Journal of Cellular Biochemistry 25S:15–22 (1996)

Genetic Susceptibility to Cancer From Exogenous and Endogenous Exposures

Heather Spencer Feigelson,[1]* Ronald K. Ross,[1] Mimi C. Yu,[1] Gerhard A. Coetzee,[2] Juergen K.V. Reichardt,[3] and Brian E. Henderson[1]

[1]Department of Preventive Medicine, University of Southern California, Los Angeles, California
[2]Department of Urology, Norris Comprehensive Cancer Center, University of Southern California, Los Angeles, California
[3]Department of Biochemistry and Molecular Biology, Institute for Genetic Medicine, University of Southern California School of Medicine, Los Angeles, California

Abstract The past four decades of epidemiological research have yielded valuable information on the risks of populations to environmental exposures such as tobacco, asbestos, and dietary components. Prevention efforts have been focused on large-scale population-based interventions to minimize exposure to such external carcinogens. While some cancers are beginning to show a decline from changing environmental exposures, hormone-related cancers, such as breast and prostate, are becoming more prevalent. The development of these cancers appears to be closely related to endogenous exposures to circulating steroid hormones. Although prevention trials using antihormone agents are proving successful in some instances, the long-term control of these cancers necessitates a clearer understanding of the metabolism and transport of the relevant hormone in vivo.

The revolution in molecular biology has provided powerful genetic tools for evaluating mechanisms of cancer causation as well as the potential to better define individual susceptibility. Using tobacco exposure as an example, we and others have demonstrated that polymorphisms in genes controlling aromatic amine metabolism provide at least a partial explanation for ethnic and individual susceptibility to bladder cancer. Similar studies have examined genetic polymorphisms in the metabolism of tobacco smoke and lung cancer risk, red meat and colorectal cancer, and aflatoxin and liver cancer.

Our current studies have pursued a similar paradigm of genetic polymorphism and individual cancer susceptibility in prostate and breast carcinogenesis. We are evaluating polymorphisms in the steroid 5α-reductase type II and androgen receptor genes in relation to prostate cancer based on the evidence that intracellular dihydrotestosterone is the critical "carcinogen." We are pursuing genetic polymorphisms affecting estradiol metabolism, including those in the 17β-hydroxysteroid dehydrogenase 2 and estrogen receptor genes as they relate to susceptibility to breast cancer. The potential role of a polymorphism in the cytochrome P450c17α gene in both breast and prostate cancers is also being examined. J. Cell. Biochem. 25S:15–22. © 1997 Wiley-Liss, Inc.

Key words: bladder cancer; breast cancer; ethnicity; polymorphism prostate cancer

The past four decades of epidemiologic research have provided valuable information on the cancer risks to populations from environmental exposures such as tobacco, asbestos, and dietary components [1]. Probable dietary risk factors include fat, certain preserved foods (such as "Cantonese" salted fish), and food contaminants (such as aflatoxin). While some cancers, such as lung and stomach cancers, are beginning to show a decline in incidence from changing environmental exposures, hormone-related cancers, such as breast and prostate cancers, are becoming more prevalent. The development of this latter group of cancers appears to be closely related to endogenous exposure to circulating steroid hormones: estrogen, and perhaps progesterone, in the case of breast cancer, and testosterone and its metabolites in the case of prostate cancer [2]. Although primary and secondary prevention trials using

Contract research sponsor: National Cancer Institute, contract grant number RO1 CA54281, contract grant number CA 17054, contract grant number CA68581; contract research sponsor: University of California Breast Cancer Research Program, contract grant number 11B-0084.

*Correspondence to: Heather Spencer Feigelson, PhD, Department of Preventive Medicine, University of Southern California, Norris Comprehensive Cancer Center, 1441 Eastlake Avenue, MS #44, Los Angeles, CA 90033-0800.

Received 12 January 1996; Accepted 3 April 1996

© 1997 Wiley-Liss, Inc.

antihormonal agents are underway for both of these cancers, their long-term control requires a clearer understanding of the metabolism and transport of the relevant hormones in vivo [3].

The revolution in molecular technology over the past decade has provided powerful genetic tools for evaluating mechanisms of cancer causation as well as the potential to better define individual susceptibility. Using tobacco exposure as an example, we and others have demonstrated that polymorphisms in genes controlling aromatic amine metabolism provide at least a partial explanation for ethnic and individual susceptibility to bladder cancer [4–7]. Similar methodology has been employed to study genetic polymorphisms in metabolic pathways for cigarette smoking and lung cancer [8], red meat and colorectal cancer [9], and aflatoxin and liver cancer [10].

In this report, we use the paradigms that have been developed from studies of bladder cancer to propose models for individual susceptibility to prostate and breast cancers. We have assumed that within and between ethnic groups there exist genetic differences that affect steroid hormone metabolism and transport. Markers (i.e., genetic polymorphisms) of these differences are likely to provide a more precise measure of risk than circulating levels of steroid hormones. The latter measurements are subject to variation by many factors, for example, time of day, age, disease status, and laboratory imprecision. The multigenic models that we have developed for the hormone-related cancers assume that there are functionally important polymorphisms in genes that encode enzymes involved in steroid hormone biosynthesis and metabolism that lead to differences in individual susceptibility to breast and prostate cancers. We have selected candidate genes based on two criteria, 1) the gene has known polymorphisms that may be functionally relevant and, more importantly, 2) the enzyme product of the gene acts in a pathway that is physiologically relevant based on our understanding of the etiology of the cancer in question. In the case of bladder cancer, cigarette smoking is an established risk factor [11]. We have hypothesized that individual variation in the ability to detoxify or activate the putative bladder carcinogens in cigarette smoke ultimately influences the lifetime risk of developing bladder cancer. Thus, the candidate genes we proposed were either known or suspected to be involved in the metabolism of carcinogens in cigarette smoke. In the case of prostate cancer, there is strong evidence that circulating levels of testosterone and its metabolites play important roles in causation [2]. Thus, the candidate genes proposed were known to be involved in the metabolism or transport of testosterone. Similarly, the primary risk factors for breast cancer can be understood as measures of the cumulative exposure of the breast to estrogen and, perhaps, progesterone [2], and the candidate genes under study are those known to be involved in the metabolism and transport of estrogen. In the following sections, we first describe our proposed model of bladder carcinogenesis and provide some recent data that support this model, then outline parallel approaches to studying individual susceptibility to prostate and breast cancers.

BLADDER CANCER MODEL

While it is widely recognized that cigarette smoking is by far the most important contributor to bladder cancer risk on a population basis [11], a number of epidemiological observations have suggested that additional factors must play a role in modifying this risk. For example, in Los Angeles, California, non-Hispanic white, African-American, and Asian men have comparable smoking habits, yet dramatically different risks of bladder cancer. Annual incidence of bladder cancer is highest among whites (31/100,000), intermediate in African-Americans (16/100,000), and lowest among Chinese and Japanese (13/100,000). These seemingly disparate observations have led to the hypothesis that differences in the metabolism of smoking-related carcinogens, e.g., arylamines, may exist across different ethnic groups.

At least three types of metabolic enzymes (and probably others) are involved in bladder carcinogenesis resulting from exposure to arylamines: the hepatic cytochrome P4501A2 isoenzyme (P4501A2), *N*-acetyltransferases (NAT-1 and NAT-2), and glutathione *S*-transferase M1 (GSTM1).

Metabolic activation is required to biotransform the arylamines into their carcinogenic forms. *N*-hydroxylation, which takes place primarily in the liver and is catalyzed by P4501A2, is generally viewed as the first critical step [12]. The enzyme is inducible by a number of environmental factors, including cigarette smoke, so

considerable individual and population variability in the activity of P4501A2 exists [13].

The metabolically active form of the arylamines (the hydroxylamines) are electrophilic and can form adducts with hemoglobin or can circulate and be excreted through the kidney [14]. In the acidic environment of the bladder lumen, these hydroxylamines, with or without further bioactivation from highly electrophilic *N*-acetoxy derivatives, can form adducts with mucosal DNA [6]. Misrepair of the damage to DNA induced by these adducts is hypothesized to lead to mutations in oncogenes and tumor suppressor genes.

Alternatively, arylamines can be catalyzed by competing detoxification pathways. The best established of these is the *N*-acetylation pathway which is regulated by *N*-acetyltransferase activity in the liver. *N*-acetyltransferase is coded by two distinct genes, NAT-1 and NAT-2. NAT-2 has long been known to exhibit polymorphism, and the NAT-2 phenotype (slow or rapid acetylators) primarily reflects enzyme activity in the liver. Recently, it was demonstrated that the NAT-2 polymorphisms are due to single base-pair changes in the coding region of the gene; individuals homozygous for mutant alleles possess the slow acetylator phenotype [15]. A number of case-control studies of bladder cancer have investigated the relationship between NAT-2 phenotype and/or genotype and bladder cancer risk. In general, these studies consistently observed a 1.5–2.0-fold excess risk for bladder cancer in slow versus rapid acetylators [16].

It was only recently that NAT-1 was shown to be polymorphic [17,18]. Badawi et al. [6] have demonstrated that a NAT-1 polyadenylation polymorphism is associated with differences in tissue NAT-1 activity and suggested that NAT-1 activity in bladder mucosa represents a major bioactivation step in the carcinogenic process. NAT-1 converts urinary *N*-hydroxyl arylamines to reactive *N*-acetoxy esters that can form covalent DNA adducts. Thus, available evidence suggests that individuals who inherit the slow NAT-2 and rapid NAT-1 genotypes would be at increased risk for bladder cancer.

GSTM1 is part of a family of enzymes that detoxify reactive chemical entities by promoting their conjugation to glutathione [19]. The GSTM1 gene is polymorphic in humans; about one-half of the U.S. white population lack both copies of the gene and hence exhibit no GSTM1 enzymatic activity (GSTM1 null). Metabolites of several polycyclic aromatic hydrocarbons that are present in cigarette smoke are known substrates of GSTM1. Metabolites of other carcinogenic compounds in cigarette smoke, including the arylamines and nitrosamines, are potential substrates [19,20]. There is some evidence that the GSTM1 null genotype is associated with an excess risk of bladder cancer [7,20].

We have assessed 133 male residents of Los Angeles County who were either non-Hispanic white, African-American, or Asian (Chinese, Japanese) and over the age of 35 years for their acetylator phenotype and levels of 3- and 4-aminobiphenyl (ABP) hemoglobin adducts [5]. We confirmed that mean 3- and 4-ABP hemoglobin adduct levels were significantly higher in cigarette smokers relative to nonsmokers, and that the level increased with increasing number of cigarettes smoked per day. More importantly, we observed varying rates of slow acetylators between the races that were in agreement with the populations' varying incidence of bladder cancer (54% in whites, 34% in African-Americans, and 14% in Asians), and that slow acetylators consistently exhibited higher mean levels of ABP-hemoglobin adducts relative to rapid acetylators regardless of race or level of cigarette smoking.

We recently expanded the above study to include GSTM1 genotype and showed that the presence of the null genotype enhances the differences in prevalence of high and low risk profiles for bladder cancer among the three populations under study [7]. There was a 10-fold difference in prevalence of the high-risk profile (slow NAT-2, null GSTM1) between high-risk whites and low-risk Asians (27% versus 2.7%), and whites had less than one-half the prevalence of the "protective" profile (rapid NAT-2, non-null GSTM1) relative to African-Americans and Asians (23% versus 57%). Moreover, mean levels of ABP-hemoglobin adducts were significantly higher in GSTM1-null versus non-null subjects independent of their acetylator phenotype.

PROSTATE CANCER MODEL

Increased cell division is the hallmark of the response of prostate epithelial cells to androgens. Specifically, cell division in the prostate is controlled by testosterone (T), following its intracellular conversion to dihydrotestosterone (DHT) as shown in Figure 1. This conversion is

catalyzed by the enzyme steroid 5α-reductase. DHT, and much less efficiently, T, bind to the androgen receptor (AR) in the cytoplasm, and the AR/ligand complexes are translocated to the nucleus for DNA binding and transactivation of target genes, including those that control cell division and death.

As in the case of bladder cancer, race-ethnicity is an important determinant of prostate cancer risk. African-Americans have higher rates than whites who, in turn, have higher rates than U.S. born Asians [21]. Differences among ethnic groups with respect to serum testosterone levels [22] and steroid 5α-reductase activities [23] have been suggested to explain, at least in part, the large ethnic variability in the incidence of prostate cancer. The genes of interest to us currently are the steroid 5α-reductase gene type II (SRD5A2), the cytochrome P450c17α gene (CYP17), and the androgen receptor gene (AR).

The Steroid 5α-Reductase Type II Gene (SRD5A2)

There are two distinct steroid 5α-reductase enzymes encoded for by different genes; the type II enzyme is active primarily in the prostate and in genital skin. Germline mutations of the type II gene (SRD5A2) have been identified, most notably in a sizable kindred in the Dominican Republic [24]. Boys with this mutated gene are phenotypically female with a persistent vaginal pouch until puberty, at which time there is some phallic enlargement and development of some secondary sex characteristics, but the prostate remains underdeveloped.

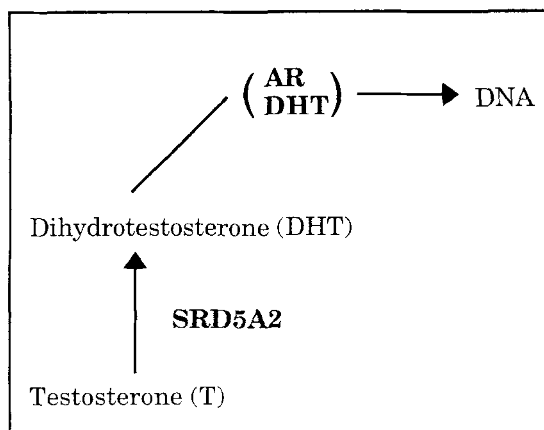

Fig. 1. Schematic presentation of steroid 5α-reductase (SRD5A2) and androgen receptor (AR) activity in testosterone metabolism in the prostate epithelial cell.

Davis and Russell [25] have described a polymorphism (TA repeat number) in the 3′ untranslated region of the SRD5A2 gene. We have confirmed this polymorphism, but find the prevalence of variant alleles using this polymorphic marker to be substantially higher than reported by Davis and Russell [25]. We noted a series of novel alleles (which cluster around the TA_{18} allele described by Davis and Russell) in African-Americans, who are at high risk for prostate cancer. A preliminary analysis of 54 African-American and 59 white prostate cancer cases and an equal number of age- and race-matched controls revealed a statistically non-significant excess in variant alleles among cases relative to controls within each ethnic group [26].

The Cytochrome P450c17α Gene (CYP17)

CYP17 codes for the cytochrome P450c17α enzyme that mediates both steroid 17α-hydroxylase and 17–20-lyase activities and functions at key branch points in human steroidogenesis [27]. 17α-hydroxylase activity converts steroids to precursors of the glucocorticoid cortisol, and 17–20-lyase activity yields precursors to estradiol and testosterone.

One polymorphism in CYP17 has been reported [28]. The 5′ promoter region contains a single base pair polymorphism that creates an Spl-type (CCACC box) promoter site (designated as the A2 allele) 34 bp upstream from the initiation of translation. Carey et al. [28] used this polymorphism to analyze the segregation of CYP17 in pedigrees with polycystic ovary disease and male pattern baldness (believed to be caused by a common underlying disorder of androgen biosynthesis or metabolism). They found an association between the affected state and the A2 allele. Since it is thought that the number of 5′ promoter elements correlates with promoter activity [29], this association with the A2 allele may reflect an increase in the rate of transcription of the gene. We are currently evaluating this polymorphism in studies of both breast and prostate cancers.

The Androgen Receptor Gene (AR)

It is likely that the AR also is involved in the racial-ethnic variation in prostate cancer incidence. The AR gene, on the long arm of the X chromosome, belongs to the superfamily of ligand-dependent transactivation factors and,

like others in this family, contains a C-terminal hormone binding domain, a central DNA binding domain, and an N-terminal transcriptional modulatory domain which, for the AR gene, is encoded entirely by exon 1 [30].

Within exon 1 there are two highly polymorphic microsatellites: one of these is a trinucleotide (CAG) repeat sequence of unknown function. Abnormal expansion of this satellite is associated with X-linked spinal and bulbar muscular atrophy (Kennedy's disease) [31]. The normal size varies from 9 to 31 repeats, whereas the mutant range associated with Kennedy's disease varies from 40 to 62. It was demonstrated that these mutant receptors have normal androgen binding, but in transfection assays they transactivate an androgen responsive reporter gene subnormally [32]. We and others have noted that African-Americans, U.S. whites and Asian-Americans show population differences in the CAG repeat polymorphism of the AR locus, with the allele frequency distribution shifted toward shorter alleles in the case of African-Americans and longer alleles in the case of Asians relative to whites [33,34]. We have hypothesized that this size polymorphism might correlate with, or cause changes in, AR function (smaller size, greater transactivity) and, therefore, possibly also be associated with prostate cancer risk [34]. We are currently evaluating this hypothesis in a multiethnic case-control study.

BREAST CANCER MODEL

Attempts to understand and quantify the role of estrogens in breast cancer have been limited to some extent by our ability to accurately and reproducibly measure steroid hormones in circulating blood. Despite the difficulty of conducting such studies, there is evidence from our group [35–37] and others [38] of significant elevations in circulating estradiol in breast cancer patients compared to controls in populations at high and low risks of breast cancer. In addition, there are reproducible differences in plasma and urine estrogen levels between healthy high-risk U.S. and low-risk Asian women [36,37]. In one such study in which cycle length was controlled, estradiol levels were 20% higher in Los Angeles compared to Shanghai premenopausal women [36]; in a companion study of postmenopausal women, estradiol levels were 36% higher in Los Angeles than in their matched Japanese counterparts [37]. Reasons for these differences remain poorly defined, but part of the explanation may be that there are genetic differences that affect metabolism or intracellular binding and transport of estradiol.

Based on the schematic presentation of estrogen metabolism in the ovaries and breast epithelium shown in Figure 2, we describe below the function of each of three candidate genes and their possible roles in breast cancer etiology. The genes of interest are the 17β-hydroxyster-

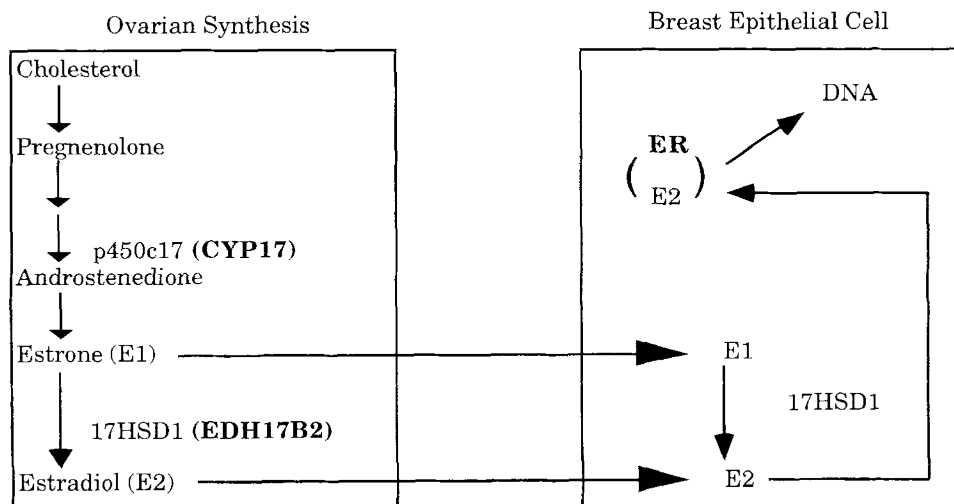

Fig. 2. Schematic presentation of 17β-hydroxysteroid dehydrogenase 2 (EDH17B2), cytochrome P450c17α (CYP1 '), and estrogen receptor (ER) genes in estrogen metabolism in the ovaries and breast epithelium.

oid dehydrogenase 2 (EDH17B2) gene, the cytochrome P450c17α (CYP17) gene, and the estrogen receptor (ER) gene.

The 17β-Hydroxysteroid Dehydrogenase 2 Gene (EDH17B2)

The EDH17B2 gene codes for the enzyme 17β-hydroxysteroid dehydrogenase type 1 (17HSD1) which catalyzes the final step of estradiol biosynthesis, namely the interconversion of estrone (E1) into the more biologically active estrogen fraction, estradiol (E2). 17HSD1 acts in the theca cells of the ovary and is expressed in both normal and malignant breast epithelium [39].

Several polymorphisms have been identified in the EDH17B2 gene [40,41]. Normand et al. [40] described 11 polymorphisms, two of which are located in the coding region and result in amino acid changes. Neither polymorphism appears to affect the catalytic or the immunological properties of the enzyme [42], but one (a serine to glycine change at position 312) has been shown to be associated with a marginally statistically significant increased risk for breast cancer [41]. We are currently evaluating this polymorphism in a case-control study of breast cancer. In addition, we are evaluating a recently identified point mutation in the TATA box in the promoter region of EDH17B2 [43]. This mutation results in an average 80% reduction of promoter activity in COS and JAR choriocarcinoma cell lines compared to cells containing the wild type EDH17B2 promoter.

The Cytochrome P450c17α Gene (CYP17)

As described in the prostate cancer model, the cytochrome P450c17α enzyme yields precursors to estradiol and testosterone and may ultimately play a role in breast cancer etiology. Figure 2 shows where P450c17α acts in the synthesis of estrone and, ultimately, estradiol in the granulosa cells of the ovary.

Estrogen Receptor Gene (ER)

Several polymorphisms in the ER gene have been reported [44–47]. The most promising evidence for an association between ER gene polymorphisms and breast cancer risk was reported by Andersen et al. [46], who examined 3 RFLPs and found that alleles having an XbaI restriction site were significantly more frequent in breast cancer patients than controls (odds ratio = 2.0). This association was limited to postmenopausal patients. del Senno et al. [47] reported a highly polymorphic $(TA)_n$ repeat upstream from the region of the ER gene originally described as exon 1 in the transactivation domain. This is a potentially interesting region since an earlier study suggested that, like other receptors, the ER may have multiple promoter regions [48]. This region is not yet well characterized, but could provide important clues for understanding the role of the ER gene in breast cancer. We have initiated studies of the XbaI polymorphism and the $(TA)_n$ repeat in breast cancer.

CONCLUSIONS

A growing body of work has contributed to a better understanding of bladder cancer etiology and how individual susceptibility, coded by multiple susceptibility genes, can explain observed population differences in risk across different ethnic groups. Hormone-related cancers also exhibit substantial unexplained differences in risk across ethnic groups. In examining these associations in multiple ethnic groups we can verify associations in even nonfunctional polymorphisms. However, such associations must be viewed with caution; ethnic differences in nonfunctional polymorphisms are not necessarily meaningful. Metabolic genes and their role in carcinogenesis is a relatively new area of research with scant information at present. Expanded knowledge in this area should lead to more refined definitions of "high-risk" individuals for specific cancers, and, hopefully, lead us to be able to better define who may benefit from specific interventions based on underlying genetic susceptibility. The multigenic susceptibility models we have proposed for breast and prostate cancers not only could further our understanding regarding the etiology of these important cancers, but could also provide important targets for prevention and control.

ACKNOWLEDGMENTS

This work was support by grants RO1 CA54281, CA17054, and CA68581 (to J.K.V.R.) from the National Cancer Institute and by grant 11B-0084 from the University of California Breast Cancer Research Program.

REFERENCES

1. Henderson BE, Ross RK, Pike MC (1991): Toward the primary prevention of cancer. Science 254:1131–1138.

2. Henderson BE, Ross RK, Pike MC, Casagrande JT (1982): Endogenous hormones as a major factor in human cancer. Cancer Res 42:3232–3239.
3. Henderson BE, Ross RK, Pike MC (1993): Hormonal chemoprevention of cancer in women. Science 259:633–638.
4. Vineis P (1992): Epidemiological models of carcinogenesis: The example of bladder cancer. Cancer Epidemiol Biomarkers Prev 1:149–153.
5. Yu MC, Skipper PL, Taghizadeh K, Tannenbaum SR, Chan KK, Henderson BE, Ross RK (1994): Acetylator phenotype, aminobiphenyl-hemoglobin adduct levels, and bladder cancer risk in white, black, and Asian men in Los Angeles, California. J Natl Cancer Inst 86:712–716.
6. Badawi AF, Hirvonen A, Bell DA, Lang NP, Kadlubar FF (1995): Role of aromatic amine acetyltransferases, NAT1 and NAT2, in carcinogen-DNA adduct formation in the human urinary bladder. Cancer Res 55:5230–5237.
7. Yu MC, Ross RK, Chan KK, Henderson BE, Skipper PL, Tannenbaum SR, Coetzee GA (1995): Glutathione *S*-transferase M1 genotype affects aminobiphenyl-hemoglobin adduct levels in White, Black, and Asian smokers and nonsmokers. Cancer Epidemiol Biomarkers Prev 4:861–864.
8. London SJ, Daly AK, Cooper J, Navidi WC, Carpenter CL, Idle JR (1995): Polymorphism of glutathione *S*-transferase M1 and lung cancer risk among African-Americans and Caucasians in Los Angeles County, California. J Natl Cancer Inst 87:1246–1253.
9. Lang NP, Butler MA, Massengill J, Lawson M, Stotts RC, Hauer-Jensen M, Kadlubar FF (1994): Rapid metabolic phenotypes for acetyltransferase and cytochrome P4501A2 and putative exposure to food-borne heterocyclic amines increase the risk for colorectal cancer or polyps. Cancer Epidemiol Biomarkers Prev 3:675–682.
10. McGlynn KA, Rosvold EA, Lustbader ED, Hu Y, Clapper ML, Zhou T, Wild CP, Xia XL, Baffoe-Bonnie A, Ofori-Adjei D, Chen GC, London WT, Shen FM, Buetow KH (1995): Susceptibility to hepatocellular carcinoma is associated with genetic variation in the enzymatic detoxification of aflatoxin B_1. Proc Natl Acad Sci U S A 92:2384–2387.
11. Doll R, Peto R (1981): The causes of cancer: Quantitative estimates of avoidable risks of cancer in the United States today. J Natl Cancer Inst 66:1191–1308.
12. Butler MA, Iwasaki M, Guengerich FP, Kadlubar FF (1989): Human cytochrome $P450_{PA}$ (P-4501A2), the phenacetin *O*-deethylase, is primarily responsible for the hepatic 3-demethylation of caffeine and *N*-oxidation of carcinogenic arylamines. Proc Natl Acad Sci U S A 86:7696–7700.
13. Kalow W, Tang BK (1991): Caffeine as a metabolic probe: Exploration of the enzyme-inducing effect of cigarette smoking. Clin Pharmacol Ther 49:44–48.
14. Bryant MS, Vineis P, Skipper PL, Tannenbaum SR (1988): Hemoglobin adducts of aromatic amines: Associations with smoking status and type of tobacco. Proc Natl Acad Sci U S A 85:9788–9791.
15. Bell DA, Taylor JA, Butler MA, Stephens EA, Wiest J, Brubaker LH, Kadlubar FF, Lucier GW (1993): Genotype/phenotype discordance for human arylamine *N*-acetyltransferase (NAT2) reveals a new slow-acetylator allele common in African-Americans. Carcinogenesis 14:1689–1692.
16. Ross RK, Jones PA, Yu MC (1996): Bladder cancer epidemiology and pathogenesis. Sem Oncol 23:536–545.
17. Kirlin WG, Trinidad A, Yerokun T, Ogolla F, Ferguson RJ, Andrews AF, Brady PK, Hein DW (1989): Polymorphic expression of acetyl coenzyme A-dependent arylamine *N*-acetyltransferase and acetyl coenzyme A-dependent *O*-acetyltransferase-mediated activation of *N*-hydroxyarylamines by human bladder cytosols. Cancer Res 49:2448–2454.
18. Frederickson SM, Messing EM, Reznikoff CA, Swaminathan S (1994): Relationship between in vivo acetylator phenotypes and cytosolic *N*-acetyltransferase and *O*-acetyltransferase activities in human urothelial cells. Cancer Epidemiol Biomarkers Prev 3:25–32.
19. Board P, Coggan M, Johnston P, Ross V, Suzuki T, Webb G (1990): Genetic heterogeneity of the human glutathione transferases: A complex of gene families. Pharmacol Ther 48:357–369.
20. Bell DA, Taylor JA, Paulson DF, Robertson CN, Mohler JL, Lucier GW (1993): Genetic risk and carcinogen exposure: A common inherited defect of the carcinogen-metabolism gene glutathione *S*-transferase M1 (*GSTM1*) that increases susceptibility to bladder cancer. J Natl Cancer Inst 85:1159–1164.
21. Ross RK: Prostate cancer. In Schottenfeld D, Fraumeni J (eds): "Cancer Epidemology and Prevention." Oxford: Oxford University Press, in press.
22. Ross RK, Bernstein L, Judd H, Hanisch R, Pike MC, Henderson BE (1986): Serum testosterone levels in young black and white men. J Natl Cancer Inst 76:45–48.
23. Ross RK, Bernstein L, Lobo RA, Shimizu H, Stanczyk FZ, Pike MC, Henderson BE (1992): 5-α-reductase activity and risk of prostate cancer among Japanese and U.S. white and black males. Lancet 339: 887–889.
24. Thigpen AE, Davis DL, Gautier T, Imperato-McGinley J, Russell W (1992): The molecular basis of steroid 5-alpha- reductase deficiency in a large Dominican kindren. N Engl J Med 327:1216–1219.
25. Davis DL, Russell DW (1993): Unusual length polymorphisms in human steroid 5-α-reductase type 2 gene (*SRD5A2*). Hum Mol Genet 2:820.
26. Reichardt JKV, Makridakis N, Henderson BE, Yu MC, Pike MC, Ross RK (1995): Genetic variablility of the human SRD5A2 gene: Implications for prostate cancer risk. Cancer Res 55:3973–3975.
27. Brentano ST, Picado-Leonard J, Mellon SH, Moore CCD, Miller WL (1990): Tissue-specific, cyclic adenosine 3',5'-monophosphate-induced, and phorbol ester-repressed transcription from the human P450c17 promoter in mouse cells. Mol Endocrinol 4:1972–1979.
28. Carey AH, Waterworth D, Patel K, White D, Little J, Novelli P, Franks S, Williamson R (1994): Polycystic ovaries and premature male pattern baldness are associated with one allele of the steroid metabolism gene CYP17. Hum Mol Genet 3:1873–1876.
29. Kadonaga JT, Jones KA, Tjian R (1986): Promoter-specific activation of RNA polymerase II transcription by SpI. Trends Biochem Sci 11:20–23.
30. Jenster G, Van der Korput HAGM, Van Vroonhoven C, Van der Kwast TH, Trapman J, Brinkmann AO (1991): Domains of human androgen receptor involved in ste-

roid binding, transcriptional activation, and subcellular localization. Mol Endocrinol 5:1396–1404.
31. La Spada AR, Wilson EM, Lubahn DB, Harding AE, Fischback KH (1991): Androgen receptor gene mutations in X-linked spinal and bulbar muscular atrophy. Nature 352:77–79.
32. Mhatre AN, Trifiro MA, Kaufman M, Kazemi-Esfarjani P, Figlewicz D, Rouleau G, Pinsky L (1993): Reduced transcriptional regulatory competence of the androgen receptor in X-linked spinal and bulbar muscular atrophy. Nature Genet 5:184–188.
33. Edwards A, Hammond HA, Jin L, Caskey CT, Chakraborty R (1992): Genetic variation at five trimeric and tandem repeat loci in four human population groups. Genomics 12:241–253.
34. Irvine RA, Yu MC, Ross RK, Coetzee GA (1995): The CAG and GGC microsatellites of the androgen receptor gene are in linkage disequilibrium in men with prostate cancer. Cancer Res 55:1937–1940.
35. Henderson BE, Gerkins V, Rosario I, Casagrande JT, Pike MC (1975): Elevated serum levels of estrogen and prolactin in daughters of patients with breast cancer. New Engl J Med 293:790–795.
36. Bernstein L, Yuan JM, Ross RK, Pike MC, Hanisch R, Lobo R, Stanczyk F, Gao YT, Henderson BE (1990): Serum hormone levels in premenopausal Chinese women in Shanghai and white women in Los Angeles: Results from two breast cancer case-control studies. Cancer Causes Control 1:51–58.
37. Shimizu H, Ross RK, Berstein L, Pike MC, Henderson BE (1990): Serum oestrogen levels in postmenopausal women: Comparison of American whites and Japanese in Japan. Brit J Cancer 62:451–453.
38. England P, Skinner L, Cottrell K, Sellwood RA (1974): Serum oestradiol-17β in women with benign and malignant breast disease. Brit J Cancer 30:571–576.
39. Poutanen M, Isomaa V, Lehto V-P, Vihko R (1992): Immunological analysis of 17β-hydroxysteroid dehydrogenase in benign and malignant human breast tissue. Int J Cancer 50:386–390.
40. Normand T, Narod S, Labrie F, Simard J (1993): Detection of polymorphisms in the estradiol 17β-hydroxysteroid dehydrogenase II gene at the EDH17B2 locus on 17q11-q21. Hum Mol Genet 2:479–483.
41. Mannermaa A, Peltoketo H, Winqvist R, Ponder BAJ, Kiviniemi H, Easton DF, Poutanen M, Isomaa V, Vihko R (1994): Human familial and sporadic breast cancer: Analysis of the coding regions of the 17β-hydroxysteroid dehydrogenase 2 gene (EDH17B2) using a single-strand conformation polymorphism assay. Hum Genet 93:319–324.
42. Puranen TJ, Poutanen MH, Peltoketo HE, Vihko PT, Vihko RK (1994): Site-directed mutagenesis of the putative active site of human 17β-hydroxysteroid dehydrogenase type 1. Biochem J 304:289–293.
43. Peltoketo H, Yunshang P, Mannermaa A, Pander BAJ, Isomaa V, Poutanen M, Winqvist R, Vihko R (1994): A point mutation in the putative TATA box, detected in nondiseased individuals and patients with hereditary breast cancer, decreases promoter activity of the 17β-hydroxysteroid dehydrogenase type 1 gene 2 (EHD17B2) in vitro. Genomics 23:250–252.
44. Castagnoli A, Maestri I, Bernardi F, del Senno L (1987): PvuII RFLP inside the human estrogen receptor gene. Nucleic Acids Res 15:866.
45. Garcia T, Sanchez M, Cox JL, Shaw PA, Ross JBA, Lehrer S, Schachter B (1989): Identification of a variant form of the human estrogen receptor with an amino acid replacement. Nuc Acids Res 17:8364.
46. Andersen TI, Heimdal KR, Skrede M, Tveit K, Berg K, Borresen A-L (1994): Oestrogen receptor (*ESR*) polymorphisms and breast cancer susceptibility. Hum Genet 94:665–670.
47. del Senno L, Aguiari GL, Piva R (1992): Dinucleotide repeat polymorphism in the human estrogen receptor (*ESR*) gene. Hum Mol Genet 1:354.
48. Keaveney M, Klug J, Dawson MT, Nestor PV, Neilan JG, Forde RC, Gannon F (1991): Evidence for a previously unidentified upstream exon in the human oestrogen receptor gene. J Mol Endocrinol 6:111–115.