

The Glutathione Biotransformation System and Colorectal Cancer Risk in Humans

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Evidence for a protective role of the glutathione biotransformation system in carcinogenesis is growing. However, most data on this system in relation to colorectal cancer originate from animal studies. Here we review the human data. In humans, a significant association was found between glutathione S-transferase (GST) activity in the mucosa along the gastrointestinal tract and the corresponding tumour incidence. Low activity was correlated with high tumour incidence and vice versa. Also, in normal colonic mucosa, GST activity is lower in patients at risk of colon cancer than in healthy controls and therefore interventions which increase the glutathione detoxification capacity may reduce cancer incidence. Consumption of vegetables and fruit is associated with a lower risk of colorectal cancer. Human intervention studies showed that (components from) vegetables induced colonic glutathione detoxification capacity. Such an effect could contribute to a lower colon cancer risk, but further data are needed. The human GSTs consist of four main classes—alpha (A), mu (M), pi (P) and theta (T)—each of which is divided into one or more isoforms. Functional polymorphisms are known for the GST genes M1, P1 and T1 and they all lead to less active enzymes compared to the wild-type gene products. However, studies that compared these GST polymorphisms in relation to colon cancer risk were not conclusive with respect to an increased or decreased risk of a particular genotype. Diet or medication can also influence the expression levels of specific isoenzymes and the effect of such interventions on cancer risk deserves more attention.

Key words: Biotransformation; colorectal cancer risk; glutathione; glutathione S-transferase; humans

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Biotransformation enzymes play a role in the metabolism of carcinogens. Glutathione S-transferases (GSTs) are a major class of biotransformation enzymes. It has been postulated that GSTs can deactivate carcinogens and thus prevent cancer. Evidence for a protective role of the glutathione biotransformation system in carcinogenesis is indeed growing. However, most data in this respect originate from animal studies. Conclusions from animal studies cannot be merely translated to the human situation.

The aim of this review is to give an overview of currently available data on the relation between the glutathione biotransformation system and colorectal cancer risk obtained from clinical and epidemiological studies in humans. Firstly we explain what biotransformation is and describe the proposed working mechanisms. Subsequently we describe the cellular levels of enzymes and substrates involved in biotransformation, known as the phenotype, as well as general aspects of the variation in genes that encode for biotransformation enzymes, known as the genotype. We also provide

arguments for a protective role of GSTs in cancer susceptibility.

What is Biotransformation?

Biotransformation is the sum of all chemical reactions in living organisms that alter the structure and aqueous solubility of undesirable compounds, in general leading to biologically less active molecules that can readily be excreted (1). Many enzyme systems are involved in biotransformation and they catalyse a wide variety of reactions. The main purpose of the biotransformation enzyme systems is to facilitate the elimination of potentially toxic compounds. This usually involves two distinct stages: phase I metabolism involves oxidation or reduction of chemical compounds and phase II involves conjugation reactions (2). In general, phase II biotransformation decreases the overall toxicity of chemical compounds, whereas phase I reactions often activate such compounds (2). The most important phase I enzymes are the cytochrome P450 enzymes, whereas the GSTs play an important role in the

phase II metabolism. The GSTs catalyse the conjugation of glutathione to a wide variety of substrates. The metabolites formed by this reaction are generally less toxic than their precursors and are more water-soluble, thus facilitating faecal or urinary excretion. It has been speculated that, in this way, GSTs can deactivate carcinogens and prevent cancer.

Biotransformation occurs in most, if not all, living organisms, including humans. The human GSTs consist of four main classes: alpha (A), mu (M), pi (P) and theta (T), each of which is divided into one or more isoforms. GSTT is, for example, divided into two isoforms: GSTT1 and GSTT2. The expression levels of GST isoenzymes are highly tissue-specific. The most important organs with respect to biotransformation are the liver and kidneys, organs particularly involved in detoxification. However, biotransformation enzymes are also active in the oesophagus, stomach, small intestine and colon.

Working Mechanism of Biotransformation

Biotransformation may be involved in the mechanism of so-called anticarcinogens, that is, compounds that inhibit tumour formation (3). The mechanisms leading to a purported anticarcinogenic action have not been elucidated. Most data originate from *in vitro* and animal studies. According to the stage in the carcinogenic process at which inhibitors are effective, three categories can be distinguished (3). The first category consists of compounds that prevent the formation of carcinogens from precursor substances. Ascorbic acid, for example, inhibits the formation of carcinogenic N-nitroso compounds (4). Secondly, some anticarcinogens, known as 'blocking agents', induce detoxifying enzyme systems that prevent carcinogens from forming or reacting with critical targets. Organosulphur compounds from garlic and onions induced an increase in GST activity and also inhibited the benzo[a]pyrene-induced neoplasia in the forestomach of mice (5). The third category comprises the so-called 'suppressing agents', agents that suppress the transformation of cells that have previously been exposed to carcinogens. Isothiocyanate, for example, inhibits carcinogen-induced neoplasia in rodents when administered after the carcinogen exposure (3).

General Aspects of Genotyping

Differences between people in terms of the risk of developing colon cancer may depend on differences in genetic make-up. An advantage of genotyping studies (studying the gene that encodes for an enzyme) is that they can be performed relatively easily: a small amount of blood is adequate to collect sufficient DNA. Polymorphisms are defined as less frequent variants of genotypes, which occur in at least 1% of a population and are caused by mutation of the gene. They may result in enzymes with enhanced or reduced activity or even in complete absence of enzyme activity, when a stop codon is introduced. The latter is seen, for example, in the GSTT1 or

GSTM1 null polymorphisms. Polymorphisms may influence cancer susceptibility in particular organs, as can be illustrated by the 'classical' N-acetylation polymorphism of N-acetyltransferase 2 (NAT2). Polymorphisms in NAT2 result in a rapid or slow acetylator phenotype. Rapid acetylators may have a slightly higher risk of colon cancer (odds ratio (OR) 1.19; 95% confidence interval (CI) 1.02–1.39), as shown in a recent meta-analysis (6), whereas slow acetylators may have an increased risk of bladder cancer (OR 1.37; 95% CI 1.20–1.57) (6). Genetic polymorphisms associated with only a slightly increased cancer risk may still be important to public health. This is especially so if the polymorphism and the specific type of cancer it is associated with are common in the general population (7).

General Aspects of Phenotyping

Phenotypic expression of enzymes can be determined by measurement of the actual enzyme levels in the target organ. This can be used to study the involvement of such enzymes in cancer susceptibility in that organ. In theory this is a better approach than studying the genotype. When only the genotype is known, hardly anything can be predicted concerning enzyme expression in the target organ. An additional advantage of direct enzyme activity measurements is the possibility to monitor changes in enzyme activities due to environmental factors. Phenotyping is the only way to really understand the action of the gene in a specific organ (8). The main disadvantage of the phenotypic approach is the difficulty of collecting adequate amounts of human target tissue. For instance, it is necessary to perform an endoscopy to collect colon samples. Studies on the phenotype of the biotransformation system, which are generally also more laborious than genotyping studies, have therefore hardly been performed.

Protective Role of GSTs in Carcinogenesis

Evidence for a protective role of GSTs in carcinogenesis is inconclusive. It is based on the hypothesis that tissues with low or reduced levels of GSTs may have a low or reduced capacity to detoxify carcinogens, resulting in more cytogenetic damage, which in turn can lead to a higher tumour risk (9–16). The first line of evidence is found in genetic epidemiology. In humans, genetic polymorphisms in GSTs are present. Such polymorphisms in GSTM1, GSTT1 or GSTP1 have been implicated in the increased risk for malignancies in the pituitary gland (17), larynx (18), bladder (18–20), stomach (14), colon (14) and lung (21–25). However, subsequent studies have often failed to confirm these associations (26, 27).

A second line of evidence comes from observations of damage to DNA in cells. Increased cytogenetic damage was observed in *in vitro* studies with human white blood cells with the GSTM1 or GSTT1 null polymorphism (15, 16, 28), and higher levels of polycyclic aromatic hydrocarbon DNA

Table I. Human intervention trials in which glutathione biotransformation in the colon was assessed

Intervention	Study type	Population	No. of subjects	Biomarker	Result	P	Reference, year
Brussels sprouts	CO; 2×1 week	Healthy controls	10	GST activity GST Alpha GST Pi Glutathione	+30% +15%	N.S. 0.005 0.05 N.S.	52, 1995
Oltipraz	1 week	Patients	26	GST activity γ -GCSmRNA Glutathione	+21% +500%	0.05 <0.001 N.S.	53, 1995
Broccoli supplement	Parallel; 2 weeks	Patients	29	GST activity		N.S.	54, 1997
Unfiltered coffee	CO; 2×2 weeks	Healthy controls	64	GST activity Glutathione	+8%	N.S. 0.01	60, 2000

CO = cross-over; GST = glutathione S-transferase; N.S. = not significant; γ -GCS = γ -glutamylcysteine synthetase.

adducts were found in the lung tissue of GSTM1 null subjects (29). High levels of DNA adducts in the human colon are associated with colorectal cancer (11).

Thirdly, human pathological tissues at high risk for malignant degeneration contain significantly lower levels of GST than normal tissues. Examples of such pathological tissues are Barrett's oesophagus (13) and small-intestinal mucosa of patients with celiac disease (30). This observation is compatible with a decreased capacity to detoxify carcinogens, resulting in more cytogenetic damage, which in turn could lead to a higher tumour risk.

A fourth line of evidence comes from the observation that normal human tissues with a low tumour incidence (liver, small intestine) contain high GST enzyme levels, whereas tissues with a high tumour risk (colon, lung, breast) have relatively low levels of GSTs (2, 13).

Fifthly, naturally occurring compounds from vegetables and fruit (31) are able to raise the levels of GSTs in several organs of rodents, including those of the gastrointestinal tract (2, 5). Some of these compounds have also been shown to inhibit chemically induced oesophageal, gastric or colorectal tumours (5, 32–37).

The sixth line of evidence results from the observation that non-steroidal anti-inflammatory drugs, which have been shown to reduce the risk of colorectal adenomas and carcinomas in humans (38, 39), are also able to enhance GSTs of the rat gastrointestinal tract (40).

Last, but not least, GSTP1/P2 knockout mice are much more susceptible to skin carcinogenesis than mice with an intact GSTP1/P2 gene (41). In addition, transgenic rats harbouring an extra rat GST P gene were less sensitive for liver tumorigenesis than non-transgenic rats (42). These results suggest that GST P protects against skin and liver carcinogenesis in rodents and this may presumably be due to an enhanced detoxification capacity.

Glutathione Biotransformation Phenotype in the Human Colon

Human studies on GST enzyme levels, activity and glu-

tathione availability in the colon are scarce. GST enzymes, in combination with their substrate glutathione, have detoxifying capacities. Our group reported a significant inverse correlation between GST activity in the mucosa along the gastrointestinal tract and tumour incidence at these sites in humans (13). In addition, the GST activity was lowest in the colon (13). We therefore suggested that the biotransformation activity may be critically low in the colon, which may partly explain the high cancer risk in this part of the gut (12). Furthermore, we showed that the colonic biotransformation capacity is even lower in patients at risk of colon cancer than in healthy controls (43). The decreased biotransformation capacity seems to be independent of hereditary factors which play a role in hereditary colon cancer syndromes, such as hereditary non-polyposis colorectal cancer (HNPCC) and familial adenomatous polyposis (FAP) (43). This observation suggests that the glutathione biotransformation system can modify the colorectal cancer risk.

The incidence rate of colorectal cancer appears to be strongly influenced by environmental factors, with diet as a major factor (44). An interesting hypothesis is that the capacity of the colonic epithelium to resist damage by mutagens or carcinogens can be upregulated and that this induction of protective enzymes could be feasible as an approach to colon cancer prevention. As discussed above, several of the non-nutrient compounds in fruits and vegetables, e.g. α -angelicalactone, coumarin, flavone, (iso)thiocyanate and indole, can induce GSTs and inhibit chemically induced colorectal tumours in rodents (2, 3, 45). In humans, diets high in fruit and vegetables are associated with a lower colorectal cancer risk (44, 46, 47). Four dietary intervention studies which focused on the induction of the glutathione biotransformation system in the colon have been published so far and a summary is given in Table I.

The anticarcinogenic properties of cruciferous vegetables have been attributed to degradation products of glucosinolates, e.g. (iso)thiocyanates and indoles (48), dithiolethiones (49, 50) and sulforaphanes (51). In a cross-over trial in 10 healthy controls, we found that daily consumption of 300 g of Brussels sprouts for 1 week resulted in 30% and 15%

Table II. Studies on colorectal cancer and GSTM1 null polymorphism. ORs are given for GSTM1 null polymorphism versus the other GSTM1 polymorphisms

Country	CRC	Controls	OR	95% CI	Reference
UK	26	49	2.3	0.9–6.1	14
UK	196	225	1.8	1.2–2.6	24
Australia	132	200	0.9	0.6–1.4	66
UK	252	577	1.0	0.7–1.3	63
Japan	103	126	1.5	0.9–2.6	72
Australia	219	200	1.0	0.7–1.4	64
USA	211	221	1.0	0.7–1.5	65
Singapore	300	183	0.8	0.5–1.1	67
USA	1567	1949	0.9	0.8–1.0	68
Egypt	63	53	0.6	0.3–1.2	69
UK	178	178	1.1	0.7–1.6	73
Poland	28	145	2.5	1.0–6.1	74
The Netherlands	32	247	1.1	0.5–2.2	*
Hungary	163	163	1.2	0.8–1.8	75
USA	1579	1898	0.9	0.8–1.1	70

* Unpublished result.

CRC = Colorectal cancer cases.

increases in rectal GSTA and GSTP1 levels, respectively (52).

O'Dwyer et al. (53) performed a study with Oltipraz, a synthetic dithiolethione, in patients at increased risk of colorectal cancer. After a single dose of 250 mg/m² Oltipraz, a 21% increase in GST activity was seen in the sigmoid colon at Day 3.

The capacity to conjugate chemical compounds with glutathione depends on its intracellular availability. The synthesis of glutathione is regulated by the activity of the key enzyme γ -glutamylcysteine synthetase. Although the colonic glutathione concentration was not influenced by Oltipraz, the γ -glutamylcysteine synthetase activity was increased 6-fold (53). In contrast, Clapper et al. (54) reported no significant difference in GST activity in the sigmoid colon of 29 patients at increased risk of colorectal cancer who consumed 3 g of broccoli supplements per day in a parallel controlled study.

Several epidemiological studies have suggested that consumption of coffee may protect against colorectal cancer (55–59). Recently we studied the effect of unfiltered coffee on glutathione and GST activity in the rectal mucosa. Sixty-four healthy controls were randomly assigned to two groups in a

placebo-controlled cross-over design. Treatments were 1 L of unfiltered coffee daily or no coffee for a period of 2 weeks. Unfiltered coffee significantly increased the glutathione content in the colorectal mucosa by 8%, whereas no effect on GST activity was noticed (60).

Comments on studies of glutathione biotransformation phenotype in the human colon

The observation of an inverse relation between the GST activity in the gastrointestinal tract and the tumour incidence at these sites (13), as well as the observation of a decreased glutathione biotransformation capacity in the colon of patients at risk of colorectal cancer (43), is compatible with a possible protective role of GSTs in colorectal cancer development.

Only four studies to date have investigated the hypothesis that the detoxifying capacity of the colon epithelium might be upregulated by dietary interventions. Although 3 of these studies were relatively small (52–54), the results are promising. All but 1 study reported a significant increase in 1 or more glutathione-related biomarkers (52, 53, 60). The negative result in the broccoli study might be explained by the low dosage used (54), which was approximately 10 times less than the quantity of Brussels sprouts (300 g/daily) given in our study (52).

Table III. Studies on colorectal cancer and GSTT1 null polymorphism. ORs are given for GSTT1 null polymorphism versus the other GSTT1 polymorphisms

Country, year	CRC	Controls	OR	95% CI	Reference
Australia, 1995	125	148	0.9	0.4–1.7	66
UK, 1996	211	509	1.9	1.3–2.7	63
Japan, 1996	103	126	1.2	0.7–2.0	72
Australia, 1997	219	200	3.3	2.1–5.2	64
USA, 1998	209	220	0.7	0.4–1.1	65
UK, 1999	178	178	1.2	0.7–2.1	73
Egypt, 1999	59	51	0.8	0.4–1.8	69
The Netherlands, 2000	32	247	0.6	0.2–1.7	*

* Unpublished result.

CRC = Colorectal cancer cases.

GST Genotypes and Colorectal Cancer Risk in Humans

Of the four main cytosolic GST isoenzyme classes that have been identified in humans, genetic polymorphisms have been described in three of them: GSTM1, GSTP1 and GSTT1. General interest in the role of polymorphisms in determining disease susceptibility has been stimulated by the relative simple methodology of genotyping, based on polymerase chain reaction, which has resulted in a considerable amount of

Table IV. Studies on colorectal cancer and GST-P1 polymorphism. ORs are given for the GST P1b and/or P1c polymorphisms versus the GST P1a polymorphism (wild type)

Country, year	CRC	Controls	OR	95% CI	Reference
UK, 1997	100	155	1.3	0.5–3.3	20
Australia, 1998	219	398	0.2	0.1–1.5	77
UK, 1999	196	178	0.6	0.3–1.2	73

CRC = colorectal cancer cases.

data on genetic polymorphisms in GSTM1, GSTP1 and GSTT1 and susceptibility to colon cancer (61).

GSTM1

Three different alleles have been described at the GSTM1 locus on chromosome 1p13.3. One of these is a partial deletion of the gene and the effect of the homozygous deletion of GSTM1, the so-called GSTM1 null genotype, is that no GSTM1 enzyme activity is present. The other two alleles do not lead to functional differences (62).

The frequency of the GSTM1 null genotype ranges from 23% to 62% in different populations worldwide, and is approximately 50% in Caucasians (27).

Until now 15 case-control studies on the role of the GSTM1 null genotype in colorectal cancer have been published. The results are inconsistent and are summarized in Table II. Three studies (63–65) found no association between the GSTM1 null genotype and colorectal cancer, 5 studies (66–70) reported a decreased risk, with ORs of 0.6–0.9, and in 7 studies (14, 71–75; Grubben et al., unpublished data) the GSTM1 null genotype was associated with an increased risk of colorectal cancer, with ORs of 1.1–2.5. In only 2 of these 7 studies was a significant increase in the risk of colorectal cancer found (71, 74). Zhong et al. (71) reported an increased risk with an OR of 1.8 (95% CI 1.2–2.6) among 196 cases and 225 controls from a Scottish population. More recently, Gawronksa et al. (74) found an OR of 2.5 (95% CI 1.0–6.1) among 28 Polish cases and 145 controls.

GSTT1

Two different alleles have been described at the GSTT1

locus on chromosome 12q11.2, one of which is a partial gene deletion. The effect of the homozygous deletion of GSTT1, the GSTT1 null genotype, is no activity of the GSTT1 enzyme (62).

The frequency of the GSTT1 null genotype ranges from 16% to 64% in different populations worldwide, and is approximately 20% in Caucasians (27).

Since 1995 eight case-control studies on the association between the GSTT1 null genotype and colorectal cancer have been published. The results are inconsistent and are summarized in Table III. Four studies (65, 66, 69; Grubben et al., unpublished data) reported a decreased risk of colorectal cancer in GSTT1 null individuals, with ORs of 0.6–0.9. In the other 4 studies (63, 64, 72, 73) the GSTT1 null genotype was associated with an increased risk of colorectal cancer, with ORs of 1.2–3.3. In 2 studies there was a significantly increased risk: Deakin et al. (63) reported an OR of 1.9 (95% CI 1.3–2.7) among 211 Caucasian cases and 509 blood-bank controls in the UK and Butler et al. (64) found an OR of 3.3 (95% CI 2.1–5.2) among 219 cases and 200 controls, all of whom were Caucasian.

GSTP1

At least four different alleles have been described at the GSTP1 locus on chromosome 11, coding for the respective GSTP1a (most common or wild type), 1b, 1c and 1d forms. The 1c and 1d forms are very rare. The effect of the GSTP1b polymorphism is a reduced activity of the enzyme compared to the wild type (76).

Only three case-control studies have assessed the colorectal cancer risk in relation to GSTP1b and/or GSTP1c polymorphisms (Table IV). One study (20) showed an increased risk (OR 1.3) and the other 2 studies (73, 77) demonstrated a decreased risk (ORs 0.2 and 0.6) of the polymorphisms compared to the wild type. In none of the studies was a statistically significant difference reported.

Comments on the GST genotype studies

The results of the influence of GST polymorphisms on colorectal cancer risk are inconsistent (Tables II–IV). The overall interpretation of these genotype studies is difficult

Table V. Studies on colorectal cancer and combinations of polymorphisms in biotransformation. ORs are given for the specific polymorphisms versus the other polymorphisms of these genes

Country, year	CRC	Controls	Polymorphisms	OR	95% CI	Reference
UK, 1996	218	448	M1-0 + T1-0	1.3	0.8–2.2	63
USA, 1998	208	221	M1-0 + T1-0	1.1	0.6–2.1	65
Singapore, 1998	37†	183	M1-0 + T1	1.9	0.9–4.1	67
Egypt, 1999	208	221	M1-0 + T1-0	0.5	0.2–1.2	69
UK, 1999	195	172	Slow NAT2 + T1-0	2.3	1.1–4.7	73
Hungary, 2000	163	163	M1-0 + CYP*	4.6	1.3–17.0	75

† Right-sided CRC.

* CYP 1A1 Val + CYP 2E1C2 polymorphism.

CRC = Colorectal cancer cases; M1-0 = GSTM1 null genotype; T1-0 = GSTT1 null genotype; T1 = GSTT1 genotype; NAT2 = N-acetyltransferase 2 genotype.

Table VI. Studies on colorectal cancer in HNPCC and GST polymorphisms. ORs are given for the specific polymorphisms versus the other polymorphisms of these genes

Country, year	CRC	Controls	Polymorphisms	OR	95% CI	Reference
Finland, 1998	182*	70	M1-0	1.4	0.7–2.4	81
	182*	70	T1-0	2.3	0.9–5.7	
	182*	159†	M1-0	0.8	0.5–1.3	
	182*	159†	T1-0	1.2	0.7–1.1	
	49* prox.	29* dist.	M1-0 + T1-0	4.0	0.5–34.2	
Poland, 1999	17‡	145	M1-0	1.1	0.4–3.1	74

* MLH1 mutation carriers.

† MLH1 non-mutation carriers from the HNPCC families.

‡ HNPCC patients according to Amsterdam I criteria.

CRC = colorectal cancer cases. M1-0 = GSTM1 null genotype; T1-0 = GSTT1 null genotype; prox = proximal CRC; dist. = distal CRC.

because of differences in study designs, and most studies are relatively small with limited statistical power. Most studies included hospital-based cases, whereas most control groups were not population-based but were recruited in diverse ways, e.g. from hospitals and blood donors, randomly selected from driver's license lists and social security lists or by means of random digit dialling, etc. Also, the populations differed with respect to racial background. Large studies with a uniform study design could probably give more consistent results.

The frequency of the GST null genotypes ranges from 16% to 64% in different populations worldwide (27). This may have an impact on the contribution of a specific GST genotype to colorectal cancer risk.

GSTP1 is the most abundant GST isoform in the colon, accounting for approximately 80% of total GST enzyme activities (78). The GSTP1b polymorphism does not lead to complete absence of enzyme activity, as found for the GSTT1 and GSTM1 null genotypes, but has decreased enzyme activity. Therefore it may be less easy to demonstrate an association between the GSTP1 polymorphisms and colorectal cancer risk than for the GST null polymorphisms. However, the associations between the GSTM1 or GSTT1 null genotypes and colorectal cancer risk are not convincing. This may be explained in part by the relatively small contribution of GSTM1 and GSTT1 to the overall colonic mucosal GST enzyme activity.

GST enzymes have an overlap of substrate specificity. A reduced activity of one GST isoenzyme may be compensated in part by another isoenzyme. Therefore GST gene–gene interactions have also been investigated. Since 1996, 6 case-control studies investigated combinations of polymorphisms in biotransformation enzymes participating in carcinogen metabolism (Table V) (63, 65, 67, 69, 73, 75). All but 1 study (69) reported an increased risk, with ORs of 1.1–4.6. Two studies reported on combinations of GST and NAT2 (73) or cytochrome P450 (CYP) (75) polymorphisms, respectively, and found a statistically significant increase in colon cancer risk. The ORs (2.3 and 4.6) in these small studies are relatively high compared to the ORs reported in Tables II–IV. Analyses of gene–gene interactions of Phase I (CYP) and Phase II (GST, NAT) biotransformation enzymes may give

additional insight into the role of GST in colorectal cancer risk.

The occurrence of colon cancer in a patient may indicate increased colon cancer risk for other family members (79, 80). Hereditary colorectal cancer aspects were not always excluded in cases and controls and may obfuscate the analysis of GST polymorphism in association with colon cancer risk in the general population. Two studies (74, 81) analysed the occurrence of GST polymorphism in cases of HNPCC (Table VI). One study (81) investigated patients with colorectal cancer who were also carriers of a mutation in 1 of the HNPCC genes called MLH1 (82). Analyses were performed with two control groups: patients with sporadic colorectal cancer and individuals from HNPCC families who did not carry the MLH1 mutation. In the HNPCC patients, the presence of GSTM1 or GSTT1 null polymorphisms was shown to be associated with an increased risk of colorectal cancer, with ORs of 1.4 and 2.3, respectively, compared to patients with sporadic colorectal cancer. Compared to individuals from the HNPCC families who did not carry the MLH1 mutation the ORs were 0.8 and 1.2, respectively. The combined null genotypes of GSTM1 and GSTT1 were associated with proximal tumours in HNPCC patients with an OR of 4.0. The second study (74) investigated the GSTM1 null genotype and reported an OR of 1.1 in HNPCC patients who were identified according to the Amsterdam I criteria (83) compared to a population-based control group. In support of the decreased glutathione biotransformation capacity in the colonic mucosa of HNPCC patients (43), these epidemiological data indicate that GSTs probably play a modifying role in colorectal cancer risk in HNPCC patients and that hereditary aspects in the design of such studies are essential in order to unravel the exact role of the GSTs.

Summary and Conclusions

Glutathione biotransformation levels in colon mucosa are low, which may be an important factor in colon carcinogenesis. Measurement of the expression levels of biotransformation enzymes, the so-called phenotype, is the preferred way to really understand the role and interaction of the biotransfor-

mation genes with respect to the process of carcinogenesis in a specific organ. So far only four human studies have been published wherein the hypothesis that the detoxifying capacity of the glutathione biotransformation system in the colon epithelium may be upregulated with dietary interventions was tested. The results are mixed: all but one study reported a significant increase in one or more components of the glutathione biotransformation system, but the effects were not consistent between the studies. Also, human data on the association between the GST genotypes and colorectal cancer risk are inconsistent with regard to a possible promoting effect of the polymorphic GST isoenzymes with reduced enzyme activity.

Future studies may focus on aspects of gene-gene interactions and their consequences for the expression levels of the various biotransformation enzymes. The feasibility of up-regulation of the detoxifying capacity of the glutathione biotransformation system in the colon may enable us to develop specific diets, dietary supplements, nutraceuticals or functional foods in order to influence health beneficially, but solid evidence for such an influence is still a long way off.

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