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# DEVELOPMENT AND VALIDATION OF METHODS FOR APPLYING PHARMACOKINETIC DATA IN RISK ASSESSMENT

# **VOLUME V OF VII: VINYL CHLORIDE**

Clement International Corporation K.S. Crump Division 1201 Gaines Street Ruston, LA 71270



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HARRY G. ARMSTRONG AEROSPACE MEDICAL RESEARCH LABORATORY HUMAN SYSTEMS DIVISION AIR FGRCE GYSTEMS COMMAND WRIGHT-PATTERSON AIR FORCE BASE, OHIO 45433-6573



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The experiments reported herein were conducted according to the "Guide for the Care and Use of Laboratory Animals," Institute of Laboratory Animal Resources, National Research Council.

This report has been reviewed by the Office of Public Affairs (PA) and is releasable to the National Technical Information Service (NTIS). At NTIS, it will be available to the general public, including foreign nations.

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#### FOREWORD

This report has been prepared by Clement International Corporation, K.S. Crump Division, for the Department of the Air Force, Harry G. Armstrong Aerospace Medical Research Laboratory, Wright Paterson Air Force Base in response to a request to investigate the incorporation of pharmacokinetic modeling into quantitative risk assessment. This report contains the results of this multiyear effort and reflects the changes in direction and priorities as this project has evolved. The Project Director was Dr. Kenny Crump and the Principal Investigator for this project was Mr. Bruce Allen; other investigators who provided technical support and internal peer review were Drs. Crump and Annette Shipp. Mr. Allen was assisted in the pharmacokinetic modeling and analyses primarily by Mr. Christopher Rambin and by Ms. Robinan Gentry. The sensitivity analyses were conducted by Mr. David Farrar, Dr. Crump, Dr. Richard Howe, and Mr. Allen. The software was developed by Ms. Cynthia Van Landingham, Mr. William Fuller, Mr. Eric Brooks, Dr. Howe, and Mr. Allen. The authors wish to acknowledge the support provided by Dr. Jeffery Fisher and Lt. Col. Harvey Clewell, who are at the Harry G. Armstrong Aerospace Medical Research Laboratory, Wright Paterson Air Force Base, and Drs. Melvin Andersen and Michael Gargas, formerly with the Harry G. Armstrong Aerospace Medical Research Laboratory and now with CIIT.

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#### PREFACE

This volume contains a description of the PBPK modeling that has been accomplished for vinyl chloride (VC). This volume consists of two parts.

Part 1 discusses preliminary VC PBPK models that were published in the literature. The models for rats were examined and model results compared to experimental results available in the literature and to closed chamber (gas uptake) studies conducted at Wright-Patterson Air Force Base (WPAFB). Alternative models are presented and also compared to the experimental data.

Part 2 presents some preliminary results documenting progress toward the extension of a rat model to other species, mice and hamsters. Strain-specific results are presented for those species and for rats. That work is based on additional gas uptake studies conducted at WPAFB which included measurement of hepatic glutathione levels in exposed and unexposed animals.

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# VOLUME V

# PART 1 OF 2 PARTS

# INITIAL PBPK MODELING FOR VINYL CHLORIDE

#### A. INTRODUCTION: PREVIOUSLY PROPOSED MODELS

Chee and Blancato (1987) proposed a physiologically based pharmacokinetic (PBPK) model for vinyl chloride (VC) similar to the styrene model of Ramsey and Andersen (1984). The Chen and Blancato model had four compartments, representing the liver, a fat group, and richly- and poorlyperfused tissue groups, with the liver as the sole metabolizing organ. Tissue compartments were connected via arterial and venous blood flows that were modeled by algebraic equations rather than the differential equations that characterized the four compartments. M. tabolism of VC was assumed to become via one saturable pathway. i.e., a pathway following hichaelis-Menten Rimetics.

Figure V-1-1 displays a diagram of a model and its associated system of equations that includes the model proposed by Chen and Blancatc as a special case. It is a generalization of their model only in the sense that it allows for an additional pathway for VC metabolism. The second pathway, a firstorder metabolism pathway, is also located in the liver. This model, which is designated the simple VC model, and the submodel corresponding to the Chen and Blancato model (obtained by setting the first-order metabolism rate constant, ktc, to zero) were examined in light of VC pharmacokinetic data discussed below.

The simple VC model was first examined with respect to its ability to predict gas uptake data obtained in experiments conducted at Wright-Patterson Air Force Base (WPAFB). Those data were used to optimize the metabolic parameters kfc, VMaxc, and km; the latter two parameters define the saturable metabolic pathway. With the value of other model parameters and the starting

 $V \cdot 1 - 1$ 

values and bounds for the metabolic parameters set as in Table V-1-1, the optimized values were estimated to be 2.30, 0.05, 4.32 for vmaxe, km, and kfc, respectively. Although the value for km leached the lower bound on the allowable raige, it was clear that the kfc parameter could not be zero and maintaic predictions close to the gas uptake results. With kfc fixed at zero, vmaxe and km optimized to 4.07 and 0.82, respectively, and all remaining parameters as shown in Table V-1-1, the least squares error term obtained with the optimized values will 36.03. Th contrast, when kfc was allowed to vary (and when it was allowed to be zero, it that value optimized the parameters) the least squares error was 19.30. Visual inspection of the resulting gas optake curves revealed that the fit with the first set of parameter values, 2.30, 0.05, and 4.32 for vmaxe, km, and kfc, respectively, was substantially better than that obtained with the second set of parameters (values of 4.07, C 82, and 0 for vmaxe, km, and kfc, cespectively) (cf. Tables V-1-2 chrough V-1-5).

Another data set was available for evaluating the simple VC model and the submodel of Chen and Blancatc. Gebring et al. (1978) reported the amount of VI metabolized in male sprague-Dawley rats in 6 hours of continuous exposure (Table V-1-6). Also in that table are the model predictions for the amount of VC metabolized, using the two optimized parameter sets discussed in the preceding paragraph. The predictions obtained when kfc was optimized along with vmaxe and km, especially at low to moderate concentrations, were more closely matched to the experimental data than the predictions obtained when kfc was zero. However, the predictions of the simple VC model (with both pathways) deviated from the experimental results presented by Gebring et al. (T-ble V-1- $\ell_T$ . At all doses above 25 ppm, the model predicted a greater

amount of VC metabolism than observed, even accounting for the standard deviation. (The exception was at 511 ppm, but that data point appears to be inconsistent with the observed amount metabolized at 1020 ppm.)

It is possible that the parameters used or estimated in the parameter optimization based on the gas uptake data were in error. Recall that the optimization returned a value for km (0.05) that was equal to the lower bound of the allowable range. Further optimization with different bounds, allowing km to be smaller than 0.05, may have yielded different estimates for all three parameters. Moreover, the strains of rats used in the two experiments were different (Fischer 344 in the gas uptake experiments and Sprague-Dawley in the Gehring et al. study), so the parameters appropriate for the gas uptake experiments may not be appropriate for the metabolism study.

A second possibility is that the metabolism data of Gehring et al. may be in error. That is, the reported amounts of VC metabolized may be inaccurate because of some measurement error or problem with the methods used to administer VC or determine metabolism. This possibility was not directly testable, aside from considering their data in light of the entire ensemble of VC metabolism data and weighing the strengths and weaknesses of each part.

The third possibility is that the model was incorrectly formulated. This possibility is the subject of much of the remainder of this investigation.

#### B. ALTERNATIVE MODELS: VINYL CHLORIDE CONJUGATION WITH GLUTATHIONE

An alternative PBPK model for VC that contained a pathway describing conjugation with glutathione was investigated.

#### 1. Model Definition

Considering that the overestimation of the Gehring et al. metabolism data by the two-pathway model becomes progressively worse as exposure concentration increases, and that a first-order pathway becomes increasingly important as exposure concentrations increase, alternative models focused initially on the first-order pathway. It was suggested (M. Andersen, personal communication) that a second-order metabolic pathway, representing the conjugation of VC with glutathione (GSH), a nonprotein sulfhydryl, may be operative and could replace the first-order pathway in the model.

It is known that GSH can be depleted (Das et al., 1981; Hogberg and Kristoferson, 1977; Sies et al., 1983; Tateishi et al., 1974). The depletion of GSH by reaction with VC and other compounds (see below), may result in the reduction in the amount of VC metabolized. Such a reduction would be consistent with the data presented in Table V-1-6. With the parameters set as estimated on the basis of the gas uptake experiments and a first-order pathway that is not dependent on GSH concentration, the model predicted amounts of VC metabolized that exceeded those observed.

The extent of GSH depletion did not parallel the extent of oxidative, saturable metabolism when the parameters defining the Michaelis-Menten equation were as estimated from gas uptake experiments (Table V-1-6, last column). If it were assumed that products of a saturable pathway were

responsible for the depletion of GSH, then one would expect that two high, near-saturating exposures (exposures that last for the same length of time so that the total amount of epoxide produced is nearly identical) would deplete GSH to almost the same extent (when GSH depletion is measured immediately following the exposures). However, Watanabe et al. (1978) observed extra GSH depletion at dose levels producing equivalent amounts of epoxide (Table V-1-6). Therefore, a model that included direct conjugation of VC with GSH was proposed.

That model, denoted the VC-GSH model, is described in Appendix V-1-A. Note that in addition to the conjugation of VC with glutathione, the (epoxide) metabolite was assumed to react with glutathione as well. The GSH conjugation of the VC epoxide has been proposed in the literature (Plugge and Safe, 1977; Vainio, 1978; Du et al., 1982) and presumably yields the thiodiacetate/ thioglycollate or N-acetyl-S-(2-hydroxyethyl)cysteine that have been observed in the urine of rats exposed to VC.

Another alternative model (the VC-FO model, Appendix V-1-B) was investigated. Since it maintained the saturable and first-order metabolic pathways of the simple VC model, the product of the first-order metabolism was assumed to react with GSH. This would explain the continued GSH depletion at saturating doses. The conflicting evidence presented by the gas uptake and the Gehring et al. data discussed above (and illustrated by the overestimation of amounts metabolized in Table V-1-6) would still have to be resolved if such a model was to be accepted.

#### 2. Parameter Estimation

Several sources were available for the estimation of parameters in the VC-GSH and VC-FO models. Described below are those sources and the manner in which they were used to define the model for rats. Apparently, very little VC experimentation has been conducted in mice, so the model development was limited to rats.

The gas upt>ke data obtained from WPAFB are illustrated in Tables V-1-2 through V-1-5. Those data were collected using Fischer 344 rats of varying weights (cf. Tables V-1-2 through V-1-5). These data were most useful in estimating the physiological parameters (volumes, flow rates) and the physicochemical parameters (partition coefficients and metabolic constants) for the parent, VC, itself. The metabolic constants that were specified to the highest degree (i.e., those to which the closed chamber concentrations were most sensitive and thus those whose values were most constrained by these data) are vmaxc, km, and kgsc or kfc (see Appendices V-1-A and V-1-B for parameter definitions). Initial model runs simulating the closed chamber environment that generated the data allowed refinement of the volume, flow, and partition coefficient estimates and determination of starting values for vmaxc, km, and kgsc or kfc.

Other metabolic parameters described the reactions of the epoxide metabolite, including its conjugation with GSH, and GSH resynthesis. These parameters were estimated from other sources.

Four published reports were the primary source for data to which the model predictions have been compared and parameter estimates adjusted. Watanabe et al. (1978) tested Sprague-Dawley rats weighing between 220 and 250 grams (assumed average, 235g, used in the model runs) exposed to fixed

concentrations ranging from 1.4 to 4600 ppm for 6 hours. As discussed above, total metabolism was reported and the extent of GSH depletion was determined immediately after exposure ceased. (The Watanabe study and all the others discussed here actually determined depletion of nonprotein sulfhydryl groups which, for the purposes of parameter estimation, was assumed to be equivalent to depletion of GSH, the most common nonprotein sulfhydryl.) Insufficient data were presented to allow for estimation of variability among the values reported in the paper.

Tarkowski et al. (1980) conducted similar experiments on male Wistar rats weighing between 180 and 220g (assumed average, 200g). Exposures lasted for 5 hours and ranged from 50 to 20,000 ppm. GSH depletion measurements were obtained between 5 and 6 hours, at 11 hours, and at 24 hours after the start of exposure. The results were reported primarily in the form of graphs so that most depletion estimates were obtained only by approximation from those graphs. However, similar presentation of standard deviations for control and exposed animal GSH concentrations allowed estimation of the variability about each of the point estimates of GSH depletion (which was calculated as the ratio of the GSH concentration in the exposed animals to that in the control

Two other short-exposure experiments were reported by Jedrychowski et al. (1984, 1985). In these experiments, male Wistar rats averaging 250 grams in weight were exposed to concentrations ranging from 5.75 to 21,149 ppm for 4 hours. In addition, pulsatile exposures were also given to animals; the exposure period lasted for 4 hours and the time-weighted average concentrations over that period were the same as the constant exposures, but peak concentrations lasting 15 minutes were alternated with 40-minute periods

of no VC exposure. At this time, these pulsatile data have not been used to estimate or validate the parameter estimates. GSH measurements were obtained 40 hours, 16 hours, and immediately post-exposure. Only in the case of the 21,149 ppm exposure was it necessary to approximate GSH concentrations from graphs. The data reported were sufficient to estimate standard deviations for the depletion estimates. Only the data obtained immediately after exposure were used to adjust parameter values. The data from other times were used to validate the parameter estimates that were considered to be satisfactory on the basis of data obtained immediately after exposure and the data from Tarkowski et al. (1980) and Watanabe et al. (1978).

Three other data sets described effects of VC exposure on GSH depletion and were similarly used as sources of validation. Hefner et al. (1975) tested male Sprague-Dawley rats weighing between 193 and 250 grams. The exposure concentrations varied between 50 and 15,000 ppm and exposures lasted from 1 hour to 5 weeks (7 hours per day, 5 days per week). In similar experiments reported by Watanabe et al. (1976), male Sprague-Dawley rats were exposed for between 1 and 7 hours to VC at levels of exposure ranging from 10 to 2000 ppm. All depletion estimates were approximated from a graphical presentation of the data. Another experiment on male Sprague-Dawley rats was reported by Du et al. (1982). The rats tested in this experiment weighed approximately 400 grams and were exposed for 2, 4, or 6 weeks to 28,000 ppm VC (7 hours per day, 5 days per week). GSH measurements were not obtained until 20 hours after the end of the last exposure, however. All three of these reports provided data sufficient for the calculation of standard deviations for GSH depletion.

Table V-1-7 presents two parameter sets for the VC-GSH model. These sets were obtained using the gas uptake data and data from Watanabe et al.

(1978), Tarkowski et al. (1980), and Jedrychowski et al. (1984, 1985) to adjust the parameter values. A variety of different sets of parameter values were tested against those data. The two that are presented in Table V-1-7 were those that came "closest" to matching the closed chamber VC concentration data, the Watanabe 2t al. metabolism data, and the GSH depletion data when appropriate body weights, exposure concentrations, and exposure times were used in the simulations. The ability of these two parameter sets to predict the data from these experiments is displayed in Figures V-1-2 through V-1-5 and Table V-1-8 for parameter set 1, and in Figures V-1-6 through V-1-9 and Table V-1-9 for parameter set 2. Recall that the data sets shown in those figures and tables were the reference sets used to adjust parameter values. The other data sets that were used only for validation of model predictions are shown in Table V-1-10 along with the predictions of the VC-GSH model with either parameter set 1 or parameter set 2.

Similar computations were done for the VC-FO model. Three sets of parameters (Table V-1-11) were examined with respect to their ability to predict the GSH depletion and metabolism data cited above. Those parameters were obtained by adjusting values to match the gas uptake data as well as the metabolism and GSH depletion data of Watanabe et al. (1978), Jedrychowski et al. (1984, 1985), and Tarkowski et al. (1980). Results are displayed in Tables V-1-12 through V-1-14 and Figures V-1-10 through V-1-21. Predictions of the VC-FO model with the three parameter sets for other exposure scenarios presented in the literature are shown in Table V-1-15.

The VC-GSH model run with the two parameter sets presented in Table V-1-7 had difficulty predicting GSH depletion at high doses (Tables V-1-8 and V-1-9). This was the case even though those high-dose data were used to

adjust the parameters. Note that at 20,000 ppm and higher, the predictions were lower than observed. This was less so with parameter set GSH2, but the differences in the predictions of the two parameter sets were not large. The major difficulty was that the predicted depletions at doses that are .n the range of 4600 to 5800 ppm (and perhaps as low as 1020 ppm) were higher than those observed. Any attempt to reduce depletion (i.e., increase the values displayed in the tables) in the 20,000 ppm range also resulted in reduction of depletion in the range from 4600 to 5800 ppm, further detracting from the match of the predictions to the observations.

Note also that the observed amount of metabolism at 4600 ppm was underpredicted. Again this was less of a problem with the GSH2 parameter set. This fact, in conjunction with the underprediction of GSH depletion at that dose, suggested increasing kgsc or vmaxc. However, while either of these adjustments might have improved the predictions for 4600 ppm, another effect would have been an increase in GSH depletion at higher doses, an effect that would have further reduced the fidelity of the predictions to the observations at those doses. Perhaps increasing kmeec, the parameter that defines the rate at which the epoxide metabolite reacts with everything other than GSH (thus defining the degree to which those pathways compete with the GSH conjugation) would offset the expected increase in GSH depletion that would accompany the suggested change.

It is also worth noting that, on the basis of the predictions of the VC-GSH model, the observed amount metabolized for the 1020 ppm exposure (Watanabe et al., 1978) appears to be more subject to error than that at 511 ppm. In the discussion of the simple VC model (Section A), it was suggested that the 511 ppm result was inconsistent, i.e., that the observed amount metabolized

was too high. In light of the VC-GSH model, it seems that, if there is a question of experimental error, the observed amount metabolized at 1020 ppm may be too low. It should be kept in mind, however, that the data may not be in error. Perhaps there is some physiological or biochemical mechanism not captured by any of the models so far discussed that could result in the apparently inconsistent metabolism results. Since VC is an anaesthetic agent, such a mechanism may be related to ventilation rate changes; if the anaesthetic (i.e., ventilation rate depressant) effect of VC is evident only at inhaled concentrations above 500 ppm, then the amount of VC reaching the liver during a 1020 ppm exposure may indeed be less than the amount of VC reaching the liver during a 511 ppm exposure. If that is the case, then total amounts metabolized might be consistent with the data of Watanabe et al. for those two concentrations (Table V-1-6). However, this explanation may not account for the substantially higher amount metabolized at 4600 ppm (compared to amounts at either 511 or 1020 ppm). Further discussion about discrepancies between the model predictions and the experimentally observed data is presented at the end of this section.

Examining the predictions of the VC-GSH model for the exposule scenarios presented in other published reports (Table V-1-10), the differences discussed above that were attributed to differences in the two parameter sets were corroborated. Parameter set GSH1 predicted slightly more GSH depletion at the end of exposure than did parameter set GSH2. This was due to the smaller values for kmeec and km in set GSH1 compared to set GSH2 which offset the fractionally larger value of kmgsc.

Unfortunately, the data related to GSH concentrations at some time following cessation of VC exposure (Jedrychowski et al., 1984; Tarkowski

et al., 1980; Du et al., 1982) did little to distinguish between the two parameter sets. With the possible exception of the ll-hour Tarkowski et al. data, all the times at which GSH concentrations were measured were so long after exposure that GSH concentrations had returned to near baseline values. An exception, data from Jedrychowski et al. (1984) for the 5796 ppm dose group, was not predicted by either of the parameter sets. The other discrepancy between observed and predicted GSH recovery following VC exposure was at 11 hours (6 hours after the end of exposure) for the 500 ppm exposure (Tarkowski et al., 1980). Tarkowski et al. observed a substantial rebound in GSH concentration at that dose that was not matched by the model predictions.

The model predictions also did not match the elevated GSH concentrations reported by Du et al. (1982). The elevated GSH concentrations observed 20 hours after the end of lengthy VC exposures were higher than the more modest rebounds predicted by either parameter set. Perhaps exposures for such a long period (2 to 6 weeks) effect changes in the GSH resynthesis pathway or trigger some other compensatory mechanisms that are not reflected in the model.

The data of Hefner et al. (1975) have some bearing on this possibility. These data appeared to be inconsistent with the results of other studies; GSH depletion after a 7-hour exposure to 50 ppm was 0.39 (Hefner et al., 1975), but Watanabe et al. (1976) observed GSH depletion of 0.86 after the same exposure. However, long-term exposure scenarios suggested that something else that may not be reflected in the models might have been occurring. The longexposure Hefner et al. data were collected immediately (or shortly) after the end of exposure, so they are not directly comparable to the Du et al. data. However, the general lack of a difference in GSH depletion across exposure concentrations after one week exposure and the suggestion of less depletion

for longer exposures than for shorter exposures (compare the 3 and 5 week results to the 1 week results, especially for the 500 ppm exposure) may be consistent with the possibility of altered GSH resynthesis or the existence of other compensatory mechanisms that become operational following extensive VC exposure. It should be noted, however, that the 1 week exposure to 50 ppm resulted in greater depletion than single exposures to similar concentrations (Watanabe et al., 1976, 1978; Tarkowski et al., 1980). This question of longterm exposure and the possibility of different responses to VC exposure for different exposure periods is an important one for cancer risk assessment because the human exposures of most interest may be chronic ones and also because the carcinogenicity bioassays from which risk estimates are frequently derived involve long-term exposure.

Parameter set GSH1 yielded slightly better matches to the gas uptake data than did set GSH2 (compare Figures V-1-2 through V-1-5 with Figures V-1-6 through V-1-9). This was especially true at low concentrations. Both parameter sets predicted well the pattern of chamber concentrations starting at 3150 ppm and 565 ppm. While both tended to underpredict concentration after about 2 hours for the exposure starting at 1230 ppm, the discrepancy appeared to be slightly worse for the GSH2 set. The same could be said about the low concentration exposure (starting at 220 ppm), although in this case both sets of predictions were high after 1 to 2 hours.

The VC-FO model predictions were similar to those of the VC-GSH model. The main difficulty was at very high doses: the amount of GSH depletion predicted exceeded that observed, but decreasing vmaxc, kfc, or kmgsc led to increased discrepancies for doses on the order of 4600 to 5800 ppm. Note that for parameter set FO1, GSH depletion associated with VC exposures at or above

20,000 ppm was as low as had been achieved and might be considered acceptable (Table V-'-12). When vmaxc and/or kfc were increased so as to bring into line the predictions associated with the 4600 to 5800 ppm exposures, the ability of the model to predict high-dose metabolism was adversely affected (compare Tables V-1-13 and V-1-14 to Table V-1-12). Moreover, the degree to which the 4600 to 5800 ppm predictions were improved was minimal. Predictions for low doses continued to be fairly close, in general, to the observations obtained in the experiments.

The information presented in Table V-1-15 again confirms the less than optimal timing of the GSH recovery data that have been reported. The predictions of the three parameter sets were very similar, to an extent beyond the resolving power of the observed data. None of the predictions obtained by the VC-FO model and the parameter sets investigated was vastly different from those obtained from the VC-GSH model and its two parameter sets.

The gas uptake data (Figures V-1-10 through V-1-21) show that the parameter sets FO2 and FO3 were less acceptable. The discrepancies arose primarily for the chamber concentrations starting at 3150 ppm. In these cases, FO2 and FO3 predicted chamber concentrations less than those observed over the entire course of exposure. These were substantially poorer fits than obtained with set FO1 or with the VC-GSH model and either of its parameter sets. These poorer predictions were also reflected in the overprediction of amounts metabolized when compared to the Watanabe et al. (1978) results. Both were probably due to values for vmaxc or kfc that were too large. Recall, however, that reducing either of these values resulted in reductions in predicted GSH depletion. The predictions for these two parameter sets with respect to GSH depletion were already too low; any changes to vmaxc or kfc

that reduce metabolism enhanced the discrepancy between observed and predicted GSH depletion.

#### U. DISCUSSION

One difficulty, alluded to above, was that the predictions of the two models, even with the total of five different parameter sets, were not sufficiently different for the data that were cited above to differentiate between them — Other data are required for refining the parameter estimation. The additional gas uptake studies conducted at WPAFB are discussed in Part 2 of this volume. It may also be possible that carcinogenility bioassay data, in the form of response rates and their differences at different doses or for different routes of exposure, can help differentiate between the models. This is an issue that should be investigated more thoroughly if this work is to progress beyond formulation of a VC PEPK model and to the uses of the model in the conduct of a risk appearent. The comments in Section B about long-term exposure and possible compensatory GSH resy: hesis are also pertinent to the proposed use of the bioassay data for model validation as well as for risk assessment.

It is important to note assumptions implicit in the VC-G5H and VC-FO models that are novel or that are not supported by the literature on VC metabolism. These assumptions should be more fully investigated.

For the VC-GSH model, as mentioned above, the conjugation of VC with GSH is a reaction that has not been postulated previously. However, two structurally similar volatile compounds, trichloroethylene and tetrachloroethylene, are thought to conjugate with GSH (Dekant et al., 1910).

Neal (1980) described three common features of compounds that are substrates for glutathione-S-tranferases: they must be hydrophobic to some degree, they must contain an electrophilic carbon atom, and they must react nonenzymatically with GSH at a measurable rate. VC is hydrophobic; however, its carbon atoms are not expected to be overly electrophilic. The nonenzymatic reaction of VC with GSH has not been reported, to our knowledge.

A number of assumptions underlie the VC-FO model. The first is that the metabolite produced by the first-order pathway is the same epoxide as produced by the saturable pathway. Actually, it seems that this assumption can be modified somewhat If the product of the first-order pathway is different, then the assumption that must be made for the VC-FO model to be valid is that the rate of reaction of that product with GSH is the same as the rate for the reaction between GSH and the epoxide. Further weakening of that assumption would lead to the introduction of more parameters in the model. While this would allow more flexibility in the fitting of the model to the GSH depletion data, it had already become apparent that the existing data may not be sufficient to characterize all of the parameters that are currently in the model. Finally, note that it was assumed that the product of the first-order pathway reacts with GSH. But, because the experiments measured total nonprotein sulfhydryl depletion, not GSH depletion per se, it was possible that some other sulfhydryl was involved with the metabolite of the first-order pathway.

The results related to the ability of the models to predict the observed data suggested another formulation of the PBPK model. That formulation would substitute a second saturable pathway for the first-order pathway that was in the VC-FO model. One effect that may be obtained from this substitution is a

reduction in the estimates of amount metabolized at high doses, yielding closer correspondence between the model predictions and the results of the gas uptake and Watanabe et al. (1978) experiments. It may also be possible to increase the extent of GSH depletion at exposures around 4600 to 5800 ppm but to maintain or even decrease the depletion predicted for the exposures to 20,000 ppm or more. A low-affinity (high km) pathway should be examined first so as not to change the predictions for low doses too much.

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Fixed Parameters	Value	
	Value	
qpc	20.0	
qcc	15.0	
qrc	0.417	
qsc	0.14	
qfc	0.07	
qlc	0.374	
NEC	0.05	
VIC	0.05	
vsc	0.73	
	0.07	
Vic	0.04	
pb	1.65	
pr	0.95	
ps	1.25	
pf	6.67	
pl	0.95	
Optimized Parameters	Initial Value	Allowable Range
kfc	3,94	(0.0. 10.0)
Vmaxc	2 46	(0,0,6,0)
km	0.095	(0,05,1,0)
	0.075	(0.00, 1.0)

# Parameter Specification for Vinyl Chloride Model Optimization<sup>®</sup>

\*The model and its parameters are defined in Figure V-1-1.

	Observed Cas	PBPK Models Predictions		
Time	Untake Data	1 Pathwav <sup>b</sup> 2 Pathwave <sup>c</sup>		
111111111111111111111111111111111111111	Uptake Data	1 Tatilway	_ 2 ratilways	
5	2968	3054	3053	
10	2888	2954	2953	
20	2800	2810	2808	
30	2731	2715	2711	
40	2676	2648	2643	
50	2624	2598	2592	
60	2593	2559	2551	
70	2546	2526	2516	
80	2514	2496	2486	
90	2493	2468	2457	
100	2457	2442	2430	
110	2427	2417	2404	
120	2394	2392	2378	
130	2391	2367	2352	
140	2339	2343	2327	
150	2315	2318	2302	
160	2293	2294	2278	
170	2263	2270	2253	
180	2228	2245	2229	
190	2208	2221	2204	
200	2195	2197	2180	
210	2171	2173	2156	
220	2147	2149	2132	
230	2130	2125	2108	
240	2116	2100	2084	
250	2089	207 <b>6</b>	2060	
260	2070	2052	2037	
270	1960	2028	2013	
280	1937	2004	1990	
290	1960	1980	1967	
300	1890	1956	1943	
310	1870	1932	1920	
320	1894	1908	1897	
330		1884	1874	
340	1840	1861	1852	
350	1787	1837	1829	
360	1788	1813	1806	

#### Observed and Predicted Chamber Concentrations Initial Concentration: 3200 ppm (Average Weight of 3 Rats is 222g)

<sup>a</sup>Data from WPAFB. <sup>b</sup>Predicted from PBPK model with kfc - 0, vmaxc - 4.07, km - 0.82. <sup>c</sup>Predicted from PBPK model with kfc - 4.32, vmax - 2.30, km - 0.05.

	Observed Gas	PBPK Models Predictions			
Time	Uptake Data <sup>*</sup>	1 Pathway <sup>b</sup>	2 Pathways <sup>c</sup>		
5		1133	1134		
10	1086	1084	1087		
15	1052	1044	1048		
25	995	982	989		
35	952	936	945		
45	916	899	911		
55	894	868	883		
65	860	840	858		
75	841	814	835		
85	820	790	814		
95	797	767	793		
105	782	744	773		
115	763	722	753		
125	743	700	734		
135	719	679	714		
145	697	657	695		
155	677	636	676		
165	662	616	657		
175	643	595	638		
185	627	575	619		
195	610	555	601		
205	594	536	582		
215	578	516	564		
225	556	497	546		
235	537	479	527		
245	519	460	509		
255	500	442	491		
265	481	425	473		
275	461	407	456		
285	443	390	438		
295	426	374	420		

# Observed and Predicted Chamber Concentrations Initial Concentration: 1200 ppm (Average Weight of 3 Rats is 255g)

<sup>a</sup>Data from WPAFB. <sup>b</sup>Predicted from PBPK model with kfc = 0, vmaxc = 4.07, km = 0.82. <sup>c</sup>Predicted from PBPK model with kfc = 4.32, vmax = 2.30, km = 0.05.

	Observed Gas	PEPK Models Predictions		
Time	Uptake Data <sup>a</sup>	l Pathway <sup>b</sup>	2 Pathways <sup>c</sup>	
5	540	516	516	
10	509	491	490	
20	468	450	450	
30	437	420	419	
40	411	394	393	
50	386	373	371	
60	362	354	351	
70	342	337	332	
80	321	320	315	
90	299	305	297	
100	282	290	281	
110	267	277	264	
120	248	263	248	
130	232	250	232	
140	214	238	217	
150	198	226	202	
160	181	214	187	
170	165	203	172	
180	149	192	158	
190	136	182	145	
200	122	172	132	
210	111	163	120	
220	97.4	154	109	
230	90.7	145	99	
240	80.9	137	89	
250	73.3	129	81	
260	64.2	122	73	
270	58.5	115	65	
280	53.7	108	59	
290	48.8	102	53	
300	45.0	96	47	

# Observed and Predicted Chamber Concentrations Initial Concentration: 550 ppm (Average Weight of 3 Rats is 256g)

Table V-1-4

<sup>a</sup>Data from WPAFB. <sup>b</sup>Predicted from PBPK model with kfc = 0, vmaxc = 4.07, km = 0.82. <sup>c</sup>Predicted from PBPK model with kfc = 4.32, vmax = 2.30, km = 0.05.

	Observed Gas	PBPK Models Predictions		
Time	Uptake Data <sup>a</sup>	l Pathway <sup>b</sup>	2 Pathways <sup>c</sup>	
5	216	206	204	
10	205	194	190	
20	181	176	168	
30	160	162	149	
40	142	150	134	
50	126	140	120	
60	112	131	108	
70	99.2	123	98	
80	87.2	116	88	
90	78.7	109	79	
100	70.4	102	71	
110	61.8	96	64	
120	55.9	91	58	
130	49.7	85	52	
140	44.4	80	46	
150	40.5	75	42	
160	36.4	71	37	
170	32.7	66	34	
180	29.4	62	30	
190	26.7	59	27	
200	23.9	55	24	
210	21.4	52	22	
220	19.0	48	19	
230	17.4	45	17	
240	15.5	43	16	
250	13.9	40	14	
260	12.9	37	12	
270	11.2	35	11	
280	10.3	33	10	

# Observed and Predicted Chamber Concentrations Initial Concentration: 220 ppm (Average Weight of 3 Rats is 253g)

<sup>a</sup>Data from WPAFB. <sup>b</sup>Predicted from PBPK model with kfc = 0, vmaxc = 4.07, km = 0.82. <sup>c</sup>Predicted from PBPK model with kfc = 4.32, vmax = 2.30, km = 0.05.

			<u>Predicted µg VC Metabolize</u>		
Exposure	Obser	ved <sup>b</sup>	<u>l Pathway</u> c	<u> </u>	athways <sup>d</sup>
Concentrationª	µg VC	GSH		Via S	aturable
(ppm)	Metabolized	<pre>§ of Control</pre>	Total	<u>Total</u>	<u>Pathway</u>
1.4	30 ± 3	104	24.3	37.6	37.5
9.0	$242 \pm 26$	89	155.3	241.3	240.4
25	557 <u>+</u> 42	93	425.7	663.9	661.0
51	1181 <u>+</u> 93	94	848.1	1337	1329
109	2406 <u>+</u> 173	81	1722	2715	2693
250	3826 <u>+</u> 345	70	3435	4442	4296
511	$6263 \pm 355$	60	5361	5215	4714
1020	$4257 \pm 765$	51	6883	6017	4789
4600	9255 <u>+</u> 1467	39	8373	11221	4844

#### VC Metabolism and GSH Depletion: Comparison of Observed and Predicted Results

\*Exposure lasting for 6 hours to the concentrations indicated.

<sup>b</sup>Results from Gehring et al. (1978) and Watanabe et al. (1978), at the end of exposure.

<sup>c</sup>Predicted from PBPK model with kfc = 0, vmaxc = 4.07, km = 0.82; at the end of exposure.

<sup>d</sup>Predicted from PBPK model with kfc = 4.32, vmaxc = 2.30, km = 0.05; at the end of exposure.

T	at	1	e	٧-	1	-7

	Parameter	Value in Set	
Parameter	GSH1	GSH2	
vmaxc*	2.9	3.1	
km <sup>*</sup>	0.05	0.1	
kgsc	0.0002	0.0002	
kmgsc*	0.20	0.19	
k-eec*	1400	1730	
kpc	0.09	0.09	
ks	30,000	30,000	
klpc	0.11	0.11	
kloc	3.04	3.04	
kls	3,000	3,000	
qpc	20.0	20.0	
qcc	15.0	15.0	
qrc	0.49	0.49	
qsc	0.15	0.15	
qfc	0.09	0.09	
qlc	0.27	0.27	
vrc	0.042	0.042	
vsc	0.760	0.760	
víc	0.060	0.060	
vlc	0.048	0.048	
,	1 ( )	1 (0	
рb	1.68	1.68	
pr	0.95	0.95	
ps	1.25	1.25	
pt	6.67	6.67	
pl	0.95	0.95	

# Parameter Sets Used with VC-GSH Model\*

<sup>a</sup>The model and its parameters are defined in Appendix V-1-A. <sup>\*</sup>An asterisk identifies parameters taking different values in the two sets.
Exposu	ure Scenario	Observed∎	Predicted <sup>b</sup>
4-hour expo	osures <sup>c</sup> :	<u>CSH</u>	Depletion <sup>d</sup> :
19.4	ppm	0.92	0.97
58.3	ppm	(0.86, 0.97) 0.79 (0.72, 0.86)	0.92
219	ppm	(0.72, 0.88) 0.72 (0.61, 0.82)	0.74
5796	ppm	(0.81, 0.82) 0.42 (0.38, 0.45)	0.50
21149	ppm	(0.38, 0.43) 0.38 (0.28, 0.47)	0.27
5-hour expo	osures <sup>e</sup> :	<u>GSH</u>	Depletion <sup>d</sup> :
50	ppm	0.80; 0.94	0.92
200	ppm	0.74	0.74
500	ppm	0.85	0.65
1000	ppm	0.63	0.62
2000	ррт	0.43; 0.47; 0.55 (0.37, 0.49); (0.30, 0.64) (0.49, 0.61)	0.32);
<u>6-hour exp</u>	osures <sup>f</sup> :	<u>CSH</u>	Depletion <sup>d</sup> :
25 51 109 250 511 1020 4600	ppm ppm ppm ppm ppm ppm	0.93 0.94 0.81 0.70 0.60 0.51 0.39	0.95 0.91 0.81 0.67 0.62 0.59 0.47

Observed and Predicted GSH Depletion and VC Metabolism: VC-GSH Model with Parameter Set GSH1

### Table V-1-8 (continued)

Exposu	are Scenario	Observed <sup>*</sup>	Predicted <sup>b</sup>
6-hour expo	sures <sup>f</sup> :	VC_N	fetabolized <sup>a</sup>
1.4	ppm	30	35
9.3	ppm	(27, 33) 242	233
25	ррт	(210, 200) 557 (515, 500)	625
51	ppm	(1088, 1274)	1267
109	ppm	(1088, 1274) 2406 (2233, 2579)	2651
250	ppm	3826	5182
511	рра	6263 (5908 6618)	6130
1020	ррш	4257 (3/92 5022)	6424
4600	ppm	9255 (7788, 10722)	7301

Observed and Predicted GSH Depletion and VC Metabolism: VC-GSH Model with Parameter Set GSH1

<sup>a</sup>Observed depletion of GSH or amount of VC metabolized as of the end of exposure. In parentheses are the values approximately one standard deviation away from the mean, when these have been estimable from reported data. In some cases, a study has reported more than one mean value for a given exposure scenario; all reported means and associated standard deviations are presented.

<sup>b</sup>Predicted, as of the end of exposure, by the VC-GSH model.

<sup>c</sup>Data from Jedrychowski et al. (1984, 1985).

<sup>d</sup>GSH depletion is presented in terms of the ratio of the GSH concentration in exposed animal livers to that in control animal livers.

\*Data from Tarkowski et al. (1980). Predicted values correspond to GSH depletion one-half hour after end of exposure because of ambiguity as to exact time of GSH measurement in this study.

<sup>f</sup>Data from Watanabe et al. (1978).

 $^{8}\text{VC}$  metabolism is presented in terms of  $\mu g$  of VC metabolized by the end of exposure.

Exposi	ure Scenario	Observed <sup>a</sup>	Predicted <sup>b</sup>
4-hour expo	osures <sup>c</sup> :	<u>GSH</u>	Depletion <sup>d</sup>
19.4	ppm	0.92	0.98
58.3	ppm	0.79	0.93
219	ppm	0.72	0.79
5796	ppm	0.42	0.52
21149	ррш	(0.38, 0.43) 0.38 (0.28, 0.47)	0.29
5-hour expo	osures <sup>•</sup> :	<u>GSH 1</u>	<u>Depletion</u> <sup>d</sup>
50	ppm	0.80; 0.94	0.93
200	Еbш	(0.07, 0.94), (0.90, 0.96 0.74 (0.70, 0.79)	0.79
500	ррш	0.85	0.70
1000	ppm	0.63	0.66
2000	ppm	(0.37, 0.43; 0.47; 0.55 (0.37, 0.49); (0.30, 0.64) (0.49, 0.61)	0.38
6-hour expo	osures <sup>f</sup> :	<u>CSH I</u>	Depletion <sup>d</sup>
25 51 109 250 511 1020 4600	ррт ррт ррт ррт ррт	0.93 0.94 0.81 0.70 0.60 0.51 0.39	0.96 0.92 0.85 0.72 0.66 0.63 0.51

Observed and Predicted GSH Depletion and VC Metabolism: VC-GSH Model with Parameter Set GSH2

#### Table V-1-9 (continued)

Exposure Cuenario	Observed <sup>a</sup>	Predicted <sup>b</sup>
<u>6-hour experies</u> t:	<u>VC N</u>	<u>letabolized<sup>8</sup></u>
1.4 ; m	30	33
9.3 ppc	(27, 33) 242	224
25 ppm	(216, 265) 557	599
51 pom	(515, 599) 1181	1210
109 ppm	(1088, 1274) 2406	2508
250 <b>Da</b>	(22 <b>33</b> , 2579) 3826	4927
511 r m	(3481, 4171)	6284
этт ррш	(5908, 6618)	0204
1020 ppm	4257 (3492, 5022)	6755
4 <b>6</b> 00 pp <b>m</b>	9255 (7788, 10722)	7745

<sup>4</sup>Observed depletion of GSH or amount of VC metabolized as of the end of exposure. In parentheses are the values approximately one standard deviation away from the mean, when these have been estimable from reported data. In some cases, a study has reported more than one mean value for a given exposure scenario; all reported means and associated standard deviations are presented.

<sup>b</sup>Predicted, as of the end of exposure, by the VC-GSH model.

'Data from Jedrychowski et al. (1984, 1985).

<sup>d</sup>GSH depletion is presented in terms of the ratio of the GSH concentration in exposed animal livers to that in control animal livers.

\*Data from Tarkowski et al. (1980). Predicted values correspond to GSH depletion one-half hour after end of exposure because of ambiguity as to exact time of GSH measurement in this study.

<sup>1</sup>Data from Watanabe et al. (1978).

<sup>8</sup>VC metabolism is presented in terms of  $\mu g$  of VC metabolized by the end of exposure.

				Predi	lcted <sup>c</sup>
Data	Set		Observed <sup>b</sup>	GSH1	GSH2
Jedrycho	wski et	al. (1984) - 4	hr exposures		<u> </u>
					• • •
19.4	4 ppm	@20 hrs <sup>a</sup>	1.02(0.94, 1.10)	1.00	1.00
5.0	2	(44 hrs	1.02(0.97, 1.07)	1.00	1.00
58.	s ppm	Q20 hrs	0.94 (0.84, 1.04)	1.00	1.00
210		(244 nrs	1 00 (1 00 1 10)	1.00	1.00
219	ЪЪш	$a_{20}$ mis	1.09 (1.00, 1.10)	1.01	1.01
5796	<b>D</b> D <b>m</b>	620 brs	0.94 (0.00, 1.01) 0.86 (0.78 0.94)	1.00	1.00
5750	քեա	044 hrs	1.02 (0.89, 1.14)	1.00	1.00
		<b>C</b> • • • • • • •		2100	1.00
<u>Tarkowsk</u>	<u>i et al</u>	<u>. (1980) - 5 hr</u>	<u>exposures</u>		
50	លា	@11 hrs	1.06 (0.91, 1.22)	0.98	0.98
50	PP-	@24 hrs	$0.94 \ (0.82, 1.06)$	1.00	1.00
500	maa	@11 hrs	1.15(1.04, 1.27)	0.92	0.93
200	rr	@24 hrs	1.04 (0.91, 1.17)	1.01	1.01
20000	ррш	@11 hrs	0.86 (0.75, 0.97)	0.89	0.90
	••	@24 hrs	1.21 (1.05, 1.37)	1.04	1.04
<u>Watanabe</u>	et al.	(1976)•			
10	ррш	@ 7 hrs	1.15 (1.09, 1.21)	0.98	0.98
50	ррш	@7 hrs	0.86 (0.45, 1.27)	0.90	0.92
150	ppm	@ 2 hrs	0.82 (0.72, 0.91)	0.88	0.91
	••	@ 4 hrs	0.74 (0.69, 0.80)	0.81	0.85
		@7hrs	0.74 (0.67, 0.81)	0.75	0.80
250	ррш	@ 1 hr	0.96 (0.78, 1.14)	0.91	0.93
	••	@ 3 hrs	0.89 (0.76, 1.01)	0.78	0.82
		@ 5 hrs	0.66 (0.52, 0.80)	0.70	0.75
		@ 7 hrs	0.63 (0.52, 0.75)	0.66	0.72
1000	ppm	@ 1 hr	0.87 (0.67, 1.05)	0.88	0.89
		@ 2 hrs	0.78 (0.71, 0.86)	0.78	0.80
		@ 3 hrs	0.71 (0.59, 0.82)	0.71	0.74
		@ 4 hrs	0.69 (0.53, 0.84)	0.66	0.69
		@ 5 hrs	0.60 (0.49, 0.72)	0.62	0.66
		@ 6 hrs	0.58 (0.45, 0.70)	0.60	0.64
		@ 7 hrs	0.62 (0.54, 0.69)	0.59	0.62
2000	ppm	@ 2 hrs	0.67 (0.56, 0.79)	0.76	0.78
		@ 4 hrs	0.53 (0.44, 0.62)	0.62	0.65
		@ 7 hrs	0.39 (0.25, 0.52)	0.55	0.58

# Observed and Predicted GSH Depletion; VC-GSH Model with Parameters Sets GSH1 and GSH2\*

#### Table V-1-10 (continued)

Data Set		Obs	served <sup>t</sup>	)	<u>Predi</u> GSH1	<u>cted</u> <sup>c</sup> GSH2
<u>Hefner et al. (19</u>	7 <u>5)</u> •					
50 ppm	@ 1 hr @ 7 hrs	0.98 0.39	(0.87, (0.21,	1.10) 0.56)	0.98 0.90	0.98 0.92
<u>Hefner et al (19</u>	7 <b>5) - 5</b> days/wk	7 hrs	/day_e>	(posure		
<u>l wk exposur</u>	e; @ 101.5 hrs					
50 ppm 500 ppm 5000 ppm 15000 ppm 3 wk exposur	e: @ 437.5 hrs	0.79 0.60 0.72 0.69	(0.62, (0.44, (0.65, (0.47,	0.95) 0.77) 0.78) 0.91)	0.91 0.63 0.48 0.32	0.93 0.67 0.50 0.33
500 ppm 5000 ppm	<u>, , , , , , , , , , , , , , , , , , , </u>	0.74 0.57	(0.60, (0.52,	0.88) 0.62)	0.63 0.48	0.67 0.50
<u>5 wk exposur</u>	<u>e: @ 773.5 hrs</u>					
500 ррт 5000 ррт		0.91 0.84	(0.76, (0.73,	1.05) 0.96)	0.63 0.48	0.67 0.50
<u>Du et al. (1982)</u>	- 5 days/wk, 7 ł	nrs/day	exposi	ire to 2800	0 ppm	
2 wk exposur 4 wk esposur 6 wk exposur	e; @ 291 hrs e; @ 627 hrs e; @ 963 hrs	1.21 1.44 1.65	(1.15, (1.08, (1.26,	1.26) 1.80) 2.04)	1.06 1.06 1.06	1.06 1.06 1.06

### Observed and Predicted GSH Depletion; VC-GSH Model with Parameters Sets GSH1 and GSH2\*

<sup>a</sup>GSH depletion is expressed as the ratio of the GSH concentration in the livers of exposed animals to that in control animals.

<sup>b</sup>Mean value and, in parentheses, estimated values one standard deviation away from the mean.

<sup>c</sup>Predicted by the model with the indicated parameter set. Study-specific body weights were used.

<sup>d</sup>The times are measured from the beginning of exposure.

\*In this study, exposure times varied. The times given are the times at which exposure ceased.

Table V-	1	-11
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	Pa	rameter Value	in Set	
Parameter	FOl	FO2	FO3	
vmaxc*	2.9	29	3 1	
km*	0.05	0.05	0 1	
kfc*	1.2	2.4	2.4	
kmgsc*	0.20	0.19	0.19	
kmeec*	1400	1730	1730	
kpc	0.09	0.09	0.09	
ks	30,000	30,000	30,000	
klpc	0.11	0.11	0.11	
kloc	3.04	3.04	3.04	
kls	3,000	3,000	3,000	
qpc	20.0	20.0	20.0	
qcc	15.0	15.0	15.0	
qrc	0.49	0.49	0.49	
qsc	0.15	0.15	0.15	
qfc	0.09	0.09	0.09	
qlc	0.27	0.27	0.27	
vrc	0.042	0.042	0.042	
VSC	0.760	0.760	0.760	
vfc	0.060	0.060	0.060	
vlc	0.048	0.048	0.048	
pb	1.68	1.68	1.68	
pr	0.95	0.95	0.95	
ps	1.25	1.25	1.25	
pf	6.67	6.67	6.67	
pl	0.95	0.95	0.95	

Parameter Sets Used with VC-FO Model\*

"The model and its parameters are defined in Appendix V-1-B.

'An asterisk marks the parameters with different values across the three sets.

Exposu	nre Scenario	Observed <sup>*</sup>	Predicted <sup>b</sup>
<u>4-hour expo</u>	osures <sup>c</sup> :	<u>GSH</u>	<u>Depletion</u> <sup>d</sup>
19.4	ppm	0.92	0.97
58.3	ppm	(0.86, 0.97) 0.79	0.92
219	ppm	(0.72, 0.86) 0.72	0.74
5796	ppm	(0.61, 0.82) 0.42	0.56
21149	ррш	(0.38, 0.45) 0.38 (0.28, 0.47)	0.35
5-hour expo	osures•:	GSH	Depletion <sup>d</sup>
50	ppm	0.80; 0.94	0.92
200	ppm	(0.87, 0.94); (0.90, 0.98 0.74 (0.70, 0.79)	0.74
500	mqq	0.85	0.66
1000	ррт	0.63	0.63
2000	ррш	0.43; 0.47; 0.55 (0.37, 0.49); (0.30, 0.64) (0.49, 0.61)	0.36
<u>6-hour exp</u>	osures <sup>f</sup> :	<u>GSH</u>	Depletion <sup>d</sup>
25 51 109 250 511 1020 4600	ррт ррт ррт ррт ррт	0.93 0.94 0.81 0.70 0.60 0.51 0.39	0.95 0.91 0.81 0.67 0.62 0.60 0.52

Observed and Predicted GSH Depletion and VC Metabolism: VC-FO Model witl. Parameter Set FC1

#### Table V-1-12 (continued)

Exposure Scenario	Observed <sup>*</sup>	Predicted <sup>b</sup>
<u>6-hour exposures</u> <sup>f</sup> :	<u>vc</u> M	letabolized <sup>s</sup>
1.4 ppm	30	35
9.3 ppm	(27, 33) 242	233
25 ppm	(216, 268) 557 (515, 500)	625
51 ppm	(515, 599) 1181	1268
109 ppm	(1088, 1274) 2406 (0022, 0570)	2651
250 ppm	(2233, 2579) 3826	5190
511 ppm	(3481, 41/1) 6263	6198
1020 ррш	(5908, 6618) 4257	6324
4600 ppm	(3492, 5022) 9255 (7788, 10722)	8563

### Observed and Predicted GSH Depletion and VC Metabolism: VC-FO Model with Parameter Set FOl

<sup>a</sup>Observed depletion of GSH or amount of VC metabolized as of the end of exposure. In parentheses are the values approximately one standard deviation away from the mean, when these have been estimable from reported data. In some cases, a study has reported more than one mean value for a given exposure scenario; all reported means and associated standard deviations are presented.

<sup>b</sup>Predicted, as of the end of exposure, by the VC-FO model.

<sup>c</sup>Data from Jedrychowski et al. (1984, 1985).

<sup>d</sup>GSH depletion is presented in terms of the ratio of the GSH

concentration in exposed animal livers to that in control animal livers. <sup>e</sup>Data from Tarkowski et al. (1980). Predicted values correspond to GSH depletion one-half hour after end of exposure because of ambiguity as to exact time of GSH measurement in this study.

<sup>f</sup>Data from Watanabe et al. (1978).

 $^{8}\text{VC}$  metabolism is presented in terms of  $\mu g$  of VC metabolized by the end of exposure.

Exposu	re Scenario	Observed®	Predicted <sup>b</sup>
<u>4-hour expo</u>	osures <sup>c</sup> :	<u>GSH Dep</u>	letion <sup>d</sup>
19.4	ppm	0.92	0.98
58.3	ppm	(0.86, 0.97) 0.79	0.93
219	ppm	(0.72, 0.86) 0.72	0.78
5796	ppm	(0.61, 0.82) 0.42	0.54
21149	ppm	(0.38, 0.45) 0.38	0.27
		(0.28, 0.47)	
5-hour expo	osures <sup>•</sup> :	<u>GSH</u> Dep	letion <sup>d</sup>
50	ppm	0.80; 0.94	0.93
200	ppm	0.74	0.78
500	ppm	0.85	0.70
1000	ppm	0.63	0.67
2000	ppm	(0.57, 0.69) 0.43; 0.47; 0.55	0.32
		(0.37, 0.49); (0.30, 0.64); (0.49, 0.61)	
<u>6-hour_exp</u>	osures <sup>f</sup> :	<u>GSH Dep</u>	letion <sup>d</sup>
25	ppm	0.93	0.96
51	ppm	0.94	0.92
109	ppm	0.81	0.84
250	ppm	0.70	U./I
110	ррш	0.60	U.08 0.64
4600	ррш РРш	0.39	0.04 5.52
	••		

# Observed and Predicted GSH Depletion and VC Metabolism: VG-FO Model with Parameter Set FO2

### Table V-1-13 (continued)

Exposure	e Scenario	Observed <sup>®</sup>	Predicted <sup>b</sup>
6-hour expos	ures <sup>f</sup> :	<u>VC_</u> 1	fetabolized <sup>8</sup>
1.4 p	pm	30	35
-	-	(27, 33)	
9.3 p	pm	242	233
		(216, 268)	
25 p	рш	557	625
		(515, 599)	
51 p	pm	1181	1268
	-	(1088, 1274)	
109 p	pm	2406	2652
	-	(2233, 2579)	
250 p	pm	3826	5206
•	•	(3481, 4171)	
511 p	pm	6263	6334
-	•	(5908, 6618)	
1020 p	pm	4257	7015
•	•	(3492, 5022)	
4600 p	DW	9255	10743
· · · · r	•	(7788, 10722)	
1020 p 4600 p	Ъш Ъш	4257 (3492, 5022) 9255 (7788, 10722)	7015 1074 <b>3</b>

#### Observed and Predicted GSH Depletion and VC Metabolism: VC-FO Model with Parameter Set FO2

\*Observed depletion of GSH or amount of VC metabolized as of the end of exposure. In parentheses are the values approximately one standard deviation away from the mean, when these have been estimable from reported data. In some cases, a study has reported more than one mean value for a given exposure scenario; all reported means and associated standard deviations are presented.

- <sup>b</sup>Predicted, as of the end of exposure, by the VC-FO model.
- <sup>c</sup>Data from Jedrychowski et al. (1984, 1985).

<sup>d</sup>GSH depletion is presented in terms of the ratio of the GSH concentration in exposed animal livers to that in control animal livers.

•Data from Tarkowski et al. (1980). Predicted values correspond to GSH depletion one-half hour after end of exposure because of ambiguity as to exact time of GSH measurement in this study.

<sup>f</sup>Data from Watanabe et al. (1978).

 ${}^{\tt g}{\sf VC}$  metabolism is presented in terms of  $\mu g$  of VC metabolized by the end of exposure.

Exposu	ure Scenario	Observed <sup>*</sup>	Predicted <sup>b</sup>			
<u>4-hour_exposures</u> <sup>c</sup> :		<u>GSH Depletion<sup>d</sup></u>				
19.4	ppm	0.92	0.98			
58.3	ppm	(0.86, 0.97) 0.79	0.93			
219	ррт	(0.72, 0.86) 0.72	0.79			
5796	ppm	(0.61, 0.82) 0.42	0.53			
21149	bbm	(0.38, 0.45) 0.38 (0.28, 0.47)	0.27			
<u>5-hour exposures</u> <sup>e</sup> :		<u>GSH D</u>	<u>epletion</u> <sup>d</sup>			
50	bbw	0.80; 0.94	0.93			
200	ppm	(0.67, 0.94); (0.90, 0.98) 0.74 (0.70, 0.70)	0.79			
500	ррт	0.85	0.70			
1000	ppm	0.63	0.66			
2000	ррт	(0.37, 0.89) 0.43; 0.47; 0.55 (0.37, 0.49); (0.30, 0.64); (0.49, 0.61)	0.32			
<u>6-hour exposures</u> <sup>f</sup> :		<u>GSH_Depletion</u> <sup>d</sup>				
25 51 109 250 511 1020 4600	ррт ррт ррт ррт	0.93 0.94 0.81 0.70 0.60 0.51 0.39	0.96 0.92 0.85 0.73 0.66 0.63 0.50			

Observed and Predicted GSH Depletion and VC Metabolism: VC-FO Model with Parameter Set FO3

#### Table V-1-14 (continued)

Exposure Scenario	Observed <sup>®</sup>	Predicted <sup>b</sup>		
<u>6-hour exposures</u> <sup>f</sup> :	<u>vc</u> M	letabolized <sup>8</sup>		
1.4 ppm	30	33		
9.3 ppm	(27, 33) 242	224		
25 ppm	(216, 268) 557	599		
51 ppm	(515, 599) 1181	1210		
109 ppm	(1088, 1274) 2406	2510		
250 ppm	(2233, 2579) 3826	4593		
511 ppm	(3481, 4171) 6263	6475		
1020 ppm	(5908, 6618) 4257	7326		
4600 ppm	(3492, 5022) 9255 (7788, 10722)	11142		

Observed and Predicted GSH Depletion and VC Metabolism: VC-FO Model with Parameter Set FO3

<sup>a</sup>Observed depletion of GSH or amount of VC metabolized as of the end of exposure. In parentheses are the values approximately one standard deviation away from the mean, when these have been estimable from reported data. In some cases, a study has reported more than one mean value for a given exposure scenario; all reported means and associated standard deviations are presented.

<sup>b</sup>Predicted, as of the end of exposure, by the VC-FO model.

<sup>c</sup>Data from Jedrychowski et al. (1984, 1985).

<sup>d</sup>GSH depletion is presented in terms of the ratio of the GSH concentration in exposed animal livers to that in control animal livers.

•Data from Tarkowski et al. (1980). Predicted values correspond to GSH depletion one-half hour after end of exposure because of ambiguity as to exact time of GSH measurement in this study.

<sup>f</sup>Data from Watanabe et al. (1978).

 ${}^{8}\text{VC}$  metabolism is presented in terms of  $\mu g$  of VC metabolized by the end of exposure.

					Predicted <sup>c</sup>			
Data S	Set			01	bserved <sup>b</sup>	F01	FO2	F03
Jedrychov	vski et	al, (198	34) - 4	4 hr expos	sures			· · · · · · · · · · · · · · · · · · ·
19.4	4 ppm	@20	hrs <sup>d</sup>	1.04	(0.94, 1.10)	1.00	1.00	1.00
		<u></u> @44	hrs	1.02	(0.97, 1.07)	1.00	1.00	1.00
58.3	3 ppm	@20	hrs	0.94	(0.84, 1.04)	1.00	1.00	1.00
		@44	hrs	0.99	(0.84, 1.14)	1.00	1.00	1.00
219	ррт	@20	hrs	1.09	(1.00, 1.18)	1.01	1.01	1.01
		@44	hrs	0.94	(0.88, 1.01)	1.00	1.00	1.00
5796	ppm	@20	hrs	0.86	(0.78, 0.94)	1.02	1.02	1.02
		@44	hrs	1.02	(0.89, 1.14)	1.00	1.00	1.00
Tarkowski	<u>i et al.</u>	(1980)	<u>- 5 h</u>	r_exposure	<u>es</u>			
50		611	hra	1 06	(0 01 1 22)	0.00	0 00	0.00
50	ppm	611	hra	1.00	(0.91, 1.22)	1 00	1 00	0.98
500		611	hra	1 15	(0.02, 1.00)	0.03	1.00	1.00
200	քքա	611	nis bra	1.15	(1.04, 1.27)	1 01	1 01	0.93
20000		(e24) (e1)	nis bee	1.04	(0.91, 1.17)	1.01	1.01	1.01
20000	րիա	@11 @24	hrs	1.21	(0.75, 0.97) (1.05, 1.37)	1.03	1.04	1.04
					(2000)			
<u>Watanabe</u>	et al.	<u>(1976)</u> •						
10	DDW	@ 7	hrs	1.15	(1.09, 1.21)	0.98	0.98	0.98
50	ppm	<b>@</b> 7	hrs	0.86	(0.45, 1.27)	0.90	0.92	0.92
150	מסמ	ē 2	hrs	0.82	(0.72, 0.91)	0.88	0.90	0.91
	r r -	<b>e</b> 4	hrs	0.72	(0.69, 0.80)	0.81	0.84	0.85
		ē 7	hrs	0.74	(0.67. 0.81)	0.75	0.79	0.80
250	מסס	à 1	hr	0.96	(0.78, 1.14)	0.91	0.92	0.93
	rr-	ê 3	hrs	0.90	(0.76. 1.01)	0.78	0.81	0.82
		a s	hrs	0.66	(0.52, 0.80)	0.70	0.74	0.75
		a 7	hrs	0.63	(0.52, 0.75)	0.66	0.71	0.72
1000	ກກຫ	a i	hr	0.87	(0.67, 1.05)	0.88	0 90	0.89
2000	PP-	a 2	hre	0 78	(0, 71, 0, 86)	0.00	0.81	0.81
		62	hre	0.70	$(0.59 \ 0.82)$	0.72	0.75	0 74
		6 J 6 A	hre	0.71	(0.53, 0.02)	0 67	0 71	0.74
		9 - 19 A - 1	hre	0.03	(0.33, 0.34)	0.07	0.71	0.70
		ر بي م د	hra	0.00	(0.43, 0.72)	0.04	0.0/	0.00
		י ש ר ה	nis bre	0.30	(0.43, 0.70)	0.01	0.00	0.04
2000	~~~	(e) (a)	nis he-	0.02	(0.43, 0.70)	0.00	0.04	0.02
2000	ЪЪщ	2 س م	nrs bre	U.0/	(0.30, 0.73)		0./9	0./8
		64	nrs	0.53	(0.44, 0.62)	0.65	0.6/	U.66
		(a /	nrs	0.39	(0.23, 0.52)	U.38	U.60	U.58

## Observed and Predicted GSH Depletion; VC-FO Model with Parameters Sets FO1, FO2 and FO3\*

#### Table V-1-15 (continued)

					Predicted <sup>c</sup>			
Data Set			Observed <sup>b</sup>			FO2	F03	
Hefner et al. (1975	)•			<u></u>				
50 ppm	@ 1 hr @ 7 hrs	0.98 0.39	(0.87, (0.21,	1.10) 0.56)	0.98 0.90	0.98 0.92	0.98 0.92	
<u>Hefner et al. (1975</u>	) – 5 days/wk,	<u>7 hr</u>	s/day e:	<u>xposu<b>re</b></u>				
<u>l wk_exposure;</u>	@_101.5 hrs							
50 ppm 500 ppm 5000 ppm 15000 ppm		0.79 0.60 0.72 0.69	(0.62, (0.44, (0.65, (0.47,	0.95) 0.77) 0.78) 0.91)	0.91 0.63 0.53 0.38	0.93 0.68 0.52 0.32	0.93 0.67 0.50 0.32	
3 wk exposure;	@ 437.5 hrs							
500 ppm 5000 ppm		0.74 0.57	(0.60, (0.52,	0.88) 0.62)	0.63 0.53	0.68 0.52	0.67 0.50	
5 wk exposure;	@ 773.5 hrs							
500 ррт 5000 ррт		0.91 0.84	(0.76, (0.73,	1.05) 0.96)	0.63 0.53	0.68 0.52	0.67 0.50	
<u>Du et al. (1982) -</u>	5 days/wk. 7 h	rs/day	y expos	ure to 2	8000 pr	om		
2 wk exposure; 4 wk esposure; 6 wk exposure;	@ 291 hrs @ 627 hrs @ 963 hrs	1.21 1.44 1.65	(1.15, (1.08, (1.26,	1.26) 1.80) 2.04)	1.05 1.05 1.05	1.06 1.06 1.06	1.06 1.06 1.06	

#### Observed and Predicted GSH Depletion; VC-FO Model with Parameters Sets FO1, FO2 and FO3\*

<sup>a</sup>GSH depletion is expressed as the ratio of the GSH concentration in the livers of exposed animals to that in control animals.

<sup>b</sup>Mean value and, in parentheses, estimated values one standard deviation away from the mean.

<sup>c</sup>Predicted by the model with the indicated parameter set. Study-specific body weights were used.

<sup>d</sup>The times are measured from the beginning of exposure.

"In this study, exposure times varied. The times given are the times at which exposure ceased.



# Simple VC Model



$$ca = (qc*cv + qp*c1) / (qp/pb + qc)$$

$$cv = (qf*cf/pf + qs*cs/ps + qr*cr/pr + q1*c1/p1) / qc$$

$$dcf = qf * (ca - cf/pf) / vf$$

$$dcs = qs * (ca - cs/ps) / vs$$

$$dcr = qr * (ca - cr/pr) / vr$$

$$dc1 = q1 * (ca - c1/p1) / vl - dcm$$

$$dcm = (vmax * c1/p1) / (km + c1/p1) / vl + kf*c1/p1$$

$$vmax = vmaxc*bw^{(0.74)}$$

$$qc = qc*bw^{(0.74)}$$

$$qp = qpc*bw^{(0.74)}$$

$$qf = qfc*qc$$

$$qs = qc*dc$$

$$qr = qrc*qc$$

$$ql = q1c*qc$$

$$vf = vfc*bw$$

$$vr = vrc*bw$$

$$vl = vlc*bw$$

$$vl = vlc*bw^{(0.70)}$$

$$kf = kfc*bw^{(0.70)}$$



V-1-43









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V-1-46





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# APPENDIX V-1-A

**VC-GSH PBPK MODEL** 

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(2)

#### Appendix V-1-A

### VC-GSH PBPK Model

ATMOSPHERIC CONCENTRATIONS

CIZONE - PULSE(0.0, DAY, TCHNG) \* PULSE(0.0, WEEK, WKEND)

CI - CIO\*CIZONE (MG/L)

CP - CI\*24450.0/MW (PPM)

ARTERIAL BLOOD CONCENTRATION, CA (MG/L)

CA = (QC\*CV+QP\*CI)/(QP/PB+QC)

CX - CA/PB

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MIXED VENOUS BLOOD CONCENTRATION, CV (MG/L)
CV = (QF\*CVF + QL\*CVL + QS\*CVS + QR\*CVR )/QC

AMOUNT IN FAT COMPARTMENT, AF (MG)

d(AF)/dt = QF\*(CA-CVF)

CVF = AF/(VF\*PF)

CF = AF/VF

AMOUNT IN SLOWLY PERFUSED TISSUES, AS (MG)

d(AS)/dt = QS\*(CA-CVS)

CVS = AS/(VS\*PS)

CS - AS/VS

AMOUNT IN RAPIDLY PERFUSED TISSUES, AR (MG)

d(AR)/dt = QR\*(CA-CVR)

CVR - AR/(VR\*PR)

CR - AR/VR

AMOUNT IN LIVER COMPARTMENT, AL (MG) d(AL)/dt = QL\*(CA-CVL) - d(AM)/dt CVL = AL/(VL\*PL) CL = AL/VL CVLP = CVL\*1000./MW (UMOLES/L)

AMOUNT METABOLIZED, AM (MG) d(AM)/dt = (VMAX\*CVL)/(KM + CVL) + d(ACPG)/dt\*MW/1000.0 AMP = AM\*1000.0/MW (UMOLES) d(AMP)/dt = d(AM)/dt \* 1000.0/MW

EPOXIDE INTERMEDIATE, AEI (UMOLES)

d(AEI)/dt = VMAXP\*CVL/(KM+CVL) - d(AEGS)/dt - d(AEOM)/dt

CMU = AEI/VL

EPOXIDE INTERMEDIATE CONJUGATED WITH GLUTATHIONE, AEGS (UMOLES) d(AEGS)/dt - KMGS\*CGS\*CMU\*VL

EPOXIDE INTERMEDIATE GOING TO EVERYTHING ELSE, AEOM (UMOLES) d(AEOM)/dt - KMEE\*CMU\*VL

GSH SYNTHETASE ACTIVITY, KT (UMOLES/HR)

d(KT)/dt = K10\*(K1S+CGSO)/(K1S+CGS) - K1P\*KT

GLUTATHIONE, AMGS (UMOLES) d(AMGS)/dt = KT\*(KS+CGSO)/(KS+CGS) - KP\*CGS\*VL - d(ACPG)/dt - d(AEGS)/dt CGS = AMGS/VL

GSD - CGS/CGSO

# AMOUNT PARENT CONJUGATED WITH GLUTATHIONE, ACGP (UMOLES)

d(ACPG)/dt = KGS\*CVLP\*VL\*CGS

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$QC = QCC * BW^{0.74}$	TOTAL CARDIAC OUTPUT (L/HR)
$QP = QPC * BW^{0.74}$	VENTILATION RATE (L/HR)
OL - OLC*OC	BLOOD FLOW TO LIVER (L/HR)
QF - QFC*QC	BLOOD FLOW TO FAT (L/HR)
$QS = 0.24 \times QC - QF$	BLOOD FLOW TO SLOWLY PERFUSED (L/HR)
QR = 0.76*QC-QL	BLOOD FLOW TO RICHLY PERFUSED (L/HR)
VL - VLC*BW	VOLUME OF LIVER TISSUE (L)
VF - VFC*BW	VOLUME OF FAT TISSUE (L)
VS - 0.82*BW-VF	VOLUME OF LIVER SLOWLY PERFUSED TISSUE (L)
$VR = 0.09 \times BW - VL$	VOLUME OF LIVER RICHLY PERFUSED TISSUE (L)
VCH - VCHC-NANIM*BW	EFFECTIVE VOLUME OF CLOSED CHAMBER
	(TOTAL VOLUME MINUS SPACE OCCUPIED BY
	NANIM ANIMALS OF WEIGHT BW)
$VMAX = VMAXC * BW^{0.7}$	(MG/HR)
VMAX = VMAXC*BW <sup>0.7</sup> VMAXP = VMAXC*BW <sup>0.7</sup> * 100	(MG/HR) 00.0/MW (UMOLES/HR)
VMAX - VMAXC*BW <sup>0.7</sup> VMAXP - VMAXC*BW <sup>0.7</sup> * 100	(MG/HR) 00.0/MW (UMOLES/HR)
VMAX = VMAXC*BW <sup>0.7</sup> VMAXP = VMAXC*BW <sup>0.7</sup> * 100	(MG/HR) DO.0/MW (UMOLES/HR)
VMAX = VMAXC*BW <sup>0.7</sup> VMAXP = VMAXC*BW <sup>0.7</sup> * 100 K1S	(MG/HR) DO.0/MW (UMOLES/HR) CONSTANT CONTROLLING ENZYME SYNTHESIS
VMAX = VMAXC*BW <sup>0.7</sup> VMAXP = VMAXC*BW <sup>0.7</sup> * 100 K1S KS	(MG/HR) DO.0/MW (UMOLES/HR) CONSTANT CONTROLLING ENZYME SYNTHESIS CONSTANT CONTROLLING RESYNTHSIS
VMAX = VMAXC* $BW^{0.7}$ VMAXP = VMAXC* $BW^{0.7}$ * 100 K1S KS KP = KPC/ $BW^{0.3}$	(MG/HR) DO.0/MW (UMOLES/HR) CONSTANT CONTROLLING ENZYME SYNTHESIS CONSTANT CONTROLLING RESYNTHSIS FIRST ORDER RATE CONSTANT FOR GSH LOSS (PER HR)
VMAX = VMAXC*BW <sup>0.7</sup> VMAXP = VMAXC*BW <sup>0.7</sup> * 100 K1S KS $KP = KPC/BW^{0.3}$ K10 = K10C*BW <sup>0.7</sup>	(MG/HR) DO.0/MW (UMOLES/HR) CONSTANT CONTROLLING ENZYME SYNTHESIS CONSTANT CONTROLLING RESYNTHSIS FIRST ORDER RATE CONSTANT FOR GSH LOSS (PER HR) CONSTANT CONTROLING SYNTHETASE FORMATION
VMAX = VMAXC*BW <sup>0.7</sup> VMAXP = VMAXC*BW <sup>0.7</sup> * 100 K1S KS KP = KPC/BW <sup>0.3</sup> K10 = K10C*BW <sup>0.7</sup>	(MG/HR) D0.0/MW (UMOLES/HR) CONSTANT CONTROLLING ENZYME SYNTHESIS CONSTANT CONTROLLING RESYNTHSIS FIRST ORDER RATE CONSTANT FOR GSH LOSS (PER HR) CONSTANT CONTROLING SYNTHETASE FORMATION (UMOLES/HR/HR)
VMAX = VMAXC*BW <sup>0.7</sup> VMAXP = VMAXC*BW <sup>0.7</sup> * 100 K1S KS KP = KPC/BW <sup>0.3</sup> K10 = K10C*BW <sup>0.3</sup> KGS = KGSC/BW <sup>0.3</sup>	(MG/HR) DO.0/MW (UMOLES/HR) CONSTANT CONTROLLING ENZYME SYNTHESIS CONSTANT CONTROLLING RESYNTHSIS FIRST ORDER RATE CONSTANT FOR GSH LOSS (PER HR) CONSTANT CONTROLING SYNTHETASE FORMATION (UMOLES/HR/HR) CONJUGATION RATE CONSTANT WITH PARENT
VMAX = VMAXC*BW <sup>0.7</sup> VMAXP = VMAXC*BW <sup>0.7</sup> * 100 K1S KS $KP = KPC/BW^{0.3}$ K10 = K10C*BW <sup>0.7</sup> KGS = KGSC/BW <sup>0.3</sup>	(MG/HR) DO.0/MW (UMOLES/HR) CONSTANT CONTROLLING ENZYME SYNTHESIS CONSTANT CONTROLLING RESYNTHSIS FIRST ORDER RATE CONSTANT FOR GSH LOSS (PER HR) CONSTANT CONTROLING SYNTHETASE FORMATION (UMOLES/HR/HR) CONJUGATION RATE CONSTANT WITH PARENT (PER HR, PER UMOLE/L GSH)
VMAX = VMAXC*BW <sup>0.7</sup> VMAXP = VMAXC*BW <sup>0.7</sup> * 100 K1S KS KP = KPC/BW <sup>0.3</sup> K1O = K1OC*BW <sup>0.3</sup> KGS = KGSC/BW <sup>0.3</sup> KMGS = KMGSC/BW <sup>0.3</sup>	(MG/HR) DO.0/MW (UMOLES/HR) CONSTANT CONTROLLING ENZYME SYNTHESIS CONSTANT CONTROLLING RESYNTHSIS FIRST ORDER RATE CONSTANT FOR GSH LOSS (PER HR) CONSTANT CONTROLING SYNTHETASE FORMATION (UMOLES/HR/HR) CONJUGATION RATE CONSTANT WITH PARENT (PER HR, PER UMOLE/L GSH) CONJUGATION RATE CONSTANT WITH EPOXIDE
VMAX = VMAXC* $BW^{0.7}$ VMAXP = VMAXC* $BW^{0.7}$ * 100 K1S KS KP = KPC/ $BW^{0.3}$ K1O = K1OC* $BW^{0.7}$ KGS = KGSC/ $BW^{0.3}$ KMGS = KMGSC/ $BW^{0.3}$	(MG/HR) DO.0/MW (UMOLES/HR) CONSTANT CONTROLLING ENZYME SYNTHESIS CONSTANT CONTROLLING RESYNTHSIS FIRST ORDER RATE CONSTANT FOR GSH LOSS (PER HR) CONSTANT CONTROLING SYNTHETASE FORMATION (UMOLES/HR/HR) CONJUGATION RATE CONSTANT WITH PARENT (PER HR, PER UMOLE/L GSH) CONJUGATION RATE CONSTANT WITH EPOXIDE (PER HR, PER UMOLE/L GSH)
VMAX = VMAXC* $BW^{0.7}$ VMAXP = VMAXC* $BW^{0.7}$ * 100 K1S KS KP = KPC/ $BW^{0.3}$ K1O = K1OC* $BW^{0.7}$ KGS = KGSC/ $BW^{0.3}$ KMGS = KMGSC/ $BW^{0.3}$ KMEE = KMEEC/ $BW^{0.3}$	(MG/HR) DO.0/MW (UMOLES/HR) CONSTANT CONTROLLING ENZYME SYNTHESIS CONSTANT CONTROLLING RESYNTHSIS FIRST ORDER RATE CONSTANT FOR GSH LOSS (PER HR) CONSTANT CONTROLING SYNTHETASE FORMATION (UMOLES/HR/HR) CONJUGATION RATE CONSTANT WITH PARENT (PER HR, PER UMOLE/L GSH) CONJUGATION RATE CONSTANT WITH EPOXIDE (PER HR, PER UMOLE/L GSH) FIRST-ORDER RATE CONSTANT FOR EPOXIDE WITH NON-GSH (PER HR)
VMAX = VMAXC*BW <sup>0.7</sup> VMAXP = VMAXC*BW <sup>0.7</sup> * 100 K1S KS $KP = KPC/BW^{0.3}$ K1O = K1OC*BW <sup>0.3</sup> KMGS = KMGSC/BW <sup>0.3</sup> KMEE = KMEEC/BW <sup>0.3</sup> K1P = K1PC/BW <sup>0.3</sup>	(MG/HR) DO.0/MW (UMOLES/HR) CONSTANT CONTROLLING ENZYME SYNTHESIS CONSTANT CONTROLLING RESYNTHSIS FIRST ORDER RATE CONSTANT FOR GSH LOSS (PER HR) CONSTANT CONTROLING SYNTHETASE FORMATION (UMOLES/HR/HR) CONJUGATION RATE CONSTANT WITH PARENT (PER HR, PER UMOLE/L GSH) CONJUGATION RATE CONSTANT WITH EPOXIDE (PER HR, PER UMOLE/L GSH) FIRST-ORDER RATE CONSTANT FOR EPOXIDE WITH NON-GSH (PER HR) FIRST-ORDER SYNTHETASE DESTRUCTION (PER HR)
VMAX = VMAXC* $BW^{0.7}$ VMAXP = VMAXC* $BW^{0.7}$ * 100 K1S KS KP = KPC/ $BW^{0.3}$ K10 = K10C* $BW^{0.7}$ KGS = KGSC/ $BW^{0.3}$ KMGS = KMGSC/ $BW^{0.3}$ KMEE = KMEEC/ $BW^{0.3}$ K1P = K1PC/ $BW^{0.3}$ K0 = K10/K1P	(MG/HR) DO.0/MW (UMOLES/HR) CONSTANT CONTROLLING ENZYME SYNTHESIS CONSTANT CONTROLLING RESYNTHSIS FIRST ORDER RATE CONSTANT FOR GSH LOSS (PER HR) CONSTANT CONTROLING SYNTHETASE FORMATION (UMOLES/HR/HR) CONJUGATION RATE CONSTANT WITH PARENT (PER HR, PER UMOLE/L GSH) CONJUGATION RATE CONSTANT WITH EPOXIDE (PER HR, PER UMOLE/L GSH) FIRST-ORDER RATE CONSTANT FOR EPOXIDE WITH NON-GSH (PER HR) FIRST-ORDER SYNTHETASE DESTRUCTION (PER HR)
VMAX = VMAXC*BW <sup>0.7</sup> VMAXP = VMAXC*BW <sup>0.7</sup> * 100 K1S KS $KP = KPC/BW^{0.3}$ K1O = K1OC*BW <sup>0.3</sup> KMGS = KGSC/BW <sup>0.3</sup> KMEE = KMEEC/BW <sup>0.3</sup> K1P = K1PC/BW <sup>0.3</sup> K0 = K1O/K1P CGSO = KO/KP/VL	(MG/HR) DO.0/MW (UMOLES/HR) CONSTANT CONTROLLING ENZYME SYNTHESIS CONSTANT CONTROLLING RESYNTHSIS FIRST ORDER RATE CONSTANT FOR GSH LOSS (PER HR) CONSTANT CONTROLING SYNTHETASE FORMATION (UMOLES/HR/HR) CONJUGATION RATE CONSTANT WITH PARENT (PER HR, PER UMOLE/L GSH) CONJUGATION RATE CONSTANT WITH EPOXIDE (PER HR, PER UMOLE/L GSH) FIRST-ORDER RATE CONSTANT FOR EPOXIDE WITH NON-GSH (PER HR) FIRST-ORDER SYNTHETASE DESTRUCTION (PER HR) INITIAL CONCENTRATION OF GSH (UMOLES/L)

# APPENDIX V-1-B

# **VC-FO PBPK MODEL**

### Appendix V-1-B

#### VC-FO PBPK Model

### ATMOSPHERIC CONCENTRATIONS

CIZONE = PULSE(0.0, DAY, TCHNG) \* PULSE(0.0, WEEK, WKEND)

CI - CIO\*CIZONE (MG/L)

CP - CI\*24450.0/MW (PPM)

ARTERIAL BLOOD CONCENTRATION, CA (MG/L)

CA = (QC\*CV+QP\*CI)/(QP/PB+QC)

CX - CA/PB

MIXED VENOUS BLOOD CONCENTRATION, CV (MG/L)

CV = (QF\*CVF + QL\*CVL + QS\*CVS + QR\*CVR)/QC

AMOUNT IN FAT COMPARTMENT, AF (MG)

d(AF)/dt = QF\*(CA-CVF)

CVF = AF/(VF\*PF)

CF = AF/VF

AMOUNT IN SLOWLY PERFUSED TISSUES, AS (MG)

d(AS)/dt = QS\*(CA-CVS)

CVS = AS/(VS\*PS)

CS - AS/VS

AMOUNT IN RAPIDLY PERFUSED TISSUES, AR (MG)

d(AR)/dt = QR\*(CA-CVR)

CVR = AR/(VR\*PR)

CR - AR/VR

AMOUNT IN LIVER COMPARTMENT, AL (MG) d(AL)/dt = QL\*(CA-CVL) - d(AM1)/dt - d(AM2)/dt CVL = AL/(VL\*PL) CL = AL/VL CVLP = CVL\*1000./MW (UMOLES/L) AMOUNT METABOLIZED BY FIRST PATHWAY, AM1 (MG)

d(AM1)/dt - (VMAX\*CVL)/(KM + CVL)

AM1P - AM1\*1000.0/MW (UMOLES)

d(AM1P)/dt - d(AM1)/dt \* 1000.0/MW

AMOUNT METABOLIZED BY SECOND PATHWAY, AM2 (MG)

d(AM2)/dt = KF\*CVL\*VL

AM = AM1 + AM2

EPOXIDE INTERMEDIATE, AEI (UMOLES)

d(AEI)/dt = VMAXP\*CVL/(KM+CVL) + KF\*CVLP\*VL - d(AEGS)/dt - d(AEOM)dt
CMU = AEI/VL

EPOXIDE INTERMEDIATE CONJUGATED WITH GLUTATHIONE, AEGS (UMOLES) d(AEGS)/dt - KMGS\*CGS\*CMU\*VL

EPOXIDE INTERMEDIATE GOING TO EVERYTHING ELSE, AEOM (UMOLES) d(AEOM)/dt - KMEE\*CMU\*VL

GSH SYNTHETASE ACTIVITY, KT (UMOLES/HR) d(KT)/dt - K10\*(K1S+CGSO)/(K1S+CGS) - K1P\*KT

GLUTATHIONE, AMGS (UMOLES)

d(AMGS)/dt = KT\*(KS+CGSO)/(KS+CGS) - KP\*CGS\*VL - d(ACPG)/dt - d(AEGS)/dt

CGS = AMGS/VL

GSD - CGS/CGSO

QC = QCC\*BW<sup>0.74</sup>TOTAL CARDIAC OUTPUT (L/HR)QP = QPC\*BW<sup>0.74</sup>VENTILATION RATE (L/HR)QL = QLC\*QCBLOOD FLOW TO LIVER (L/HR)QF = QFC\*QCBLOOD FLOW TO FAT (L/HR)QS = 0.24\*QC-QFBLOOD FLOW TO SLOWLY PERFUSED (L/HR)QR = 0.76\*QC-QLBLOOD FLOW TO RICHLY PERFUSED (L/HR)VL = VLC\*BWVOLUME OF LIVER TISSUE (L)VF = VFC\*BWVOLUME OF FAT TISSUE (L)VS = 0.82\*BW-VFVOLUME OF LIVER SLOWLY PERFUSED TISSUE (L)VR = 0.09\*BW-VLVOLUME OF LIVER RICHLY PERFUSED TISSUE (L)VCH = VCHC-NANIM\*BWEFFECTIVE VOLUME OF CLOSED CHAMBER<br/>(TOTAL VOLUME MINUS SPACE OCCUPIED BY<br/>NANIM ANIMALS OF WEIGHT BW)

CONSTANT CONTROLLING ENZYME SYNTHESIS CONSTANT CONTROLLING RESYNTHSIS
FIRST ORDER RATE CONSTANT FOR GSH LOSS (PER HR)
CONSTANT CONTROLING SYNTHETASE FORMATION (UMOLES/HR/HR)
FIRST ORDER RATE OF EPOXIDE FORMATION (PER HR)
CONJUGATION RATE CONSTANT WITH EPOXIDE
FIRST-ORDER RATE CONSTANT FOR EPOXIDE WITH NON-GSH (PER HR)
FIRST-ORDER SYNTHETASE DESTRUCTION (PER HR)
INITIAL CONCENTRATION OF GSH (UMOLES/L)

# **VOLUME V**

# PART 2 OF 2 PARTS

# PRELIMINARY EXTENSIONS OF A PBPK MODEL TO MICE AND HAMSTERS

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#### A. INTRODUCTION

The VC-GSH model discussed in Part 1 of this volume was investigated further. Additional data from WPAFB were made available so that strainspecific analyses could be performed. The analyses based on those data are presented here.

#### B. DATA

The following data were obtained from WPAFB.

Tissue/air partition coefficients were estimated by vial equilibration techniques (Gargas et al., 1989) for four tissues (blood, liver, muscle, and fat) from males and females of three strains of rats (F-344, CDBR, and Wistar) and one strain of homster (Golden Syrian). Blood/air partition coefficients were estimated for two strains of mice (B6C3F1 and CD-1). The measurements obtained are listed in Table V-2-1.

Gas uptake experiments were conducted using the same rodent strains. In those studies, concentrations of VC in closed chambers were monitored during the course of exposure to various initial concentrations. Exposure lasted up to 6 hours.

In addition, the hepatic glutathione content of the animals exposed in the closed chamber experiments was measured immediately post-exposure. Control animals were cacrificed for GSH determination either at the time the exposure started (pre-exposure controls) or at the time exposure ended

V-2-1

(concurrently with the exposed animals; post-exposure controls). The results are summarized in Table V-2-2.<sup>1</sup>

### C. ANALYSES AND RESULTS

Initially, the gas uptake results were used to estimate the parameters vmaxc and kfc of the simple VC model discussed in Part 1 of this volume. The species-specific physiological parameters used for the estimation are displayed in Table V-2-3. The parameter km was set to the value 0.1 for all runs (all strains). The strain-specific partition coefficients shown in Table V-2-1 were used to derive tissue/blood partition coefficients.<sup>2</sup> The optimized estimates of vmaxc and kfc are displayed in Table V-2-4.

The estimated vmaxc and km values were used unchanged in the next step of the analysis. That step was the elaboration of the VC-GSH model discussed in Part 1 of this volume, for each of the strains tested. The kfc estimates were used as follows. As shown in Appendix V-1-A, the rate of GSH conjugation of VC in the VC-GSH model was

(1) kgs  $\star$  CGS  $\star$  CVLP  $\star$  VL,

<sup>&</sup>lt;sup>1</sup>Michael Cargas, who supervised the experiments described, has informed us that he believes the glutathione measurements to be unreliable. Limited use was made of those measurements.

<sup>&</sup>lt;sup>2</sup>Certain exceptions apply. All rat blood/air coefficients were set to 3.0; male F-344 rat coefficients were set to 2.6 for liver/air, 2.04 for muscle/air, and 11.6 for fat/air.

where CGS is the concentration of GSH ( $\mu$ M/L), CVLP is the concentration of VC in the venous blood leaving the liver ( $\mu$ M/L), and VL is the volume of the liver (L). The rate constant, kgs ( $hr^{-1}/\mu$ M GSH/L), was scaled to body weight:

(2) kgs - kgsc \* 
$$bw^{-0.3}$$
.

If the first-order rate of VC metabolism estimated by the simple VC model (- kf\*CVLP\*VL) approximated the second-order conjugation of VC and GSH, then the rates should be approximately the same for those two pathways; i.e., the following would be approximately true:

This implies that kgs\*CGS was approximately equal to kf. The parameter, kf, was also scaled according to body weight  $(kf - kfc*bw^{-0.3})$ , so that kgsc could be approximated by kfc/CGS. The values of kfc were estimated for each of the strains of rats, mice, and hamsters tested in the gas uptake studies (Table V-2-4), so it was theoretically possible to obtain strain-specific estimates of kgsc for use in the VC-GSH model.

The concentration of GSH in the liver is not constant during exposure to VC, as shown in the results discussed in Part 1 of this document. Thus, selection of GSH concentrations to use in the estimation of kgsc was problematic. The average of the pre-exposure control and post-exposure control values (averaged over all exposure groups; see Table V-2-2) was used as a first approximation for each strain. The estimates of kgsc obtained in this manner are shown in Table V-2-5.

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It is of interest to note that the kgsc values for F-344 and CDBR (Sprague-Dawley) rats (averaging  $2.39 \times 10^{-4}$ ) are not essentially different from the value of  $2 \times 10^{-4}$  that was used in parameter sets GSH1 and GSH2. The kgsc estimate in those parameter sets was estimated from a combination of results from the literature; both F-344 and Sprague-Dawley rats were used to get those results.

Finally, closed chamber concentrations were predicted using the strainspecific estimates of vmaxc and kgsc (with km set equal to 0.1, the partition coefficients set equal to the values shown in Table V-2-1,<sup>3</sup> and the physiological parameters set equal to the values shown in Table V-2-3). The other parameters of the VC-GSH model were set to values listed for parameter set GSH2 (Table V-1-7). The predictions were in fairly good agreement with the observed concentrations for all of the strains (Figures V-2-1 through V-2-10).

#### D. DISCUSSION

The results presented above represent a first step toward defining strain-specific VC-GSH models. Additional work would be required before those models could be claimed to be complete. Some of that work is outlined here.

The gas uptake experiment results can be further exploited to help refine some of the parameters, especially those related to the metabolism of the parent compound. The parameter vmaxc can be reoptimized using the gas uptake experiment results and the first approximations of kgsc. Strain-

<sup>&</sup>lt;sup>3</sup>Certain exceptions apply. All rat blood/air coefficients were set to 3.0; male F-344 rat coefficients were set to 2.6 for liver/air, 2.04 for muscle/air, and 11.6 for fat/air.

specific initial GSH concentrations (based in the glutath one measurements of Table V-2-2) could also be used. The initial concentration was determined by the parameters kpc, kloc, and klpc in the VC-GSH model (see Appendix V-1-A), so there is an opportunity to adjust these parameters to approximate the strain-specific initial concentrations and also affect the predictions of kinetic behavior of GSH depletion and resynthesis.

Now that estimates of parameters are available for specific strains of rats, the data on glutathione depletion in the literature can also be reexamined. Thus, it may be possible to take advantage of the strain-specific estimates discussed in the preceding paragraph (obtained from the gas uptake results) to estimate values for kmeec and kmgsc (and refine estimates of kpc, kloc, klpc, kls and ks) based on the published glutathione data.

The modeling for humans is still uncertain. It does not appear that there exist data comparable to those available for rodents (making possible estimation of all of the parameters in the VC-GSH model). Species scale-up (allometry) may have to suffice for a number of the parameters.

### REFERENCES

Gargas, M., Burgess, R., Voisard, D., et al. (1989). Partition coefficients of low molecular weight. Volatile chemicals in various liquids and tissues. Toxicol Appl Pharmacol 98:87-99.

Table	V-2	2-1
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			Partition Coefficients				
<u>Species</u>	Strain	Sex	Blood	Liver	Muscle	Fat	
Rat	F-344	М	1.60 <u>+</u> 0.328	1.99 <u>+</u> 1.96	2.06 <u>+</u> 0.703	11.8 <u>+</u> 0.811	
		F	1.55 <u>+</u> 0.11	2.05 <u>+</u> 0.17	2.39 <u>+</u> 0.46	21.1 <u>+</u> 1.3	
	CDBR	м	1.79 <u>+</u> 0.216	3.0 <u>+</u> 0.407	2.18 <u>+</u> 0.470	14.6 <u>+</u> 0.917	
		F	2.12 <u>+</u> 0.437	1.66 <u>+</u> 0.429	1.28 <u>+</u> 0.245	19.2 <u>+</u> 0.956	
	Wistar	м	2.10 <u>+</u> 0.313	2.69 <u>+</u> 0.555	2.72 <u>+</u> 0.575	10.2 <u>+</u> 1.61	
		F	1.62 <u>+</u> 0.0664	1.48 <u>+</u> 0.28	1.06±0.221	22.3 <u>+</u> 0.542	
Mouse	B6C3F1	м	2.83+0.22	•••	•		
		F	2.56 <u>+</u> 0.14			•••	
	CD-1	М	2.27 <u>+</u> 0.0725		* * *		
		F	2.37 <u>+</u> 0.16				
Hamster	Golden	М	2.74 <u>+</u> 0.151	3.38 <u>+</u> 0.362	2.56 <u>+</u> 0.457	14.3 <u>+</u> 5.32	
	Syrian	F	2.21 <u>+</u> 0.47	1.31 <u>+</u> 0.28	1.96 <u>+</u> 0.28	21.10 <u>+</u> 2.01	

Tissue/Air Partition Coefficients for Rodent Tissues\*

<sup>a</sup>Data obtained from WPAFB.

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## Hepatic Glutathione Measurements from Control and Exposed Rodents<sup>a</sup>

			<u>Average Her</u> Cont	<u>(µM/g)</u>		
<u>Species</u>	Strain	Sex	Exposure <sup>b</sup>	Pre-exposure	Post-exposure	Exposed
Rat	F-344	м	300	4 40	5 04	3.35
nue	1 0 1 1		500	5.12	4 36	3.64
			1000	3 92	3 56	2 47
			3000	4.75	3,49	1.95
		F	300	2.22	2.53	2.77
			500	7.25	5.41	4.41
			1000	4.50	4.71	2.79
			3000	5.06	4.59	2.36
	annad	_	200	( 70		0 70
	CDBR	F	300	4.72	3.11	2.70
			500	3.62	3.98	4.10
			1000			3.91
			3000	5.25	4.15	3.10
	Wistar	м	300	5.10	4 40	4.72
		••	500	5 13	4 45	3 72
			1000	4 07	3 71	2 41
			3000	6.04	5.44	3.16
		F	300	3.63	3.69	4.15
			500	3.49	4.16	3.34
			1000	3.28	4.46	3.12
			3000	4.28	3.97	2.26
Mauro	RECORI	м	300	10.04	6 04	1. 67
Mouse	DOCOLT	гі	500	7 66	0.04	4.07
			1000	7.00	5.90	2.99
			1000	8.76	4.10	2.61
			3000	/.82	4.80	2.61
		F	300	4.71	5.40	3.77
			500	4.87	3.63	3.25
			1000	7.66	5.09	3.17
			3000	7.33	4.11	1.63
	<b>.</b> .		200		0.07	< 17
	CD-1	ň	300	10.40	2.3/	0.1/
			500	12.56	5,83	6.49
			1000	10.28	6,90	7.06
			3000	11.52	7.58	2.07

#### Table V-2-2 (continued)

### Hepatic Glutathione Measurements from Control and Exposed Rodents<sup>®</sup>

							<u>Average Her</u> Cont	<u>e (µM/g)</u>
Species	Strain	Sex	Exposure <sup>b</sup>	Pre-exposure	Post-exposure	Exposed		
Mouse	CD-1	F	300	8.78	7.37	6.05		
			500	8.16	8.61	5.22		
			1000	7.05	8.85	6.48		
			3000	9.67	6.94	2.09		
Hamster	Golden <sup>®</sup>	м	300 <sup>r</sup>	3.52	4.15	3.32		
	Syrian		500	5.04	4.63	1.07		
	, <u> </u>		1000	4.98	5.85	1.84		
			3000	4.60	5.83	3.11		

 Data from WPAFB. Exposed animals were those used in gas uptake experiment, expressed for up to 6 hours.

<sup>b</sup> Exposures listed are approximate initial concentrations, in ppm.

<sup>c</sup> Pre-exposure control values were determined at the time the exposed animals began their closed-chamber exposures. Post-exposure control values were determined at the same time as the exposed values, i.e., at the end of the exposure period

<sup>d</sup> Male CDBR rats were not used for glutathione determination.

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• Female hamsters were not used for glutathione determination.

<sup>f</sup> The reported values for this exposure level are the averages of two sets of control and exposed GSH measurements. The 300 ppm exposure was run twice, about a week apart.

Parameter	Rats	Mice	Hamsters
qpc	14	23-35 <b>*</b>	13
qcc	14	23-35*	13
qlc	.25	. 24	.24
qrc	. 51	. 52	. 52
qfc	. 09 <sup>b</sup>	.05	.09
qsc	.15 <sup>b</sup>	. 20	.15
vlc	.04	. 04	.04
vrc	. 05	.05	.05
vfc	.071°	.04	.07
vsc	.7275°	.78	.75

### Physiological Parameters Used for Estimation of Vmaxc and Kfc from Gas Uptake Experiments

 Qpc was 23 for male B6C3F1, 25 for female B6C3F1, 28 for female CD-1, and 35 for male CD-1. Qcc equals qpc in each case.

<sup>b</sup> Male Wistar and CDBR rats had qfc=.08 and qsc=.16.

<sup>c</sup> Female F-344 and female Wistar rats had vfc=.07 and vsc=.75; male F-344 and female CDBR rats had vfc=.08 and vsc=.74; male Wistar and male CDBR rats had vfc=.1 and vsc=.72.

 Species	Strain	Sex	Vmaxc	Kfc	
Rat	F-344	м	3.17	1.08	
		F	2.95	1.03	
	CDBR	м	2.50	0.63	
		F	2.47	1.00	
	Wistar	M	3.11	0.45	
		F	2.97	1.55	
Mouse	B6C3F1	M	5.89	5.50	
		F	5.53	8.93	
	CD-1	м	6.99	5.10	
		F	5.54	6.62	
Hamster	Golden	M	4.94	1.67	
	Syrian	F	4.76	2.06	

Estimates of Vmaxc and Kfc Obtained from Gas Uptake Experiments\*

\* These estimates were completed by M. Gargas, then at WPAFB.

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Species	Strain	Sex	Kgsc	
Rat	F-344	M	0.000249	
		F	0.000227	
	CDBR <sup>b</sup>	F	0.000241	
	Wistar	м	0.000093	
		F	0.000400	
Mouse	B6C3F1	M	0.000827	
		F	0.00167	
	CD-1	м	0.000563	
		F	0.000809	
Hamster	Golden <sup>b</sup>	M	0.000330	
	Syrian			

Estimates of Kgsc Obtained from Gas Uptake Experiments\*

<sup>a</sup> See text for a discussion of the estimation procedure.

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<sup>b</sup> No GSH data were available for male CDBR rats or female hamsters.



V-2-13





V-2-14



Closed Chamber Concentrations vs. Time (Female CDBR Rats)





V-2-15





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Figure V-2-4





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Figure V-2-6

Figure V-2-7













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Figure V-2-10