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

Janet Rea Hardy

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/m^2 to 810mg/m^2 (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/m^2.

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/m^2 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

ACKNOWLEDGEMENTS

This work was supported in part by grants from the Auckland division of the Cancer Society of N.Z., the Medical Research Council of N.Z., and the Park-Davis Pharmaceutical Division, Warner-Lambert company, Ann Arbor, Michigan, U.S.A. (who also kindly supplied the CI-921 for use in these trials).

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.

Christine Thoreau provided expert assistance in the animal studies and Dr Margaret Sage kindly prepared and reported on the histological sections.

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 sulphoximine
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
LD_{10}	lethal dose in 10% of treated mice
LD_{50}	lethal dose in 50% of treated mice
LD_{90}	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/m^2	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

QSAR	quantitative-molecular-structure-activity-relationship
QTc	corrected Q-T interval of electrocardiogram
r	Pearson's correlation coefficient
s.c.	subcutaneous
SC	seeded quality control (plasma)
SCU	seeded quality control (urine)
STI	systolic time interval
TEAP	triethylamine phosphate
$t_{1/2}$å	distribution half-life
$t_{1/2}$ß	terminal half-life
UE	urinary excretion
Tt	last time point
Vss	volume of distribution

LIST OF FIGURES

LIST OF TABLES

APPENDICES

CHAPTER 1

INTRODUCTION AND LITERATURE REVIEW

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.

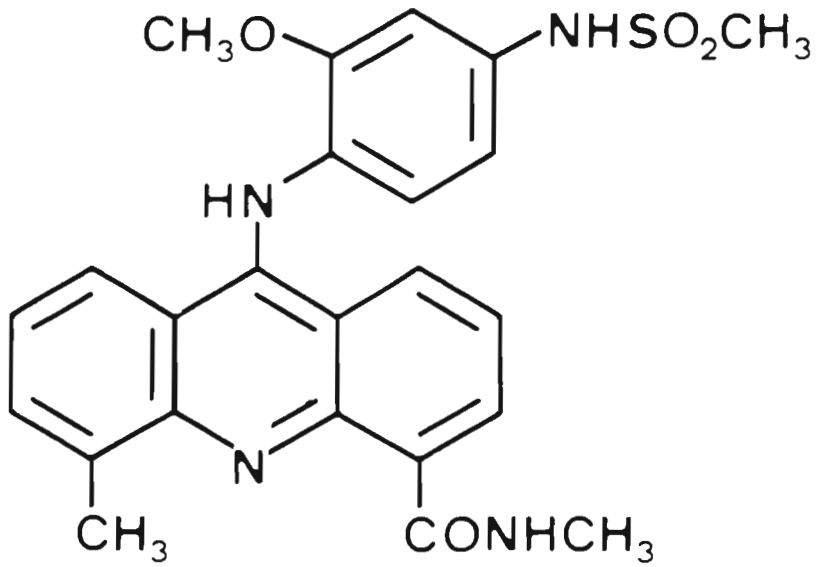
The anilino-acridines were selected as potential anti-tumour agents in the early 1970s and following extensive QSAR studies of this series, amsacrine was developed and entered into clinical trial. By the early 1980s, amsacrine had been shown to be effective against haematological malignancies (Arlin 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 <u>in vivo</u> and <u>in vitro</u> tumour screens (Denny <u>et al</u> 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.

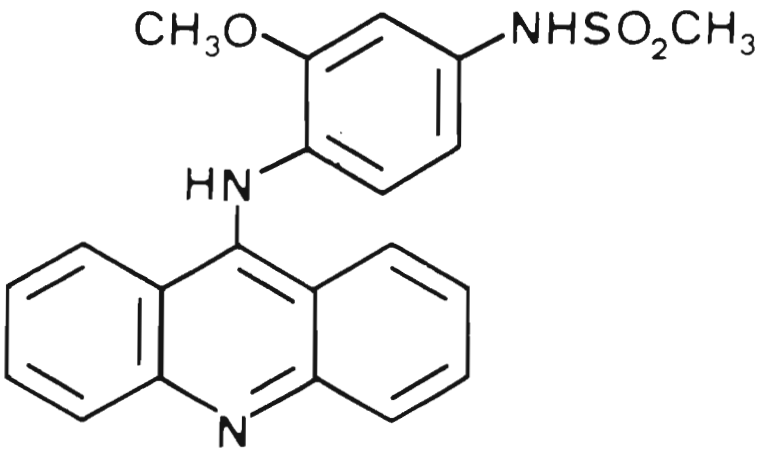
FIG. 1.

CI-921



Empirical formula:	$C_{26}H_{30}N_4S_2O_8$
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**



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