Turnbull DM, Jankowski JA, Wright NA. Mechanisms of field cancerization in the human stomach, the expansion and spread of mutated gastric stem cells. Gastroenterology. 2008; 134:500–510. [PubMed: 18242216]

[95]. Hou PC. The relationship between primary carcinoma of the liver and infection with *Clonorchis sinesis*. J. Pathol. Bacteriol. 1956; 72:293–346.

[96]. Rim HJ. Clonorchiasis in Korea. Kor. J. Parasitol. 1990; 28:63–78. Suppl.

[97]. Lee JH, Rim HJ, Bak UB. Effect of *Clonorchis sinesis* and dimethylnitrosamine on the induction of cholangiocarcinoma in syrain golden hamsters. Korean J. Parasitol. 1993; 1:21–30. [PubMed: 8390293]

[98]. Lee JH, Rim HJ, Sell S. Promotion of cholangiocarcinomas by *Clonorchis sinesis* infection in hamsters after induction by dimethynitrosamine. J. Tumor Marker Oncol. 1995; 10:7–16.

[99]. Ohshima J, Bartwch H. Chronic infections and inflammatory processes as cancer risk factors, possible role of nitric oxide in carcinogenesis. Mutation Res. 1994; 305:254–265.

[100]. Haswell-Elkins, Satarug S, Tsuda M, Mairiang E, Esumi H, Sithitaworn PM, Moaiiang P, Saitoh M, Youngvanit P, Elkins DB. Liver fluke infection and cholangiocarcinoma, model of endogenous nitric oxide and extragastric nitrosation in human carcinogenesis. Mutation Res. 1994; 305:241–252. [PubMed: 7510035]

[101]. Lee JH, Rim HJ, Sell S. Heterogeneity of the “oval cell” response in the hamster liver during cholangiocarcinogenesis following *Clonorchis sinensis* infection and dimethylnitrosamine treatment. J. Hepatol. 1997; 26:1313–1323. [PubMed: 9210619]

[102]. Sell S, Salman J. Light- and electronmicroscopic autoradiographic analysis of proliferating cells during the early stages of chemical hepatocarcinogenesis in the rat induced by feeding N-2-fluornylacetamide in a choline deficient diet. Am. J. Pathol. 1984; 114:287–300. [PubMed: 6696045]

[103]. Sell S. Liver stem cells. Modern Pathol. 1994; 7:104–107.

[104]. Sell S. Heterogeneity and plasticity of liver stem cells. Hepatology. 2001; 33:738–750. [PubMed: 11230756]