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CI-921: A CLINICAL, PHARMACOKINETIC AND METABOLIC STUDY OF A POTENTIAL NEW CYTOTOXIC AGENT.

Being a thesis submitted for
the degree of
Doctor of Medicine
from the
University of Auckland

by

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Department of Clinical Oncology
Auckland Hospital
Auckland, New Zealand
1989
ABSTRACT

CI-921, an analogue of the antileukaemic agent amsacrine, was produced in an attempt to develop a cytotoxic agent with a broader spectrum of activity. CI-921 was selected for clinical trial on the basis of superior in vivo and in vitro solid tumour activity.

Sixteen patients with histologically documented cancer for which there was no conventional cytotoxic treatment were entered into a phase I trial. The dose of CI-921 was escalated from 39mg/m² to 810mg/m² (total dose divided over 3 days) and repeated 3 weekly. Neutropenia was the major dose limiting toxicity and defined a maximum tolerated dose of 810 mg/m².

Pharmacokinetic studies revealed a biexponential pattern of drug distribution with a distribution half-life of 2.6 h. The kinetics appeared linear over the dose range tested. Less than 1% of total drug was excreted in the urine.

Nineteen patients were entered into a limited phase II trial in non-small cell lung cancer using CI-921 at a dose of 648 mg/m² in the same 3-day schedule. One of the 16 evaluable patients achieved a partial response lasting five months. Myelosuppression was the predominant toxicity as in the phase I trial, but the degree of toxicity confirmed this dose as being suitable for further phase II trials. One patient had a grand mal seizure temporally associated with three of four courses of CI-921 raising the possibility of neurotoxicity.
Although drug-induced cardiotoxicity has been reported with the parent drug amsacrine, there was no evidence of this in the current study.

It has been suggested that CI-921 undergoes hepatic metabolism and biliary excretion following conjugation with glutathione. There was no fall in whole blood glutathione levels in patients following CI-921 infusion, although a transient decrease in mouse hepatic GSH was demonstrated following both amsacrine and CI-921. The toxicity of CI-921 in mice was markedly increased following depletion of hepatic glutathione with BSO but was not affected by pre-treatment with morphine or the glutathione "protector" N-acetyl cysteine.
CONTENTS

Abstract\hfill i

Contents\hfill iii

Acknowledgements\hfill iv

Glossary of abbreviations\hfill vi

List of figures\hfill ix

List of tables\hfill xiii

List of appendices\hfill xvi

Chapter 1 - Introduction and literature review\hfill 1

Chapter 2 - Phase I clinical trial\hfill 72

Chapter 3 - Phase II clinical trial in non-small cell lung cancer\hfill 90

Chapter 4 - Pharmacokinetic studies\hfill 104

Chapter 5 - The assessment of CI-921 for cardiotoxicity\hfill 153

Chapter 6 - Metabolic studies\hfill 172

Chapter 7 - Conclusions\hfill 220

Appendices\hfill 228

References\hfill 252

Associated publications\hfill 295
ACKNOWLEDGEMENTS

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I am indebted to the patients who took part in these studies in the knowledge that CI-921 was unlikely to help them personally and to the nursing staff who cared for them, especially staff nurse Shelba Smith. I wish to thank my principal supervisor, Dr V.J. Harvey, without whose enthusiastic help and encouragement this work could never have been undertaken.

Dr Baguley is the Director of the Cancer Research Laboratory in which both amsacrine and CI-921 were developed. I am very grateful to have had the opportunity of working in association with Dr Baguley and his team and hope that the "bridging" between clinical and laboratory cancer research will continue.

To Dr J. Paxton and Professor D. Paton who appointed me as honorary research fellow in the Department of Pharmacology, I offer special thanks.

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I am very grateful to Dr W. Wilson for providing the equipment, information, advice and materials necessary for the glutathione studies.

Sincere thanks to my mother Joan Hardy and to Jill Simpson and Noeline Pou for the preparation of this manuscript.

Most special thanks to Pandora Evans, my friend and mentor, whose technical, emotional and social support throughout this study was invaluable.
GLOSSARY OF ABBREVIATIONS

AGC  absolute granulocyte count
ALP  alkaline phosphatase
AST  aspartate transferase
AUC  area under the concentration time curve
AUMC area under the first moment of the concentration/time curve
BSO  buthionine sulfoximine
ß    slope of the terminal portion of the log concentration/time curve
cf   compared with
CHO  chinese hamster ovary
CCNU 1-(-2-chloroethyl)-3-cyclohexyl-1-nitrosourea
CL   plasma clearance
Cmax maximum concentration achieved in plasma
CNS  central nervous system
Ct   concentration at last time point
cv   coefficient of variation
DEM  diethyl maleate
DNA  deoxyribose nucleic acid
ECOG Eastern co-operative oncology group
EORTC European organization for research and the treatment of cancer
FS   fractional shortening
g/dl grams per deciliter
GSH glutathione (reduced form)
GSSH glutathione (oxidized form)
HPLC high performance liquid chromatography
%ILS percent increased life span
i.c. intra-cerebral
i.p. intra-peritoneal
I.S. internal standard
i.v. intra-venous
Ka association constant
LD₁₀ lethal dose in 10% of treated mice
LD₅₀ lethal dose in 50% of treated mice
LD₉₀ lethal dose in 90% of treated mice
LVEF left ventricular ejection fraction
m-AQDI quinine diimine of amsacrine
m-AQI quinine monoimine of amsacrine
mg/m² milligrams per square metre (surface area)
MIS misonidazole
MTD maximum tolerated dose
NAC N-acetyl cysteine
NCI National Cancer Institute
NSCLC non-small lung cancer
P probability
PCV packed cell volume
<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>QSAR</td>
<td>quantitative-molecular-structure-activity-relationship</td>
</tr>
<tr>
<td>QTc</td>
<td>corrected Q-T interval of electrocardiogram</td>
</tr>
<tr>
<td>r</td>
<td>Pearson's correlation coefficient</td>
</tr>
<tr>
<td>s.c.</td>
<td>subcutaneous</td>
</tr>
<tr>
<td>SC</td>
<td>seeded quality control (plasma)</td>
</tr>
<tr>
<td>SCU</td>
<td>seeded quality control (urine)</td>
</tr>
<tr>
<td>STI</td>
<td>systolic time interval</td>
</tr>
<tr>
<td>TEAP</td>
<td>triethylamine phosphate</td>
</tr>
<tr>
<td>$t_{1/2\alpha}$</td>
<td>distribution half-life</td>
</tr>
<tr>
<td>$t_{1/2\beta}$</td>
<td>terminal half-life</td>
</tr>
<tr>
<td>UE</td>
<td>urinary excretion</td>
</tr>
<tr>
<td>Tt</td>
<td>last time point</td>
</tr>
<tr>
<td>Vss</td>
<td>volume of distribution</td>
</tr>
</tbody>
</table>
# LIST OF FIGURES

<table>
<thead>
<tr>
<th>FIGURE</th>
<th>Description</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>FIGURE 1</td>
<td>CI-921</td>
<td>4</td>
</tr>
<tr>
<td>FIGURE 2</td>
<td>Amsacrine</td>
<td>5</td>
</tr>
<tr>
<td>FIGURE 3</td>
<td>Comparative structures of CI-921 and amsacrine</td>
<td>6</td>
</tr>
<tr>
<td>FIGURE 4</td>
<td>The metabolism of amsacrine</td>
<td>12</td>
</tr>
<tr>
<td>FIGURE 5</td>
<td>Intercalation of drug between base-pairs and subsequent distortion of the DNA helix</td>
<td>14</td>
</tr>
<tr>
<td>FIGURE 6</td>
<td>Mechanism of topoisomerase-mediated DNA cleavage by amsacrine and related drugs</td>
<td>19</td>
</tr>
<tr>
<td>FIGURE 7</td>
<td>The chemical structure of glutathione</td>
<td>64</td>
</tr>
<tr>
<td>FIGURE 8</td>
<td>Pathway of synthesis and metabolism of glutathione</td>
<td>66</td>
</tr>
<tr>
<td>FIGURE 9</td>
<td>Glutathione oxidation reduction cycle</td>
<td>66</td>
</tr>
<tr>
<td>FIGURE 10</td>
<td>Sample HPLC tracing for CI-921 and internal standard (IS)</td>
<td>113</td>
</tr>
<tr>
<td>FIGURE 11</td>
<td>Sample standard curve for back-calculation of CI-921 standards during assay validation</td>
<td>115</td>
</tr>
<tr>
<td>FIGURE 12</td>
<td>Plasma CI-921 concentration after the first day infusion over the dose range in three patients</td>
<td>121</td>
</tr>
<tr>
<td>FIGURE 13</td>
<td>Relationship between pharmacokinetic parameters and dose in individual patients</td>
<td>126</td>
</tr>
<tr>
<td>FIGURE 13.1</td>
<td>Relationship between dose and AUC of CI-921 after 65 infusions in 16 patients during a phase I trial</td>
<td>126</td>
</tr>
</tbody>
</table>
FIGURE 13.2  Relationship between dose and Cmax of CI-921 after 65 infusions in 16 patients during a phase I trial 126

FIGURE 13.3  Relationship between dose and Vss of CI-921 after 65 infusions in 16 patients during a phase I trial 127

FIGURE 13.4  Relationship between dose and CL of CI-921 after 65 infusions in 16 patients during a phase I trial 127

FIGURE 13.5  Relationship between dose and MRT of CI-921 after 65 infusions in 16 patients during a phase I trial 128

FIGURE 13.6  Relationship between dose and $t_{1/2}\beta$ of CI-921 after 65 infusions in 16 patients during a phase I trial 128

FIGURE 14  Relationship between dose of CI-921 and percent unchanged drug excreted in urine 129

FIGURE 15  Relationship between toxicity (nadir absolute granulocyte count) and pharmacokinetic parameters 131

FIGURE 15.1  Relationship between toxicity of CI-921 and area under curve (AUC) 131

FIGURE 15.2  Relationship between toxicity of CI-921 and maximum plasma concentration 131

FIGURE 15.3  Relationship between toxicity of CI-921 and dose 132

FIGURE 16  Relationship between toxicity (nadir absolute granulocyte count) and pharmacokinetic parameters - Phase II 136

FIGURE 16.1  Relationship between toxicity and AUC 136

FIGURE 16.2  Relationship between toxicity and CL 136
FIGURE 16.3  Relationship between toxicity and Cmax  137
FIGURE 17  Relationship between dose and mean area under the curve: protocol 921-1: single dose study  140
FIGURE 18  Relationship between dose and mean area under the curve; combined data for the 3 phase I study centres  141
FIGURE 19  Relationship between clearance and dose; combined data from 3 phase I study centres  142
FIGURE 20  The relationship between dose and pharmacokinetic parameters for individual patients  144
FIGURE 21  Baseline and post-treatment cardiac parameters following treatment with CI-921 - phase I clinical trial  160
FIGURE 22  Cardiac parameters of patient no. 005 over 6 cycles of CI-921  162
FIGURE 23  Baseline and post-treatment cardiac parameters following treatment with CI-921- phase II trial  165
FIGURE 24  Cardiac parameters of patient no. 025 over 5 cycles of CI-921  167
FIGURE 25  Standard curve for GSH assay  182
FIGURE 26  Whole blood GSH concentrations in patients receiving CI-921  192
FIGURE 27  Whole blood GSH concentrations in healthy controls  195
FIGURE 28  Mouse hepatic GSH following amsacrine  199
FIGURE 29  Mouse hepatic GSH following CI-921  200
FIGURE 30  Mouse hepatic GSH following BSO  202
FIGURE 31  Mouse hepatic GSH following morphine

FIGURE 32  Mean weight of surviving mice following treatment with CI-921 and amsacrine

FIGURE 33  Survival curves of mice following treatment with CI-921 at 40-60 mg/kg i.v.

FIGURE 34  Survival curves of mice following treatment with amsacrine at 30-60 mg/kg i.v.

FIGURE 35  Survival curves of mice pretreated with morphine, NAC and BSO compared to the survival of mice treated with CI-921 alone

FIGURE 36  Mean weight of surviving mice pretreated with BSO, morphine sulphate and N-acetyl cysteine prior to CI-921

FIGURE 37  Survival curves of mice following treatment with CI-921 and NAC compared to CI-921 alone
LIST OF TABLES

TABLE 1.1: LD_{50} values (mM) of CI-921 and related agents
TABLE 1.2: Activity against i.p. innoculated P388 leukaemia
TABLE 1.3: Activity against i.v. innoculated Lewis lung carcinoma
TABLE 1.4: Modified Fibonacci search scheme for dose escalation in a phase I study
TABLE 2.1: Dose escalation in the phase I trial
TABLE 2.2: Patient characteristics - Phase I clinical trial
TABLE 2.3: Frequency and grade of toxicity at dose levels ≥ 228 mg/m² (total dose) - Phase I
TABLE 2.4: Dose escalation methods used in each of the three trial centres for the phase I study of CI-921
TABLE 2.4.1: Trial centre 1: single dose repeated every 3 weeks
TABLE 2.4.2: Trial centre 2: weekly dose for 3 consecutive weeks repeated every 5 weeks
TABLE 2.4.3: Trial centre 3: daily dose for 3 consecutive days repeated every 3 weeks
TABLE 3.1: Patient characteristics - Phase II clinical trial in NSCLC
TABLE 3.2: Haematological toxicity - Phase II
TABLE 3.3: Non-haematological toxicity - Phase II
<p>| TABLE 4.1. | Quality control of CI-921 plasma assay - Phase I | 119 |
| TABLE 4.2 | Quality control of CI-921 plasma assay - Phase II | 120 |
| TABLE 4.3 | Mean model-independent pharmacokinetic parameters in the phase I trial | 122 |
| TABLE 4.4 | Mean ratios of day 3/day 1 pharmacokinetic parameters after the first and third infusion of a three-day course with increasing dose | 123 |
| TABLE 4.5 | Mean (SD) kinetic parameters at increasing i.v. dose | 124 |
| TABLE 4.6 | Relationship between dose (mg/m²) and pharmacokinetic parameters - Phase I | 125 |
| TABLE 4.7 | Relationship between toxicity (nadir absolute granulocyte count) and pharmacokinetic parameters - Phase I | 130 |
| TABLE 4.8 | Mean model-independent pharmacokinetic parameters in the phase II trial | 134 |
| TABLE 4.9 | Relationship between toxicity (nadir absolute granulocyte count) and pharmacokinetic parameters - Phase II | 135 |
| TABLE 4.10 | Mean terminal half-life following day 1 and day 3 infusions | 145 |
| TABLE 5.1 | Pre- and post-treatment cardiac parameters - Phase I | 159 |
| TABLE 5.2 | Pre- and post-treatment cardiac parameters - Phase II | 164 |
| TABLE 6.1 | GSH levels in whole blood of patients receiving CI-921 | 190 |
| TABLE 6.2 | GSH levels in whole blood of healthy controls | 193 |</p>
<table>
<thead>
<tr>
<th>Table 6.3</th>
<th>GSH levels in whole blood of mice following CI-921</th>
<th>196</th>
</tr>
</thead>
<tbody>
<tr>
<td>Table 6.4</td>
<td>Mouse hepatic GSH following amsacrine</td>
<td>197</td>
</tr>
<tr>
<td>Table 6.5</td>
<td>Mouse hepatic GSH following CI-921</td>
<td>198</td>
</tr>
<tr>
<td>Table 6.6</td>
<td>Mouse hepatic GSH following BSO</td>
<td>201</td>
</tr>
<tr>
<td>Table 6.7</td>
<td>Mouse hepatic GSH following morphine</td>
<td>203</td>
</tr>
<tr>
<td>Table 6.8</td>
<td>Pre-clinical toxicology studies</td>
<td>217</td>
</tr>
</tbody>
</table>
APPENDICES

APPENDIX 1.1  Performance status (ECOG)  229
APPENDIX 1.2  Consent form for phase I trial  230
APPENDIX 1.3  Toxicity grading  232
APPENDIX 1.4  Consent form for phase II trial  233
APPENDIX 1.5  Patient information sheet for phase II trial  234

APPENDIX 2.1  Assay validation. Back calculated concentrations of plasma standards  235
APPENDIX 2.2  Assay validation. Back calculated concentrations of seeded quality controls (SC) in plasma  237
APPENDIX 2.3  Assay validation. Back calculated concentrations of urine standards  238
APPENDIX 2.4  Assay validation. Back calculated concentrations of seeded quality controls (SCU) in urine  240

APPENDIX 3.1  Mean CI-921 pharmacokinetic parameters calculated from the first and third infusion after different doses in all patients in the phase I trial  241
APPENDIX 3.2  CI-921 pharmacokinetic parameters calculated from the first infusion (216 mg/m²) - phase II trial  243
APPENDIX 3.3  CI-921 pharmacokinetic parameters calculated from those patients in the phase I trial treated at 216 mg/m²  244

xvi
<table>
<thead>
<tr>
<th>Appendix</th>
<th>Description</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>4.1.1</td>
<td>Mouse hepatic GSH concentration following amsacrine (Assay 1)</td>
<td>245</td>
</tr>
<tr>
<td>4.1.2</td>
<td>Mouse hepatic GSH concentration following amsacrine (Assay 2)</td>
<td>246</td>
</tr>
<tr>
<td>4.2.1</td>
<td>Mouse hepatic GSH concentration following CI-921 (Assay 3)</td>
<td>247</td>
</tr>
<tr>
<td>4.2.2</td>
<td>Mouse hepatic GSH concentration following CI-921 (Assay 4)</td>
<td>248</td>
</tr>
<tr>
<td>4.3.1</td>
<td>Mouse hepatic GSH concentration following BSO (Assay 5)</td>
<td>249</td>
</tr>
<tr>
<td>4.3.2</td>
<td>Mouse hepatic GSH concentration following BSO (Assay 6)</td>
<td>250</td>
</tr>
<tr>
<td>4.4</td>
<td>Mouse hepatic GSH concentration following morphine (Assay 7)</td>
<td>251</td>
</tr>
</tbody>
</table>
CHAPTER 1

INTRODUCTION AND LITERATURE REVIEW

Preface 2

Amsacrine
- Clinical efficacy 7
- Clinical toxicity 9
- Metabolism 10
- Pharmacokinetics 13

Mechanism of action of amsacrine - topoisomerase II 14
- Amsacrine and topoisomerase II 14
- The function of topoisomerase in the cell 16
- The interaction between topoisomerase and anti-neoplastic agents 18

The cardiotoxicity of amsacrine 23

The development of CI-921 27
- QSAR in the design of CI-921
- Pre-clinical evaluation 33

Experimental tumour models and early clinical trials in the development of cytotoxic drugs 43

Non-small cell lung cancer and phase II trials 57

The relevance of pharmacokinetics in clinical trials 62

The metabolism of CI-921 - Glutathione 64
- The role of glutathione in the cell
- The interaction of GSH with anti-neoplastic therapy 67
- The effect of changing GSH concentrations on anti-neoplastic therapy 68
PREFACE

Historically, the discovery of anti-neoplastic agents has been largely serendipitous, with the chance finding of cytotoxic activity in a natural product, or drug originally developed for some other purpose. Recently, there has been a more logical approach to the development of new anti-cancer agents by the chemical manipulation of compounds already known to have some activity. By changing various parts of a molecule to alter such physico-chemical properties as lipid solubility, stability and DNA binding strength, the aim is to improve the anti-neoplastic activity of the compound whilst preserving those parts of the molecule already known to confer a desired property. This is known as the quantitative molecular-structure-activity-relationship (or QSAR) approach to drug design.

The anilino-acridines were selected as potential anti-tumour agents in the early 1970s and following extensive QSAR studies of this series, amsacrine was developed and entered into clinical trial. By the early 1980s, amsacrine had been shown to be effective against haematological malignancies (Arlin 1983) but had no significant activity against solid tissue tumours in man (Zittoun 1985). Using the QSAR approach, it was hoped to produce a drug with both a wider spectrum of action and physico-chemical properties more suited to solid tumour activity. To this end hundreds of analogues of amsacrine were produced and tested against in vivo and in vitro tumour screens (Denny et al 1984).
The murine leukaemias L1210 and P388 were retained as convenient initial screens of cytotoxic activity but were used in association with the Lewis lung tumour model in an attempt to select out those agents with greater solid tumour activity. The Lewis lung system is a particularly valuable in vivo tumour model. It is one of the few murine tumour lines in the National Cancer Institute's tumour panel which differentiates those agents known to be active against solid tumours (for example cyclophosphamide, doxorubicin, 5-fluorouracil and methotrexate) and those known to be active against haematological malignancies (e.g. daunorubicin, cytosine arabinoside and L-asparaginase) (Goldin et al 1981). The agent showing the most cytotoxic activity against the Lewis lung tumour model whilst retaining its anti-leukaemic activity was the 4-methyl, 5-methyl carboxy derivative of amsacrine, known as CI-921 or "AMSALOG" (Baguley 1984) (Fig.1).

CI-921 is being developed by the Parke-Davis Pharmaceutical Research Division, Warner-Lambert Company. Three phase I and limited phase II studies of CI-921 were undertaken in Puerto Rico, Ohio and Auckland, each centre giving the drug according to a different schedule. The phase I and II clinical and pharmacokinetic studies of CI-921 completed in Auckland form the basis of this thesis.
Empirical formula: $\text{C}_{26}\text{H}_{30}\text{N}_4\text{S}_2\text{O}_8$

Molecular weight: 590.5 (monohydrate, 608.7)

Chemical name: 9-[[2-methoxy-4-[(methyl-sulfonyl) amino]-phenyl]amino]-N, 5-dimethyl-4-acridine-carboxamide, 2-hydroxyethane sulfonate (1:1)

Common name: "AMSALOG"

Other designations: CI-921
NSC 343,499.
FIG. 2.  

**Amsacrine**

![Chemical Structure of Amsacrine](image)

- **Empirical formula:** $C_{21}H_{19}N_3O_3S$
- **Molecular weight:** 393.5
- **Chemical name:** $N\{4\text{-}(9\text{-acridinylamino})\text{-}3\text{-methoxyphenyl}\}\text{-methanesulfonamide.}$
- **Common name:** m-AMSA, acridinyl anisidide.
- **Trade name:** Amsidine (Europe)
  Amsidyl (USA)
- **Other designations:** CI-880
  NSC-249992.