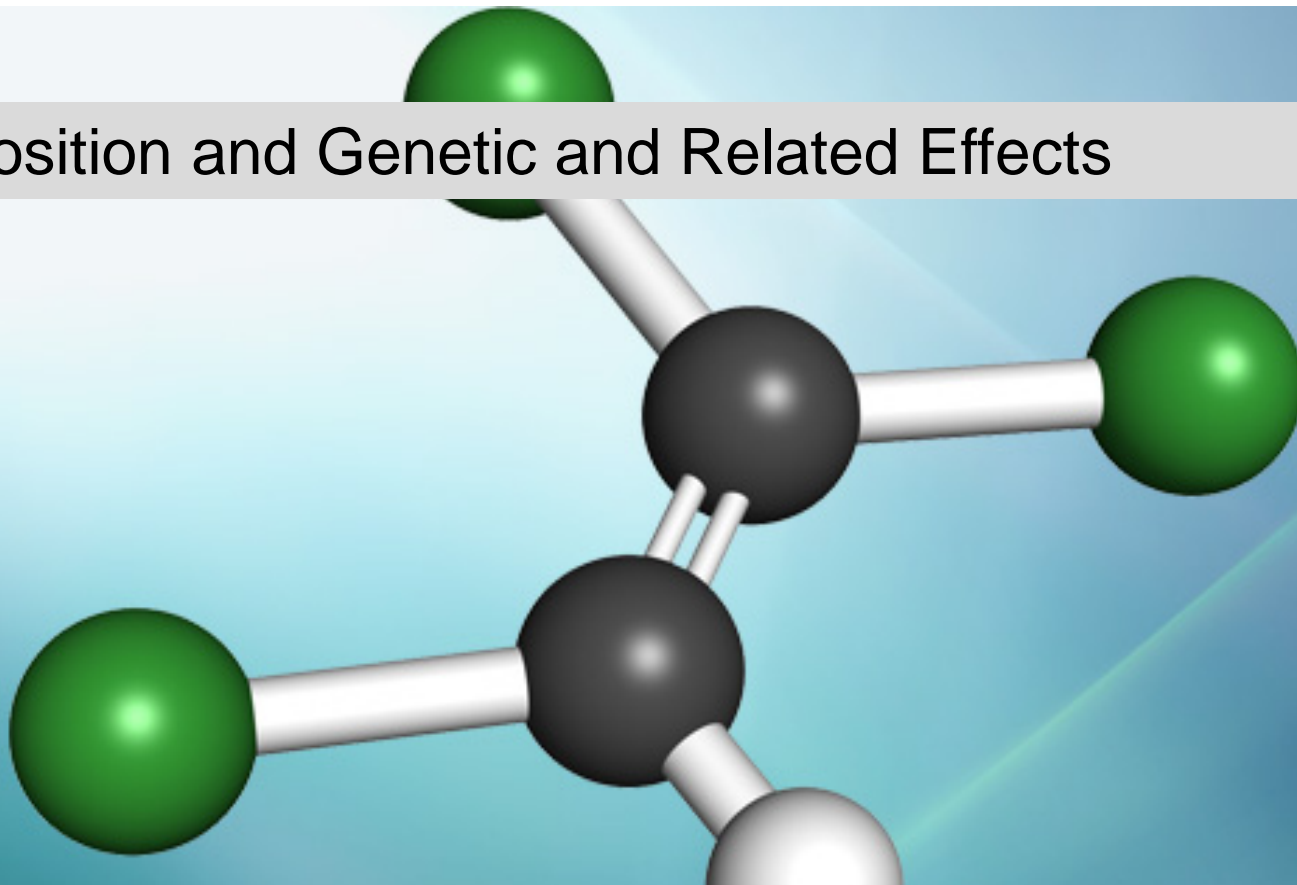




NTP

National Toxicology Program

Disposition and Genetic and Related Effects



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TCE: Disposition and Genetic Effects

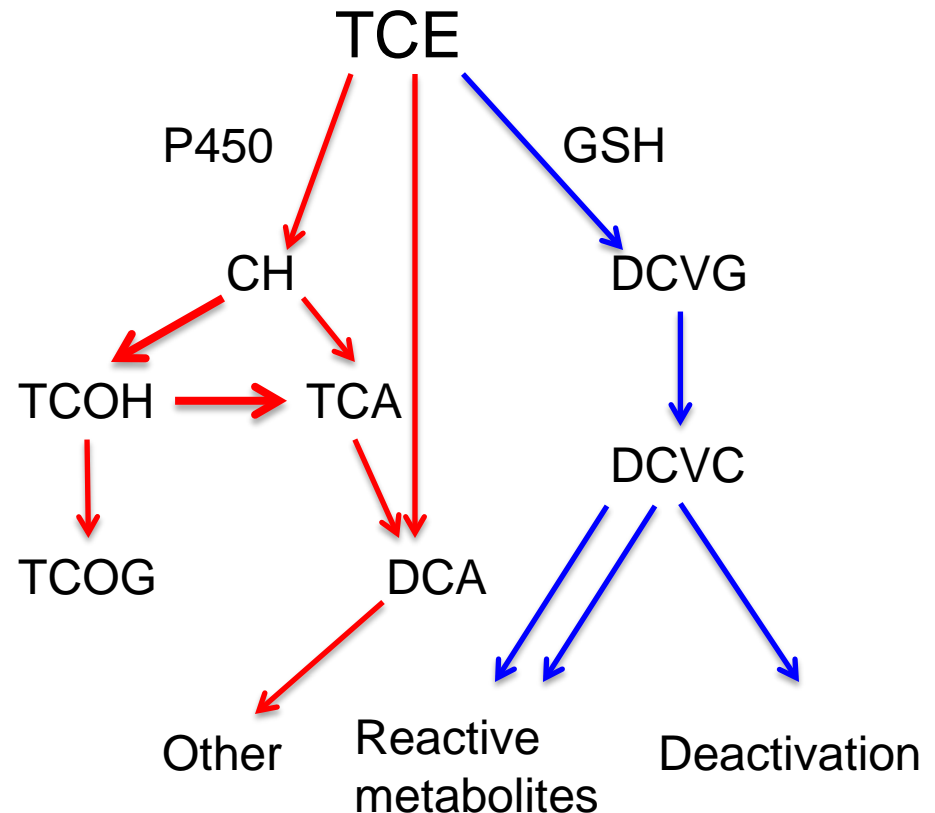
- Disposition
 - Absorption, distribution, excretion
 - Metabolism
- Genetic and related effects
 - TCE
 - Metabolites

Absorption, distribution and excretion

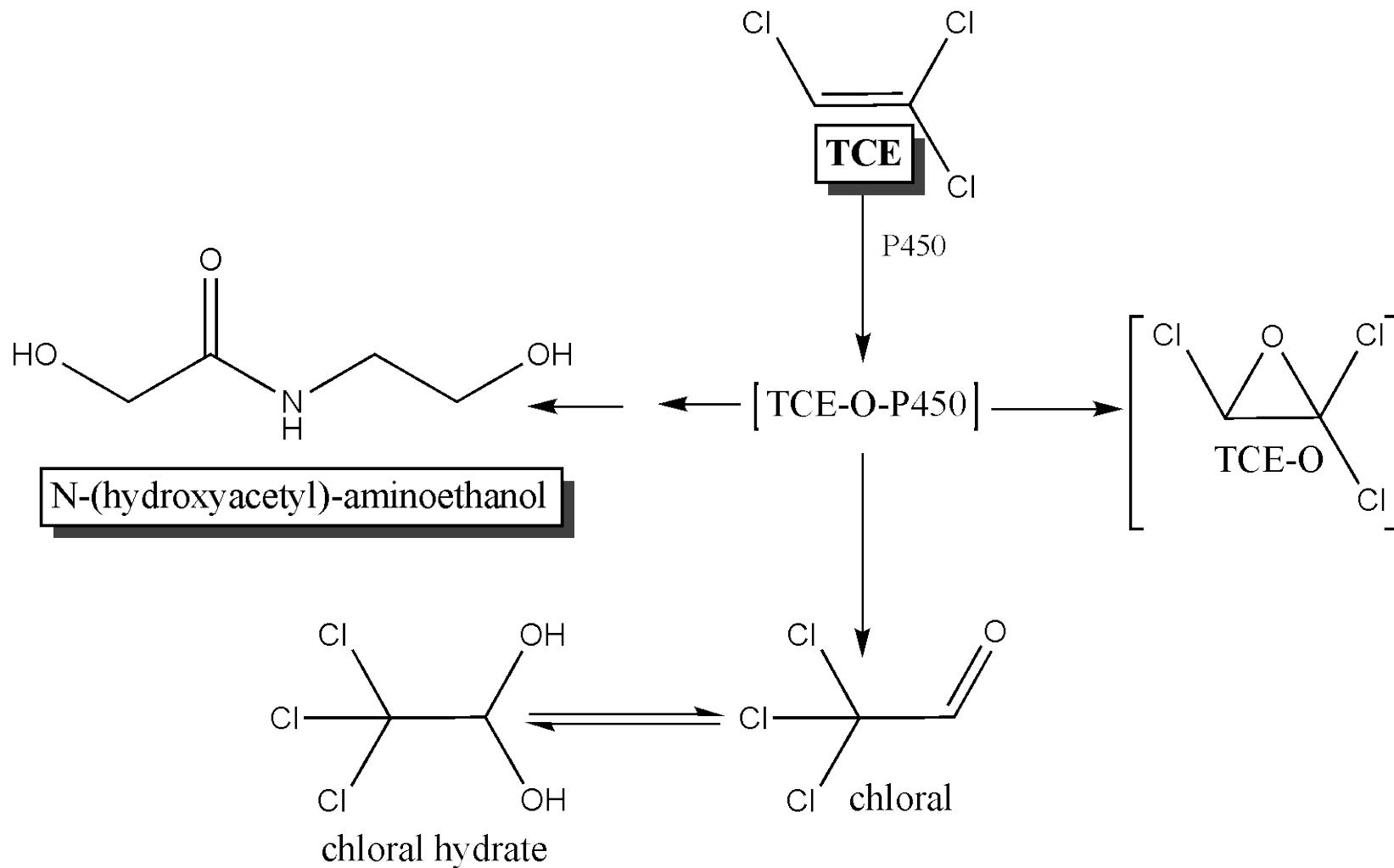
- Small lipophilic molecule
- Well absorbed from all routes
- Rapidly distributed to all tissues
 - Determined by blood:tissue partition coefficients
 - Highest concentrations in adipose tissue
- Excretion
 - Unchanged or CO₂ in exhaled breath
 - Metabolites in urine
 - Feces, sweat, saliva, milk

TCE metabolism is complex

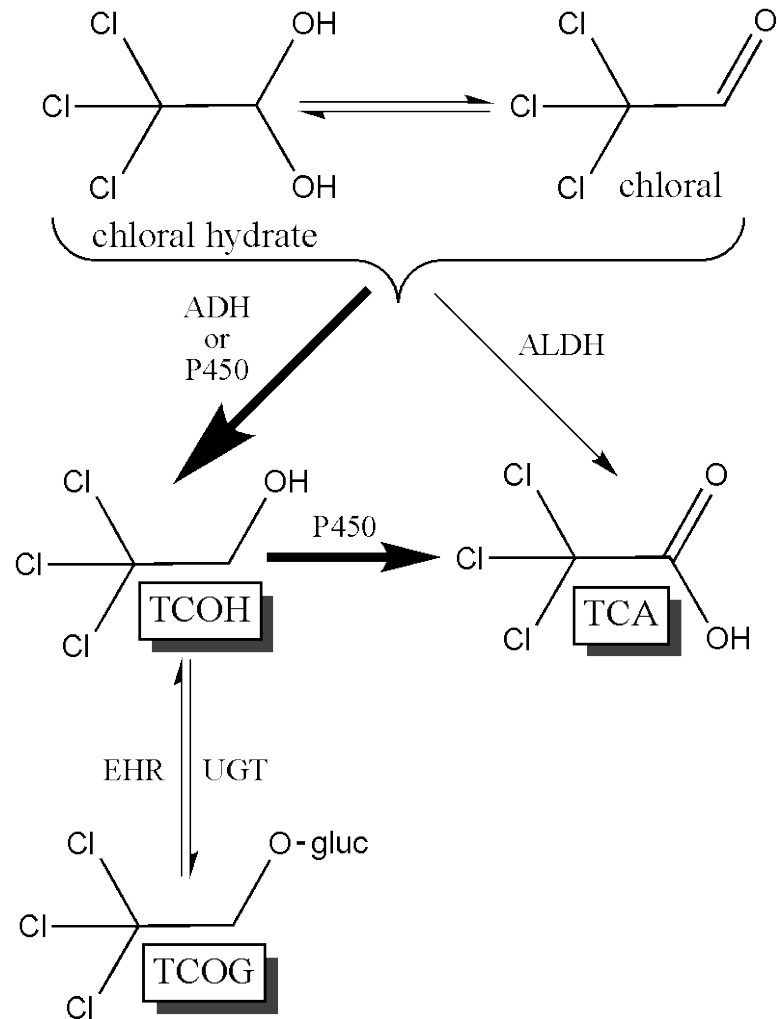
- Two key pathways
 - P450 (liver)
 - Dominant pathway
 - Several stable metabolites
 - GSH (liver/kidney)
 - Many reactive metabolites
 - One urinary metabolite
 - Flux uncertain (variable and can be altered)
 - Qualitatively similar in rodents and humans



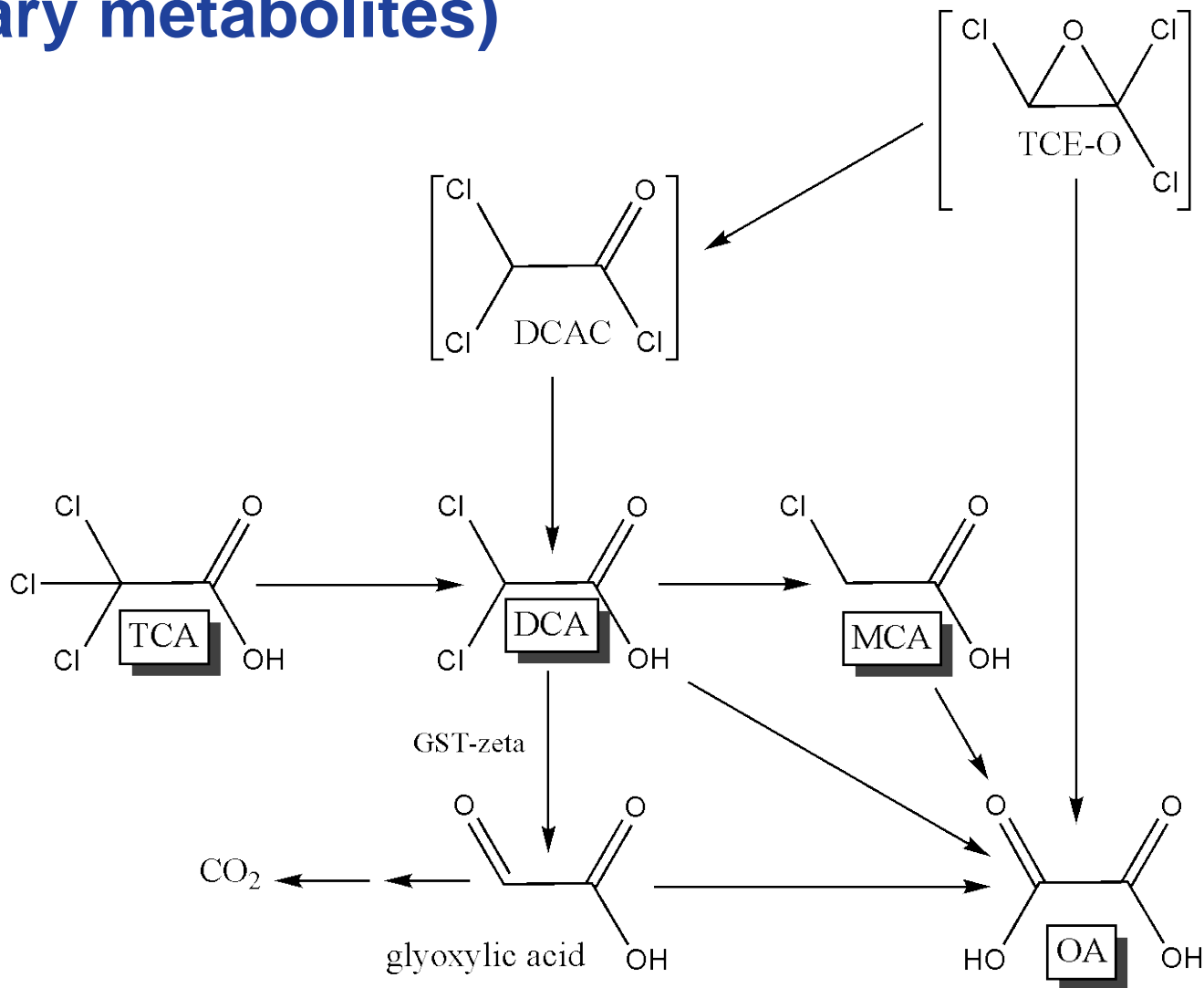
TCE metabolism: Oxidation (initial reactions)



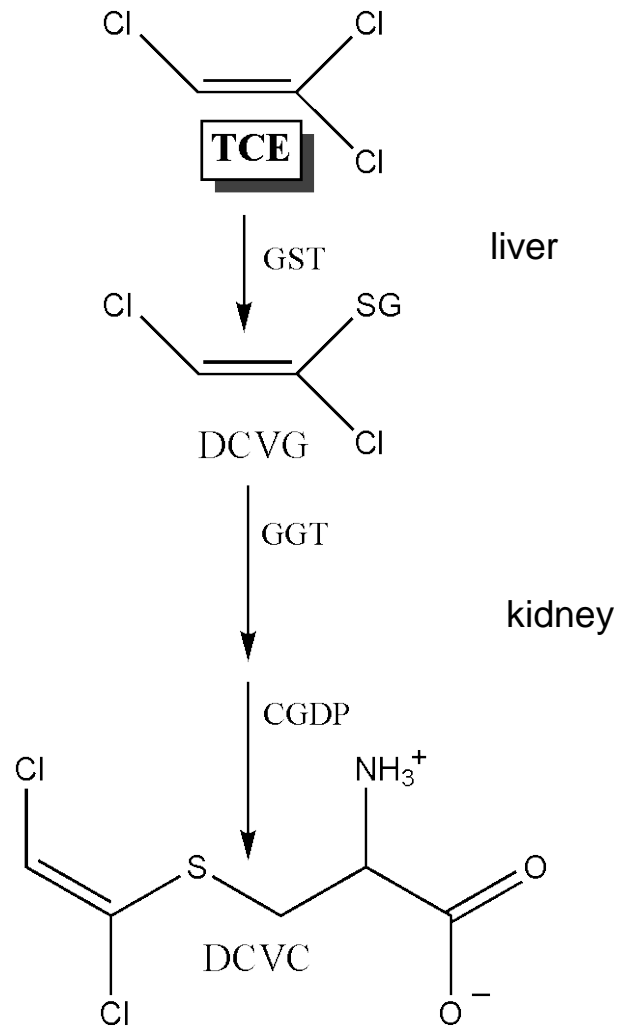
TCE metabolism: Chloral pathway (primary urinary metabolites)



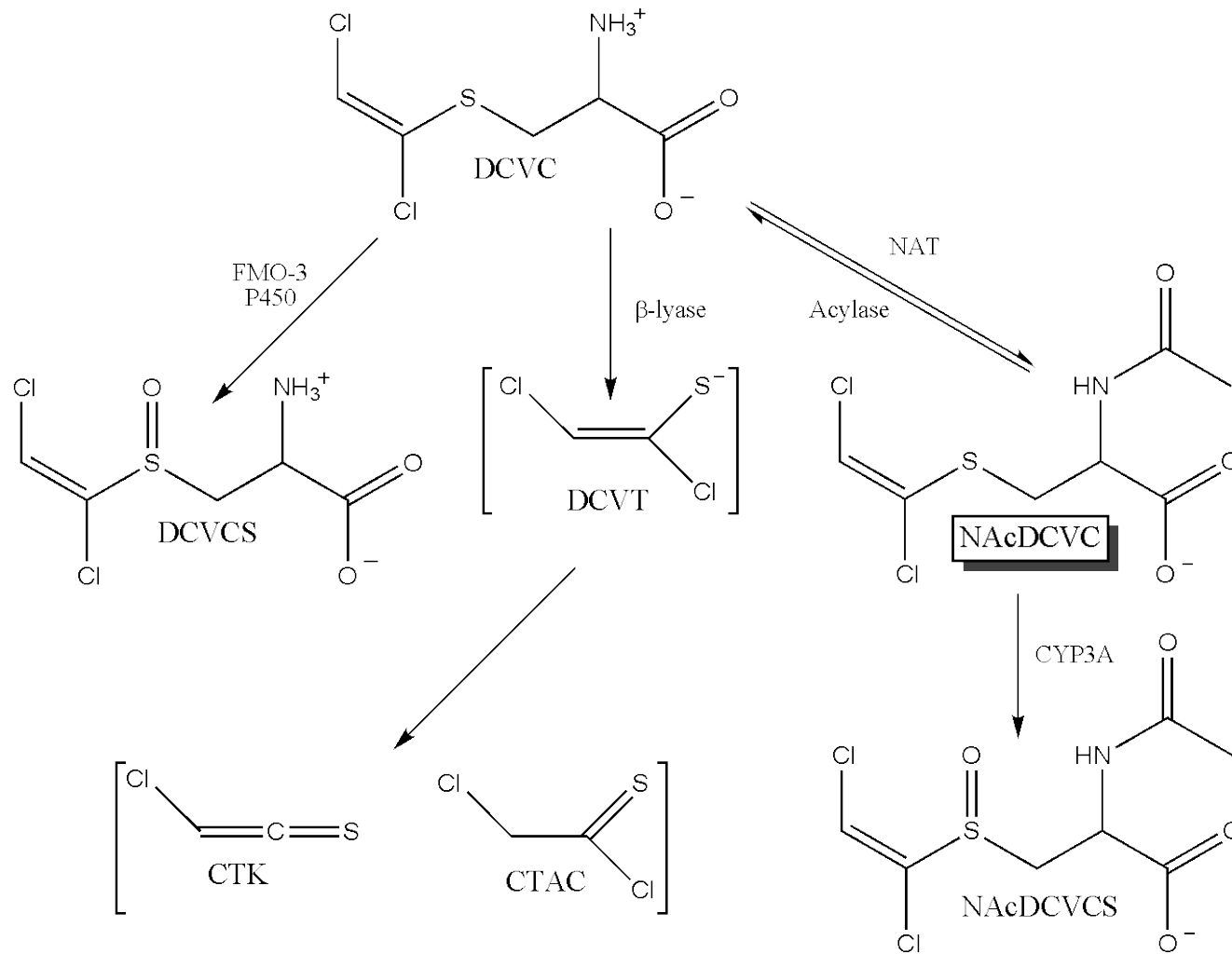
TCE Metabolism: TCE-oxide pathway (minor urinary metabolites)



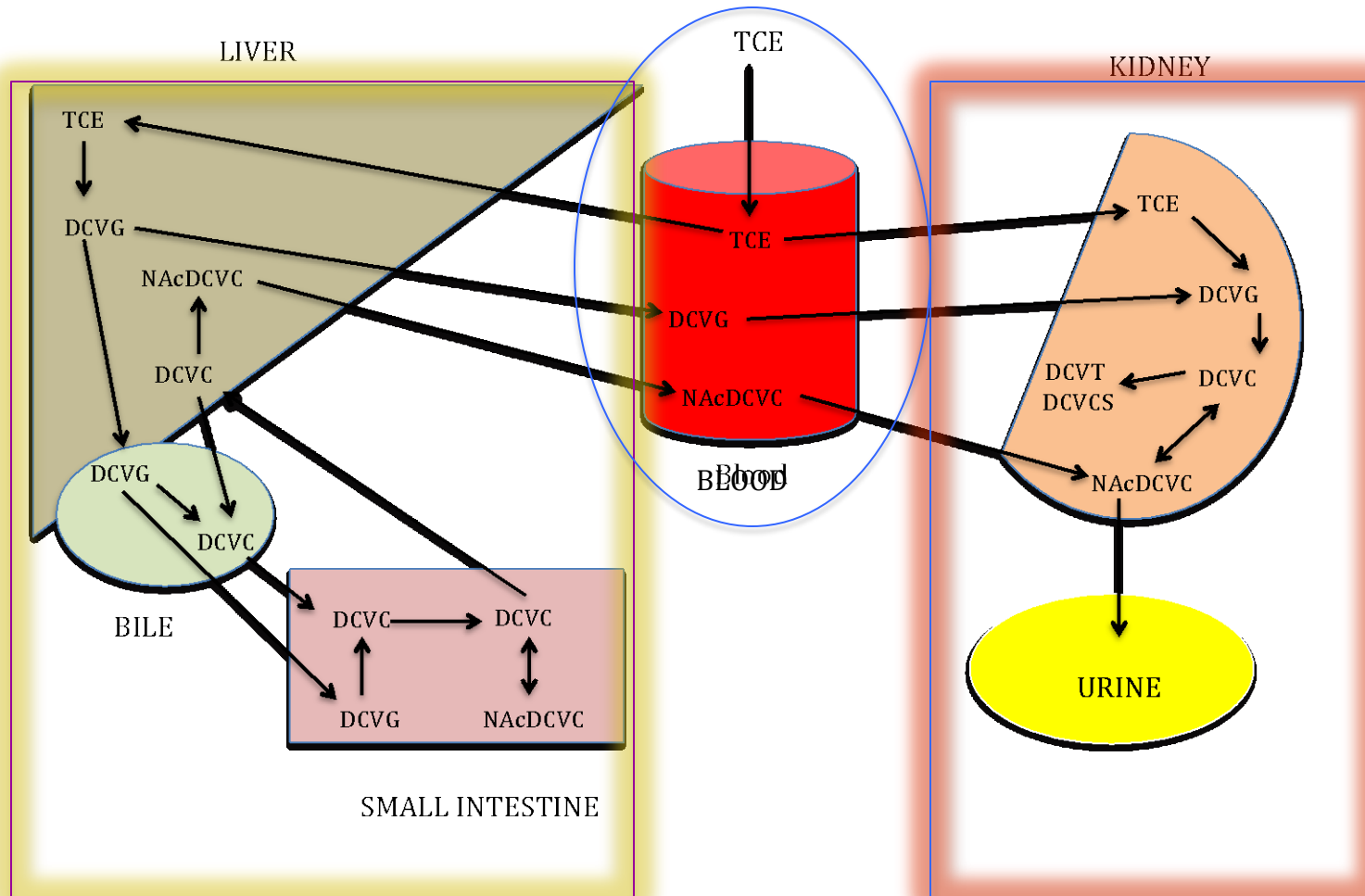
TCE Metabolism: GSH conjugation



TCE Metabolism: DCVC pathways (kidney)



TCE GSH Conjugates: Interorgan transport



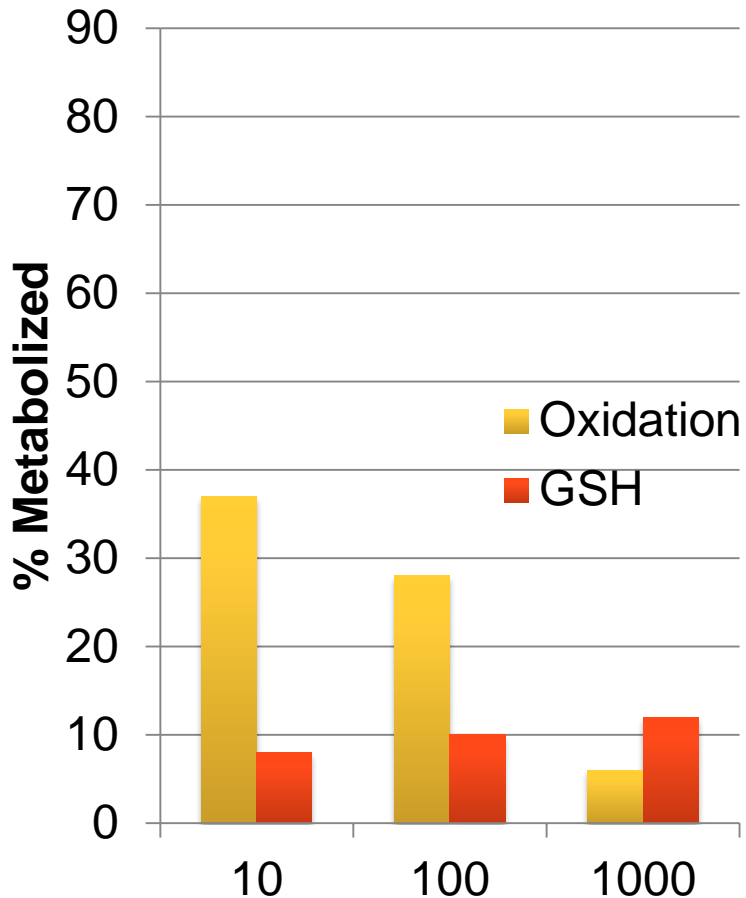
Adapted from Lash *et al.* 2014

TCE: GSH pathway is important

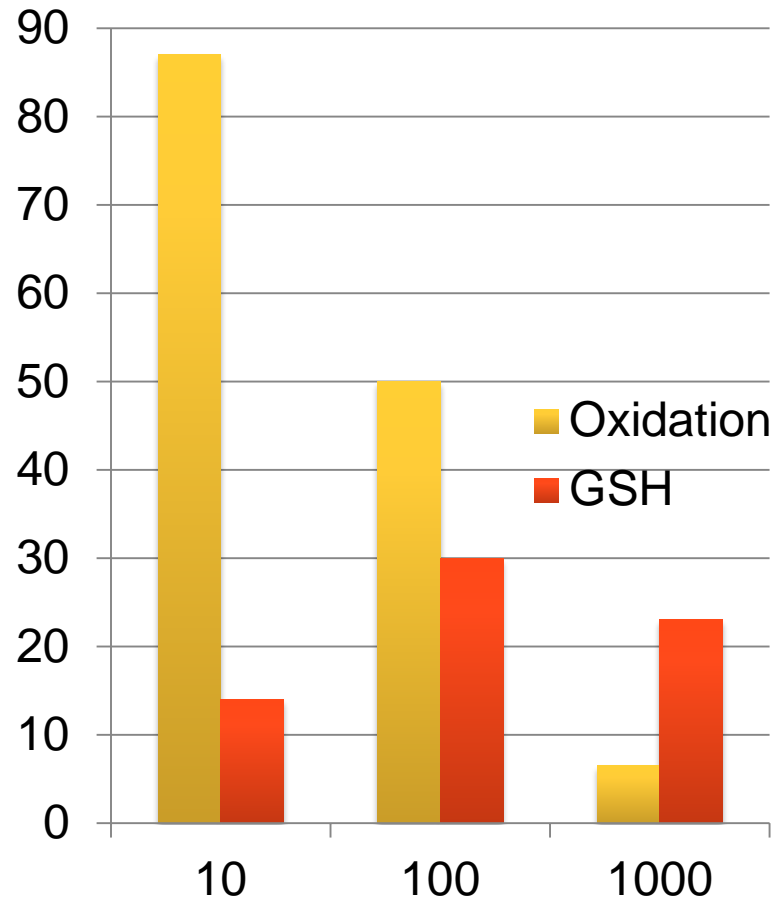
- Produces several reactive metabolites
- Flux estimates uncertain but may be higher than previously thought
 - Estimates of (TCA + TCOH)/NAcDCVC \geq 1000:1 but NAcDCVC in urine is a small fraction of total flux
 - DCVG blood levels similar to TCA/TCOH in human volunteers
 - *In vitro* K_m and V_{max} values for oxidation and conjugation in human hepatocytes and subcellular fractions show overlap
- Genetic polymorphisms in GSTs/P450s/other
- Exposure to P450 inducers/inhibitors
 - Impact likely greater at high substrate concentrations

PBPK model predictions TCE oxidation vs. GSH conjugation - humans

Inhalation (ppm)



Oral (mg/kg/day)



TCE/metabolites cause genetic effects *in vitro*

Endpoint	TCE	CH	TCA	DCA	DCVG/DC VC
Mutations	(-)	+	(-)	(+)	+ ^b
Aneuploidy	+	+	NT	-	NT
DNA strand breaks	+ ^a	-	(-)	-	+ ^c
UDS or DNA damage	(-)	+	-	+	+ ^c
Clastogenic effects	+ ^a	+	+	±	-
Cell transformation	±	+	NT	NT	+ ^c
DNA/protein binding	+	-	NT	NT	+

- = negative, (-) = probably negative, ± = mixed results, (+) = probably positive, + = positive; NT = not tested

^a Included strand breaks and MN in primary cultures of rat and human kidney cells and human HepG2 cells

^b Effects increased with a kidney-derived activation system and diminished with β-lyase inhibitor

^c Included effects in rodent kidney cells

TCE/metabolites cause genetic effects *in vivo*

Endpoint	TCE	CH	TCA	DCA	DCVG/DCVC
Mutations	-	NT	NT	+	NT
Aneuploidy	NT	± ^a	NT	NT	NT
DNA strand breaks	± ^b	± ^c	± ^{c,d}	(-)	± ^e
Clastogenic effects	(+) ^f	+ ^g	+	(-)	NT
DNA/protein binding	±	-	+	NT	+

- = negative, (-) = probably negative, ± = mixed results, (+) = probably positive, + = positive; NT = not tested

^a Positive in 2 of 4 studies in mouse blood or sperm

^b Generally positive in liver in rats/mice, kidney in mice, inconsistent findings in rat kidney

^c Positive in one study in liver (rats/mice) but not in another

^d Also positive in human HepG2 cells

^e Positive in rat proximal tubule cells (10 mg/kg at 2 hr); negative at 16 hr and at 1 mg/kg. Positive in male albino rabbit *in vivo* and in rabbit isolated kidney and proximal tubules (*ex vivo*).

^f Included MN in rat kidney

^g Included MN and SCE in peripheral blood lymphocytes of treated infant.

TCE Disposition and Genetic Effects: Summary

- Well absorbed from all routes and rapid distribution
- Highest tissue concentrations in fat
- Excreted unchanged in exhaled breath and as urinary metabolites
- Complex metabolism
 - Similar in humans and rodents
 - P450 pathway: TCA, TCOH, TCOG (liver)
 - GSH pathway: DCVC and other reactive metabolites (kidney)
- Genetic effects attributed to metabolites
 - Mutations
 - DNA and chromosome damage (MN)

TCE Disposition and Genetic Effects: Reviewer's questions/discussion

- Comment on whether the information on ADME and toxicokinetics is clear, technically correct, and objectively presented.
 - Identify any information that should be added or deleted.
- Comment on whether the genotoxicity data presented in the cancer evaluation component for TCE are clear, technically correct, and objectively presented.
 - Provide any scientific criticisms of the NTP's interpretation and application of the genotoxicity data from the cited studies for assessing effects of TCE.