Chemoprevention of Cancer

Anne S. Tsao, MD; Edward S. Kim, MD; Waun Ki Hong, MD

Dr. Tsao is Medical Oncology Fellow, Division of Cancer Medicine, University of Texas MD Anderson Cancer Center, Houston, TX.

Dr. Kim is Assistant Professor, Director of Educational Programs, Department of Thoracic & Head and Neck Medical Oncology, University of Texas MD Anderson Cancer Center, Houston, TX.

Dr. Hong is Head, Division of Cancer Medicine, Professor/Chair, Department of Thoracic & Head and Neck Medical Oncology, University of Texas MD Anderson Cancer Center, Houston, TX.

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ABSTRACT Cancer chemoprevention is defined as the use of natural, synthetic, or biologic chemical agents to reverse, suppress, or prevent carcinogenic progression to invasive cancer. The success of several recent clinical trials in preventing cancer in high-risk populations suggests that chemoprevention is a rational and appealing strategy. This review will highlight current clinical research in chemoprevention, the biologic effects of chemopreventive agents on epithelial carcinogenesis, and the usefulness of intermediate biomarkers as markers of premalignancy. Selected chemoprevention trials are discussed with a focus on strategies of trial design and clinical outcome. Future directions in the field of chemoprevention will be proposed that are based on recently acquired mechanistic insight into carcinogenesis. (*CA Cancer J Clin 2004;54:150–180.*) © *American Cancer Society, 2004.*

INTRODUCTION

Epithelial carcinogenesis is a multistep process in which an accumulation of genetic events within a single cell line leads to a progressively dysplastic cellular appearance, deregulated cell growth, and, finally, carcinoma. Cancer chemoprevention, as first defined by Sporn in 1976, uses natural, synthetic, or biologic chemical agents to reverse, suppress, or prevent carcinogenic progression.[1] It is based on the concepts of multifocal field carcinogenesis and multistep carcinogenesis. In field carcinogenesis, diffuse epithelial injury in tissues, such as the aerodigestive tract, results from generalized carcinogen exposure throughout the field and clonal proliferation of mutated cells. Genetic changes exist throughout the field and increase the likelihood that one or more premalignant and malignant lesions may develop within that field. Multistep carcinogenesis describes a stepwise accumulation of alterations, both genotypic and phenotypic. Arresting one or several of the steps may impede or delay the development of cancer. This has been described particularly well in studies involving precancerous and cancerous lesions of the head and neck, which focus on oral premalignant lesions (leukoplakia and erythroplakia) and their associated increased risk of progression to cancer. In addition to histologic assessment, intermediate markers of response are needed to assess the validity of these therapies in a timely and cost-efficient manner.

THE BIOLOGIC BASIS OF EPITHELIAL CARCINOGENESIS

Field Carcinogenesis

The concept of field carcinogenesis was originally described for the upper aerodigestive tract in the early 1950s.[2] Here, the surface epithelium, or field, is chronically exposed in large amounts to environmental carcinogens, predominantly tobacco smoke. Multifocal areas of cancer develop from multiple genetically distinct clones (field carcinogenesis) and lateral (intraepithelial) spread of genetically related preinvasive clones.[3] Pathologic evaluation of the epithelial mucosa of the upper aerodigestive tract located adjacent to carcinomas frequently reveals hyperplastic and dysplastic changes. These premalignant changes found in areas of carcinogen-exposed epithelium adjacent to tumors are termed field carcinogenesis and suggest that these multiple foci of premalignancy could progress concurrently to form multiple primary cancers. Second primary tumors (SPTs) are the leading cause of mortality in head and neck cancer. This best illustrates the concept of field carcinogenesis.

Warren and Gates defined SPTs in 1932 as new lesions that can arise either from the same genetically altered "field" as the first tumor or independently from a different clone.[4–7] Multiple genetic abnormalities have been detected in normal and premalignant epithelium of the lung and upper aerodigestive tract in high-risk patients. In limited studies, when primary tumors and SPTs are analyzed for *p53* mutations, evidence supports the independent origin of these tumors. Mutations of *p53* may occur in only one of the tumors, or distinct mutations can occur in the primary and SPT. Multifocal field carcinogenesis effects have been observed in head and neck, lung, esophagus, vulva, cervix, colon, breast, bladder, and skin cancers.[4, 8–16] Continued work in analyzing molecular characteristics of primary and second primary cancers is needed.

Multistep Carcinogenesis

The pathological observations in field carcinogenesis gave rise to the hypothesis of multistep carcinogenesis, which proposes that neoplastic changes evolve over a period of time due to the accumulation of somatic mutations in a single cell line, resulting in phenotypic progression from normal to hyperplastic to dysplastic, and finally, to fully malignant phenotypes.[16–18] Figure 1 illustrates this schematically with respect to lung cancer based on identification of genetic abnormalities in premalignant and malignant epithelial cells.[19] Genetic damage from accumulated carcinogenic exposure becomes evident during neoplastic transformation. Specific genes have been discovered that, when altered, may play a role in epithelial carcinogenesis. These include both tumor suppressor genes and proto-oncogenes, which encode proteins that are involved in cell-cycle control, signal transduction, and transcriptional regulation. These affect different stages of carcinogenesis including initiation, promotion, and progression. Initiation involves direct DNA binding and damage by carcinogens, and it is rapid and irreversible. Promotion, which involves epigenetic mechanisms, leads to premalignancy and is generally irreversible. Progression, which is due to genetic mechanisms, is the period between premalignancy and the cancer and is also generally irreversible. With rare exceptions, the stages of promotion and progression usually span decades after the initial carcinogenic exposure.

CLINICAL AND BIOLOGIC APPROACHES TO PREVENTION

Patient Populations

Primary prevention strategies seek to prevent de novo malignancies in an otherwise healthy population. These individuals may have high-risk features, such as prior smoking histories or particular genetic mutations predisposing them to cancer development. Secondary prevention involves patients who have known premalignant lesions (ie, oral leukoplakia, colon adenomas) and attempts to prevent the progression of the premalignant lesions into cancers. Tertiary prevention focuses on the prevention of SPTs in patients cured of their initial cancer or individuals definitively treated for their premalignant lesions. Chemoprevention trials are based on the hypothesis that interruption of the biological processes involved in carcinogenesis will inhibit this process and, in turn, reduce cancer incidence.[20] This hypothesis provides a framework for the design and evaluation of chemoprevention trials, including the rationale for the selection of agents that is likely to inhibit biological processes and the development of intermediate markers associated with carcinogenesis. When considering which populations to test chemopreventive agents, enrolling patients in the highest-risk subgroups would enhance the efficiency of controlled chemoprevention trials. These populations would be targeted for primary, secondary, and tertiary prevention.

Intermediate Biomarkers

Development of intermediate markers for chemoprevention trials is crucial. Improvements in cancer incidence among populations receiving a chemopreventive intervention may require years to evaluate. Monitoring intermediate markers that correlate with a reduction in cancer incidence would allow a more expeditious evaluation of potentially active chemopreventive agents. Premalignant lesions are a

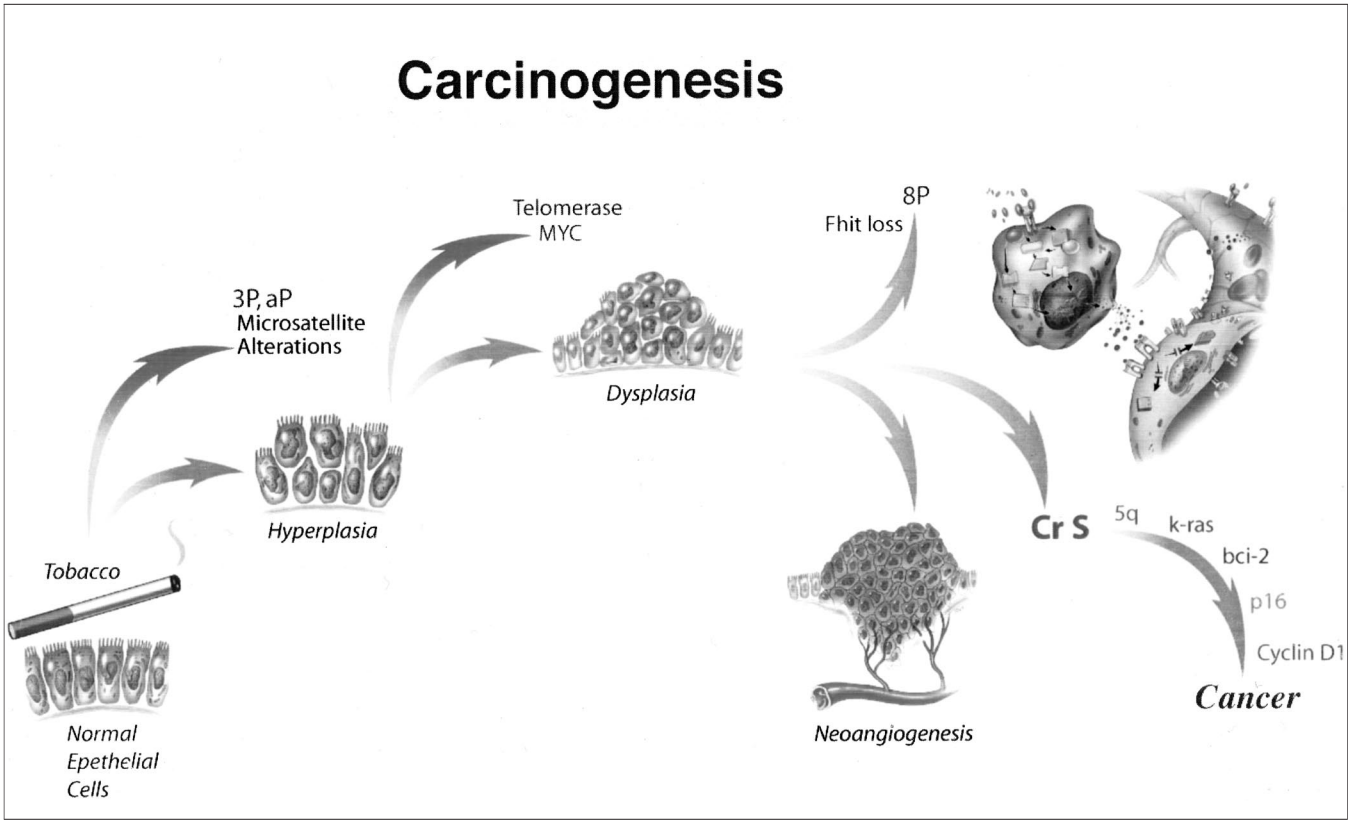

FIGURE **1** Multistep Carcinogenesis Model.
Adapted from Soria JC, Kim ES, Fayette J, et al.[19] with permission from Elsevier.

potential source of intermediate markers. If disappearance of these lesions can be correlated with a reduction in cancer incidence, then markers of premalignancy may serve as intermediate endpoints for chemoprevention trials. One example is intraepithelial neoplasia (IEN). IEN is defined as a noninvasive lesion that has genetic abnormalities, loss of cellular control functions, and some phenotypic characteristics of invasive cancer, and that predicts a substantial likelihood of developing invasive cancer.[21] The American Association of Cancer Research Task Force defined prevention and regression of IEN as being an important clinical trial endpoint. Future studies in chemoprevention will continue to test this hypothesis.

As discussed above, a series of defects occur before the development of frank carcinoma. This can be caused by a variety of factors that will be discussed, including genetic and epigenetic changes in oncogenes and tumor suppressor genes, growth factor imbalances, and dysregulation of other enzymes or targets including the cyclooxygenase pathway, telomerase activity, and the retinoic acid pathway. Alterations in one or several of these factors may expedite the change from normal histology to atypia and cancer. Strategies to prevent these abnormal signals must be developed to delay or detour carcinogenesis (Figure 2).[19]

Genetic Changes During Multistep Carcinogenesis

Genetic susceptibility differences are relevant to the process of multistep carcinogenesis in that, for example, 85% of smokers do not develop aerodigestive tract cancers.[22] Study of genes implicated in activation or detoxification of tobacco carcinogens showed that enzymatic genetic polymorphism such as a high level of, or specific mutations with, *P450* cytochrome activity[23,24] may play a role in the incidence of lung and head and neck cancers. The null genotype of detoxification enzyme glutathione S-transferase (GST) and GSTM1, as an AG or GG genotype of GSTP1, also seems to be a risk factor for lung and

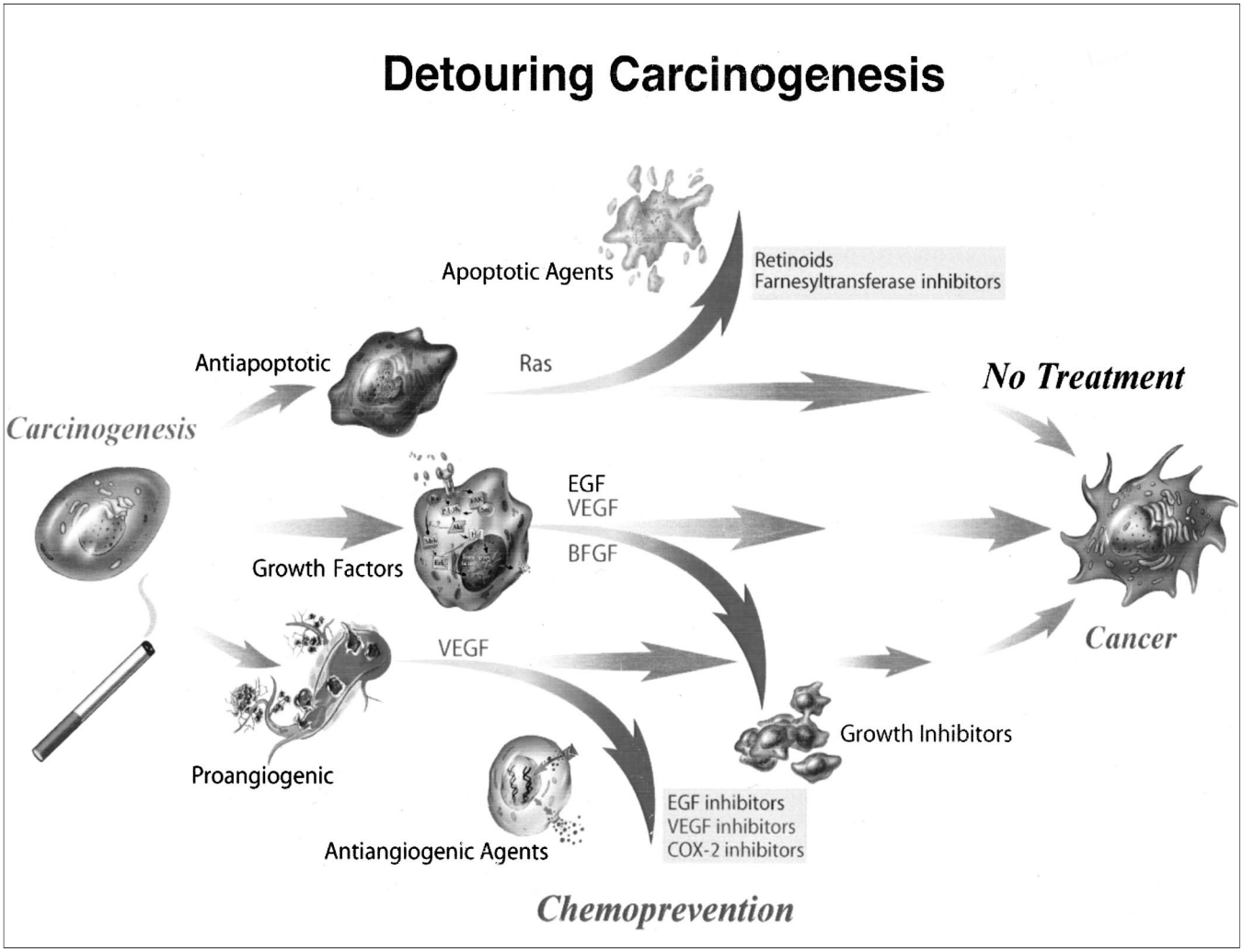

FIGURE **2** Biological Approaches to Preventing Cancer Development.
Adapted from Soria JC, Kim ES, Fayette J, et al.[19] with permission from Elsevier.

head and neck cancers.[25–27] Case-control studies have shown that defective repair of genetic damage, increased sensitivity to mutagens, and sequence variations in DNA repair genes (ie, *XPD*) have been associated with increased susceptibility to lung cancer.[28,29]

Chromosomal abnormalities can occur in tumor cells and also in adjacent histologically normal tissues[30] in a majority of cancer patients. The common chromosomal abnormalities include allelic deletions or loss of heterozygosity (LOH) at sites where tumor suppressor genes map: *3p* (*FHIT* and others), *9p* (*9p21* for $p16^{INK4}$, $p15^{INK4B}$ and $p19^{ARF}$), *17p* (*17p13* for *p53* gene and others), and *13q* (*13q14* for retinoblastoma gene *Rb* and others). Especially important are *3p* and *9p* losses, which have been associated with smoking and are recognized as early events of lung carcinogenesis. They remain detectable many years after smoking cessation.[31] Progression of chromosomal abnormalities parallels the phenotypic progression from premalignant lesion to invasive cancer.[32] Deletions affecting *3p*, *5q*, *8p*, *9p*, *17p*, and *18q* chromosomal regions are among the common changes in epithelial cancers.

Tumor suppressor gene inactivation can be caused by a mutation, loss of chromosomal material (one or two alleles), or methylation. A common tumor suppressor gene, *p53*, acts as a

transcription factor in the control of G_1 arrest and apoptosis. It reduces *Rb* phosphorylation and induces a stop at the G_1-S checkpoint to allow cells to undergo DNA repair or *Bax/Bcl-2*-mediated apoptosis. Its properties are abrogated as a result of mutation or inhibition of *p53* pathway alterations.[33,34] Another region where there is a high prevalence of LOH is *5q*, near the *APC* gene. Although LOH at the *APC* locus occurs, for example, in 80% of dysplastic oral epithelia, 67% of in situ oral carcinomas, and 50% of invasive oral cancers, the tumor suppressor gene located at *5q* has not been identified definitively.[35]

Activation of oncogenes, which drive the cell to multiply and migrate, may be due to genetic modification (mutation, amplification, or chromosomal rearrangement) or to epigenetic modification (hyperexpression). More than 100 oncogenes have been identified to date, and many among them have been implicated in carcinogenesis, including *Ras*, c-*myc*, epidermal growth factor receptor (EGFR, erb-B1), and erb-B2 (HER-2/neu).

The *ras* family of genes encodes 21-kDa proteins, which bind GTP to form a *ras*-GTP complex, which tranduces proliferation signals. Activation of the *ras* genes in *ras*-GTP induces transcription factors C-fos, C-jun, and C-myc and DNA synthesis. Activating *ras* mutations, which are mostly identified at codon 12 of the K-*ras* gene, more rarely at codons 13 and 61, and infrequently in the N- and H-*ras* genes, are induced by tobacco carcinogens such as benzo[a]pyrene and nitrosamine. *Ras* mutations are detected more frequently in adenocarcinomas, large-cell lung carcinomas, and carcinoid tumors rather than squamous cell carcinomas.[36,37]

C-myc plays a necessary role in cellular proliferation triggered by growth factors that act as inducers of proliferation and inhibitors of differentiation. C-myc is also able to induce apoptosis in normal cells through the *p53* pathway, whereas in lung cancer, despite c-myc overexpression, apoptosis is blocked by several deregulators of apoptotic pathways, including Bcl-2. Oncogenic activation of *myc* occurs in 20% of small cell lung carcinoma (SCLC) and 10% of nonsmall cell lung carcinoma (NSCLC) in relation with genetic amplification. Whether L- and N-*myc* are exclusively amplified in aggressive neuroendocrine lung cancer, one of the *myc* genes, C-, L-, or N-, is overexpressed in 45% of NSCLC.[38] Patients with lung cancer present with a high c-myc level in histologically normal or altered lung surgical margins.[39] This suggests that c-*myc* expression is an early event in lung carcinogenesis.

C-erb-B1 (EGFR) and c-erb-B2 (HER-2/neu) are tyrosine kinase receptors both overexpressed in NSCLC and are involved in lung cancer progression. This overexpression is bound to increases of both transcription and translation, with only a low percentage of tumors presenting with gene amplification. C-erb-B1 overexpression has been associated with poor survival rate, advanced stage, poor differentiation, high proliferation index, and increased risk of metastasis.[40] C-erb-B2 (HER-2) overexpression is also a pejorative prognostic factor, especially if associated with a high degree of chemoresistance.[41]

Cyclins *E*, *D1,* and *B1* may be important oncogenes in cancer.[42–44] Cyclin D1 and/or cyclin E overexpression is responsible for deregulation of *Rb* phosphorylation in about 50% of lung carcinomas and is an early event in the preinvasive process; it can be detected by immunohistochemical techniques in half of dysplasias, increasing in frequency with their grade.[45]

Cyclooxygenases (COX) catalyze the synthesis of prostaglandins from arachidonic acid. There are two identified cyclooxygenase enzymes, COX-1 and COX-2. Most tissues express COX-1 constitutively. COX-2 is inducible, and increased levels are seen with inflammation and in many types of cancer. The COX-2 gene is an immediate, early response gene that is induced by growth factors, oncogenes, carcinogens, and tumor-promoting phorbol esters.[46,47] The constitutive isoform is essentially unaffected by these factors.

A large body of evidence from a variety of experimental systems suggests that COX-2 is important in carcinogenesis. COX-2 is upregulated in transformed cells and in malignant tissue.[46–52] In addition to the genetic evidence implicating COX-2 in tumorigenesis, the majority of studies

investigating the role of prostanoids in epithelial malignancy have concentrated on colon cancer and suggest that COX-2 expression and prostaglandin production are crucial to the growth and development of these tumors.[53,54]

Telomeres are highly complex terminal chromosome structures that correct function and are crucial for normal cell survival. Telomerase is the key enzyme stabilizing the telomeres. Telomerase is preferentially expressed in tumor cells with short telomeres and is not expressed in most somatic cells, which usually have longer telomeres. Telomerase is expressed in various epithelial cancers, including in 80% to 85% of NSCLC and in almost all of SCLC.[55,56] Telomerase activity is detected in precancerous lesions of the lung, reflecting the early involvement of the molecule in lung tumorigenesis.[57] Telomerase is a prognostic factor in early-stage NSCLC.[58] Furthermore, telomerase activity has been correlated with cell proliferation, higher tumor-node-metastasis tumor stage, and node invasion.[59]

Retinoids (vitamin A and its analogs) are modulators of differentiation and proliferation of epithelial cells. They are able to invert cancerous progression in the airway by complex mechanisms. These mechanisms essentially depend on the retinoids' capacity to regulate gene expression through nuclear transduction signal modulation mediated by nuclear retinoid receptors. These receptors act as ligand-activated transcription factors. It has been demonstrated that expression of retinoic acid receptor (RAR-β), one of these receptors, is inhibited in early stages of head and neck carcinogenesis (premalignant lesions of the oral cavity and tumors adjacent to dysplastic tissues) and in lung carcinogenesis.[60]

As further biomarkers are studied in epithelial cancers (Tables 1 and 2),[31, 61–112] they will be able to complement the current histologic standard of assessment and response. The following sections will discuss specific tumor types, biomarkers of interest, premalignant development, and clinical trials of chemoprevention.

TABLE 1 Common Biomarkers in Solid Tumors*

p53
EGFR†
PCNA‡
RAS
COX-2§
Ki-67
DNA aneuploidy
DNA polymerase-α

*References 61–83.
†EGFR = Epidermal growth factor receptor.
‡PCNA = Proliferating cell nuclear antigen.
§ = Cyclooxygenase 2.

BREAST CANCER

Breast cancer is a leading cause of morbidity and mortality worldwide. It is estimated in the United States that 217,440 new cases and 40,580 deaths will occur in 2004.[113] The lifetime risk of developing breast cancer is 12.6% for women, and the estimated rate of SPT is 0.8% per year.[114,115] The associated risk factors include older age, higher body mass index, alcohol consumption, hormone replacement, prior radiation exposure, nulliparity, family history, gene carrier status of *BRCA1* and *BRCA2*, and prior history of breast neoplasia.[116–119]

Premalignant Process

There is currently no obligate precursor to invasive breast cancer.[120] The most commonly known benign breast lesions with potential to transform into frank malignancy are atypical ductal hyperplasia, atypical lobular hyperplasia, ductal carcinoma in situ (DCIS), and lobular carcinoma in situ (LCIS).[121,122] Although none of these lesions themselves have invasive or metastatic potential, these lesions have high proliferative rates and have been associated with an increased risk of invasive breast cancer.

Risk Models

There are several proposed risk models for breast cancer. The most commonly used one is the Gail risk model, which was utilized in the National Surgical Adjuvant Breast and Bowel Project (NSABP) trials.[123] The Claus model, which was used in the Cancer and Steroid

TABLE 2 Tumor-specific Biomarkers

Cancer Site	Biomarkers
Breast[69,84]	ER Her2*neu* E-cadherin
Head and Neck[72,83,85–91]	RARβ hTERT *p16*^{INK4a} FHIT (*3p14*) Bcl-2 VEGF-R HPV infection* LOH *9p21* LOH *17p*
Lung[31,92–99]	p-AKT hTERT RARβ *hn*RNP A2/B1 FHIT RAF Myc VEGF-R c-KIT cyclin D1, E, and B1 IGF1 bcl-2 *p16* LOH *3p21.3* LOH *3p25* LOH *9p21* LOH *17p13* LOH *13q* LOH *8p*
Colorectal[70,100–102]	*h*MSH2 APC DCC DPC4 JV18 BAX
Prostate[103–105]	PSA GSTP1 Telomerase
Skin[106]	NF-kB AP1
Cervix[107–111]	D3S2 HPV infection LOH *3p25* LOH *3p14* LOH *4q* LOH *5p*
Bladder[112]	BTA† BTA TRAK‡ Urinary tract matrix protein 22 Fibrin degradation product Autocrine motility factor receptor BCLA-4 Cytokeratin 20 Telomerase Hyaluronic acid Urinary bladder cancer test CYFRA 21-1 Chemiluminescent hemoglobin Hemoglobin dipstick Urinary TPS antigen§ BCA¶ Beta-human chorionic Gonadotropin TPA** Microsatellite analysis

*HPV = Human papilloma virus.
†BTA = Bladder tumor antigen.
‡Manufactured by Alidex, Inc., Redmond, WA.
§TPS = Tissue polypeptide-specific antigen.
¶BCA = Bladder cancer antigen.
**TPA = Tissue polypeptide antigen.

Hormone Study, accounts for both second- and first-degree relatives but not other risk factors. Other models use family history/genetic, reproductive/hormonal, proliferative benign breast pathology, mammographic density,[124] high-risk gene mutations (ie, *BRCA1/2*), and ER+/PR+ status for breast cancers most susceptible for tamoxifen prevention.[125]

Chemoprevention Trials

Breast cancer chemoprevention trials have set the standard for other disease types to follow. This successful research has shown that tamoxifen prevents the development of SPTs and de novo breast cancer in high-risk patients. Tamoxifen is an oral selective antiestrogen agent or SERM (selective estrogen receptor modulator). Its use in breast cancer chemoprevention began with meta-analyses from prior adjuvant trials showing that tamoxifen reduced the rate of contralateral breast cancers by 40% to 50%.[126–130] This effect was observed in women with estrogen receptor positive (ER+) tumors but not in estrogen receptor negative (ER-) tumors. These positive results prompted several large primary chemoprevention trials, including the Breast Cancer Prevention Trial (BCPT) or NSABP P-1 (Table 3).[126, 131–141]

TABLE **3** Selected Breast Cancer Chemoprevention Trials

Trial	Year	Patients (n)¶	Prevention	Population	Endpoint	Compounds*	End Result
Breast Cancer Prevention Trial[131,132]	2000	13,388	Primary	Healthy but positive Gail model risk factors	Breast cancer	Tamoxifen (20 mg)	Positive for ER+† tumors
Royal Marsden Hospital Tamoxifen Chemoprevention Trial[133,134]	1998	2,494	Primary	Healthy volunteers	Breast cancer	Tamoxifen (20 mg)	Negative
Italian Randomized Trial of Tamoxifen[135]	1998	5,408	Primary	Healthy with prior hysterectomies	Breast cancer	Tamoxifen (20 mg)	Positive
International Breast Cancer Intervention Study[136]	2002	7,152	Primary	Healthy but increased risk	Breast cancer	Tamoxifen (20 mg)	Positive
NSABP B-24[137]	2000	1,804	Tertiary	DCIS‡	Breast cancer	Tamoxifen (20 mg)	Positive
NSABP B-14[126]	2001	4,000+	Tertiary	Prior Stage I breast cancer ER+	Breast cancer	Tamoxifen (20 mg)	Positive
Multiple Outcomes of Raloxifene Evaluation (MORE) Trial[138]	2001	7,705	Primary	Postmenopausal women with osteoporosis	Fracture risk, breast cancer	Raloxifene (60 mg)	Positive
Veronesi et al.[139]	1999	2,972	Tertiary	Prior Stage I breast cancer or DCIS	Breast cancer	4-HPR (200 mg)§	Negative
Arimidex, Tamoxifen Alone or in Combination (ATAC) Trial[140]	2003	9,366	Tertiary	Postmenopausal, prior operable breast cancer	Breast cancer	Anastrozole (1 mg) Tamoxifen (20 mg)	Positive
Goss et al.[141]	2003	5,187	Tertiary	Postmenopausal, prior adjuvant tamoxifen therapy for five years	Breast cancer	Letrozole (2.5 mg)	Positive

*Doses are daily regimens unless specified.
†ER+ = Estrogen receptor positive.
‡DCIS = Ductal carcinoma in situ.
§4-HPR = N-[4-Hydroxyphenyl] retinamide.
¶ = Number of patients.

The BCPT (NSABP P-1) was a placebo-controlled trial of tamoxifen in 13,000 women at high risk for breast cancer. This trial was closed early after the interim analysis showed a 49% reduction in incidence of invasive breast cancer in the tamoxifen arm (two-sided, $P < 0.00001$). The BCPT results also confirmed the conclusion from the meta-analysis that only ER+ tumors were affected (69% reduction) by tamoxifen; the incidence of ER- tumors was unaffected. The study reported an increased risk of invasive endometrial cancer and thrombotic events, with women aged 50 and older at highest risk from these complications.[126] Therefore, the conclusions from this trial suggested that the use of tamoxifen in a chemoprevention setting should be highly individualized. The highest level of benefit was seen in patients (mostly premenopausal) with LCIS (relative risk = 0.44) and atypical ductal hyperplasia (relative risk = 0.14).[126] Tamoxifen appeared to reduce the breast cancer incidence in healthy BRCA2 carriers by 62% but did not affect incidence among women aged 35 years or older with BRCA1 mutations.[142] Most additional trials have confirmed the use of tamoxifen in primary prevention. The Italian Randomized Trial of Tamoxifen was a double-blind, placebo-controlled trial with 5,408 healthy women with prior hysterectomies.[135,143,144] After a median follow-up of 81.2 months, women with high-risk features were found to have the most benefit from tamoxifen ($P = 0.003$). The incidence of breast cancer was 0.93% in the tamoxifen arm compared with 4.9% in the placebo arm.[144] Women with low-risk features did not have significant benefit from tamoxifen intervention (1.47% versus 1.52%). The International Breast Cancer Intervention Study 1 enrolled 7,152 healthy women at high risk.[136] After a

median follow-up of 50 months, a risk reduction of 32% was seen with tamoxifen intervention ($P = 0.013$).[136] The International Breast Cancer Intervention Study 1 showed a significant increase in thromboembolic events ($P = 0.001$), especially after surgery.

On the other hand, the Royal Marsden Hospital (RMH) Tamoxifen Chemoprevention trial did not report any benefit of tamoxifen use in healthy women.[134] This trial was a smaller study (n = 2,494) and enrolled patients with strong family histories of breast cancer. The negative results from this trial may be accounted for by the population of tamoxifen-resistant patients enrolled to the RMH trial. The NSABP P1 showed that patients with LCIS and atypical hyperplasia were the most responsive to tamoxifen therapy, and these patients were not studied in the RMH trial. Also, because a strong family history of breast cancer was required for the RMH trial, many women were likely carriers of familial breast cancer genes and may have had an intrinsically different response to estrogen antagonism.[7]

Based on the positive data from the large randomized trials, tamoxifen was approved by the Food and Drug Adminstration (FDA) for use in the primary prevention of breast cancer in high-risk patients. Tamoxifen has also been explored in the secondary and tertiary settings. The NSABP conducted trials in patients with DCIS and in those with resected early-stage breast cancers and reported a positive benefit from using tamoxifen in both settings.[126,137] However, the benefit of tamoxifen remains only in ER+ tumors; no effect on ER- tumors has been shown.

Because tamoxifen increases the risk of endometrial cancer and thromboembolic events, the search for less toxic therapies has looked at other SERMS.[115] The Multiple Outcomes of Raloxifene Evaluation Trial was a multicenter, randomized, placebo-controlled trial evaluating raloxifene, a second generation SERM.[138] Raloxifene has positive estrogenic effects on bone and lipid metabolism and antiestrogenic effects on breast tissue. It doesn't appear to increase risk of endometrial cancer. Although this trial was designed to assess raloxifene's effect on bone density, a 65% reduction in risk of both in situ and invasive breast cancer was observed ($P < 0.001$). Raloxifene is currently being evaluated in the ongoing Study of Tamoxifen and Raloxifene (STAR, or NSABP-P2).[145] Eligibility criteria require inclusion of postmenopausal women with an increased Gail model risk. The treatment arms will receive either 20 mg of oral tamoxifen or 60 mg of raloxifene for five years.

Other agents targeting the estrogen pathway have been investigated and have shown promise in chemoprevention. Aromatase inhibitors prevent estrogen synthesis from androgens and are used in postmenopausal women. Two studies in the tertiary chemoprevention setting are notable. Goss et al. recently reported in an interim analysis that letrozole given for five years after patients with hormone-dependent tumors received definitive treatment and five years of tamoxifen had improved disease-free survival rates ($P \leq 0.001$).[141] The endpoint in this double-blind, placebo-controlled trial included local or metastatic recurrences or new primary cancer in the contralateral breast. An additional agent, anastrozole (Arimidex) is a nonsteroidal aromatase inhibitor and was studied in the Arimidex, Tamoxifen Alone or in Combination trial.[140] In this trial, patients enrolled on the anastrozole arm had longer disease-free survival and fewer primary contralateral breast cancers. In comparison with the tamoxifen arm, there was also a decreased incidence of endometrial cancer ($P = 0.02$), cerebrovascular accidents ($P = 0.0006$), and venous thrombotic events ($P = 0.0006$) but not musculoskeletal disorders ($P < 0.0001$) and fractures ($P < 0.0001$) in the anastrozole arm.

Retinoids are vitamin A derivatives and affect gene expression by modulating nuclear retinoic acid receptors and retinoid X receptors.[86] N-[4-hydroxyphenyl] retinamide (4-HPR, fenretinide) has been studied in women with prior early breast cancer or DCIS. 4-HPR showed benefit in premenopausal women for both contralateral (hazard ratio = 0.66) and ipsilateral (hazard ratio = 0.65) breast cancer.[139,146]

Summary

The FDA's approval of tamoxifen for breast cancer prevention was a landmark achievement that crowned over 20 years of progress in che-

moprevention research. Tamoxifen has demonstrated efficacy in preventing both breast cancer in healthy but high-risk women and SPTs in the adjuvant settings. However, the toxicities of endometrial cancer and thromboembolic events preclude tamoxifen use in certain populations. Several newer agents with potentially less toxicity have shown promise. Studies of second-generation SERMs, aromatase inhibitors (International Breast Cancer Intervention Study II), and retinoids are ongoing in the breast cancer chemoprevention setting. The Study of Tamoxifen and Raloxifene (NSABP-P2) trial will compare tamoxifen to raloxifene in 19,000 postmenopausal women with high-risk factors. Other chemopreventive agents under investigation include luteinizing hormone-releasing hormone agonists in high-risk premenopausal women. Three trials are ongoing that combine the luteinizing hormone-releasing hormone agonist goserelin (Zoladex) with antiosteoporotic agents: raloxifene (RAZOR), tibolone (TIZER), and bisphosphonate ibandronate (GISS).[115] Future studies will also test inhibitors of cyclooxygenase, polyphenol E (green tea extract) with low-dose aspirin, angiogenesis (vascular endothelial growth factor [VEGF]), epidermal growth factor receptors, and ras.

COLORECTAL CANCER

Colon cancer is the third leading cause of cancer-related death in both men and women.[113] Although specific causes of colon cancer are not known, environmental and nutritional factors have been associated with the development of colon cancer. Among these associated risks are diets high in processed meats and low in fruits and vegetables, smoking, and alcohol intake. Stronger, albeit less prevalent, risk factors that are more significant include inflammatory bowel disease and genetic disorders such as familial adenomatous polyposis (FAP) and hereditary nonpolyposis colorectal cancer (HNPCC).

Premalignant Process

In nonheritable colon cancer, at least seven independent genetic events are needed over decades and in the correct order to develop colorectal cancers.[70] This process begins with a normal colonic epithelial cell developing an adenomatous polyposis coli (*APC*) mutation, migrating to the top of the colonic crypt, expanding, and then forming an early adenoma.[147,148] Accumulation of a K-*ras* mutation then promotes intermediate adenoma formation followed by the transition to a late adenoma after mutations on chromosome *18q21* (candidate genes DCC, DPC4, JV18) occur. Mutations in the *p53* gene then transform the premalignant lesion to invasive carcinoma, and other additional genetic hits lead to metastasis.[100]

There are two heritable forms of colon cancer: HNPCC and FAP. In HNPCC, germ line mutations in two genes are commonly found, *h*MSH2 and *h*MLH1.[100] These genes encode for mismatch repair proteins, which when abnormal will lead to genomic microsatellite instability and a two- to three-times higher mutation rate.[101, 149–151] FAP is defined by an autosomal dominant germline mutation in the *APC* gene.[152] Patients with FAP develop hundreds to thousands of adenomatous polyps in the colorectum by their teenage years and colorectal carcinoma by the fourth decade of life.[153]

Chemoprevention Studies

Despite promising data in epidemiologic studies, most dietary changes have not been successful in preventing colorectal cancer (Table 4).[154–163] Specifically, clinical trials have shown no benefit with fiber, beta carotene, vitamin A, C, and E interventions.[160,161,163–166] Other studies suggest that calcium may prevent colorectal carcinoma by binding bile and fatty acids and inhibiting the proliferation of colonic epithelial cells.[167] The Calcium Polyp Prevention Study evaluated calcium carbonate (3 g [1,200 mg of elemental calcium] daily) supplementation in 930 patients for four years and reported a decrease in the recurrence rate of colorectal adenomas (adjusted risk ratio = 0.85; $P = 0.03$).[162] Although calcium supplementation led to a moderate reduction in risk of colorectal adenomas, it remains unclear whether this translates into prevention of invasive colorectal malignancies and a survival benefit.

TABLE 4 Selected Colorectal Chemoprevention Trials

Trial	Year	Patients (n)§	Prevention	Population	Endpoint	Compounds*	End Result
Alpha-Tocopherol Beta Carotene (ATBC) Study[154]	2000	29,133	Primary	Male smokers	Colon cancer	α-tocopherol (50 mg) Beta carotene (20 mg)	Negative
Physician's Health Study[155]	1996	22,071	Primary	Male physicians	Colon cancer	Beta carotene (50 mg every other day) Aspirin (325 mg every other day)	Negative
Giardiello et al.[156]	1993	22	Secondary	FAP†	Polyp regression	Sulindac (150 mg twice a day)	Positive
Steinback et al.[157]	2000	77	Secondary	FAP	Polyp regression	Celecoxib (100 or 400 mg)	Positive
The Aspirin/Folate Polyp Prevention Study[158]	2003	1,121	Tertiary	Prior colorectal adenoma	Recurrence or cancer	Aspirin (81 or 325 mg/day) Folate (1 mg/day)	Positive for aspirin
The Colorectal Adenoma Prevention Study[159]	2003	635	Tertiary	Prior colorectal carcinoma	Adenoma	Aspirin (325 mg/day)	Positive
The Polyp Prevention Trial[160]	2000	2,079	Tertiary	Prior colorectal adenomas	Recurrence	Fiber (18 g/1000kcal)	Negative
Pheonix Colon Cancer Prevention Physician's Network[161]	2000	1,429	Tertiary	Prior colorectal adenomas	Recurrence	Fiber (2 to 13.5 gm)	Negative
The Calcium Polyp Prevention Study[162]	1999	930	Tertiary	Prior colorectal adenomas	Recurrence	Calcium carbonate (3 gm)	Positive
Cascinu et al.[163]	2000	90	Tertiary	Prior Duke's B-C cancer	PCNA‡	Vitamin A (30,000 IU) Vitamin C (1 gm) Vitamin E (70 mg)	Negative

*Doses are daily regimens unless specified.
†FAP = Familial adenomatous polyposis.
‡PCNA = Proliferating cell nuclear antigen.
§ = Number of patients.

Colon cancer prevention has now focused on novel targeted therapies, such as nonsteroidal antiinflammatory agents (NSAIDs). Aspirin, an inhibitor of COX-1 and -2, has been studied in several large randomized studies, but the effect on colorectal cancer prevention is unclear. The US Physician's Health Study, which enrolled 22,071 physicians as participants, reported that aspirin had no effect on the incidence of polyps or colon cancer.[155] However, Baron et al. conducted the Aspirin/Folate Polyp Prevention Study, a randomized, double-blind, placebo-controlled trial of daily aspirin (325 mg and 81 mg) and daily folate (1 mg) in 1,121 patients with a recent history of colon adenomas.[158] This trial demonstrated that the 81-mg dose of aspirin prevented recurrence of colorectal adenomas (47% placebo versus 38% aspirin 81 mg versus 45% aspirin 325 mg; $P = 0.04$). This translated into a relative-risk reduction of 19% in the 81-mg aspirin group and a nonsignificant reduction of 4% in the 325-mg aspirin group. This study also reported a relative-risk reduction of 40% in the 81-mg aspirin group for advanced lesions. Analysis of the folate intervention is ongoing. Also, Sandler et al. reported the Colorectal Adenoma Prevention Study, which randomized 635 patients with prior colorectal cancer to 325 mg aspirin or placebo.[159] Twenty-seven percent of the placebo group developed recurrent adenomas compared with 17% in the aspirin arm ($P = 0.0004$), for an adjusted relative risk of 0.65. Aspirin intervention delayed the development of recurrent adenoma and also decreased the number of recurrent adenomas.

Although the role of aspirin remains debated, the benefit of NSAIDs in chemoprevention has clearly been defined in certain high-risk subgroups. Aspirin and sulindac have been shown to reduce microsatellite instability in HNPCC cell lines carrying *h*MLH1, *h*MSH2, and *h*MSH6 mutations.[168] In clinical trials of patients with FAP, sulindac (150 mg twice a day for nine months) was shown to decrease the number of polyps by 44% and decrease the diameter of the polyps by 35% ($P = 0.014$ and

$P < 0.001$, respectively).[156] In a study from the University of Texas MD Anderson Cancer Center and St. Mark's Hospital, United Kingdom, 77 patients with FAP (more than five polyps 2 mm in size) were randomized to receive placebo, 100 mg, or 400 mg of celecoxib twice daily.[157] Celecoxib is a selective COX-2 inhibitor. Response to treatment was reported as the mean percent change from baseline. After six months, the 30 patients assigned to 400 mg of celecoxib had a 28% reduction in the mean number of colorectal polyps ($P = 0.003$) and a 30.7% reduction in the polyp burden ($P = 0.001$) compared with 4.5% and 4.9% in the placebo group, respectively. This positive result led to the FDA's approval of celecoxib in the treatment of patients with FAP.

Other agents under investigation in colorectal chemoprevention include difluoromethylornithine (DFMO), which irreversibly inhibits ornithine decarboxylase and blocks cell proliferation. Ursodeoxycholic acid reduces the concentration of secondary bile acid deoxycholic acid in the colon and affects arachidonic acid metabolism.[169–171] 3-hydroxy-3-methylglutaryl Coenzyme A reductase inhibitors are usually used in the setting of lowering cholesterol but also have antioxidant antiinflammatory properties and inhibit cell proliferation.[172] Preclinical work in mutant APC murine models have show that sulindac in combination with EGFR inhibitor EKI-785 can decrease intestinal polyps.[173] Almost one-half the mice treated with the combination agents did not develop polyps. With the recent success of bevacizumab, an antibody to the VEGF-receptor in metastatic colorectal cancer, and cetuximab, an antibody to EGFR, further strategies will be applied to prevention.

Summary

Advances in delaying the development of colorectal carcinoma have been shown in patients with FAP with celecoxib treatment. However, the use of COX-2 inhibitors in the primary prevention of sporadic colorectal cancer is being studied in several ongoing trials. Current and future trials using celecoxib alone or in combination with chemotherapy and other biologic therapies are targeting several cohorts, including children with APC mutations, patients with FAP, HNPCC, prior colorectal adenoma, or prior history of sporadic adenomas.[174] The use of celecoxib in the prevention of polyps has resulted in continued efforts to define a high-risk population and to implement a chemopreventive agent in the treatment of cancer. With regard to aspirin use in the prevention of colon adenomas, two large randomized, placebo-controlled trials showed benefit. However, although the Aspirin/Folate Polyp Prevention Study and the Colorectal Adenoma Prevention Study reported positive results, a certain percentage of patients receiving aspirin intervention still developed colon adenomas. This suggests that aspirin use cannot be a substitute for colon surveillance and that further studies are necessary for effective colon cancer chemoprevention.

HEAD AND NECK CANCERS

Head and neck squamous cell cancers (HNSCC) are the sixth most common cancers in the world and are a major cause of significant morbidity. In the United States, 38,530 new cases and 11,060 deaths are estimated for 2004.[113] Advances in locoregional control with combined modality therapy have improved morbidity, but the five-year survival rates have only moderately improved. In patients with definitively treated early-stage or locally advanced tumors, 10% to 40% will develop recurrence or SPTs.[175,176] SPTs occur at a rate of 1.2% to 4.7% per year following the initial therapy. Some of the associated risk factors for HNSCC include tobacco use, betel nut use, alcohol consumption, frequent mouthwash use, and exposure to human papillomavirus (HPV).[177] HPV has been detected in 31% to 74% of oral cancers and is also associated with papillomas, condyloma, verrucous leukoplakia, and carcinoma.[83,178–181]

Risk Models

A standard risk model does not exist for HNSCC, but several have been proposed. We have attempted to study characteristics of to-

bacco intake as a risk model, but the specific genetic changes have been shown to have greater prognostic value. Lee et al. successfully analyzed multiple biomarkers and have been able to predict cancer development in patients with oral premalignancy.[72] In 70 patients with advanced oral premalignancy enrolled on an isotretinoin chemoprevention trial, premalignant histology, prior cancer history, and three biomarkers (chromosomal polysomy, *p53* protein expression, and LOH at chromosome *3p* or *9p*) predicted high risk for cancer development.[182,183] The strongest predictors for malignancy were histology ($P = 0.0003$) and the combined biomarker score of chromosomal polysomy, p53, and loss of heterozygosity ($P = 0.0008$).

Premalignant Process

Oral premalignant lesions or leukoplakia are "predominantly white lesions of the oral mucosa that cannot be characterized as any other definable lesion; some oral leukoplakias will transform into cancer."[184] Leukoplakia occurs in 0.1% to 0.2% of the normal population, and 2% to 3% of these cases develop into carcinoma.[16] The spontaneous regression rate is approximately 30% to 40%. Other more advanced premalignant lesions include erythroleukoplakia and dysplastic leukoplakia. Advanced oral premalignant lesions are associated with a 17.5% overall rate of malignant transformation at eight years for dysplastic lesions.[185] An associated higher risk for malignant transformation is seen with erythroplasia (erythroleukoplakia), verrucous-papillary hyperkeratotic pattern, and being a nonsmoker.

In oral premalignancy, dysplastic tissue has been found to have alterations in *9p*, *3p* and *17p*, indicating that these are "early" events in carcinogenesis.[87] Leukoplakia lesions often contain genetic aberrations such as microsatellite alterations at *9p21* and *3p14*, which predict progression to invasive cancer.[88,186] Frequently, inactivation of $p16^{INK4a}$ has also been shown.[90] Polysomy carries increased risk of development to invasive oral cancer.[89]

Chemoprevention Trials

HNSCC has been one of the most studied tumor types in chemoprevention. Several chemoprevention trials studying different settings have been conducted (Table 5).[182,187–197] There are two major areas of focus: reversal of premalignancy and prevention of SPTs.

Reversal of Premalignancy

Hong et al. reported the first successful randomized, placebo-controlled oral leukoplakia trial in 1986. This trial used high-dose 13-cis retinoic acid (13cRA) for three months and showed a major reduction in size of oral leukoplakia in 67% of patients receiving the retinoid versus 10% of patients receiving placebo ($P = 0.002$).[187] This trial also demonstrated the common toxicities to retinoid therapy as cheilitis, facial erythema, skin dryness, conjunctivitis, and occasionally hypertriglyceridemia. A second trial in patients with oral leukoplakia compared isotretinoin with beta carotene.[182] This trial had two phases, the high-dose isotretinoin (1.5 mg/kg/day) for three months followed by a maintenance phase, in which patients were randomized to beta carotene (30 mg/day) or a low-dose isotretinoin (0.5 mg/kg/day) for nine months. Patients were required to have a response or stable disease before beginning the maintenance phase. This study concluded that low-dose isotretinoin maintenance was significantly more active against leukoplakia than beta carotene (92% versus 45% response or stable disease; $P < 0.001$) in patients who responded initially to high-dose isotretinoin. However, the beneficial effects from retinoid therapy diminished over time.[183] This trial also reported a dose-related toxicity to isotretinoin. In the induction arm, 34% of patients experienced Grade 3 or 4 toxicity compared with only 12% receiving low-dose isotretinoin in the maintenance arm. Toxicity included dry skin, cheilitis, conjunctivitis, and hypertriglyceridemia.[182]

Stich et al. compared 100,000 IU of vitamin A twice weekly with placebo in 65 patients with oral leukoplakia from tobacco or betel nut use.[188] Vitamin A users had higher complete

TABLE **5** Selected Head and Neck Chemoprevention Trials

Trial	Year	Patients (n)§§	Prevention	Population	Endpoint	Compounds*	End Result
High-risk Patients with Oral Leukoplakia							
Hong et al.[187]	1986	44	Secondary	Oral leukoplakia	Response	Isotretinoin (1–2mg/kg)	Positive
Lippman et al.[182]	1993	70	Secondary	Oral leukoplakia	Response	Isotretinoin (1.5 mg/kg)† Beta carotene (30 mg)	Positive‡
Stich et al.[188]	1988	65	Secondary	Oral leukoplakia from tobacco or betel nut use	Response	Vitamin A (100,000 IU) twice weekly	Positive
Chiesa et al.[189]	1993	137	Secondary	Oral leukoplakia	Recurrence	4-HPR (200 mg)§	Positive
Han et al.[190]	1990	61	Secondary	Oral leukoplakia	Response	4-HPR (40 mg)	Positive
Adjuvant Trials							
Hong et al.[191]	1990	103	Tertiary	Prior HNSCC	Recurrence SPT Survival	Isotretinoin (50 to 200 mg/m^2)	Positive
EUROSCAN[192]	2000	2,592	Tertiary	Prior lung or HNSCC¶	SPT Survival	Retinyl palmitate§ (300,000 IU)** N-Acetylcysteine (600 mg)	Negative††
Bolla et al.[193]	1994	316	Tertiary	Prior early-stage oral/oropharynx cancer	SPT‡‡ Survival	Etretinate (50 mg for one month, then 25 mg for 24 months)	Negative
NCI C91-002[194]	2003	1,384	Tertiary	Prior Stage I–II HNSCC	Recurrence SPT Survival	Isotretinoin (30 mg)	Negative
Biochemoprevention for Advanced Premalignant Lesions							
Papadimitrakopoulou et al.[195]	1999	36	Secondary	Advanced dysplasia	Response	Interferon-α (3 MU/m^2 twice weekly) α-Tocopherol (1200 IU) Isotretinoin (100 mg/m^2)	Positive for laryngeal lesions but not oral
Shin et al.[196,197]	2001	44	Tertiary	Prior head and neck cancer	Survival	Interferon-α (3 MU/m^2 three times weekly) α-Tocopherol (1200 IU) Isotretinoin (50 mg/m^2)	Positive

*Doses are daily regimens unless specified.
†Isotretinoin was given as 1.5 mg/kg for three months followed by randomization to maintenance treatment with daily beta carotene (30 mg) or isotretinoin (0.5 mg/kg).
‡In patients who responded to induction isotretinoin, low-dose maintenance isotretinoin conferred a 92% response rate compared with only 45% in the beta carotene arm ($P < 0.001$).
§4-HPR = N-[4-Hydroxyphenyl] retinamide.
¶HNSCC = Head and neck squamous cell cancer.
**Patients received 300,000 IU of retinyl palmitate for 12 months, then were decreased to 150,000 IU for another 12 months.
††EUROSCAN enrolled 60% patients with head and neck cancer and 40% with lung cancer who were treated surgically for their primary tumors. Both disease types had negative results.
‡‡SPT = Second primary tumor.
§§ = Number of patients.

remissions (57% versus 3%) and no progression of their lesions when compared with placebo (0% versus 21%). Stitch et al. also showed that beta carotene combined with retinol led to higher response rates than beta carotene alone.[198] Han et al. reported a randomized trial in 61 patients with oral leukoplakia receiving 4-HPR (40 mg/day orally and 40 mg/day topically) or placebo for four months. The 4-HPR arm had an 87% complete response compared with 17% in the placebo arm ($P < 0.01$).[190] Chiesa et al. randomized patients who received laser resection of oral leukoplakia to receive adjuvant 4-HPR (200 mg/day) or placebo for

one year.[189] An 18% failure rate (local relapse or new lesion) was seen in the fenretinide arm compared with 29% in the placebo-control arm ($P = 0.01$). Other nonretinoid studies have been conducted. Benner et al. performed a nonrandomized Phase II trial using α-tocopherol in patients with oral leukoplakia. Twenty patients (47%) had a clinical response, with nine (21%) showing histologic effect.[199]

Prevention of SPTs

Hong et al. performed a randomized, placebo-controlled chemoprevention trial of high-dose 13-cRA (50 to 100 mg/m^2/day for one year) in 103 patients with a prior HNSCC (larynx, pharynx, or oral cavity).[191] At a median follow-up of 32 months, fewer SPTs were seen in the high-dose 13cRA-treated patients compared with placebo (4% versus 24%; $P = 0.005$). This preventive effect for aerodigestive SPTs also persisted after the one-year intervention. At 54.5 months follow-up, the isotretinoin effect compared with placebo was 14% versus 31% SPT, respectively ($P = 0.042$), with greater preventive benefit in SPT of the aerodigestive tract ($P = 0.008$).[200] However, overall survival was not significantly different between the two arms (57% versus 52%; $P = 0.39$), and the annual SPT rate was also similar. Based on the compelling results from the Hong et al. trial, an effort to reduce toxicity with a lower dose of isotretinoin was initiated. This trial (NCI C91–002) randomized 1,218 patients with prior HNSCC to low-dose 13cRA (30 mg/day) for three years versus placebo. Although the interim analysis was promising, the report at the American Society of Clinical Oncology 39th Annual Meeting in 2003 indicated that low-dose 13cRA did not have an impact on SPT rates but may delay recurrence.

Additional trials have studied retinoids alone and in combination with other agents. EUROSCAN enrolled 2,592 patients definitively treated for their primary tumors (60% head and neck cancer, 40% lung cancer) and randomized them to receive retinyl palmitate, N-acetylcysteine, both agents, or placebo for two years. This study did not show any survival benefit or decrease in SPT with the agents in either disease type.[192] Bolla et al. compared etretinate with placebo in 316 patients with prior early-stage squamous cell cancers of the oral cavity or oropharynx and reported no difference in five-year survival rate, disease-free survival rate, or in SPT rates.[193]

Biochemoprevention

Advanced premalignant lesions have a high risk of transformation to malignancy as well as resistance to single-agent retinoid therapy. Biochemoprevention, which combined retinoids with interferon (IFN) and α-tocopherol,[195] was therefore designed to target this group. Papadimitrakopoulou et al. conducted a nonrandomized clinical trial in 36 patients with advanced premalignant lesions using IFN-α, α-tocopherol, and 13cRA for one year.[195] Biochemoprevention prevented laryngeal lesions but had no effect on oral cavity lesions ($P = 0.009$). From biopsy specimens at different time points in this trial, it was discovered that patients with high p53 expression had lower complete response rates ($P = 0.04$) and higher disease progression rates ($P = 0.02$) than patients with low p53 expression.[196] Based on this study, another trial using biochemoprevention induction therapy for one year followed by two years of maintenance fenretinide or placebo is underway.

In another biochemoprevention trial, patients with prior HNSCC were given one year of IFN-α, α-tocopherol, and 13cRA.[197] At a median of 24 months, 86% of patients had completed treatment and only 14% had developed recurrent disease. Only one patient had developed an SPT (acute promyelocytic leukemia). Overall survival at one year and two years was 98% and 91%, respectively. The toxicity seen in this trial included fatigue (40% of patients), mild to moderate mucocutaneous side effects, flu-like symptoms (arthralgia or myalgia, transient fever, or headache), anorexia, weight loss, peripheral neuropathy (11% of patients), and hypertriglyceridemia (30% of patients). Although minor hematologic side effects were seen, no patients required transfusions or growth factor support. This study suggested that biochemoprevention is a feasible

adjuvant therapy. A randomized trial is underway to confirm these results.

Summary

It is thought that patients with head and neck premalignant changes consist of a diverse population and should be treated differently depending on their molecular genotype.[201] Patients with minimal genetic changes may be treated with single-agent retinoids or other agents. Those with more accumulated genetic changes will require combination chemoprevention therapies. Lesions that have advanced genetic changes with mutant *p53* may benefit from targeted *p53* therapy, and those lesions that express EGFR and COX-2 may require inhibitors of EFGR and COX-2.[201] Other novel strategies include the oncolytic adenovirus dl1520 (ONYX-015), which selectively targets p53-deficient cells.[202] Ongoing trials and future strategies include studying EGFR inhibitors, VEGF-R inhibitors, demethylating agents, farnesyltransferase inhibitors, celecoxib, vitamin E, and Bowman-Birk inhibitors.[203]

LUNG CANCER

Lung cancer is the leading cause of cancer death in the world and is one of the most preventable diseases. For the year 2004, 173,770 new cases and 160,440 deaths are anticipated in the United States.[113] Primary intervention with smoking cessation is a major priority, especially in teenagers and young adults. As the population of former smokers is markedly growing, chemopreventive agents are needed. Risk factors for development of lung cancer include smoking; exposure to radon, polycyclic aromatic hydrocarbons, nickel, chromate, arsenic, asbestos, chloromethyl ethers, and benzo[a]pyrene; radiation exposure; and chronic obstructive pulmonary disease with airflow obstruction. There is no current standard screening modality, as there is not sufficient evidence indicating that the practice improves mortality.[204,205] Ongoing studies in new techniques, including autofluorescence bronchoscopy, molecular markers in sputum analysis, and low-dose spiral computed tomography, are under investigation.[204] Risk models for this disease are needed, but, to date, none are used as the standard of care.

Premalignant Process

Multiple genetic mutations are needed to develop an invasive lung cancer.[82,206–210] Defining genetic susceptibility in lung cancer is difficult because there are no well-documented familial syndromes, a chromosomal translocation break point, or a gatekeeper gene. The most common site of chromosomal allelic loss is *3p21.3*, although additional areas between *3p12* and *3p25* and areas on *3q* are also highly deleted in lung cancer.[211]

A variety of premalignant processes lead to the different histologies in NSCLC. The World Health Organization reports three separate preinvasive neoplastic lung lesions: squamous dysplasia and carcinoma in situ (CIS), atypical adenomatous hyperplasia (AAH), and diffuse idiopathic pulmonary neuroendocrine neoplasia.[212] In squamous cell carcinoma (SCC), the multistep process is suspected to be similar to that of head and neck but arises from stem cells located in the bronchial mucosa. In adenocarcinoma, the AAH lesion precursor cells are not clearly defined. The origin of small cell carcinoma also remains unknown, but there is some speculation that this disease may arise de novo from the bronchial epithelium without a precursor lesion.[213]

Chemoprevention Trials

In primary prevention trials, three large studies demonstrated that neither α-tocopherol nor beta carotene had a preventive effect in lung cancer (Table 6).[192,214–223] The Alpha-Tocopherol Beta Carotene Study enrolled 29,133 male smokers (smoked five or more cigarettes a day) in Finland and treated patients with four daily regimens: α-tocopherol, beta carotene, both α-tocopherol and beta carotene, or placebo. After five to eight years of follow-up, none of the treatment arms showed benefit, and, in fact, patients who received beta carotene supplementation (either as a single agent or combined) had an 18% increase in lung cancer incidence and 8% more overall

TABLE 6 Lung Cancer Chemoprevention Trials

Trial	Year	Patients (n)¶¶	Prevention	Population	Endpoint	Compounds*	End Result
Alpha-Tocopherol Beta-carotene (ATBC) Study[214]	1994	29,133	Primary	Male smokers	Lung cancer	α-Tocopherol (50 mg) Beta carotene (20 mg)	Negative†
Physician's Health Study[215]	1996	22,071	Primary	Male physicians	Lung cancer	Beta carotene (50 mg every other day) Aspirin (325 mg every other day)	Negative
Beta-carotene and Retinol Efficacy Trial (CARET)[216]	1996	18,314	Primary	Smokers Asbestos exposure	Lung cancer	Retinyl palmitate (25,000 IU) Beta carotene (30 mg)	Negative‡
Arnold et al.[217]	1992	150	Secondary	Smokers	Sputum atypia	Etretinate (25 mg)	Negative
Lee et al.[218]	1994	86	Secondary	Smokers	Bronchial biopsy metaplasia index	Isotretinoin (1 mg/kg)	Negative
McLarty et al.[219]	1995	755	Secondary	Asbestos workers	Sputum atypia	Beta carotene (50 mg) Retinyl palmitate (25,000 IU)	Negative
Kurie et al.[220]	2000	82	Secondary	Smokers	Bronchial biopsy dysplasia, RARβ¶	4-HPR (200 mg)§	Negative
Lam et al.[221]	2002	112	Secondary	Smokers	Sputum and bronchial dysplasia	ADT (25 mg three times a day)**	Positive
Pastorino et al.[222]	1993	307	Tertiary	Prior Stage I NSCLC	SPT†† Survival	Retinyl palmitate (300,000 IU)	Positive for SPT but negative for survival
Intergroup Study 91025[223]	2001	1,166	Tertiary	Prior Stage I NSCLC	SPT	Isotretinoin (30 mg)	Negative
EUROSCAN[192]	2000	2,592	Tertiary	Prior lung or head and neck cancer	SPT Survival	Retinyl palmitate (300,000 IU)‡‡ N-Acetylcysteine (600 mg)	Negative§§

*Doses are daily regimens unless specified.
†In the ATBC trial, beta carotene was found to increase the incidence of lung cancer in male smokers for an overall harmful effect.
‡In the CARET, a 28% higher rate of lung cancer incidence and 17% higher overall death rate were seen in the beta carotene and retinyl palmitate arm.
§4-HPR = N-[4-Hydroxyphenyl] retinamide.
¶RARβ = Retinoic acid receptor beta.
**ADT = 5-[*p*-Methoxyphenyl]-1,2-dithiole-3-thione; anethole dithiolethione.
††SPT = Second primary tumor.
‡‡Patients received 300,000 IU of retinyl palmitate for 12 months, then were decreased to 150,000 IU for another 12 months.
§§EUROSCAN enrolled 60% head and neck cancer and 40% lung cancer patients who were treated surgically for their primary tumors. Both disease types had negative results.
¶¶ = Number of patients.

deaths.[214] In the Physician's Health Study, 22,071 male physicians from the United States were randomized to receive beta carotene or placebo on alternate days and aspirin or a placebo on alternate days. This trial was monitored for 12 years and showed no benefit or harm from beta carotene on cancer incidence or overall mortality rate.[215] The Beta-carotene and Retinol Efficacy Trial enrolled 18,314 patients with at least a 20-pack-per-year smoking history or extensive occupational exposure to asbestos (n = 4,060 men).[216] This trial randomized patients to receive the combination of daily beta carotene and retinyl palmitate or placebo. After an interim analysis was performed, a 28% higher rate of lung cancer incidence and 17% higher overall death rate was seen in the beta carotene and retinyl palmitate arm.[216] The trial was concluded due to the potential harmful effect.

Secondary lung cancer prevention trials have focused on surrogate endpoints. Unfortunately, these trials have mostly been negative for any beneficial effect (Table 6). Arnold et al. found no effect of six months of etretinate therapy on sputum atypia in 150 smokers with a greater than 15-pack-per-year smoking history.[217] Lee et al. randomized 86 heavy smokers with bron-

chial dysplasia or metaplasia to isotretinoin or placebo for six months but found no benefit in the metaplasia index.[218] McLarty et al. found no benefit to beta carotene with retinol on sputum atypia of former asbestos workers.[219] Kurie et al. looked for reversal of premalignant bronchial histology in 82 patients treated with 200-mg daily 4-HPR for six months.[220] In this study, no effect on bronchial squamous metaplasia or dysplasia was seen in the current smokers nor was there any effect on the genetic (LOH at chromosome *3p*, *9p*, or *17p*) or phenotypic (mRNA levels of retinoic acid receptor beta) markers.[220] Soria et al. also conducted a negative randomized trial of six months of daily 4-HPR therapy compared with placebo.[85]

In secondary prevention, one randomized trial has shown positive results. In this trial, 112 smokers were treated with six months of 25-mg anethole dithiolethione (5-[*p*-methoxyphenyl]-1,2-dithiole-3-thione) three times a day or placebo.[221] Sputum samples and autofluorescence bronchoscopy were performed pretherapy and posttherapy. In the patients with anethole dithiolethione therapy, a 19% lower rate of progression of dysplastic lesions was seen, with a 21% increase in complete response rate, as defined by histopathologic grade and nuclear morphometry index.

In the tertiary prevention setting, only one of three controlled trials reported a beneficial effect. Pastorino et al. randomized 307 patients with resected Stage I NSCLC to daily 300,000 units of retinol palmitate or placebo for 12 months.[222] The median follow-up was 46 months and showed 18 SPTs in the treatment arm compared with 29 in the control arm. While the rate of SPT was decreased, the estimated survival rate at five years was not significantly different (62% versus 54%; $P = 0.44$). The Intergroup Study 91025 (NCI 191–0001) randomized 1,166 patients with Stage 1 NSCLC to 30-mg daily isotretinoin or placebo.[223] There was no difference between the isotretinoin or placebo arms in either SPT or survival rates. In fact, this study showed an adverse effect in smokers who received retinoids. The EUROSCAN trial, as discussed in the Head and Neck Cancer section of this article, did not show any difference in overall or event-free survival in the treatment arms.[192] A nonstatistically significant lower incidence of SPT was seen in the arm that received no intervention.[192] However, this trial did not differentiate between current and former smokers.

Summary

Currently, there are no chemoprevention agents that have clearly shown clinical benefit in lung cancer. There are several ongoing trials in secondary and tertiary prevention. At the University of Texas MD Anderson Cancer Center, a large trial with celecoxib is underway in current and former smokers. This trial utilizes a surrogate endpoint of reversal of bronchial histology. In the adjuvant setting, the Intergroup E5597 is using daily 200-mcg selenium in 1,960 patients with resected early-stage lung cancers, and the Lung Cancer Biomarkers Chemoprevention Consortium is planning to study gefitinib, an approved agent for NSCLC, in a Phase IIB multicenter trial. The Lung Cancer Biomarkers Chemoprevention Consortium trial is utilizing surrogate endpoints of reversal of bronchial premalignant lesions and Ki-67 levels. Additional studies of adjuvant therapy in high-risk groups such as in early-stage NSCLC and HNSCC are needed. MD Anderson Cancer Center, in cooperation with selected centers through the Department of Defense Grant mechanism, plans on initiating a clinical program called the Vanguard Trial of Investigational Therapeutics in the Adjuvant Treatment of Lung Cancer. With the implementation of novel agents in the treatment of advanced NSCLC and improved safety profiles, further studies in the chemoprevention setting will continue.

BLADDER CANCER

Bladder cancer is the fourth most common cancer in the United States. For the year 2004, it is estimated to account for 60,240 new cases and 12,710 deaths.[113] In most cases, bladder cancer will present as a superficial transitional cell carcinoma that is easily

resectable. However, high local recurrence rates are seen (66% at five years and 88% at 15 years), and approximately 10% to 30% will progress to invasive cancer.[105,112] Screening for bladder cancer requires cystoscopy and the use of urine tumor markers.[112] Risk factors for bladder cancer include cigarette smoking, lower levels of vitamin A intake, infrequent milk and carrot intake, infrequent consumption of cruciferous vegetables, low serum carotene and retinal levels, aromatic amines from rubber or paint occupational exposure, schistosomiasis, and chronic bladder infections.[224–226]

Premalignant Process

Bladder cancer can develop from a low-grade, highly recurrent superficial papillary lesion or as a high-grade flat CIS lesion. In the low-grade tumors, abnormalities on chromosome 9 have been reported to be an initiating event. There are currently no identified gatekeeper candidate genes for the high-grade lesions. The World Health Organization defined several categories for flat urinary bladder lesions: reactive atypia, atypia of unknown significance, dysplasia, and CIS. The classification is based on the growth pattern (papillary or flat) and cytologic and architectural changes, with dysplasia considered as low grade and CIS as high grade.[227]

Chemoprevention Trials

Bladder cancer chemoprevention trials have often focused on nutritional supplementation (Table 7).[228–233] In a primary prevention study, Shibata et al. followed 11,580 retirement community residents who were cancer-free at enrollment. At eight years, an inverse relationship between vitamin C supplement use and bladder cancer risk was seen.[228] However, studies in retinoid and vitamin B6 therapy have been conflicting (Table 7). The National Bladder Cancer Collaborative Group A found no benefit in using 13cRA in patients with rapidly recurring bladder cancer, yet other studies using etretinate, a synthetic retinoid, were shown to decrease the recurrence rates and lengthen the mean time interval to tumor recurrence in superficial papillary bladder tumors.[230,234,235] Another study showed that 4-HPR can reverse abnormal cytology in patients with suspicious or positive flow cytometry.[236] Vitamin B6 was reported in an early randomized study[231] to

TABLE 7 Selected Bladder Cancer Chemoprevention Trials

Trial	Year	Patients (n)§	Prevention	Population	Endpoint	Compounds*	End Result
Shibata et al.[228]	1992	11,580	Primary	Healthy elderly	Bladder cancer	Vitamin C (dietary)	Positive
National Bladder Cancer Collaborative Group[229]	1992	20	Tertiary	Prior T_{a-1} superficial bladder cancer	Bladder cancer	13cRA (0.5 to 1 mg/kg)	Negative
Studer et al.[230]	1995	90	Tertiary	Prior T_{a-1} superficial bladder cancer	Bladder cancer	Etretinate (25 mg)	Positive
Byar et al.[231]	1977	121	Tertiary	Prior Stage I bladder cancer	Bladder cancer	Pyridoxine (25 mg)	Negative
EORTC Genito-Urinary Cooperative Group[232]	1995	291	Tertiary	Prior T_{a-1} superficial bladder cancer	Bladder cancer	Pyridoxine (20 mg)	Negative
Lamm et al.[233]	1994	65	Tertiary	TCC bladder cancer† Receiving I-BCG‡	Bladder cancer	Vitamin A (40,000 IU) Vitamin B_6 (100 mg) Vitamin C (2000 mg) Vitamin E (400 U) Zinc (90 mg)	Positive

*Doses are daily regimens.
†TCC = Transitional cell carcinoma.
‡I-BCG = Intra-vesicle bacillus Calmette-Guérin.
§ = Number of patients.

decrease tumor recurrence rates in patients with Stage I bladder cancer; however, later trials showed no benefit.[232]

Combinations of high doses of vitamins were reported to have a beneficial effect on preventing superficial and low-grade bladder tumor recurrence.[233] Sixty-five patients with prior bladder cancer were randomized to the recommended daily allowance of multiple vitamins or to the recommended daily allowance plus 40,000 IU of vitamin A, 100 mg of vitamin B6, 2,000 mg of vitamin C, and 400 IU of vitamin E.[233] Although the first 10 months showed no difference in time to recurrence, the five-year estimates favored the megavitamin arm ($P = 0.0014$). Recurrence rates were 80% in the control arm compared with 40% in the megavitamin arm ($P = 0.0011$). Further confirmation of this study is needed before using this approach.

Summary

Bladder cancer chemoprevention trials demonstrate conflicting findings. Although some trials using dietary vitamin C in healthy patients and megavitamins and etretinate in the adjuvant setting have been positive, further confirmation is needed before accepting this into practice. Dietary fat, soy protein, garlic, and selenium have been reported to have anticancer properties in the bladder but remain largely unstudied in humans. As nutritional supplementation has not shown definitive benefit, ongoing trials using targeted agents are underway. NSAIDs and oltipraz (4-methyl-5-[2-pyrazinyl]-1,2-dithiole-3-thione) are currently at the Phase I clinical trial stage, and the University of Texas MD Anderson has two National Cancer Institute (NCI)-sponsored Phase III trials using 4-HPR and celecoxib. The 4-HPR trial is designed for early-stage superficial bladder cancer patients, and the celecoxib trial is geared for more advanced stage patients receiving bacillus Calmette-Guérin adjuvant therapy. The NCI is currently conducting a Phase II trial to assess the efficacy of DFMO in patients with superficial bladder cancer.

PROSTATE CANCER

Prostate cancer is the most common cancer that occurs in men. In the United States, the estimated incidence for the year 2004 was 230,110 new cases and 29,900 deaths.[113] The lifetime risk of developing prostate cancer is 19% in the United States. Risk factors include older age, family history, race and ethnicity, and possibly dietary fat.[237] Screening for prostate cancer depends on digital rectal exam and prostate-specific antigen.[238] Clinicians should offer both tests to their patients yearly beginning at age 50 years. Men with positive family histories or other risk factors should begin at age 40 to 45 years.[238]

Premalignant Process

Prostatic intraepithelial neoplasia (PIN) is an intraluminal proliferation of secretory cells of the prostate duct-acinar system.[239] Common genetic alterations in PIN and prostate cancer have been identified: gain of chromosome 7, loss of *8p*, gain of *8q*, and loss of *10q*, *16q*, and *18q*.[237] While low-grade PIN has unclear predictive value for malignancy, high-grade PIN is suspected to be the precursor to prostatic carcinoma because of the similarities in histologic diagnosis. High-grade PIN has a high predictive value for adenocarcinoma originating from the peripheral zone of the prostate.[240] AAH has been considered to be the premalignant lesion of the transition zone, but it is not well defined.

Chemoprevention Trials

Vitamin A and its derivatives have shown a protective effect against various cancers, but the data on prostate cancer is conflicting (Table 8).[241,242] Some studies have a statistically significant trend of increased prostatic cancer risk associated with decreasing serum retinol levels, and others report that vitamin A has no benefit and can be harmful.[243,244] Vitamin D deficiency was reported to increase the risk of prostate cancer, and sunlight exposure is inversely proportional to prostate cancer mortal-

TABLE 8 Selected Prostate Cancer Chemoprevention Trials

Trial	Year	Patients (n)‡	Prevention	Population	Endpoint	Compounds*	End Result
Prostate Cancer Prevention Trial (PCPT)[241]	2003	18,882	Primary	Male	Prostate cancer	Finasteride (5 mg)	Positive
McConnell et al.[242]	2003	3,047	Secondary	BPH†	Progression BPH	Doxazosin (4 or 8 mg) Finasteride (5 mg)	Positive

*Doses are daily regimens.
†BPH = Benign prostatic hyperplasia.
‡ = Number of patients.

ity.[105,245,246] Low plasma levels of vitamin E were related to an increased risk of prostate cancer.[225] Although the ATBC was designed to evaluate the effects of α-tocopherol and beta carotene on lung cancer, the study showed that men receiving vitamin E had a 34% lower incidence of prostate cancer during the six-year period.[214,247] These studies have inspired ongoing large multicenter trials such as the Selenium and Vitamin E Cancer Prevention Trial (SELECT).

Aside from nutritional intervention, hormonal therapy has led to promising results. Finasteride is a steroidal analog of testosterone that competitively inhibits 5-α-reductase and leads to a reduction in serum dihydrotestosterone and intraprostatic dihydrotestosterone levels. The Prostate Cancer Prevention Trial enrolled 18,882 men aged 55 years and older with normal digital rectal exams and prostate-specific antigen levels less than 3 ng/mL and randomized patients to finasteride or placebo for seven years.[241,248,249] This trial reported 24.4% prostate cancer incidence in the placebo arm compared with 18.4% in the finasteride arm, for a 24.8% reduction over seven years ($P < 0.001$). In the patients who received finasteride, the prostate cancers that developed were of higher Gleason grade (7, 8, 9, or 10), and more adverse sexual side effects were reported. When compared with the placebo group, patients in the finasteride arm had fewer urinary symptoms. In a separate finasteride trial, McConnell et al. randomized 3,047 men with benign prostatic hyperplasia to finasteride (5 mg), doxazosin (4 mg or 8 mg), combination, or placebo. The mean follow-up was 4.5 years and showed that the combination therapy prevented progression of benign prostatic hyperplasia.[242]

Summary

Hormonal therapy with finasteride is promising in prostate cancer prevention. However, although finasteride decreased the incidence of prostate cancer, the malignancies that did develop with the intervention were histologically more aggressive. Finasteride was also associated with sexual side effects. Therefore, finasteride intervention should be cautiously considered for primary prevention. Several large multiinstitutional trials are underway investigating other promising nutritional agents. SELECT is a Phase III study designed to study 32,400 men treated with selenium and vitamin E.[250] This trial will take 12 years to complete. Two other Southwest Oncology Group studies complementary to SELECT include a randomized pharmacodynamic study in presurgical prostatectomy patients and a Phase III trial in patients with high-grade PIN. Prostate cancer and prevention is another area of high interest to implement novel biologic strategies.

SKIN CANCER

Skin cancer accounts for approximately 40% of all new cancer diagnoses.[251] Most skin cancers (80%) result from basal cell carcinomas (BCC); another 16% are SCC, and 4% are melanomas.[252] A high percentage of patients with SCC develop second primary skin cancers within five years.[253–255] Associated risk factors for skin cancer include childhood and chronic sun exposure, individual

susceptibility with red or blond hair and fair-skinned phenotype, older age, polycyclic aromatic hydrocarbon, immunocompromised status, or xeroderma pigmentosum. SCC is especially associated with cigarette smoking, organ transplant recipients on immune suppressive therapy, or receiving photochemotherapy (psoralen-ultraviolet A).[256–261] Xeroderma pigmentosum is a rare autosomal recessive disease that is characterized by defective DNA repair and a greater than a 1,000-fold risk of developing melanomas and nonmelanoma skin cancers (NMSC).

Premalignant Process

Actinic keratosis (AK; solar keratosis, senile keratosis) is the precursor lesion to SCC.[262,263] The rate of malignant transformation is 0.075% to 0.096% per lesion per year.[264] Patients with multiple AK have a lifetime risk of progression to SCC of 6% to 10%.[265] A proposed grading system for AK has renamed the premalignant lesion as keratinocyte intraepidermal neoplasia (KIN).[266,267] KIN is divided into three groups: KIN I, II, and III based on clinical features, degree of cytologic atypia of the keratinocytes, and extent of abnormal epidermis. KIN III is often called SCC in situ or Bowen disease.[267]

Chemoprevention Trials

Skin cancer prevention trials have focused on nutritional supplements with limited success (Table 9).[268–276] Trials in both the primary prevention and adjuvant settings have shown no benefit for either beta carotene or selenium.[268,271,272] Systemic retinoids have had mixed results. The Southwest Skin Cancer Prevention Study Group treated 525 patients with at least four prior NMSC with daily retinol, isotretinoin, or placebo for three years and found no beneficial chemopreventive effect from either retinol or isotretinoin.[275] The Isotretinoin-Basal Cell Carcinoma Prevention Trial, which treated 981 patients with two or more biopsy-proven BCC within five years of enrollment with daily low dose isotretinoin (10 mg) or placebo for three years, confirmed that isotretinoin did not prevent BCC.[273,274]

However, several other trials have shown benefit in preventing progression of AK or development of SCC. Moriarty et al. reported a 84% partial or complete response rate in patients with AK receiving two months of etretinate compared with 4.7% in the placebo arm.[269] The Skin Cancer Prevention-Actinic Keratosis trial randomized 2,297 subjects at moderate risk to oral retinol (25,000 IU/day) or placebo for up to five

TABLE **9** Selected Nonmelanoma Skin Cancer Chemoprevention Trials

Trial	Year	Patients (n)¶	Prevention	Population	Endpoint	Compounds*	End Result
Green et al.[268]	1999	1,621	Primary	Healthy sun-exposed volunteers	Skin cancer	Beta carotene (30 mg)	Negative
Moriarty et al.[269]	1982	50	Secondary	Actinic keratosis	Response	Etretinate (75 mg)	Positive
Skin Cancer Prevention-Actinic Keratosis Trial[270]	1997	2,297	Secondary	Moderate-severe actinic keratosis	Skin cancer	Retinol (25,000 IU)	Positive for SCC†
Skin Cancer Prevention Group Trial[271]	1990	1,805	Tertiary	Prior NMSC‡	Skin cancer	Beta carotene (50 mg)	Negative
Nutritional Prevention of Cancer Study Group[272]	1996	1,312	Tertiary	Prior BCC/SCC§	Skin cancer	Selenium (200 mcg)	Negative
Isotretinoin-Basal Cell Carcinoma Prevention Trial[273,274]	1990	981	Tertiary	More than two prior BCC	BCC	Isotretinoin (10 mg)	Negative
Southwest Skin Cancer Prevention Study Group[275]	1997	719	Tertiary	At least four prior skin cancers	Skin cancer	Retinol (25,000 IU) Isotretinoin (5 to 10 mg)	Negative
Kraemer et al.[276]	1988	5	Tertiary	Xeroderma Pigmentosum	Skin cancer	Isotretinoin (2 mg/kg)	Positive

*Doses are daily regimens.
†SCC = Squamous cell carcinoma.
‡NMSC = Nonmelanoma skin cancer.
§BCC = Basal cell carcinoma.
¶ = Number of patients.

years and found that it prevented SCC ($P = 0.04$) but not BCC.[270] In a nested cohort study in high-risk patients with psoriasis treated with oral psoralen-ultraviolet A, systemic retinoids had no benefit in BCC prevention but did have a 30% reduction in SCC development ($P = 0.002$).[277] In the larger skin cancer prevention trial, adverse toxicity with retinoid intervention was not reported to be significant.[270]

Although the role of systemic retinoids in NMSC is unclear, patients with xeroderma pigmentosum have been shown to benefit from retinoid therapy. Kraemer et al. treated five patients with xeroderma pigmentosum who were surgically cleared of all skin tumors with oral isotretinoin at 2 mg/kg/day for two years.[276] Although an average risk reduction of 63% was seen with isotretinoin therapy ($P = 0.019$), the positive effect tapered once therapy was discontinued ($P = 0.007$).[276] This trial was followed by a low-dose isotretinoin study where patients received an initial dose of 0.5 mg/kg/day isotretinoin, and then the dose escalated to 1.0 or 1.5 mg/kg/day.[278] This study reported significant variability in the lowest effective dose that was tolerable in this patient population.[278]

Summary

In skin cancer chemoprevention, retinoids appear to be more beneficial in patients with earlier stages of premalignancy than in patients with resected or overt malignancies. The studies done in patients with xeroderma pigmentosum suggest that retinoids have a preventive role. However, while the FDA has approved the use of topical diclofenac in AK, no systemic therapies have been approved to date. Future promising agents that regulate the AP1 and NF-kB pathways include NSAIDs (COX-2 inhibitors), green and black tea polyphenols (catechins, theaflavins), resveratrol, isothiocyanates, and inositol hexaphosphate (phytic acid).[106]

CERVICAL CANCER

Cervical cancer is a major cause of morbidity in women worldwide. In the United States, 10,520 new cases and 3,900 deaths are estimated for the year 2004.[113] Cervical cancer is characterized by a premalignant condition, cervical intraepithelial neoplasia (CIN). Screening is performed with the Papanicolaou test and has a 70% to 80% sensitivity for CIN lesions.[279] Risk factors for cervical cancer include HPV infection (HPV 16, 18, 45, and 56),[280–282] early age at first intercourse, ethnicity, immunosuppression, multiple sexual partners, smoking, oral contraceptive use, and beta carotene deficiency. HPV infection has been reported in 77% of high-grade CIN and 84% to 100% of invasive cervical cancers.[280,283,284]

Premalignant Process

Premalignant cervical lesions are classified cytologically as squamous epithelial lesions or histologically as CIN. CIN is divided into three groups: CIN 1 (mild dysplasia), CIN 2 (moderate dysplasia), and CIN 3 (severe dysplasia). These histologic findings are confined to the squamous epithelium and lie on a continuum to CIS, and then invasive cervical cancer.[285] Most often, CIN is treated with surgical excision, cryotherapy, laser therapy, or loop excision. In these cases of definitive therapy, the two-year response rate, as defined by clinical remission, is 80%.[110] The risk of recurrence is higher in women aged 30 and older, those with HPV 16 or 18 infection, or patients with prior therapy.[286]

Chemoprevention Trials

Despite promising results in earlier Phase I/IIa trials, almost all large Phase IIb/III trials showed no benefit in systemic chemoprevention for this cancer type (Table 10).[287–294] Beta carotene, folic acid, and systemic retinoid supplementation have been studied in several cervical cancer chemoprevention trials, but no benefit has been seen.[289,290,295–300] Using systemic retinoids, Kim et al. treated 45 patients with a prior history of high-grade CIN or high-grade squamous intraepithelial lesion with 13cRA for six months and reported no benefit in preventing CIN recurrence.[292] Another negative trial evaluated 12 weeks of high-dose 9cRA (50 mg), low-dose 9cRA (25 mg), or placebo daily in 114 patients

TABLE **10** Selected Cervical Cancer Chemoprevention Trials

Trial	Year	Patients (n)¶	Prevention	Population	Endpoint	Compounds*	End Result
Meyskens et al.[287]	1994	141	Secondary	CIN 2†	Dysplasia	Topical ATRA‡	Positive in moderate dysplasia patients
Follen et al.[288]	2001	39	Secondary	CIN 2-3	Dysplasia biomarkers	4-HPR (200 mg)§	Negative
Mackerras et al.[289]	1999	141	Secondary	Atypia to CIN 1	Dysplasia	Beta carotene (30 mg) Vitamin C (500 mg)	Negative
Butterworth et al.[290]	1992	177	Secondary	CIN 1-2	Dysplasia	Folic acid (10 mg)	Negative
Alvarez et al.[291]	2003	114	Secondary	CIN 2-3	Dysplasia	9cRA (25 to 50 mg)	Negative
Kim et al.[292]	2003	45	Secondary	CIN 1-3	Dysplasia	13cRA (1 mg/kg)	Negative
Bell et al.[293]	2000	27	Secondary	CIN 2-3	Dysplasia	Indole-3-carbinol (200 to 400 mg)	Positive
Mitchell et al.[294]	1995	30	Secondary	CIN 3	Dysplasia	α-Difluoromethylornithine (0.06 to 1 gm/m^2)	Positive

*Daily doses are depicted.
†CIN = Cervical intraepithelial neoplasia.
‡ATRA = All-*trans*-retinoic acid.
§4-HPR = N-[4-hydroxyphenyl] retinamide.
¶ = Number of patients.

with CIN 2 or 3.[291] No difference was seen between the three arms in the rate of histologic regression. Follen et al. studied the effect of 4-HPR in 39 patients in a randomized, double-blinded Phase II trial.[288] Patients received placebo or 4-HPR at 200 mg a day for six months with a three-day-per-month drug holiday. Biopsies were performed on the patients at six months, and then the patients were monitored for an additional six months. An interim analysis demonstrated that 4-HPR intervention led to a worse prognosis.[288]

Although systemic retinoid use was not beneficial, topical application has led to CIN regression. Meyskens et al. treated women with moderate to severe dysplasia with a collagen sponge insert and cervical cap delivering all-transretinoic acid or placebo.[287] All-transretinoic acid treatment had higher complete regression rates than placebo (43% versus 27%, $P = 0.041$) in the moderate dysplasia group but had no effect in severe dysplasia.[287]

Indole-3-carbinol (I-3-C), a DNA adduct reducer, has been studied in 30 patients with CIN 2 or 3.[293] Complete regression was seen in four of eight patients in the 200 mg/day arm and four of nine patients in the 400 mg/day arm. None of the patients with placebo had complete regression. This translated into a relative risk of 0.5 for the 200 mg/day I-3-C treatment ($P = 0.023$) and a relative risk of 0.55 in the 400 mg/day I-3-C ($P = 0.032$).[293] Another promising agent is DFMO, an inhibitor of ornithine decarboxylase. Mitchell et al. conducted a Phase I trial in 30 patients with Grade 3 CIN and reported an overall response rate of 50%.[294] A Phase II trial is underway to evaluate these promising results.[301]

Summary

In CIN, several agents have been studied: retinoids, micronutrients, α-difluoromethylornithine, and I-3-C. Unfortunately, the promising findings from early Phase I/IIA studies ultimately were negative in larger Phase IIB and III trials. Chemoprevention trials in cervical cancer have faced certain challenges. The lack of reliable surrogate biomarkers, high and variable regression rates, the effect of colposcopic biopsy, and the natural history of the lesion grade are important variables that need to be accounted for in future chemoprevention trials.[302] Also, because HPV is the major causative agent in most cases of CIN and cervical cancer, agents should be able to demonstrate efficacy against HPV viral protein expression or tumorigenesis in preclinical work before investments into large clinical trials should be made.[301] Studies in HPV vaccines are underway.[301,303,304] Future trials will need to account for the high regression rate in the sample sizes and power the trials accordingly. Uniform biopsy procedures will also need to be issued.

FUTURE DIRECTIONS IN CANCER CHEMOPREVENTION

The future of cancer chemoprevention remains open to innovation, with a specific need for emphasizing cancer prevention in public health policy. In the case of lung cancer, smoking cessation campaigns need to continue because tobacco exposure remains the most important causative agent. However, statistics have shown an increased risk of lung cancer even after smokers quit. With the increasing number of former smokers, these patients are certainly appropriate for chemoprevention because they remain at risk of cancer development. It is paramount to identify appropriate at-risk patient populations (eg, previous history of cancer, heavy smoking history, dysplastic lesions) to apply chemopreventive interventions.

As our current regimens combining chemotherapy, radiation therapy, and surgery for the treatment of cancer continue to expand, the mechanisms underlying tumor biology are becoming better understood. The continued study of tumor biology and natural history through controlled trials focusing not only on efficacy endpoints but also on biologic markers in tissue and serum will help develop detailed risk models. Chemopreventive agents appear thus far to have efficacy in several tumor types, and we hope to define their future role in treating and preventing other cancers in high-risk individuals.

Despite curative local therapy, SPTs have emerged as an increasingly important problem, underscoring the principle of field cancerization. Chemopreventive agents have affected this arena as well, and as further studies are performed, their role in preventing SPTs will be further defined. Combination regimens targeting specific molecular defects show early promise. A multidisciplinary approach involving clinicians and basic researchers is needed to study the biology of cancers before chemoprevention can be incorporated into a societal standard of care.

CONCLUSIONS

The goals for the success of chemoprevention include devising a tumor-specific risk model for identifying high-risk cohorts, increasing preclinical drug-testing models (ie, gene targeting/knockout models), developing translational/mechanistic studies to develop novel chemopreventative agents, identifying surrogate endpoints using molecular alterations, and locating promising new targets of drug activity and further study of existing candidate surrogate-endpoint markers. Only through these measures can we attempt to make a meaningful impact in the survival of our patients at risk.

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