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The role of glutathione-*S*-transferase in anti-cancer drug resistance

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Glutathione-*S*-transferases (GSTs) are a family of Phase II detoxification enzymes that catalyse the conjugation of glutathione (GSH) to a wide variety of endogenous and exogenous electrophilic compounds. GSTs are divided into two distinct super-family members: the membrane-bound microsomal and cytosolic family members. Microsomal GSTs are structurally distinct from the cytosolic in that they homo- and heterotrimerize rather than dimerize to form a single active site. Microsomal GSTs play a key role in the endogenous metabolism of leukotrienes and prostaglandins. Human cytosolic GSTs are highly polymorphic and can be divided into six classes: α, μ, ω, π, θ, and ζ. The π and μ classes of GSTs play a regulatory role in the mitogen-activated protein (MAP) kinase pathway that participates in cellular survival and death signals via protein : protein interactions with c-Jun N-terminal kinase 1 (JNK1) and ASK1 (apoptosis signal-regulating kinase). JNK and ASK1 are activated in response to cellular stress. GSTs have been implicated in the development of resistance toward chemotherapy agents. It is plausible that GSTs serve two distinct roles in the development of drug resistance via direct detoxification as well as acting as an inhibitor of the MAP kinase pathway. The link between GSTs and the MAP kinase pathway provides a rationale as to why in many cases the drugs used to select for resistance are neither subject to conjugation with GSH, nor substrates for GSTs. GSTs have emerged as a promising therapeutic target because specific isozymes are overexpressed in a wide variety of tumors and may play a role in the etiology of other diseases, including neurodegenerative diseases, multiple sclerosis, and asthma. Some of the therapeutic strategies so far employed are described in this review.
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GSTs As Phase II Detoxification Enzymes

Glutathione *S*-transferases (GSTs) are a family of Phase II detoxification enzymes that function to protect cellular macromolecules from attack by reactive electrophiles. Specifically, GSTs catalyse the conjugation of glutathione (GSH) to a wide variety of endogenous and exogenous electrophilic compounds (Figure 1). Glutathione conjugation is the first step in the mercapturic acid pathway that leads to the elimination of toxic compounds. GSTs have evolved with GSH, and are abundant throughout most life forms. GSTs are divided into two distinct super-family members: the membrane-bound microsomal and cytosolic family members. Cytosolic GSTs are subject to significant genetic polymorphisms in human populations. They are divided into six classes, which share $\sim$30% sequence identity, and are designated by Greek letters α, μ, ω, π, θ, and ζ (Table 2). Although risen from a single common ancestor, their substrate specificity and diversity have been reshaped by gene duplication, genetic recombination, and an accumulation of mutations. These have accorded GSTs properties consistent with promiscuous substrate specificities geared to act on functional groups rather than specific compounds (Tables 1 and 2). In general, substrates of GST are hydrophobic and have carbon at the electrophilic center; however, some contain nitrogen, oxygen, or sulfur.

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GSTs as regulators of the MAP kinase pathway

The first GST described was originally identified as 'ligandin' due to its ability to interact covalently and noncovalently with various compounds that are not substrates for enzymatic activity, including steroids, thyroid hormones, bile acid, bilirubin, and heme (Litwack *et al*., 1971; Tipping *et al*., 1978; Danielson and Mannervik, 1985). While the ligand-binding function remains unclear, sequestering molecules may serve a regulatory role, preventing cytotoxic ligands from interacting with their targets. Supporting this conclusion, recent studies have demonstrated a regulatory role for the π and μ classes of GSTs in the mitogen-activated protein (MAP) kinase pathway that participates in cellular survival and death signalling.

For example, GSTπ plays a key role in regulating the MAP kinase pathway via protein : protein interactions. Specifically, GSTπ was shown to be an endogenous inhibitor of c-Jun N-terminal kinase 1 (JNK1), a kinase involved in stress response, apoptosis, and cellular proliferation (Adler *et al*., 1999; Yin *et al*., 2000). In nonstressed cells, low JNK activity is observed due to the sequestration of the protein in a GSTπ : JNK

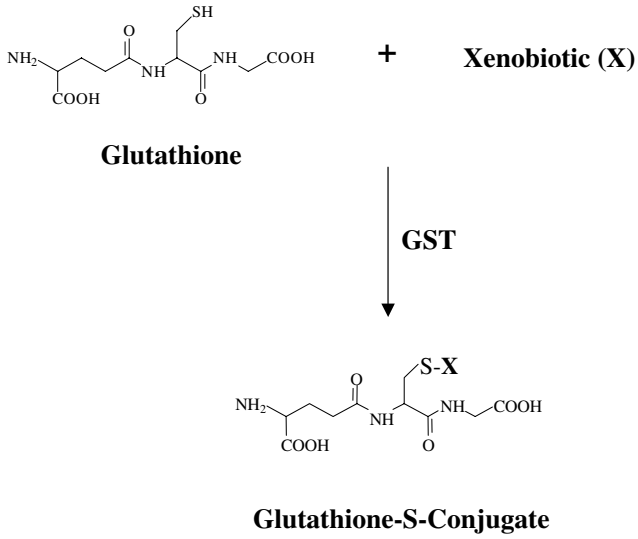

Figure 1 Glutathione conjugation to a generic xenobiotic (X) via GST results in the formation of a glutathione-*S* conjugate

Table 1 Substrates of GSTs

Endogenous
O-Quinones of catecholamines and dopamine (μGSTs)
Prostaglandins (microsomal GSTs, αGSTs, μGSTs)
Lipid peroxidation products generated by reactive oxygen species (αGST, μGST)
Exogenous
Polycyclic aromatic hydrocarbons (πGSTs)
α, β unsaturated aldehydes (πGSTs)
Molecules with epoxide groups (θGSTs)
Chemotherapeutic agents (πGSTs, αGSTs)

Table 2 Cytosolic GSTs

Class	*Gene*	*Alleles*	*Chromosome location*
Alpha (α)	GSTA1–2	GSTA1*A–B GSTA2*A–B	6
Mu (μ)	GSTM1-4	GSTM1*A–B, null GSTM3*A–B GSTM4*A–B	1, 6, 13
Omega (ω)	GSTO1		
Pi (π)	GSTP1	GSTP1*A–D	11
Theta (θ)	GSTT1-2	GSTT1*A, null	22
Zeta (ζ)	GSTZ1	GSTZ1*A–D	14

complex (Adler *et al*., 1999). Direct protein : protein interactions between the C-terminal of JNK and GSTπ were reported with a binding constant of approximately 200 nM (Wang *et al*., 2001). However, suppression of JNK activity is reversed by conditions of oxidative stress (UV irradiation or hydrogen peroxide treatment), resulting in the dissociation of the GSTπ : JNK complex, oligomerization of GSTπ, and induction of apoptosis (Adler *et al*., 1999). While the precise mechanism for the disruption of the ligand-binding interaction between these two proteins is not defined, the presence of an aggregation of GST monomers following oxidative stress implicates intermolecular disulfide bridge formation between available cysteine residues. In particular, it has been previously shown that cysteine residues at positions 14, 47, 117, and 169 of GSTπ are particularly susceptible to dimerization (Shen *et al*., 1993). Although it was determined that residues 47 and 101 were critical to the maintenance of catalytic activity, the protein : -protein interactions between GSTπ and JNK appear to be distal from, and independent of, the catalytic site of GSTπ. This novel, nonenzymatic role for GSTπ has direct relevance to the GST-overexpressing phenotypes of many drug-resistant tumors. As an endogenous switch for the control of signaling cascade pathways, elevated expression of GSTπ can alter the balance of regulation of kinase pathways during drug treatment, thereby conferring a potential selective advantage. This process can also provide a plausible explanation for the numerous examples of drug resistance linking GST overexpression with agents that are not substrates for these enzymes.

Additional support for the model of GST regulation is provided by the observations that either immunodepletion of GSTπ, or its inhibition by a rationally designed GSH-based-peptidomimetic inhibitor, -γ-glutamyl-*S*-(benzyl)cysteinyl-*R*(-)-phenyl glycine diethyl ester (TLK199), also results in the activation of JNK. The rational design and synthesis of TLK199 was based on the principle that modulation of drug resistance could be utilized as a viable clinical approach to cancer treatment. Earlier preclinical and later clinical protocols made use of ethacrynic acid as a broad-spectrum inhibitor of GST activity. In principle, GST-mediated drug resistance would be reversed by the addition of a reasonably nontoxic modulator. Indeed, studies in both tumor cell lines and animals confirmed that a therapeutic advantage was obtainable with combinations of ethacrynic acid and alkylating agents (Tew *et al*., 1988; Clapper *et al*., 1990). These preclinical results led to a clinical trial in chronic lymphocytic leukemia (CLL), where chlorambucil-resistant patients achieved further remission through ethacrynic acid cotreatment with the alkylating agent (Petrini *et al*., 1993). An analysis of GST levels in CLL patients showed an elevated expression of GST in individuals responding to chlorambucil and corticosteroids, and this expression was further enhanced in patients who had received multiple rounds of therapy and whose disease was classified as resistant to chlorambucil/steroid treatment (Schisselbauer *et al*., 1990). This analysis is one of the few correlates to show a cause/effect relationship for clinical drug resistance. However, the long-term utility of ethacrynic acid was compromised by dose-limiting toxicities related to diuresis and subsequent fluid imbalance (O'Dwyer *et al*., 1991).

ASK1 (apoptosis signal-regulating kinase) is a MAP kinase kinase kinase that activates the JNK and p38 pathways leading to cytokine- and stress-induced apoptosis (Ichijo *et al*., 1997). ASK1 is activated in response to oxidative stress and heat shock. Like JNK, the activity of ASK1 is low in nonstressed cells due to its sequestration via protein : protein interactions with

GSTM1 (GSTM1 : ASK1 complex), and/or thioredoxin (Trx : ASK1 complex) (Cho *et al*., 2001; Saitoh *et al*., 1998). The mechanism by which ASK1 is released from and activated by either of these two proteins is distinct. GSTM1 plays a regulatory role in the heat shock-sensing pathway, while thioredoxin plays a regulatory role in the oxidative stress-sensing pathway that leads to p38 activation. For example, oxidative stress triggers dissociation of the Trx : ASK1 complex by oxidizing thioredoxin, resulting in the activation of ASK1 (Tobiume *et al*., 2002). In contrast, dissociation of the GSTM1 : ASK1 complex is heat shock dependent. (Saitoh *et al*., 1998; Dorion *et al.,* 2002). Forced expression of GSTM1 blocked ASK1 oligomerization and repressed ASK1-dependent apoptotic cell death (Cho *et al*., 2001). GST-mediated regulation of the kinase pathways adds a new dimension to their known role in metabolism and cellular homeostasis. This link with kinase-mediated signaling could provide a plausible explanation for the increased expression of GSTπ in drug-resistant cells, even when the selecting drug is not a substrate for GST-mediated conjugation to GSH.

Resistance

Development of drug resistance is a key element in the failure of chemotherapy treatment. Exposure to anticancer agents may lead to the induction and expression of gene products that protect the cell. GSTs have been implicated in the development of resistance toward chemotherapy agents, insecticides, herbicides, and microbial antibiotics (Tew, 1994; McLellan and Wolf 1999; Tang and Tu, 1994; Ranson *et al*., 1997). It is plausible that GSTs serve two distinct roles in the development of drug resistance via direct detoxification as well as acting as an inhibitor of the MAP kinase pathway. Hence, it is not surprising that high levels of GSTs have been reported in a large number of tumor types (Tew, 1994). A survey of the NCI cancer drug screening panel of cell lines showed a correlation between GST expression and sensitivity toward alkylating agents (Tew *et al*., 1996). Table 3 is a list of anticancer agents to which resistance has been associated with elevated levels of GSTs. Some of these agents are substrates of GSTs and can be directly inactivated through catalytic conjugation to GSH through thioether bond formation. Many cancer drugs that decompose to produce electrophilic species can be detoxified via glutathione metabolism (Kauvar *et al*., 1998). It is frequently considered that increased resistance to a specific drug must be shown in transfected cells in order for the gene product to be causatively linked to a resistant phenotype. This one area has provided disparate and sometimes controversial results for GSTs. While earlier reviews have dealt in some detail with this issue (see for example, Tew (1994)), there are examples where GST isozyme transfections yield mild increases in resistance (mostly in the 2–5 fold range) to a number of different anticancer drugs. This has even been achieved in lower eukaryotes such as *Saccharomyces cerevisiae,* where a significant resistance to chlorambucil and doxorubicin (eight- and 16-fold, respectively) was reported in cells transfected with mammalian GST isozymes (Black *et al*., 1990). Interpretation of these earlier data did not take into account the more recent observations on the involvement of GSTs in kinase regulation.

Table 3 Anticancer agents associated with increased levels of GST and resistance

Substrates of GST
Chlorambucil
Melphalan
Nitrogen mustard
Phosphoramide mustard
Acrolein
Carmustine
Hydroxyalkenals
Ethacrynic acid
Steroids
Not characterized as substrates
Antimetabolites*
Antimicrotubule drugs*
Topoisomerase I & II inhibitors*
Bleomycin
Hepsulfam
Mitomycin C*
Adriamycin*
Cisplatin*
Carboplatin

*Require JNK activation for cytotoxicity

The link between GSTs and the MAP kinase pathway provides a rationale as to why many of the selecting drugs are neither subject to conjugation with GSH, nor substrates for GSTs (Tew, 1994). Many anticancer agents induce apoptosis via activation of the MAP kinase pathway, specifically via JNK and p38 (Davis, 2000; Ono and Han, 2000). The anticancer agents that require JNK activation for cytotoxicity are listed in Table 3 (Tew, 1994; Fan and Chambers, 2001). Cisplatin is an example of a drug whereby JNK activity is required for maximal cytotoxicity. Inhibition of the JNK signaling pathway leads to a decrease in cisplatin-induced apoptosis, while overexpression of c-jun increases the sensitivity of cells toward cisplatin (Potapova *et al*., 1997).

Elevated levels of GST are associated with increased resistance to apoptosis initiated by a variety of stimuli (Kodym *et al*., 1999; Voehringer *et al*., 2000; Cumming *et al.,* 2001). These data are consistent with GSTs acting as inhibitors of the MAP kinase pathway.

Aberrant cellular signaling is also a hallmark of the malignant phenotype, and thus high levels of GSTπ in many tumors may be either a cause or effect of the transformation process. The pathology of prostate cancer strongly supports these conclusions. Hypermethylation of the GSTπ regulatory region is the most common somatic alteration identified in human prostate cancer (Lin *et al*., 2001). This alteration results in the loss of GSTπ expression, and is proposed to occur during pathogenesis of the disease (Lee *et al*., 1994). Recently, a methyl-CpG-binding domain (MBD) protein that mediates hypermethylation of the GSTπ

regulatory region has been identified (Bakker *et al.,* 2002). These findings provide a possible target for restoration of GSTπ activity. GST expression and/or activity of specific isoforms is lost in some individuals with allelic variation. Although it has been speculated that reduced detoxification of possible carcinogens may be causal to malignant transformation and disease progression, a more plausible link may be through an altered capacity to regulate kinase-dependent proliferation pathways.

GSTs as a therapeutic target

GSTs have emerged as a promising therapeutic target because specific isozymes are overexpressed in a wide variety of tumors and may play a role in the etiology of other diseases, including neurodegenerative diseases, multiple sclerosis, and asthma (Tew, 1994; Baez *et al*., 1997; Mapp *et al*., 2002). Some of the therapeutic strategies so far employed will be described in this section. An initial approach was the design of GST inhibitors to act as reasonably non-toxic modulatory agents, useful in situations where conventional anticancer agents are detoxified by GSTs. Alternatively, compounds designed to disrupt the protein : protein interactions of GST with stress kinases also provide a possible therapeutic approach with respect to modifying proliferative responses. A further strategy is to exploit the elevated expression of GST in tumors, particularly GSTπ, through design of GST-activated prodrugs.

Inhibitors

A variety of GST inhibitors were shown to modulate drug resistance by sensitizing tumor cells to anticancer drugs (Tew *et al*., 1988; Hall *et al*., 1989; Ford *et al*., 1991). The first clinical modulatory studies focused on an approved drug, ethacrynic acid (EA). EA inhibits GST-α, -μ, and -π by binding directly to the substrate-binding site of the isozyme, as well as by depleting its cofactor, GSH, via conjugation of the Michael addition intermediate to the thiol group of GSH (Mulder *et al*., 1997; Oakley *et al*., 1997). EA has been reported to potentiate the cytotoxic effects of chlorambucil in human colon carcinoma cell lines and melphalan in human colon tumor xenografts in SCID mice (Tew *et al*., 1988; Clapper *et al*., 1990). The therapeutic value of EA as a chemosensitizer has also been demonstrated in patients (O'Dwyer *et al.,* 1991; Petrini *et al*., 1993). Since the clinical utility of EA was limited both by its diuretic properties, and its lack of isozyme specificity, efforts were made to develop inhibitors with more optimal isozyme specificity and clinical application. TLK199, one such lead compound, is a peptidomimetic glutathione analogue that is a low micromolar inhibitor of GSTπ (Figure 2) (Lyttle *et al*., 1994a,b). TLK199 acts as a chemosensitizer, and was shown to potentiate the toxicity of numerous anticancer agents in different tumor cell lines. In animal studies, sensitivity to melphalan was enhanced in xenograft models with elevated GST levels (Morgan *et al*., 1996). TLK199 has also been shown to be an inhibitor of the multidrug resistance-associated protein-1 (MRP-1), reversing the resistance to a variety of agents in NIH3T3 cells transfected with MRP-1 (O'Brien *et al*., 1999).

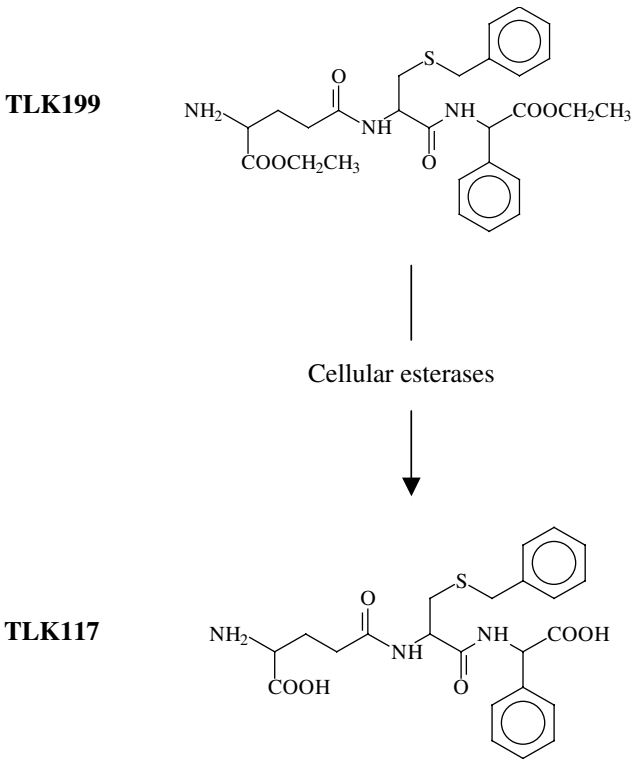

Figure 2 Structure of TLK199 and activation via cellular esterases to the active GST inhibitor TLK117

A serendipitous outcome to the preclinical studies with TLK199 was the observation that TLK199 behaves as a small molecular myeloproliferative agent in rodents (Ruscoe *et al*., 2001). Although this was an entirely unexpected observation, a plausible mechanism for this drug-induced myelostimulation is provided by the subsequently generated data that show that TLK199 can disrupt protein : protein interactions in the GST : JNK complex. In theory, this disruption of the kinase pathways can influence mitogenic response in marrow progenitor cells. Elevated levels of JNK were identified in HL60 cells made resistant to TLK199 via chronic exposure to the drug (Ruscoe *et al*., 2001). This corresponded to increased proliferation under stress conditions that induced apoptosis in the parental cell line. *In vivo* studies in myelosuppressed rodents showed a dose-dependent increase in the peripheral platelet and neutrophil counts within 24 h of drug treatment (Carver-Moore *et al*., 1996; Morgan *et al*., 1996). The collective preclinical results have now been translated into Phase I clinical trials in patients with myelodysplastic syndromes. These trials are ongoing.

GSTs may also be viable drug targets in disease states unrelated to cancer. For example, GSTs homologous to mammalian GST isozymes are present in many parasitic

organisms and provide targets for intervention, with the potential advantage of not compromising the human host. At a time when many of the standard drugs used to treat such diseases are frequently ineffective, these new targeting directives provide a timely opportunity. Many antiparasitic drugs (for example, chloroquinone (CQ), an antimalarial agent) form free radicals that may be inactivated by parasitic GST-mediated conjugation to GSH. (DeCampo and Moreno, 1984; Davioud-Charvet *et al*., 2001). The efficacy of CQ can be enhanced by GSH depletion and GST inhibition. Hence, inhibition of parasitic GSTs or destabilization of intraparasitic pools of GSH provide two means of combination therapy with CQ (Davioud-Charvet *et al*., 2001; Harwaldt *et al*., 2002). Investigations are underway to utilize the X-ray crystal structure of the malarial parasite's GST in the development of a structure-based drug design (Harwaldt *et al*., 2002).

Schistosomiasis, a debilitating tropical disease caused by the parasite *Schistosoma japonicum*, affects over 200 million people worldwide and results in about 500 000 deaths annually (Capron *et al*., 1995; Harwaldt *et al*., 2002). Present therapy for the disease uses oltipraz, a drug that binds directly to the schistosome GST in the integument of the trematode (Nare *et al*., 1992). Development of an effective vaccine is underway for long-term prevention. In fact, a variety of schistosome antigens are capable of protecting experimental animals from challenge. One such antigen that is a potential vaccine is the 28-kDa *S. mansoni* GST (Sm28GST) that confers protective immunity in transgenic mice expressing Sm28GST (Nare *et al*., 1992; Xu *et al*., 1997).Vaccination with Sm28GST was shown to decrease parasite fecundity and egg viability, thereby decreasing host pathology in rats, mice, and baboons. Following vaccination in human populations, an inverse correlation was found between IgA antibody production to Sm28GST and a decrease in parasitic egg production (Capron *et al*., 1995). An alternative vaccine directed against the *S. haematobium* GST (Sh28GST) has been shown to be well tolerated in Phase I and II clinical trials, and demonstrated the ability to block transmission of the parasite (Capron *et al*., 2001).

GST-activated prodrugs

Traditional chemotherapeutics are cytotoxins that target rapidly dividing cells. The therapeutic index is compromised because normal tissues, such as bone marrow, the gastrointestinal mucosa, and hair follicles, receive exposure equivalent to the tumor. An attractive treatment approach is provided by prodrug therapy. Prodrugs are rationally designed inactive agents that are converted to cytotoxins preferentially and specifically in target tissues by enzymes that are specifically elevated in the tumor. This strategy allows for an increased delivery of active agent to the tumor tissue, while minimizing the toxicity toward normal tissues. GSTs are a promising target because expression is enhanced in many tumors and high levels are sometimes correlated with poor prognosis. In addition, GSTP1-1 is frequently elevated in drug-resistant tumors. Thus, a two-pronged attack may be afforded by such an activation strategy. A number of synthetic endeavors have been instigated and are at various stages of development. Most have attempted to exploit the capacity of GSTs to catalyse GSH conjugation to electrophilic intermediates. For example, cis-3-(9H-purin-6-ylthio)acrylic acid (PTA) is a prodrug of the antitumor and immunosuppressive antimetabolite 6-mercaptopurine that requires GSH conjugation and subsequent metabolism for activation (Gunnarsdottir and Elfarra, 1999). Renal and hepatic GSTs enhance activation nearly two-fold – *in vivo* and *in vitro* (Gunnarsdottir and Elfarra, 1999). At this time, this drug is in early preclinical development. However, future clinical applications may be limited by the somewhat narrow spectrum of activity and limited efficacy of the parent drug 6-MP. A further approach has been to design prodrugs that exploit GST's ability to mediate cleavage of sulfonamides by promoting a β-elimination reaction. Hence, established agents as well as novel agents can be synthesized as inactive compounds via GSH conjugation through a sulfone linkage. One of these is a GSH analogue of cyclophosphamide that was shown to enhance toxicity selectively via GST activation in both cell and animal models (Kauvar, 1996).

The most advanced of the prodrug candidates is TLK286, γ-glutamyl-α-amino-β-(2-ethyl-*N,N,N′,N′*-tetrakis (2-chloroethyl) phosphorodiamidate)-sulfonyl-propionyl-(*R*)-(−) phenylglycine). This is the lead compound from a novel class of latent drugs activated in cancer cells by GSTP1-1 (Lyttle *et al.,* 1994b). GSTP1-1 promotes a β-elimination reaction that cleaves TLK286 into a GSH analogue and nitrogen mustard that can alkylate cellular nucleophiles (Figure 3) (Morgan *et al*., 1998). Sensitivity to the drug is correlated with GSTP1-1 expression both in cell culture and in animal models (Morgan *et al*., 1998; Rosario *et al.,* 2000). In contrast, downregulation of GSTP1-1 at the protein and transcript level was observed as an adaptive mechanism of resistance in a human promyelocytic cell line through long-term chronic exposure to TLK286. Furthermore, mouse embryo fibroblast cells from GSTπ null mice (deficient in both GSTP1-1 and P2-2) were less susceptible to TLK286 than their wild-type counterparts (Henderson *et al*., 1998). In addition, transfection of GSTπ into recipient cells increased their sensitivity to the drug (Rosario *et al*., 2000). These data support the rationale that tumors expressing high levels of GSTP1-1 will be more sensitive to the cytotoxic effects of TLK286. The efficacy of TLK286 was examined both *in vitro* and *in vivo*. Clonogenic assays showed that TLK286 had significant activity against 15 of 21 lung tumors and 11 of 20 breast tumors (Izbicka *et al*., 1997). Reports on Phase I clinical trials showed minor drug-related side effects that included fatigue, nausea, vomiting, and hematuria with no indications of cumulative toxicity. These results were combined with antitumor activity and/or disease stabilization in

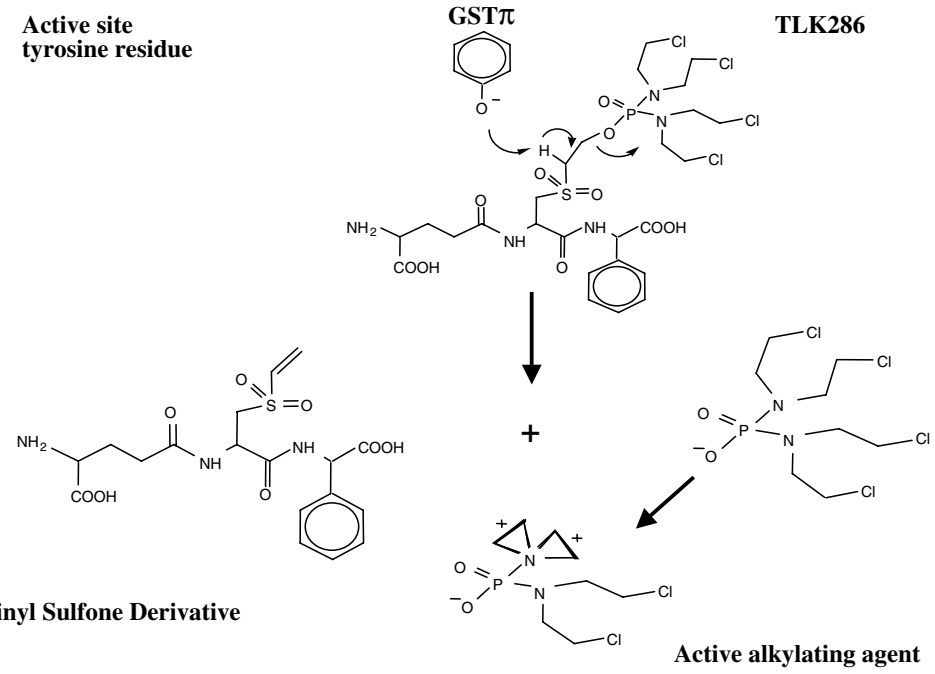

Figure 3 Structure of TLK286 and its activation by GSTπ

patients with various advanced malignancies (Brown *et al*., 2001; Rosen *et al*., 2003). None of the toxicities were grade IV, and there were no indications of sufficient seriousness to limit further clinical development. Clinical benefit has now been reported in single-agent Phase II trials currently underway in patients with advanced platinum-resistant non-small-cell lung cancer, platinum-resistant ovarian-cancer and 5-FU, leucovorin and irinotecan-resistant colon cancer (Kavanagh *et al*., 2002; Papadimitrakopoulou *et al*., 2003; Rosen *et al*., 2003). Such results have encouraged the formulation and application of Phase III studies, which will initially focus on combinations of TLK286 and taxotere in the treatment of drug-resistant NSCLC.

In summary, over the last two decades, a significant body of data has accumulated, linking the aberrant expression of GST isozymes with the development and expression of drug resistance. The initial confusion presented by the fact that not all drugs used to select for resistance were substrates for thioether bond catalysis by GSTs has been alleviated by the explanation that certain GST isozymes also possess the unexpected capacity to regulate MAP kinases. It is ironic to reflect that the recently reported protein ligand binding of GSTπ with JNK, and of GSTμ with ASK1, presents properties reminiscent of the 'ligandin' functionality ascribed to the capacity of liver GST to bind reversibly to heme and bilirubin (Litwack *et al*., 1971). This dichotomy of function has provided additional opportunities to use GSTs as plausible pharmaceutical targets. As GST isozymes (in particular GSTπ) are frequently upregulated in many solid tumors and lymphomas, prodrugs activated by GST-mediated catalysis have become a viable drug design concept. Additionally, the regulatory properties of GSTπ in kinase cascades have provided a translational opportunity to target GSTs in myeloproliferative pathways, with the consequent clinical testing of new agents in myelodysplastic syndrome.

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