

**TOWARDS SUSTAINABLE HARVESTING OF SENECA SNAKEROOT
(*Polygala senega* L.) ON MANITOBA HYDRO RIGHTS-OF-WAY**

BY

CANDACE L. TURCOTTE

A Thesis

**Submitted to the Faculty of Graduate Studies
in Partial Fulfillment of the Requirements
for the Degree of**

MASTER OF SCIENCE

**Department of Botany
University of Manitoba
Winnipeg, Manitoba**

© April, 1997



**National Library
of Canada**

**Acquisitions and
Bibliographic Services**

**395 Wellington Street
Ottawa ON K1A 0N4
Canada**

**Bibliothèque nationale
du Canada**

**Acquisitions et
services bibliographiques**

**395, rue Wellington
Ottawa ON K1A 0N4
Canada**

Your file Votre référence

Our file Notre référence

The author has granted a non-exclusive licence allowing the National Library of Canada to reproduce, loan, distribute or sell copies of this thesis in microform, paper or electronic formats.

The author retains ownership of the copyright in this thesis. Neither the thesis nor substantial extracts from it may be printed or otherwise reproduced without the author's permission.

L'auteur a accordé une licence non exclusive permettant à la Bibliothèque nationale du Canada de reproduire, prêter, distribuer ou vendre des copies de cette thèse sous la forme de microfiche/film, de reproduction sur papier ou sur format électronique.

L'auteur conserve la propriété du droit d'auteur qui protège cette thèse. Ni la thèse ni des extraits substantiels de celle-ci ne doivent être imprimés ou autrement reproduits sans son autorisation.

0-612-23536-X

THE UNIVERSITY OF MANITOBA
FACULTY OF GRADUATE STUDIES

COPYRIGHT PERMISSION PAGE

TOWARDS SUSTAINABLE HARVESTING OF SENECA SHAKEROOT
(Polygala senega L.) ON MANITOBA HYDRO RIGHTS-OF-WAY

BY

CANDACE L. TURCOTTE

A Thesis/Practicum submitted to the Faculty of Graduate Studies of The University
of Manitoba in partial fulfillment of the requirements of the degree
of
MASTER OF SCIENCE

Candace L. Turcotte 1997 (c)

Permission has been granted to the Library of The University of Manitoba to lend or sell copies of this thesis/practicum, to the National Library of Canada to microfilm this thesis and to lend or sell copies of the film, and to Dissertations Abstracts International to publish an abstract of this thesis/practicum.

The author reserves other publication rights, and neither this thesis/practicum nor extensive extracts from it may be printed or otherwise reproduced without the author's written permission.

TABLE OF CONTENTS

TABLE OF CONTENTS.....	i
ABSTRACT	v
ACKNOWLEDGEMENTS.....	vii
LIST OF FIGURES	ix
LIST OF TABLES	x
CHAPTER 1 INTRODUCTION AND OBJECTIVES.....	1
CHAPTER 2 BIOLOGY, ECOLOGY AND ETHNOBOTANY OF SENECA	
SNAKEROOT (<i>Polygala senega</i> L.).....	3
2.1 Introduction	3
2.2 Taxonomy, morphology and botanical relationships.....	3
2.3 Geographical distribution.....	5
2.4 Habitat description.....	8
2.5 Reproductive mechanisms.....	8
2.6 Seed production.....	9
2.7 Seed dispersal.....	11
2.7.1 Pre-dispersal.....	11
2.7.2 Seed rain and seed dispersal	11
2.7.3 Seed dispersal by animals.....	12
2.7.4 Seed dispersal by ants	12
2.7.5 Post-dispersal.....	14
2.8 The seed bank.....	14
2.9 Seed dormancy and germination.....	15
2.10 Cultivation	20
2.11 Biochemistry.....	20
2.12 Ethnobotany	23
2.12.1 Native uses	23

3.4.1 Nutrient experiment.....	68
3.4.2 Competition experiment.....	68
3.4.3 Sowing of seed under field conditions	69
3.5 Data analysis	69
3.5.1 Competition and nutrient experiments.....	69
3.5.2 Multivariate analysis of seneca habitats.....	69
CHAPTER 4 RESULTS AND DISCUSSION.....	70
4.1 Ecology of seneca snakeroot.....	70
4.1.1 Habitat preferences.....	70
4.1.2 Soil relations.....	71
4.1.3 Associated vegetation.....	71
4.2 Population biology of seneca snakeroot	75
4.2.1 Phenology.....	75
4.2.2 Population density	77
4.2.3 Demography - size and spatial structure.....	77
4.2.4 Biomass allocation	81
4.2.5 Flowering and seed production.....	81
4.3 Harvesting and mortality in wild populations	86
4.4 Nutrient experiment	86
4.5 Competition experiment.....	88
4.6 Seed germination - field experiment.....	88
4.7 Soil seed bank.....	88
4.8 Laboratory seed germination trials.....	92
4.8.1 Germination in petri dishes	92
4.8.2 Germination in soil.....	96
4.9 Vegetative propagation.....	97
4.9.1 Root cuttings.....	97

4.9.2 Shoot cuttings	97
4.9.3 Whole-plant division.....	99
CHAPTER 5 SUMMARY, SUSTAINABILITY AND RECOMMENDATIONS.....	101
5.1 Summary	101
5.2 Sustainability of seneca snakeroot populations	102
5.3 Recommendations	104
REFERENCES.....	106
Appendix I Phytochemical constituents of <i>Polygala senega</i> L. (from Duke 1992).....	114
Appendix II Mean cover values per species per transect.....	115
Appendix III Two year phenological profile of seneca snakeroot.....	123
Appendix IV Spatial patterns of seneca snakeroot.....	127

ABSTRACT

The root of seneca snakeroot (*Polygala senega* L.), a perennial herb native to North America, has been used medicinally by Europeans for over 300 years. Seneca snakeroot is valued, particularly for its expectorant properties. Lately, the demand for wild seneca root has increased due to an escalation of interest in herbal medicine. Increased harvesting pressure has raised concerns as to the sustainability of native seneca snakeroot populations in Manitoba. This study investigated the biology, ecology, ethnobotany and economics of seneca snakeroot in order to address the question of the sustainability of seneca snakeroot populations.

Ten study sites were sampled along Manitoba Hydro rights-of-way in the northern Interlake and Grand Rapids region to obtain baseline information on the population biology and ecology of seneca snakeroot. The baseline information revealed that seneca snakeroot populations in the Northern Interlake region seem to be successfully regenerating under current harvesting pressures. Investigations into the economics of seneca snakeroot revealed that an entire network of people is involved in the seneca industry including; diggers, exporters, brokers, pharmaceutical companies and consumers. Results from the germination and propagation experiments indicate that seneca snakeroot can be propagated either vegetatively or by seed. Two seed germination and two vegetative propagation methods proved successful.

The information gained from the study lead to four recommendations concerning the sustainability of seneca snakeroot. The cultivation of seneca snakeroot should be promoted in order to relieve harvesting pressure on existing wild populations. Harvesting and marketing strategies should be developed in Manitoba for the direct benefit of local harvesters and exporters. The rights-of-way in the Interlake region should be maintained

(using mechanical vegetation removal) as they provide excellent habitats for snakeroot populations. Experiments and long-term monitoring of seneca populations could be continued utilizing the semi-permanent plots of this study.

ACKNOWLEDGEMENTS

Funding for this project was in the form of a grant from the Manitoba Hydro Research and Development Committee. Thanks to Roy Bukowsky and Bob Mann of Manitoba Hydro for their assistance over the duration of the project. The views expressed in this thesis are those of the author, and do not necessarily reflect the policies and/or views of Manitoba Hydro.

Thank you to the people of Grand Rapids for their cooperation and advice on the project. I am particularly grateful to Albert Campbell, Henry Chartier and Clarence Campo. The cooperation of three seneca snakeroot brokers, Dave Buck, Doug Elsasser and Ken Hooper, is also greatly appreciated.

I would like to thank my thesis committee members (Dr. Briggs (Pharmacy), Dr. Stewart (Botany), and Dr. Staniforth, (Biology, U. of W.)) for their help and advice. I appreciate the advice and assistance I received from the Botany department, in particular, Keith Travis, Margaret Smith and Dr. Punter. Mark Elliot (Botany Greenhouse) provided valuable assistance especially in the seed and vegetative propagation of seneca snakeroot. Dr. I. Waters offered advice on germination using gibberellic acid, while Dr. B. Ford and G. Keleher helped in the identification of plants. W. Fraser provided soil maps and site information. Thanks to Drs. T. and J. Shay for their advice and encouragement.

Valuable field assistance was provided by Philip Northover, Kristin Olson, Anne Moulton, Andrew Park, Kerri Cook, Kelly Graham, Jen Barker, Derek Ebner, Rod Lastra and Heidi Wiebe. Special thanks to my husband Ron, who was conned many times into making the long trip to the study sites. Funding for field assistance was supplemented by a

Natural Sciences and Engineering Research Council of Canada individual operating grant to Dr. N. Kenkel, and CareerStart Manitoba.

Dave Walker and Michael Shaw provided valuable computer assistance, particularly in the preparation of figures and slides.

I would like to thank my parents, Jack and Karen Jorgenson, for their support and encouragement, and my brother Jason for occasionally watering my plants and taking many of the photographs.

Finally, I would like to extend my deepest gratitude to Professor N. C. Kenkel (my advisor) for introducing me to the project and for helping me (for the past three years) to see it through to its completion.

LIST OF FIGURES

Figure 2.1 (1). Side view of <i>Polygala senega</i> L. Photo taken in mid June 1994 (with dollar coin for size comparison) (2). Top view of <i>Polygala senega</i> L. Photo taken in mid June 1994 (with dollar coin for size comparison).	4
Figure 2.2 Distribution of seneca snakeroot in North America (adapted from Catling and Small 1994).	6
Figure 2.3 Distribution of <i>Polygala senega</i> L. in Manitoba.....	7
Figure 2.4 Seneca snakeroot seeds. Each seed is <i>ca.</i> 2.5 mm in length. Note bilobed elaiosome	10
Figure 2.5 Senegin saponins present in <i>Polygala senega</i> L. (adapted from Shibata 1976).	22
Figure 2.6 Native groups in Manitoba who harvested seneca snakeroot in the 1950's (adapted from Weir 1960).	33
Figure 3.1 Map of Manitoba showing study area.	46
Figure 3.2 Mean temperature and total precipitation data for the Grand Rapids area, 1966-1990 (Environment Canada 1995).	49
Figure 3.3 Mean temperature and total precipitation data for the Grand Rapids area, 1994 (Environment Canada 1995).	50
Figure 3.4 Mean temperature and total precipitation data for the Grand Rapids area, 1995 (Environment Canada 1995).	51
Figure 3.5 Map of study area showing site locations.	53
Figure 3.6 Scarified seeds of seneca snakeroot.	63
Figure 3.7 (1) Cold-stratification trays of seneca snakeroot seeds. The seeds are layered between moist sand and placed in a cold room (2°C) for two to six months. (2) Diagrammatic illustration of seneca snakeroot seed stratification.	65
Figure 4.1 Ordination biplot of species and sites with characteristic species labelled.	74
Figure 4.2 Phenological profile of <i>Polygala senega</i> L. (1994, 1995 pers. obs.).	76
Figure 4.3 Size-frequency histograms for the 10 study sites.	79
Figure 4.4 Log-log plot of root vs. shoot biomass in grams (n=56). Line indicates a one-to-one relationship between root and shoot.	84

LIST OF TABLES

Table 2.1 Results of germination experiments by Merilee Teresa and Avita (1989) on <i>Polygala chinensis</i> L.	18
Table 2.2 Use of seneca snakeroot by native group (from Arnason <i>et al.</i> 1981, Densmore 1913, 1928, Johnston 1970, Kindscher 1992, Moerman 1986, Weiner 1980, Zieba 1990).	25
Table 2.3 Canadian export of seneca snakeroot (adapted from Hlady and Poston 1959).	32
Table 2.4 Herbal products manufactured abroad utilizing seneca snakeroot (Reynolds 1993).	36
Table 2.5 Products manufactured in Canada which contain seneca snakeroot (compiled from the Compendium of Non-Prescription Products, Canadian Pharmaceutical Association 1995).	39
Table 3.1 Description of techniques used in seed germination trials.	61
Table 3.2 Summary of vegetative propagation experiments.	66
Table 4.1 Results of soil analysis.	72
Table 4.2 Species commonly associated with seneca snakeroot.	73
Table 4.3 Population density and mean number of shoots of seneca snakeroot at various sites.	78
Table 4.4 Number of shoots per marked seneca plants in 1994 and 1995. A dash (-) refers to a plant that was missing due to harvesting, natural mortality or road construction.	82
Table 4.5 Number of seeds in a random sample of 50 seneca snakeroot shoots.	85
Table 4.6 Loss of seneca plants from plots due to harvesting, construction or mortality.	87
Table 4.7 (1) Species that germinated from seed bank soil collected in mid-summer 1994. (2) Species that germinated from seed bank soil collected in mid-summer 1995. (3) Species that germinated from seed bank cold-treated soil collected in mid-summer 1995	89
Table 4.8 Seed germination trials. In all cases petri dishes were used along with filter paper dampened with water or gibberellic acid (GA), if used. W.S. refers to window sill, G.H. refers to greenhouse and G.C. refers to growth chamber. All seeds were pre-soaked in water for 24 hours prior to scarification and/or GA treatments except trials in 1994. Germination results with a question mark (?) were only qualitatively noted.	93
Table 4.9 Propagation experiments involving root and shoot cuttings and whole plant division.	98

CHAPTER 1

INTRODUCTION AND OBJECTIVES

The root of seneca snakeroot (*Polygala senega* L.) has been used for centuries by aboriginal peoples in North America as a treatment for various ailments. After its introduction into European medicine during the early 1700's, seneca became a highly sought after remedy for the treatment of respiratory problems. Presently, Manitoba provides the vast majority of the global supply of wild seneca root. The recent resurgence of interest in natural remedies has greatly increased the demand for seneca root, raising concerns as to whether natural populations in Manitoba could be over-harvested as occurred in eastern North America in the last century. In addressing the question of the sustainability of seneca snakeroot populations in Manitoba, aspects of biology, ecology and economics of seneca snakeroot need to be considered. This leads to the four main objectives of this study:

- to summarize the biology, ecology, economics and history of use of seneca snakeroot.

A complete review of the use of seneca snakeroot by aboriginal North Americans, as well as its historical and current use in European and Asian medicine, was undertaken. Research on the pharmaceutical and biochemical aspects of seneca snakeroot was summarized. The ecology and population biology of seneca snakeroot, and available information on seed germination, propagation and cultivation of the species, was considered. Economic aspects related to the harvesting, export and marketing of wild seneca snakeroot were summarized.

- to collect baseline information on seneca snakeroot populations in Manitoba.

Baseline information on seneca snakeroot was obtained from ten populations located along Hydro rights-of-way in the Manitoba's northern Interlake (Grand Rapids) region. Information was collected on habitat preferences of seneca snakeroot, including soil type and nutrient status, drainage, degree of shading, fire and disturbance history, and associated vegetation. Demographic (size-structure) and phenological (life-history) profiles for the species were also obtained. Individual plants were marked to determine rates of growth, or harvested to determine biomass allocation to above-ground and below-ground plant parts. Seeds were collected from field plants for use in germination experiments.

- to determine the current and potential economic benefits of seneca snakeroot harvesting.

Available literature on the harvesting and exporting of seneca snakeroot in Manitoba was summarized. Individuals involved in the industry, including seneca root diggers, exporters, brokers, pharmaceutical companies, and retailers were contacted. An attempt was made to follow the 'route of the roots' from digging to retailing.

- to conduct field experiments and propagation studies on seneca snakeroot.

Soil was collected to examine the seed bank in areas where seneca snakeroot occurs naturally. Seeds were sown into prepared field plots to determine the amount of germination under natural conditions. Field experiments were conducted to determine whether the addition of nutrients, or the removal of competing vegetation, would result in increased growth of seneca root. Laboratory seed germination experiments were conducted to determine the conditions required to break seed dormancy. Vegetative propagation experiments (including root and shoot cuttings, and whole-plant division) were also undertaken. Recommendations are made as to the most efficient way to propagate seneca, and whether currently harvested populations are sustainable.

CHAPTER 2

BIOLOGY, ECOLOGY AND ETHNOBOTANY OF SENECA SNAKEROOT (*Polygala senega* L.)

2.1 Introduction

This chapter provides a literature review of the biology (taxonomy, habitat, distribution, reproduction), ecology (ant - plant interactions), and ethnobotany (history of use) of seneca snakeroot. Aspects of the biochemistry and economics of seneca are also included. Concepts of plant population biology are integrated when relevant.

2.2 Taxonomy, morphology and botanical relationships

Seneca snakeroot (*Polygala senega* L.) is a member of the Polygalaceae (milkwort) family. This family contains seventeen genera and over one thousand species. The genus *Polygala* L. is the largest, containing about 400-450 species (Eriksen 1993). Eight species are native to Canada (Gillett 1968). *Polygala* means 'much milk' in reference to the milky secretions produced by many members of the genus (Grieve 1967).

Polygala senega (pictured in Fig. 2.1) is an erect, low-growing (10-30 cm high) perennial herb, which each spring produces a circular spray of vertically-oriented shoots from a single knotty root crown (Catling and Small 1994). The aromatic root is woody and twisted, and has numerous lateral branches (Gillett 1968). Each shoot consists of a large number of alternate, lance-shaped leaves. Most leaves are pale green below and dark green above. The lowest leaves are reduced or scale-like and purplish in colour (Great Plains Flora Association 1986; Gillett 1968). The inflorescence is a dense, terminal, spike-

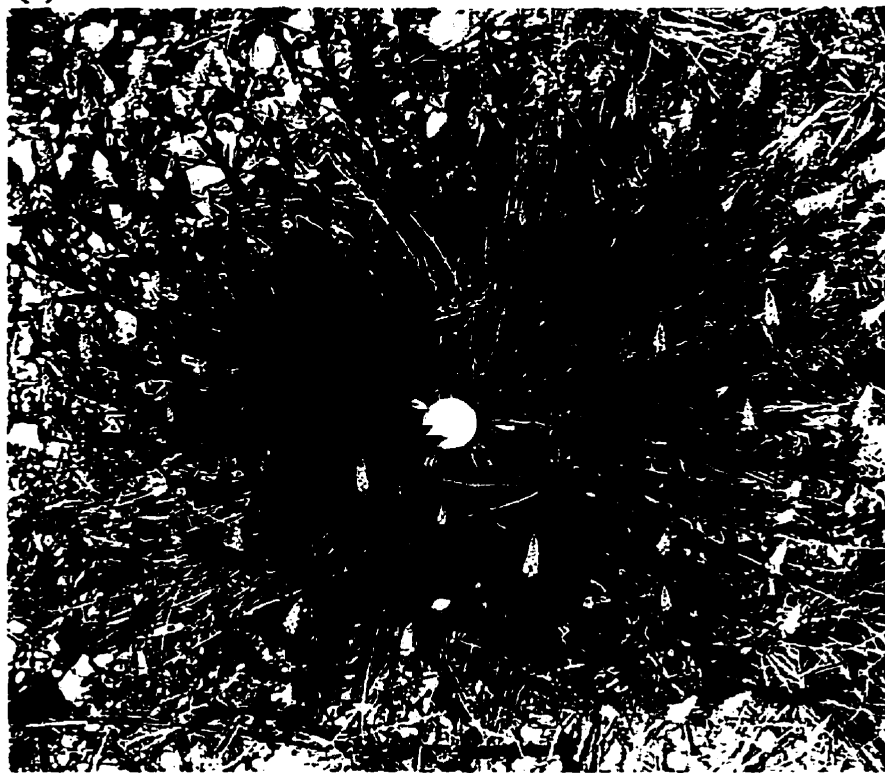
Figure 2.1(1). Side view of *Polygala senega* L. Photo taken in mid June 1994 (with dollar coin for size comparison).

(2). Top view of *Polygala senega* L. Photo taken in mid June 1994 (with dollar coin for size comparison).

(1)



(2)

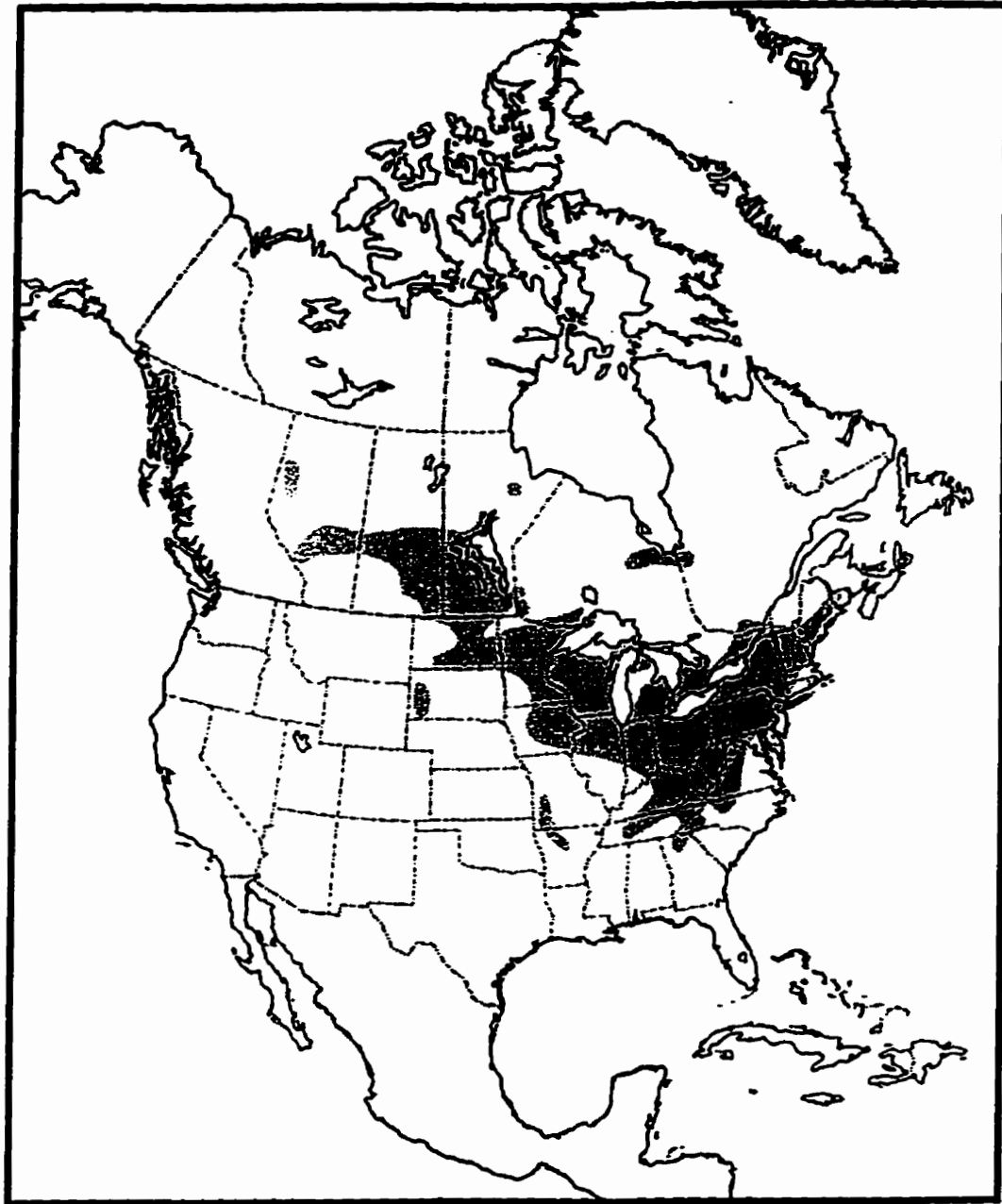


like panicle which tapers at the apex (Gillett 1968). The flowers are initially greenish-white, but turn pinkish-white as the corolla ages (Miller 1971). The calyx is composed of five petaloid sepals (Gillett 1968), while the corolla consists of three hypogynous petals united into a tube (Gillett 1968; Great Plains Flora Association 1986). The seeds are black, sparsely white-pubescent, reticulate and arillode (Great Plains Flora Association 1986; Montgomery 1977).

Seneca snakeroot is also known as 'senega snakeroot', 'seneca (senega) root', 'black snakeroot', or simply 'snakeroot'. Unfortunately, the name 'snakeroot' has been applied to a number of other medicinal plants, including *Aristolochia serpentaria*, *Actaea pachypoda*, *Asarum canadense*, *Cimicifuga racemosa*, *Eupatorium rugosum*, *Rauvolfia serpentina*, *Sanicula europaea* and *Senecio aureus* (Thomson 1978; Tyler 1993). It is therefore important to include the modifying adjective 'seneca' when referring to *P. senega*. The Plains Cree refer to the species as 'wisak', while the Swampy Cree use the name 'wincekes' (Zieba 1990).

2.3 Geographical distribution

Seneca snakeroot is native to North America (Gillett 1968). In Canada, the species is particularly common in Manitoba and Saskatchewan. It is also found in Alberta, Ontario (primarily south of the Canadian Shield), south-western Québec, and in the St. John River valley of western New Brunswick. In the United States, it is found from North Dakota and Maine in the north, to Georgia and Tennessee in the south. The distribution of seneca snakeroot in North America is illustrated in Fig. 2.2 (adapted from Catling and Small 1994) and its distribution in Manitoba is shown in Fig. 2.3 (based on information from herbarium collections [U.S. National Museum, National Herbarium of Canada, Institut de recherche en biologie végétale, Ottawa Department of Agriculture, University of Manitoba



**Figure 2.2. Distribution of seneca snakeroot in North America
(adapted from Catling and Small 1994).**

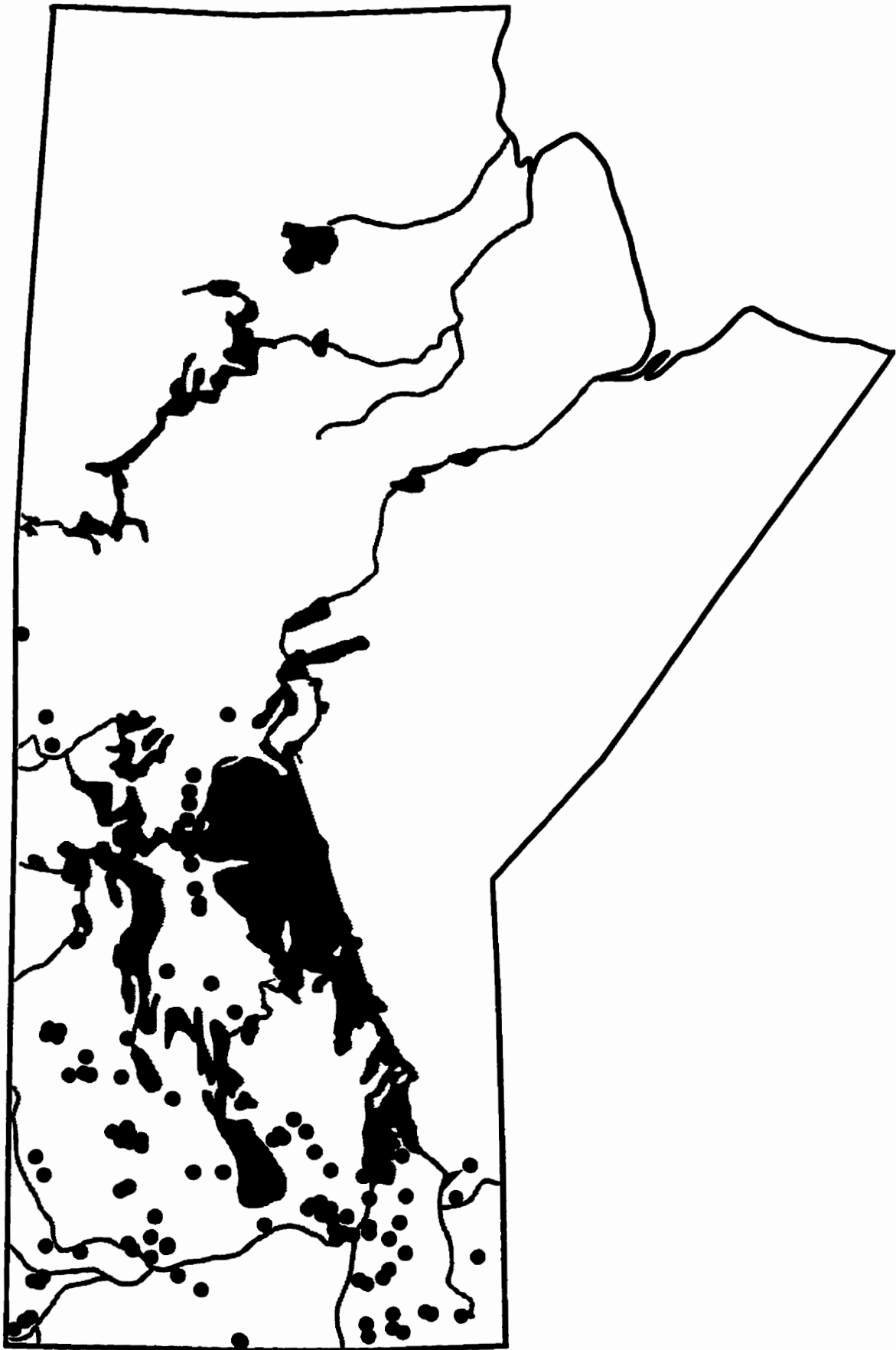


Figure 2.3. Distribution of *Polygala senega* L. in Manitoba.

Herbarium] and personal observations). The species is widely distributed in southern and west-central Manitoba. It occurs at least as far north as Flin Flon and Grand Rapids. According to Hlady and Poston (1959), the species has been collected as far north as Gillam and York Factory (although no herbarium specimens were found to support this claim). Scoggan (1957) reports the species from near Gillam, and along the western shore of James Bay.

In the Canadian prairies, seneca snakeroot was probably once more widely distributed than it is today. Many native elders describe collecting seneca root from areas that are now under intensive cultivation (Hlady and Poston 1959). Agricultural practices have greatly reduced the land available for wild seneca growth. Trottier (1974) suggests that Riding Mountain National Park is one area that seneca root may be protected, as harvesting is not permitted in the park.

2.4 Habitat description

Seneca snakeroot generally occurs in open to partially shaded habitats such as prairies, open woods and roadsides. It prefers limestone-based (calcareous) soils (Catling and Small 1994; Kindsher 1992; personal observations 1994). Seneca snakeroot is particularly abundant in the Interlake region of Manitoba, since soils in this region are calcareous and open habitats (such as roadsides, recently burned areas, and rights-of-way) are common.

2.5 Reproductive mechanisms

Literature pertaining to the reproductive mechanisms of *Polygala senega* L. is scant but some information does exist on the pollination mechanisms for related species and genera. Autogamy (self-fertilization) appears to occur in some species of *Polygala*, and in two other

genera in the family Polygalaceae (*Salomonina* Lour. and *Muraltia* D.C.) (Miller 1971). Research on four Indian species of *Polygala* by Venkatesh (1956) revealed that in these species the style is curved bringing the stigma in close proximity to the pollen, thus facilitating self-pollination. In another species, *P. lutea*, self-pollination appears to occur since the stigma terminates in a tuft of hairs which catches pollen from the closely arranged anthers (Miller 1971). In many annuals (which have short life cycles and high mortality rates) self-pollination regularly occurs to ensure the production of a large amount of seeds (Grime 1979). *P. senega* produces numerous seeds and possesses a brush-like style (Gillett 1968). This may be interpreted as a safety mechanism to ensure seed production by selfing if cross-pollination fails to occur. Bee pollination is suggested to be suited to the large, showy flowers of *P. paucifolia* and the related *P. chamaebuxus* (Miller 1971). Both bees and ants have been observed on *Polygala senega* plants (pers.obs. 1995).

2.6 Seed production

Seed production refers to the probability of a plant producing seeds and the number of viable seeds produced by a plant. In some species, such as *Polygala senega*, an estimate of seed production can be derived from the number of mature capsules multiplied by the mean number of seeds per capsule. Viability of seeds may be determined by the combination of hand sorting (to remove undeveloped or decaying seeds), germination experiments and/or chemical tests (Moore and Chapman 1986).

Each seneca snakeroot shoot produces a dense, many-flowered inflorescence. Each flower produces a capsule containing two seeds. The seeds (pictured in Fig. 2.4), which take about 35 days to ripen (Hlady and Poston 1959), are small (ca. 2.8 x 1.3 x 1.3 mm) and black, obliquely obovate and reticulate (Montgomery 1977). The seed surface is sparsely white-pubescent. A bi-lobed appendage (elaiosome or aril) is present (Gillett 1968).

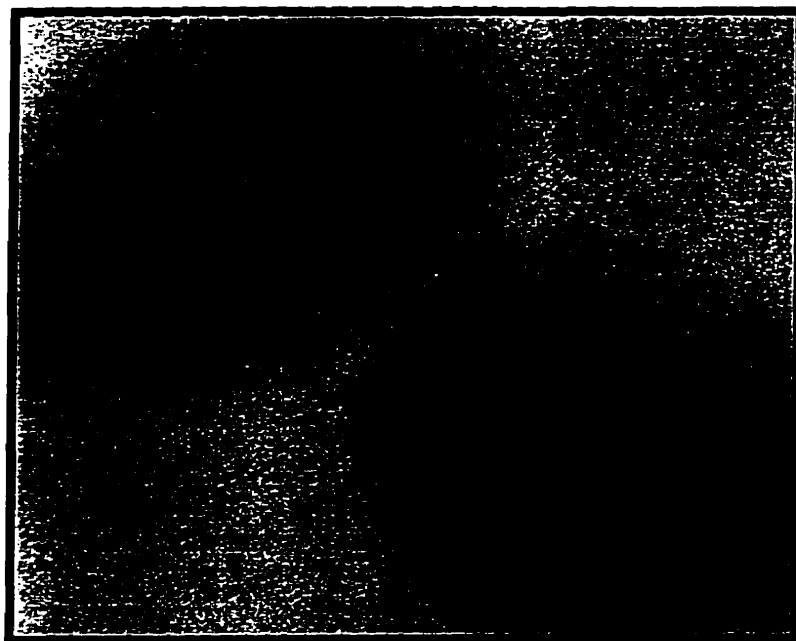


Figure 2.4. Seneca snakeroot seeds. Each seed is ca. 2.5 mm in length. Note bi-lobed elaiosome.

2.7 Seed dispersal

2.7.1 Pre-dispersal

Soon after fertilization (and before dispersal), seeds become potential targets for predators and pathogens. Depredation and pathogen attacks are parameters of seed population dynamics which could adversely affect population numbers (Moore and Chapman 1986).

2.7.2 Seed rain and seed dispersal

If seeds escape depredation and pathogen attack in pre-dispersal, they will contribute to the seed rain. Seeds are generally initially deposited close to the parent plant (with the exception of wind or water dispersed seeds). In most cases, with an increase in distance from the parent plant there is a decrease in the density of seeds deposited (Moore and Chapman 1986).

Seed dispersal is important in population biology since: 1) seeds entering an area would increase population size, whereas dispersal out of the area may result in a decrease in population size. 2) Seed dispersal over a considerable distance could result in the establishment of a new population, which under favorable conditions, could grow to a substantial size over time (Silvertown 1987).

Seed dispersal may occur through the actions of animals, wind or water, or through the actions of the plant itself (Silvertown 1987). Autochory (self-dispersal) may occur by ballochory or creeping diaspores (van der Pijl 1972). Seed dispersal may be a two phase

process: (1) primary dispersal from the inflorescence to the soil, possibly aided by an animal or the wind; (2) secondary dispersal when water or animals move the seed further along the soil surface (Moore and Chapman 1986).

2.7.3 Seed dispersal by animals

Dispersal distance, location and the deposition pattern of seeds can be determined by the behavior of dispersal agents such as bats, birds, mammals, insects, earthworms and even fish. Dispersal by animals is extremely common in temperate woodland herbs and shrubs. Seeds enclosed in attractive fleshy fruits (berries for example) are typically dispersed by animals. Alternatively, fruits may have spines which attach to animal coats and are carried away. Fleshy fruits and spines are considered as examples of the coevolution between plant and animal species (Silvertown 1987; Begon *et al.* 1990).

Seed dispersal by animals increases the distance between offspring (seed) and parent, but not necessarily between the offspring themselves (seeds could be concentrated [clumped] in one area). The end result of animal seed dispersal, in terms of plant survival, depends upon whether or not the seed is damaged by the disperser (Silvertown 1987).

2.7.4 Seed dispersal by ants

Some plants generate seeds or fruits with external structures (elaiosomes) that are attractive or useful to ants. A plant which produces diaspores that are attractive to ants is termed a myrmecochore. As ants collect the diaspores, they act indirectly as dispersal agents (Berg 1975). In general, the process of myrmecochory involves forager ants which carry seeds back to the ant nest where the elaiosome is removed (often fed to larvae or adult worker ants) and the undamaged seed is discarded (Beattie 1985; Holldobler and Wilson

1990). Myrmecochorous plants are found nearly all over the world, and in as many as sixty-seven different plant families (Beattie 1983). Myrmecochory is especially common in early-flowering north-temperate herbs, some Australian and south African perennials, and in a diverse group of tropical species (Holldobler and Wilson 1990).

The attractive external structures on the seeds or fruits are collectively termed elaiosomes when referring to ants (arils if discussing birds). Elaiosomes contain lipids, proteins, vitamins, sugars and starch which provide a conveniently-packaged energy source to the ants which feed on them (Holldobler and Wilson 1990; Hughes *et al.* 1994). Elaiosomes, which vary in size, shape and colour, are derived from various tissues such as the raphe, pericarp, and receptacle (Holldobler and Wilson 1990). Like many members of the Polygalaceae, elaiosomes are present in the genus *Polygala*. In literature pertaining to *Polygala*, these outgrowths at the micropylar end of the ovule have also been referred to as arils, arillodes, caruncles, or strophioles (Catling and Small 1994; Merlee Teresa and Avita 1989; Verkerke and Bouman 1980). The elaiosomes in *Polygala* are separated from the seed by a thick-walled structure, which may have evolved to ensure that only the elaiosome is consumed and the seed itself is rejected (Verkerke 1985). Seed hairs present on *Polygala* species probably aid the ants in carrying the seed (Oostermeijer 1989). Like most ant-dispersed (myrmecochorous) plants, the seeds of *Polygala* species are released from their capsules after they have fallen to the ground, resulting in a concentration of seeds near the parent plant (Oostermeijer 1989).

Myrmecochory is considered to be a mutualistic interaction between plants and ants, since the ants receive a food source while the plant's seeds are dispersed. Five adaptive advantages of myrmecochory (to the plant) identified by Beattie (1985) are: (1) interspecific competition avoidance, (2) fire avoidance, (3) additional nutrients in microsites (ant nests), (4) parental competition avoidance (dispersal for distance), and (5) predator avoidance.

Violet species (e.g. *Viola papilionacea*, *V. nuttallii*) benefit from myrmecochory, as ant dispersal relocates a large portion of seeds that would otherwise be vulnerable to bird and rodent attacks (Silvertown 1987). Myrmecochory is advantageous for *Carex pedunculata*, as greenhouse experiments show that seedling growth is greatly improved when the seeds are removed from the parent plant and away from similarly-aged seedlings (Handel 1976). The myrmecochore *Corydalis aurea* benefits from directed dispersal as its seeds are deposited in sites favorable for survival and growth (Hanzawa *et al.* 1988).

2.7.5 Post-dispersal

The fate of seeds after dispersal could be: (1) to remain where they land, (2) to move along the soil, (3) to become buried, (4) to be consumed by a predator, (5) to be picked up and moved by dispersal agents, (6) to be killed, (7) to die or (8) to germinate (Moore and Chapman, 1986). A seed may experience more than one of these factors with the exception of predation, death and germination which are end-points of the seed phase.

2.8 The seed bank

A seed is a dormant or resting stage of a plant's life. The seed bank is defined as a storage area for seed populations buried in or on the soil (Moore and Chapman 1986). The seed bank is subject to depredation and pathogen attacks, a factor which affects the overall population numbers (Silvertown and Lovett Doust 1993).

The number of seeds in the soil is dependent upon the rate of input from the seed rain, loss rates due to depredation and disease, the rate of loss due to germination, and the dispersal of seeds into and out of an area. Seed densities are highest in frequently disturbed habitats (such as cultivable fields) and lowest in relatively undisturbed habitats. The species

most heavily represented in the soil tend to be those with the shortest lifespan (these are the species that produce large numbers of small seeds which are often capable of dormancy). Seed banks in highly disturbed areas contain seeds which reflect the above ground vegetation whereas the seed bank below perennial vegetation can be rather unrepresentative. The seed bank below perennial vegetation often consists primarily of seeds of herbs, and pioneer trees and shrubs (Silvertown and Lovett Doust 1993; Moore and Chapman 1986).

The spatial distribution of seeds in the seed bank is patchy, due to factors mentioned earlier which cause seeds to be deposited in clumps. Seeds are rarely evenly distributed in the soil profile, instead they tend to be concentrated near the surface. Seeds that are located away from the surface are often the result of redistribution by soil invertebrates who may carry seeds to considerable depths. In general, shallow burial of seeds often increases the chances of germination, whereas deep burial often prevents germination (Silvertown and Lovett Doust 1993).

2.9 Seed dormancy and germination

Seed dormancy is of selective value to plants as it allows them to delay germination until environmental conditions become favorable (Silvertown and Lovett Doust 1993). Many studies have been conducted to investigate mechanisms which inhibit and trigger germination. A variety of factors influence germination, including light intensity and quality, temperature and temperature fluctuations, nitrates, O₂ and CO₂ levels, pH, moisture, abrasion of the seed coat (scarification), hormones and stratification (Silvertown 1987). Dormant seeds possess hormones which either initiate or inhibit germination. Seed dormancy may be broken by; the change or loss of inhibitors, an increased permeability to

water and/or oxygen, or an increase in the activity of growth promoting hormones (Merlee Teresa and Avita 1989).

Long-lived species, such as trees, often have short-lived (ephemeral) seeds, whereas short-lived species (annuals) frequently have seeds capable of extended dormancy. Long-lived seeds often undergo annual cycles of dormancy caused by seasonal temperature changes (e.g. may require a cold treatment) which allow them to germinate at a favorable time of the year (Silvertown and Lovett Doust 1993).

Seneca snakeroot germinates readily under natural conditions, as evidenced by its abundance in disturbed sites (e.g. Hydro rights-of-way) in central Manitoba. Overcoming the dormancy factor seems to be the biggest problem facing researchers attempting to germinate seeds of seneca snakeroot (MacArthur 1994). According to Howarth and Keane (1995a), seneca snakeroot requires a prolonged stratification period (2-3 years), and even then seeds have low viability. The seeds are thought to lose their vitality quickly following collection and drying (Holm 1929), which could account for observed low germination rates. According to Bailey (1975), seneca snakeroot is best grown in sandy peat soil under partially shaded conditions. Seeds may be sown in early fall or spring.

Seneca snakeroot was apparently cultivated in England by 1739 following its introduction by John Tennent (Grieve 1967). An American farmer's bulletin originally published in 1915 reported that (quoted from Sievers 1948):

" Senega can be grown in any soil that contains a fair proportion of leafmold. Shade is not essential, although the plant thrives in partial shade in open hardwood forests. To propagate from seed it is necessary to plant seed that has been stratified by mixing it with sand and burying it in boxes or flowerpots in moist soil until the following spring,

when it may be sown in seedbeds or shallow boxes of loam and leafmold. The seedlings when old enough to be handled safely can be readily transplanted to permanent beds and set in rows to facilitate cultivation. The plant can also be propagated from roots, which may be obtained from dealers or collected from the wild plants in fall or early spring. In cold situations the plants will probably need protection during the first winter after transplanting. A light covering of straw or pine needles will be sufficient to protect them from severe frost. The plants grow slowly, and experiments thus far indicate that about 4 years are required to obtain marketable roots."

Attempts by university researchers and agriculturists to transplant or cultivate seneca snakeroot in the 1950's were apparently unsuccessful (Shipley 1956). The species was also found to grow slowly under cultivation (Hlady and Poston 1959). Doug Elsasser, a herb broker from Saskatchewan working under a grant from a Saskatchewan Agricultural Development fund, found that only 3 of 500 planted seeds germinated (MacArthur 1994). Elsasser contacted eleven other researchers in an attempt to solve the germination problem. Larry Gusta (Crop Development Centre, University of Saskatchewan) has been experimenting with different methods of triggering germination, including cold treatment and plant hormones. Prairie Plant Systems in Saskatchewan are experimenting with micro-propagation reproduction (MacArthur 1994; Elsasser 1996). Howarth and Keane (1995b) indicate that the species can also be propagated by root division.

Merlee Teresa and Avita (1989) undertook a series of experiments in an effort to break dormancy in *P. chinensis* (Indian seneca), the seeds of which are similar to those of seneca snakeroot. The results of their experiments are summarized in **Table 2.1**. Fresh seed did not germinate. The most effective treatment used gibberellic acid and/or mechanical scarification to break dormancy. The authors concluded that a combination of innate dormancy, a thick seed coat, and chemical inhibitors in the seed coat probably accounted

**Table 2.1. Results of germination experiments by Merlee Teresa and Avita (1989)
on *Polygala chinensis* L.**

Seed Treatment	% Germination
Untreated freshly collected seeds	0
Superficial scarification with "0" number sand paper	5
Deep scarification, slits on either side (mechanically treated)	70
Removal of strophiole	0
Removal of strophiole with superficial scarification	10
Complete removal of seed coat	100
Seed coat and endosperm removed	100
Seeds under running water - 2 days	0
Seeds under running water - 6 days	30
Seeds under running water - 8 days	50
Seeds under running water - 10 days	60
Seeds under running water - 15 days	60
Hot water treatment	0
Heat treatment	0
Sulfuric Acid pretreatment - 2 minutes	0
Sulfuric Acid pretreatment - 5 minutes	0
Sulfuric Acid pretreatment - 10 minutes	10
Sulfuric Acid pretreatment - 15 minutes	30
Sulfuric Acid pretreatment - 20 minutes	5
Sulfuric Acid pretreatment - 30 minutes	5
Temperature(range) and untreated seeds	0
Temperature (6°C) and mechanically treated seeds	20
Temperature (25°C) and mechanically treated seeds	80
Temperature (28°C) and mechanically treated seeds	90
Temperature (37°C) and mechanically treated seeds	50
Temperature (50°C) and mechanically treated seeds	20
Laboratory diffuse daylight- untreated	0
Laboratory diffuse daylight - mechanically treated	60
Continuous light - untreated	0
Continuous light - mechanically treated	50

Table 2.1. continued. Results of germination experiments by Merlee Teresa and Avita (1989) on *Polygala chinensis* L.

Seed Treatment	% Germination
Continuous darkness - untreated	0
Continuous darkness - mechanically treated	40
Black light - untreated	0
Black light - mechanically treated	30
White light - untreated	0
White light - mechanically treated	30
Green light - untreated	0
Green light - mechanically treated	20
Yellow light - untreated	0
Yellow light - mechanically treated	50
Red light - untreated	0
Red light - mechanically treated	60
Blue light - untreated	0
Blue light - mechanically treated	50
KNO ₃ - untreated	0
KNO ₃ - mechanically treated	10
KMnO ₄ - untreated	0
KMnO ₄ - mechanically treated	10
CuSO ₄ - untreated	0
CuSO ₄ - mechanically treated	0
Thiourea - untreated	0
Thiourea - mechanically treated	0
GA ₃ (500 ppm) - untreated	70
GA ₃ (500 ppm) - mechanically treated	100
GA ₃ (50 ppm) - strophiole removed	30
GA ₃ (100 ppm) - strophiole removed	30
GA ₃ (200 ppm) - strophiole removed	40
GA ₃ (250 ppm) - strophiole removed	60
GA ₃ (500 ppm) - strophiole removed	70
GA ₃ (1000 ppm) - strophiole removed	70

for the low germination of fresh seed.

2.10 Cultivation

Successful cultivation of seneca snakeroot and related species has been reported from Japan, China, India and Russia (Hlady and Poston 1959; Gillett 1968; Prescott-Allen and Prescott-Allen 1986; Briggs 1988; Catling and Small 1994). Seneca is apparently cultivated in southern Russia, where experimenters are examining its potential in treating respiratory disorders (Hutchens 1992). Seneca is not currently under cultivation in North America.

Significant amounts of seneca snakeroot (*P. senega* var. *latifolia*) are produced annually in Japan (Briggs 1988). Catling and Small (1994) report that annual production in Japan is 8 to 10 tonnes. More recently, the Canadian Embassy in Tokyo reported that Japan produces about 6 tonnes per year, but that the cultivated seneca root is not as popular as imported wild root (Commercial Officer, Canadian Embassy in Japan, pers. comm. 1996). European and American purchasers also prefer roots harvested in the wild (Gillett 1968; Briggs 1988). Cultivation of seneca snakeroot in North America seems possible provided that the optimal growth environment can be determined. The species is being considered as a potential new 'alternative' crop for Manitoba and other provinces (Manitoba Agriculture 1993; Catling and Small 1994). Seneca snakeroot takes about 4 to 5 years to produce a taproot of marketable size (Howarth and Keane 1995a; Gillett 1968).

2.11 Biochemistry

The two principally active constituents of seneca snakeroot (the drug is known pharmaceutically as *Radix Senegae* [Shibata 1976]) are the triterpenoid saponin glycosides

polygalic acid and senegin, which make up *ca.* 5% and 4% respectively of the dried root (Allport 1944; Wallis 1967). The root also contains a small amount of methyl salicylate, which gives the root a wintergreen-like odour (Allport 1944). Other chemicals in the root include sterols, fats, sucrose, oligosaccharides (Senegoses A-E, F-I, J-O), polygalitol, and *ca.* 5% fixed oils (Briggs 1988; Saitoh *et al.* 1993a,b 1994; Wallis 1967). The known phytochemical constituents of *P. senega* are listed in Appendix I (Duke 1992).

The saponin glycosides in seneca snakeroot are responsible for its pharmaceutical efficacy. Saponin glycosides are compounds that yield a foaming aqueous solution (soap-like froth) when mixed with water (Allport 1944; Shibata 1976). These glycosides occur naturally in certain higher plants and marine organisms. Saponins form precipitates with cholesterol in alcohol, and have anti-microbial (primarily anti-fungal) properties (Shibata 1976). They also have hemolytic properties and can be poisonous to fishes, shells and insects. Chemically, there are two types of saponins, steroidal and triterpenoid, which are based upon the structure of their sapogenins. Steroidal saponins are known as the precursors to steroid hormone production, whereas triterpenoid saponins are recognized for their pharmaceutical effects. Triterpenoid saponins are often the main ingredient in Chinese herbal products, and are responsible for the efficacy of these drugs (Shibata 1976). The structure of the senegin saponins present in *Polygala senega* L. are shown in Fig. 2.5.

A number of studies have examined the biochemistry of seneca snakeroot and related species. Some examples include:

- Fujita and Itokawa (1961) concluded that *P. senega* var. *latifolia* and *P. tenuifolia* contain the same sapogenins.
- Corner *et al.* (1962) isolated at least five hydroxycinnamoyl esters from seneca.

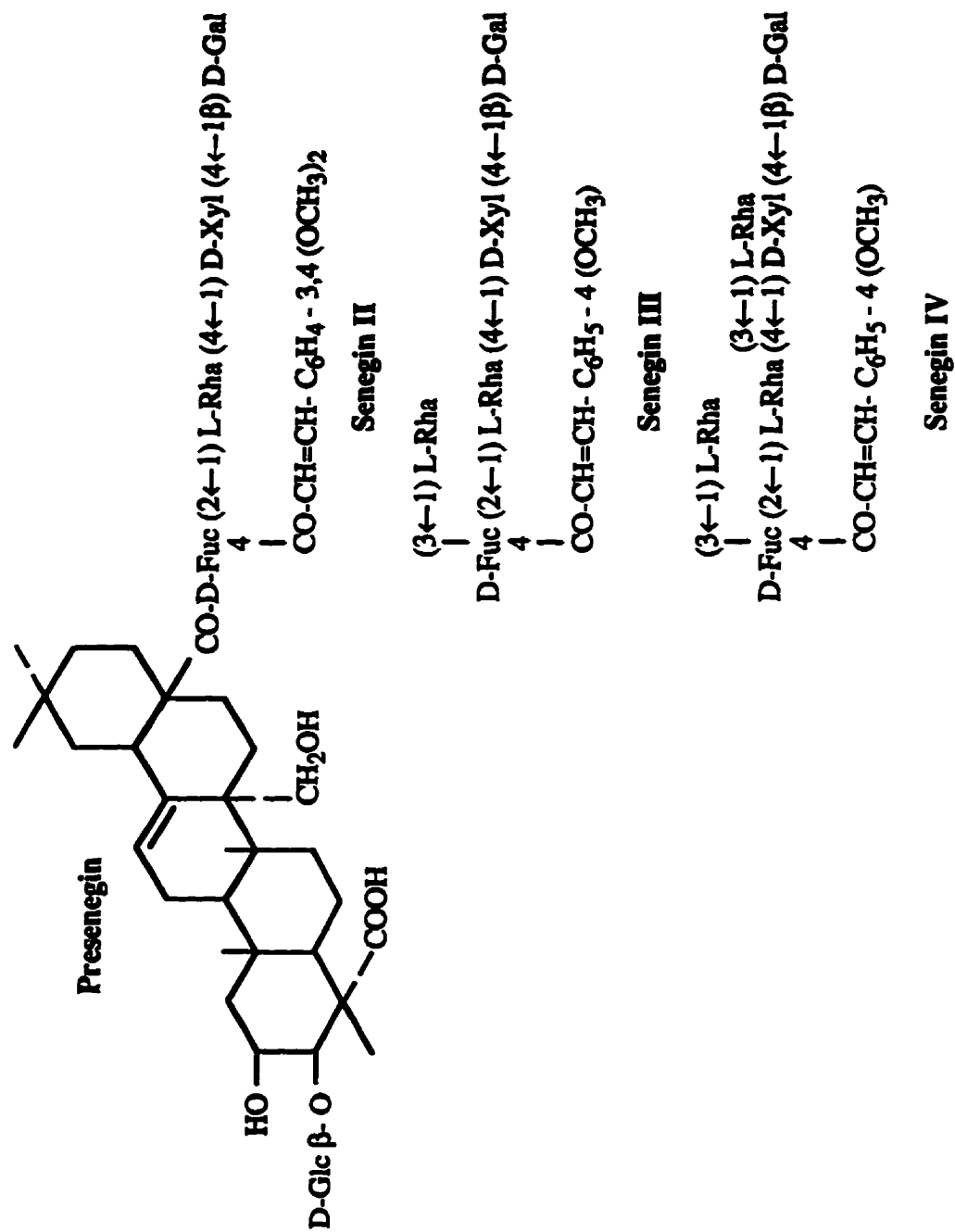


Figure 2.5. Senegin saponins present in *Polygala senega* L.
(adapted from Shibata 1976).

- Dugan *et al.* (1964) depict the functional groups and structure of the sapogenin senegenin of *Polygala senega* L.
- Pelletier *et al.* (1964) discuss the unusual structure of senegenic acid.
- Yosioka *et al.* (1966) describe a new method for soil bacterial hydrolysis as a possible mechanism for the structural study of saponins.
- Shoji *et al.* (1971, 1972) describe the chemical structure of the senegin saponins.
- Saitoh *et al.* (1993a,b; 1994) isolated new oligosaccharides (senegoses A-E, F-I, J-O).
- Yoshikawa *et al.* (1995) describe experiments on *Senegae radix* and its inhibitory effects on alcohol absorption and hypoglycemic activity.
- Masuda *et al.* (1996) discuss the how senegin-II of *Senegae radix* reduced the levels of blood triglycerides in normal mice.

The Saskatchewan Herb Research Centre, University of Saskatchewan began working on the phytochemistry of seneca in May of 1995. Their objective is to develop an analytical procedure for evaluating seneca snakeroot potency using saponins as marker compounds. They are also examining seasonal variation in the yield and composition of snakeroot saponins (B. Barl, pers. comm. 1995).

2.12 Ethnobotany

2.12.1 Native uses

The use of plants by the native peoples of North America may have changed as a result of European influences. According to Arnason *et al.* (1981):

"Indian uses of plants have been recorded, compiled, and recompiled so that their original importance are often difficult to ascertain. The importance of specific plants has been exaggerated by commercial interests."

Nonetheless, historical evidence indicates that seneca snakeroot has long been utilized by a variety of native groups in eastern-central North America for the treatment of specific ailments (summarized in Table 2.2). Seneca snakeroot was most notably used in the treatment of rattlesnake bites, the root being first chewed and then applied to the bite as a paste. The resemblance of the root to a coiled snake ("Doctrine of signatures") could perhaps explain its use by the Seneca Indians to treat snakebite (Briggs 1988; Weiner 1980). The root was also used to treat snakebite by the Winnebago, Dakota, Cherokee and Iroquois peoples (Kindscher 1992; Moerman 1986). Snakeroot was also used in the treatment of insect stings and poisoning by the Winnebago and Dakota peoples.

The Seneca Indians also used the root to make a tea, which was drunk as a treatment for coughs, sore throat and colds (Kindscher 1992). The boiled root 'bark' was made into a tea and used as an abortifacient by the Ottawa and Chippewa (Weiner 1980). The Nishinam boiled the entire plant and drank the liquid as a diarrhetic. The boiled root was used to treat heart trouble by the Mesquakies and Potawatomis (Kindscher 1992). The dried root of seneca snakeroot was used as a charm and carried as a talisman by the Chippewa and other native peoples (Densmore 1928).

In his memoirs, John Dunn Hunter mentions that the Kickapoo, Osage and Kansas peoples used seneca snakeroot:

"... in cold infusions, during the remission of fevers, which are attended with great prostration of strength, and in diseases of the pulmonary organs. They also gave it

Table 2.2. Use of seneca snakeroot by native group (from Arnason *et al.* 1981, Densmore 1913, 1928, Johnston 1970, Kindscher 1992, Moerman 1986, Weiner 1980, Zieba 1990).

Use	Blackfoot	Cherokee	Chippewa	Dakota	Fox	Iroquois	Kansas	Kickapoo	Malecite	Menome
Abortifacient		X	X							
Aches & Pains										
Anticonvulsive			X							
Antirheumatic		X								
Cathartic		X								
Charm			X							
Colds & Coughs		X								
Congestion							X	X		X
Diaphoretic		X								
Diarrhea										
Diuretic		X								
Earache										
Expectorant	X	X								
Female complaints							X	X		
Gastrointestinal aid							X	X		
General medicine										
Heart trouble			X							
Hemostat			X		X					
Inflammation			X							
Insect stings				X						
Kidney aid		X								
Pulmonary aid		X					X		X	
Snakebite		X		X		X				
Sore throat										
Stimulant										
Tonic										

Table 2.2 continued. Use of seneca snakeroot by native group (from Arnason *et al.* 1981, Densmore 1913, 1928, Johnston 1970, Kindscher 1992, Moerman 1986, Weiner 1980, Zieba 1990).

Use	Mesquakie	Micmac	Nishinam	Ojibwa	Osage	Ottawa	Plains Cree	Potawatomi	Sioux	Seneca	Winnebago
Abortifacient						X					
Aches & Pains				X							
Anticonvulsive				X							
Antirheumatic				X							
Cathartic											
Charm											
Colds		X			X						
Congestion				X						X	
Diaphoretic				X							
Diarrhea											
Diuretic											
Earache											
Expectorant											
Female complaints					X				X		
Gastrointestinal aid					X						
General medicine				X							
Heart trouble				X							
Hemostat								X			
Inflammation											
Insect stings											
Kidney aid											X
Pulmonary aid											
Snakebite					X						
Sore throat										X	X
Stimulant				X						X	
Tonic											

warm, in combination with various other drugs, with a view to promote the sweating process, or to discharge the collection of mucus from the trachea and lungs. They esteem it very highly in their female complaints, and also in disease of their children when there is great difficulty of breathing" (quoted from Kindscher 1992).

Seneca snakeroot was often used in mixture with other plant species. The Great Lakes Chippewa made a medicine consisting of the roots of seneca snakeroot, sagebrush, ground plum, milkvetch and Arkansas rose. Seneca snakeroot, called Bi'jikiwuck' (literally, "cattle herb"), was the principal ingredient. The roots were washed, scraped, dried and then pounded into a powder. The dried powder of seneca snakeroot was kept separate, while the other herbs were ground into a mixture (Densmore 1913). The mixture, which was used as a tonic and stimulant, was traditionally prepared in the following manner:

"A quart of water is heated in a pail and about 1/3 of a teaspoon of the mixed ingredients is placed on the surface of the water at the 4 sides of the pail (representing north, south, east and west). A very little of the first [principal ingredient] is placed on top of each. The ingredients soon dissolve. A stronger decoction was secured by boiling. The medicine was taken 4 times a day, the dose being small at first, and gradually increased to about a tablespoonful." (quoted from Densmore 1913).

In Manitoba and adjacent regions, the Plains Cree use a decoction of seneca snakeroot as a general remedy. The plant may be used alone or in mixture with other herbal remedies. Seneca snakeroot has also been used by the Cree and Sioux to treat earaches. Zieba (1990) reports that the Cree name for seneca snakeroot is 'wisak', while the Swampy Cree use the name 'wincekes'. The root is collected in the summer, dried, and stored for later use. The root is steeped in hot water, and the lukewarm solution used to cure earaches.

2.12.2 Early European uses

The first dated account of seneca snakeroot by Europeans was that of Rev. J. Clayton in 1687, in which he mentions snakeroot as one of 40 herbs "*of great secret*" shown to him by natives in Virginia (Erichsen-Brown 1979). Later accounts in Virginia mention that native healing herbs were generally concealed from European settlers, but that certain herbs such as 'rattlesnake root' were allowed to be known as they need to be applied immediately. Seneca snakeroot was one of the first native healing plants used by European settlers (Kindscher 1992). William Byrd of Virginia (1728) claimed the best medicines for gout were those that:

"... clear a passage through the narrow vessels, that are the seat of this cruel disease. Nothing will do this more suddenly than rattle-snake's oil, which will even penetrate the pores of glass when warm'd in the sun." (quoted from Coffey 1993).

As there was no rattlesnake oil available on one of their expeditions, Byrd continued:

"... but, lately the Seneca Rattle-Snake-Root has been discover'd in this country, which being infus'd in wine, and drank every morning and evening, has in several instances had a very happy effect upon the gout, and enabled cripples to throw away their crutches and walk several miles." (quoted from Coffey 1993).

In the early 1700's, a Virginia doctor named John Tennent began using the root as a treatment for pleurisy and pneumonia. He had observed the Seneca Indians using the root on rattlesnake bites, and noted that the symptoms of the bites were similar to respiratory disorders (Millspaugh 1974). Tennent wrote to Dr. R. Mead, a physician in London:

"At last I was informed... that there was a Root discovered by the Seneca Indians which was a certain remedy against the Bite of the Rattlesnake...and was distinguished ...by the name of Seneca Rattle-Snakeroot." (quoted from the file on Seneca Snakeroot at the Hudson Bay Archives, Wpg.).

In 1736 Tennent published "An Essay on the Pleurisy" in which he described and promoted the medicinal virtues of seneca snakeroot. At the time, pleurisy was the most epidemic disease in colonial Virginia (Jellison 1963). Although respected by many Virginian colonists, Tennent's experiments with seneca snakeroot caused considerable controversy amongst his fellow physicians. Unfortunately, other medicinal plants with the same common name were often mistaken for seneca snakeroot by the Virginia colonists. In response, Tennent began supplying the root free of charge, which pleased the colonists but further alienated his colleagues.

In 1737, Tennent travelled to London with a supply of seneca snakeroot. He was well received, and some of his material was sent to the Royal Society of Paris where its effectiveness in the treatment of pleurisy was again demonstrated. Tennent returned to Virginia in the fall of 1737, but his personal and financial situations did not improve. He later returned to England, where he died in 1748 a bitter and broken man. In 1760, his son petitioned the Virginia House of Burgesses to reward his father's findings, but the petition was rejected (Jellison 1963).

Seneca snakeroot came to be widely used in North America and Europe following Tennent's death (Crellin and Philpott 1990). By the early 1800's the plant had attracted a great deal of attention from the medical public, and was exported in large quantities to European apothecaries (Erichsen-Brown 1979). Seneca snakeroot was used as an effective diuretic and expectorant, and in the treatment of rheumatism, dropsy, typhus, asthma and

many other diseases (Holm 1907; Crellin and Philpott 1990). German physicians praised seneca in treating ophthalmia, preventing the formation of cataract, and promoting pus formation in hypopyon (Millsbaugh 1974). In 1870, C. J. Cowle spoke of seneca as *"one of the few roots and seeds that you cannot get enough of [for resale]"* (quoted from Crellin and Philpott 1990).

By 1887, seneca snakeroot was over-harvested to the point of near extinction in the eastern North America, but it remained abundant in the Northwest (Trease 1966; Coffey 1993). Seneca snakeroot was widely harvested in Manitoba and Saskatchewan. In 1883, N. M. W. J. McKenzie wrote of the harvesting of seneca snakeroot by the Plains Cree:

"... I saw that senega or Snakeroot was in great demand in the States. It seemed to be the chief composition in nearly all patent medicines and we had many acres of it growing all over the reserve; in fact there was an abundance of it all over the country. I knew the root well, as the Indians always used it for their own medicinal preparations...Senega root was worth all the way from 25 to 85 cents per pound...some of them made as much as \$5 a day...Snakeroot digging soon became a great industry all over the country....for several years later when I would be travelling by rail, at any of the little stations from which butter and eggs were shipped, you would always see a few sacks of Snakeroot in the shipment." (McKenzie 1921).

In a letter to the Right Honourable Sir John A Macdonald, McColl (1886) wrote that the aboriginal peoples of the Rosseau River reserve (ca. 60 km south of Winnipeg) *"... have been for a number of years extensively engaged in gathering seneca Snakeroot, for which they annually received about ten thousand dollars"*, a considerable sum in 1886.

In 1909, the dried root sold for fifty-five to seventy cents per pound (Wiener 1980). Canadian exports and prices for wild seneca snakeroot from 1919 to 1957 are summarized in **Table 2.3** (from Hlady and Poston 1959). Harvesting peaked in 1930 at *ca.* 730,000 lbs of dried root, but dropped off to about 150,000 lbs by the mid-1950's. During the 1950's, seneca root was collected by at least 18 different native groups in southern and central Manitoba (illustrated in **Figure 2.6**). At this time, seventy-five percent of the world's supply of the root was harvested from the Interlake region of Manitoba, providing an annual income of \$150,000 to the local aboriginal peoples (Shipley 1956). The towns of Hodgson, Ashern and Gypsumville were the main depots for receiving roots. Seneca root was an important source of seasonal income to some native families in the Interlake region. The dried root was purchased by the R.S. Robinson company in Winnipeg, which in the 1950's annually exported 150,000 pounds of the dried root to England, Cuba, Japan and Switzerland, as well as supplying the United States and Canada (Shipley 1956).

Seneca snakeroot was listed in the official United States Pharmacopoeia from 1820 to 1936 (Weiner 1980), and on the U.S. National Formulary from 1936 to 1960 (Kindscher 1992). In 1868, the species was included in a list of Canadian medicinal plants in the Canadian Pharmaceutical Journal (Anonymous 1868). The species was also included on a drug buyer's list in the Appalachians (Krochmal 1968).

In the 1920's, seneca snakeroot was used in patent medicines to treat bronchitis, often in combination with other natural expectorants. In the mid-1950's, seneca snakeroot was the main ingredient in a number of patent medicines and cough syrups. Demand for seneca snakeroot declined after 1960, so that by the mid-1960's the harvest in Canada was no longer commercially important (Gillett 1968). The reduction in demand was largely attributable to the introduction of cheaper, chemically-synthesized expectorants (Tyler 1981).

Table 2.3. Canadian export of seneca snakeroot (adapted from Hlady and Poston 1959).

Year	Quantity (pounds)	Total dollar value (\$)	Price per pound (\$)
1919	340,148	281,875	0.83
1920	415,223	594,088	1.43
1921	268,363	283,830	1.06
1922	181,894	124,748	0.69
1923	415,018	281,032	0.68
1924	383,505	229,275	0.60
1925	508,099	266,447	0.52
1926	294,110	166,262	0.57
1927	212,850	140,873	0.66
1928	271,885	278,157	1.02
1929	524,119	593,017	1.13
1930	728,221	660,284	0.91
1931	183,392	103,950	0.57
1932	346,263	131,335	0.38
1933	225,907	68,745	0.30
1934	339,305	118,558	0.35
1935	337,657	91,990	0.27
1936	308,033	95,303	0.26
1937	376,054	175,917	0.47
1938	340,627	151,286	0.45
1939	397,034	203,571	0.51
1940	346,268	231,653	0.67
1941	341,020		
1942	76,572		
1943	107,940	values missing due to war-time	
1944	202,227		
1945	224,975		
1946	369,000	818,436	2.21
1947	258,000	507,405	1.95
1948	146,096	246,948	1.70
1949	175,492	207,792	1.19
1950	221,005	411,761	1.86
1951	257,918	576,238	2.23
1952	127,443	215,309	1.69
1953	118,458	198,101	1.68
1954	193,470	426,381	2.21
1955	180,539	450,119	2.50
1956	158,271	364,238	2.30
1957	166,603	361,915	2.22

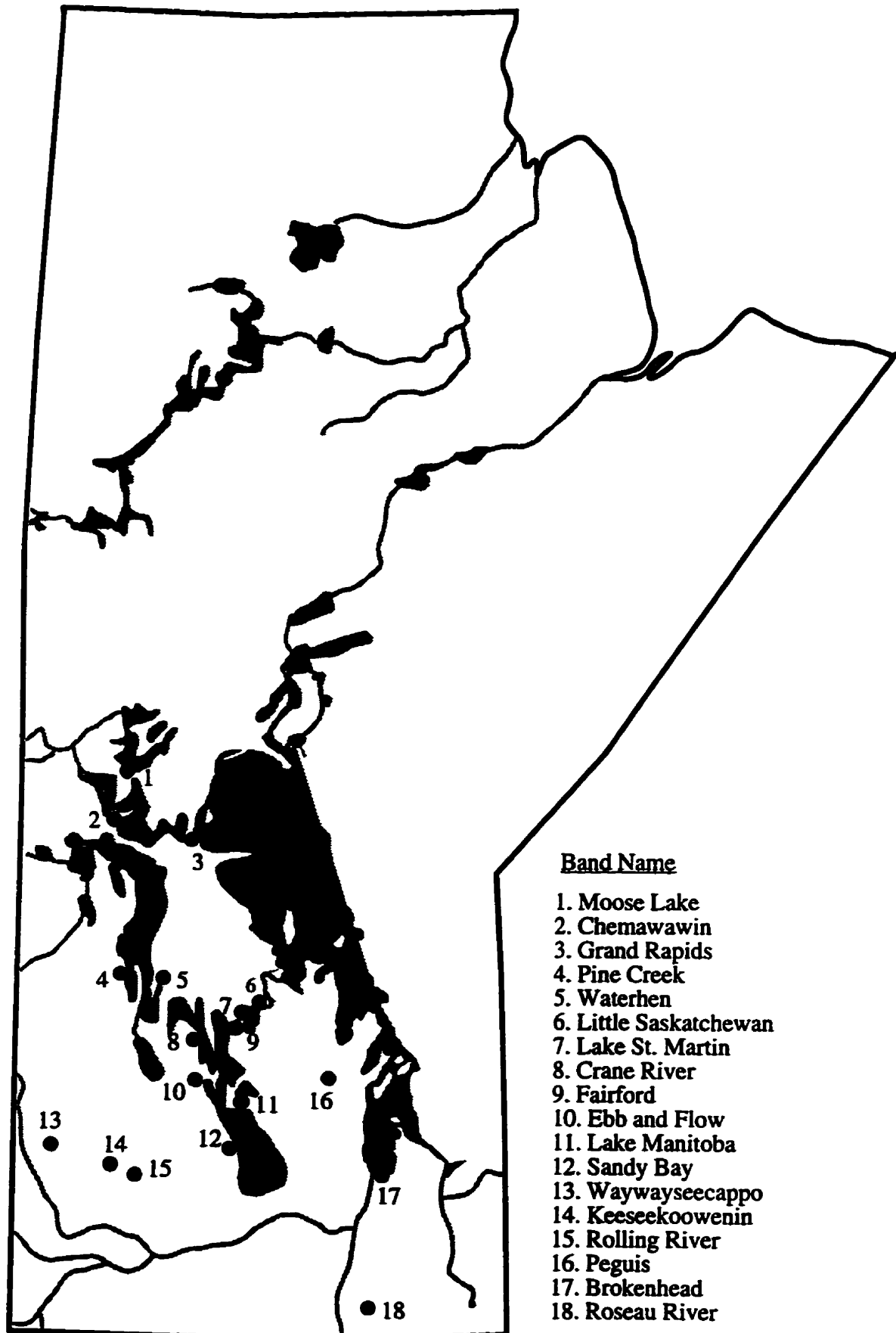


Figure 2.6. Native groups in Manitoba who harvested seneca snakeroot in the 1950's (adapted from Weir 1960).

2.12.3 Modern uses

(1) Utilization and pharmaceutical activity

Seneca snakeroot is used as an expectorant, diaphoretic, sialagogue and emetic in the treatment of colds, asthma and bronchitis (Tyler 1981). It is generally administered as an infusion, liquid extract or tincture (Pharmaceutical Society of Great Britain, 1952). Recent research in Japan indicates that seneca snakeroot extracts are able to inhibit alcohol absorption by routing the alcohol to the large intestine before it can be absorbed into the blood stream (Conlon 1995; Yoshikawa *et al.* 1995).

The principal pharmaceutically-active chemicals in seneca snakeroot, as described previously, are a mixture of triterpenoid saponins, which make up 5-10% of the dried root (Briggs 1988; Tyler 1994). Saponins are irritating to the gastric mucosa, causing secretion of mucus in the bronchioles (Pharmaceutical Society of Great Britain, 1979). Seneca is therefore classified as an expectorant and is recommended in the treatment of respiratory disorders. Vomiting and purging can occur if recommended dosages are exceeded (Tyler 1981).

(2) Medicinal preparations (pre-1960)

The 1936 American Pharmaceutical Association's National Formulary describes the preparation of a fluid extract of seneca snakeroot that was used as an important medicinal base. The Pharmaceutical Recipe Book (American Pharmaceutical Association, 1943) outlines the preparation of an ammoniated mixture of seneca, ipecac, and paregoric to be administered to children. By 1955, the National Formulary included only one fluid extract preparation and a single formula for seneca syrup. The recommended preparation was a

diluted ammonia solution of 100 g of powdered seneca root, 2 volumes of alcohol, and 1 volume of water. This was administered as a decoction in 1 ml dosages.

In Great Britain, the 1953 *Pharmaceutical Formulas* (12th edition) listed seneca snakeroot as one of the 37 chief decoctions in use. It was employed as a fluid extract, glyceextract (liquid extracts without alcohol) or infusion. The material was used fresh or in concentrated form, usually in mixture with other products.

In Europe and North America, interest in seneca snakeroot declined in early 1960's. The plant was delisted from *The U.S. National Formulary* in 1961. Tyler (1981) noted that the *Handbook of Non-Prescription Drugs (United States)* did not list a single cough syrup containing seneca snakeroot.

(3) Medicinal preparations (1980-present)

A resurgence of interest in natural product medicine led to a considerable increase in exports of seneca snakeroot from Manitoba in the late 1980's (Briggs 1988). Seneca snakeroot continues to be used mainly as an effective expectorant and emetic (*British Pharmacopoeia*, 1993). The root material is obtained either from the harvesting of wild material in North America, or from plants cultivated in Japan (Prescott-Allen and Prescott-Allen 1986). *Martindale's Extra Pharmacopoeia* (Reynolds 1993) has recently listed seneca snakeroot as an ingredient in thirty-five drug products, for use mainly in the treatment of coughs, colds and respiratory disorders. These products are manufactured in Europe (mainly Spain, France and Switzerland), Australia, Sweden and South Africa (Table 2.4).

Table 2.4. Herbal products manufactured abroad utilizing seneca snakeroot (from Reynolds 1993).

No.	Product name	Use	Country produced
1	Antibron	coughs	UK
2	Broncovital	respiratory tract disorders	Spain
3	Bronquiasmol	respiratory tract disorders	Spain
4	Chest mixture	coughs, catarrh	UK
5	Cocilix	coughs	South Africa
6	Cocillana-etyfin	coughs	Sweden
7	Combitorax	respiratory tract infections	Spain
8	Cosanyl	coughs	Australia
9	Dinacodi	coughs	France
10	Expectoran	coughs and associated respiratory tract disorders	Switzerland
11	Expectoran avec Codeine	coughs and associated respiratory tract disorders	Switzerland
12	Hederix	coughs	Switzerland
13	Hortepulmo	respiratory tract infections	Spain
14	Iodocafedrina	respiratory tract disorders	Spain
15	Lasa infantil		Spain
16	Makatussin	coughs	Switzerland
17	Makatussis forte	coughs, catarrh	Switzerland
18	Neo-codion sirop Nourrissons		France
19	Passedy	bronchopulmonary secretory disorders	France
20	Pastillas Pectoral	respiratory tract disorders	Spain
21	Patussol	coughs	Switzerland
22	Pectosan	coughs	France
23	Polery	coughs	France
24	Polery-enfants	coughs	France

Table 2.4 continued. Herbal products manufactured abroad utilizing seneca snakeroot (from Reynolds 1993).

No.	Product name	Use	Country produced
25	Pulmo Hidratol Codeina	respiratory tract disorders	Spain
26	Pulmofasa	respiratory tract disorders	Spain
27	Pulmofasa Antihist	upper respiratory tract disorders	Spain
28	Pulmothiol	coughs	France
29	Pilmothiol Enfants	coughs	France
30	Senamon	respiratory tract disorders	Australia
31	Senepius	coughs and colds	Australia
32	Silphoscalin		Germany
33	Sirop Pectoral adulte	coughs	France
34	Tuberol	coughs	France
35	Tusolone	respiratory tract infections	Spain

Prescott-Allen and Prescott-Allen (1986) list seneca snakeroot as an ingredient in 13 drug products manufactured in Canada, while the Compendium of Non-Prescription Products (Canadian Pharmaceutical Association, 1995) lists seneca snakeroot as an ingredient in six cough syrups manufactured in Canada (Table 2.5). Therapex (Québec) produces a bronchial cough syrup containing seneca root. Their main suppliers of the root are EMIL Flachsmann, Switzerland (who did not cooperate with my request for information) and Active Organics of California (who have since gone out of business). Trans Herb (Québec) incorporates seneca snakeroot into a herbal toothpaste sold in Canada and Europe. The company purchases seneca root directly from Canadian suppliers, though they indicated that obtaining sufficient amounts of root is sometimes difficult. A homeopathic cold remedy containing a tincture of seneca root is manufactured by Homeocan (Québec). Jeunique International (Québec) incorporates seneca into their N-R-G Plus vitamin-mineral supplement.

(4) Herbalists and herbal preparations

Herbalists describe seneca snakeroot as a herb "that's coming back" into popularity (Crellin and Philpott 1990). Tyler (1981) notes that although herbalists have praised the virtues of seneca root, overdoses can cause severe stomach upset and vomiting. Seneca root has been used by herbalists in treating coughs and colds, and it has been used as a stimulant. It has also been recommended in the treatment of rheumatism, sore throat, and as a blood purifier. Crellin and Philpott (1990) note that seneca snakeroot has also been used in the treatment of acute bronchitis, asthma, blood poisoning, chronic catarrh, chronic croup, dropsy, lung congestion, pleurisy, pneumonia, rheumatism, smallpox, and whooping cough.

Table 2.5. Products manufactured in Canada which contain seneca snakeroot (compiled from the Compendium of Non-Prescription Products, Canadian Pharmaceutical Association 1995).

Product Name	Manufacturer	Province	Dosage form	Seneca content
Bronchial Cough Syrup	Theralab	Quebec	250 mL syrup	1mg/mL
Bronchial Cough Syrup	Drug Trading	Ontario	250 mL syrup	0.015 mL/mL
Bronchidia Cough Syrup IDA	Drug Trading	Ontario	250 mL syrup	0.015 mL/mL
Bronchozone Cough Syrup Certified	Drug Trading	Ontario	250 mL syrup	0.015 mL/mL
Sirop Cocillana Codeine	Lab Atlas	Quebec	250, 500 mL syrup	0.57 mg/mL
Wampole Bronchial Cough Syrup	Wampole	Ontario	250 mL syrup	5.5 mg/mL

Seneca snakeroot is widely used in Europe in the form of cough drops, herbal teas or syrups for treating coughs, colds and throat irritations (Tyler 1981; Dwyer and Rattray 1986). Unprocessed dry root is sold in Europe for herbal tea preparations. In Vienna, Austria, the root is sold at the retail level for 1283 Austrian schillings per kg (ca. C\$250 per lb., Mike Hauser, pers. comm. 1996).

(5) Asian markets

The cultivated form of seneca snakeroot (*Polygala senega* var. *latifolia*, Japanese seneca) is used as an expectorant in Japan (Wallis 1967; Saitoh *et al.* 1993a). *Polygala tenuifolia* ('yuanzhi' in Chinese, 'onji' in Japanese), which is closely related to seneca snakeroot, is mentioned in the ancient Chinese herbal literature. Traditional processing methods specified that only the root 'bark' be used, although modern preparations generally utilize the entire root. Interestingly, studies have indicated that the root core contains only a small amount of the active saponins (Chang and But 1986). The product was traditionally used as an expectorant, sedative and resuscitating agent in both China (Tang and Eisenbrand 1992; Huang 1993) and Japan (Saitoh *et al.* 1993a). In China, it is also used to strengthen the nervous system, as an anti-swelling medicine (Shibata 1976; Tang and Eisenbrand 1992), and as a cancer treatment (American Herbal Pharmacology Delegation, 1975). Clinical studies in China indicate that it is also useful (in mixture with other herbs) in the treatment of chronic bronchitis, insomnia and fatigue (Chang and But 1986; Keys 1976). Related species (*Polygala tenuifolia* and *P. chinensis*) are valued in China for their medicinal properties, especially in treating coughs (Tang and Eisenbrand 1992).

It has been demonstrated that the saponins of *P. senega* and *P. tenuifolia* are almost identical (Fujita and Itokawa 1961; Shibata 1976). The saponins of Indian seneca (*Polygala*

chinensis) and Siberian seneca (*P. sibirica*), which have also been used in traditional medicine, contain closely-related saponin compounds (Wallis 1967; Shibata 1976; Huang 1993).

(6) Veterinary medicine

Seneca snakeroot has been used in veterinary medicine as an antitussive in sedative expectorant complexes (Morton 1977; Rossoff 1974). Personal communications (1995) with a sample of veterinarians in Winnipeg revealed an unfamiliarity with the drug.

(7) Modern uses by native peoples in Manitoba

Snakeroot is also used medicinally in homes in Manitoba. A seneca digger from the Grand Rapids area currently uses the root as a cure for colds and arthritis. He boils about three root crowns in water, strains the mixture and then drinks it (H. Chartier, pers. comm. 1995). Seneca root is well known among the Nithawitniw of Canoe Portage. The root is collected, dried and stored for future use. The root is steeped in hot water, and the lukewarm solution placed in the ear to cure earaches (Zieba 1990).

2.13 Economics of seneca snakeroot in Manitoba

2.13.1 Seneca diggers

Canada (particularly the provinces of Manitoba and Saskatchewan) has long been the major supplier of wild seneca snakeroot (Gillett 1968). Plants from the Canadian prairie provinces are known commercially as the 'Northern' or 'Manitoba' variety, and were held in high esteem due to their large size (Gillett 1968).

Seneca snakeroot is generally dug in the early summer, when the plants are in flower and more easily recognized. In Manitoba populations, it takes *ca.* 30-40 roots to make one dry pound (Hlady and Poston 1959). Roots dry to about one-third of their fresh weight (Elsasser, pers. comm. 1996). Diggers of seneca snakeroot often travel along roadsides in the Interlake region of Manitoba, equipped with a spade and a burlap bag tied around their waist. In the summer of 1995 two seneca diggers, Clarence Campo and Henry Chartier, were interviewed. Both were observed collecting seneca snakeroot along Provincial Highway 6 near Grand Rapids.

Clarence Campo has been digging seneca root for about fifteen years. He claims that not many people bother digging for seneca root anymore. He uses a small, narrow spade to loosen the plant from the soil. Once the plant is out of the ground, the shoots are twisted off and discarded. The fresh root is then placed in a shoulder bag.

Henry Chartier, 69, is a retired Manitoba Hydro employee who digs seneca root throughout the summer. He uses a large pick-axe tool which he believes works better than a shovel. He places the roots in a bag which he keeps tied around his waist. In two hours Henry can dig about three pounds of root. When he gets home he washes off the roots with water and then lays them out on a screen to dry (either in the sun or in a small shed). The drying process may take up to two weeks, depending on the weather.

2.13.2 Seneca exporters

Three seneca snakeroot exporters were interviewed: Ken Hooper (Winnipeg), Dave Buck (The Pas), and Doug Elsasser (Togo, Saskatchewan). They all stated that the current problem with the industry is the lack of seneca root diggers.

Mr. Hooper purchases seneca root primarily from diggers in the Interlake region. He remembers that seneca root digging used to be a family affair. Entire families would go out for the day or week to collect seneca root. He feels that today many people believe that the time and effort involved in collecting the root is too great. Mr. Hooper exports seneca to the United States and Europe.

Mr. Buck purchases snakeroot from diggers in the Grand Rapids, Gypsumville and Moose Lake areas. He sells the dried material to an unnamed major pharmaceutical company in the United States that has international connections. Mr. Buck believes that most if not all the root eventually ends up in Europe.

Mr. Elsasser is the owner of Parkland Botanicals. He purchases seneca root from diggers in eastern Saskatchewan and adjacent western Manitoba (Mafeking, Swan River and Boggy Creek regions). The dried root is sold to a pharmaceutical company in Montreal, as well as to brokers in Vancouver and North Carolina.

In the summer of 1994, exporters were paying *ca.* five dollars a pound for the dried root. This increased to *ca.* six to eight dollars a pound in 1995, *ca.* ten to twelve dollars a pound in 1996, and it is anticipated that the price will increase to *ca.* ten to fifteen dollars a pound in 1997 (B. Barl, pers. comm.; D. Elsasser, pers. comm.).

2.13.3 Overseas exports

It appears that a large portion of Manitoban seneca root ends up overseas. According to the Canadian Embassy in Tokyo, Japan imports *ca.* six metric tons of seneca root each year. Their largest supplier is Germany. Since the Japanese prefer roots harvested in the

wild, it is very unlikely that such a large amount of seneca root is coming from cultivated sources in Germany. In all likelihood, German exporters are acting as intermediaries between Canada and Japan.

CHAPTER 3

MATERIALS AND METHODS

3.1 Description of study sites

3.1.1 Study area

The study area incorporates Manitoba Hydro rights-of-way (ROW) corridors in the northern Interlake and Grand Rapids regions of Manitoba (Fig. 3.1). The study sites were located either on the high voltage direct current (450 kv HVDC) transmission line, or the adjacent lower voltage alternating current (230 kv AC) transmission line.

Construction of the HVDC line began in 1968, and it was fully operational by 1972 (MacLellan 1982). It is a 895 km double-line system running from Gillam to the Dorsey receiving station near Winnipeg (Walker 1994). The AC line parallels the DC line from the Minago River to Dorsey. The two AC line sections included in this study are the bipole line section from Ashern to Grand Rapids (operational since 1964), and the unipole section between Grand Rapids to William River (operational since 1966; R. Bukowsky, pers. comm.). The AC and DC transmission lines generally parallel Provincial Highway No. 6, allowing ready access to most sites.

3.1.2 Geology, physiography and soils

The study area falls entirely within the Boreal Plains ecozone, and the Mid-Boreal Lowland ecoregion (Smith *et al.* 1995). This ecoregion is underlain by Palaeozoic limestone bedrock that is covered with glacial deposits. Limestone outcrops are frequent

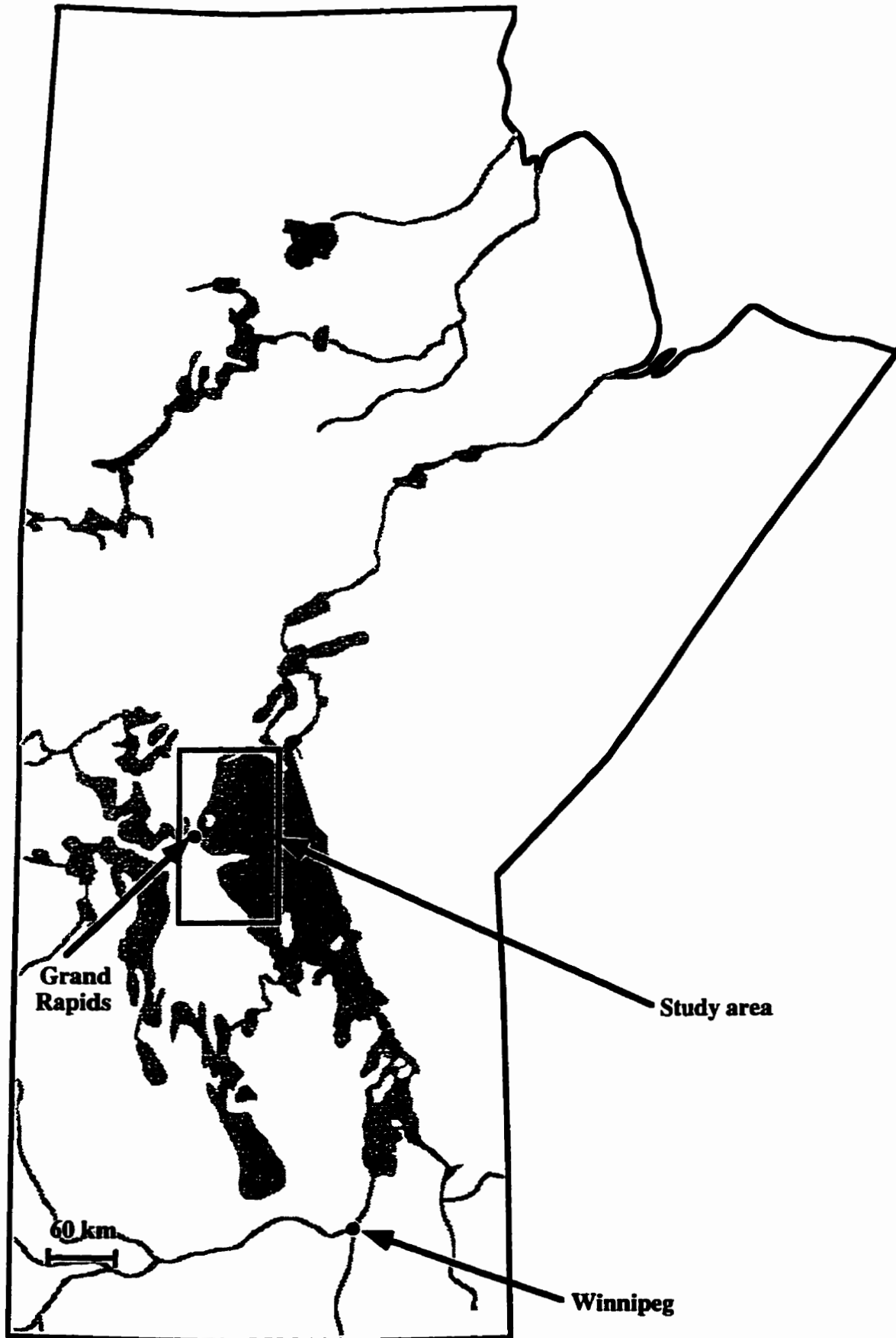


Figure 3.1 Map of Manitoba showing study area.

throughout the area. Elevation within the study area ranges between 218 m a.s.l. (Lake Winnipeg) and 310 m a.s.l. (The Pas moraine). The topography consists of a ridge and swale pattern trending from north to south. Soils in the area are mainly Eutric Brunisols, Organic Mesisols and Fibrisols. The study area includes three ecodistricts (Smith *et al.* 1995):

Cedar Lake ecodistrict

This ecodistrict occurs north of The Pas moraine, and includes the town of Grand Rapids. This region is characterized by a low-relief limestone plain overlain by a thin layer of glacial till. Mean elevation is 259 m a.s.l. The district is dominated by limestone bedrock, and small limestone sinkholes are common. Relief changes are *ca.* 0.6 m per km.

The Pas Moraine ecodistrict

The Pas Moraine, a distinct topographical feature of the mid-Boreal Lowland ecoregion, extends from Lake Winnipeg (Long Point) west between Lake Winnipegosis and Cedar Lake, ending south of The Pas. The moraine is characterized by southwesterly trending ridges and swales with a mean elevation of 279 m a.s.l. The southern edge of the moraine consists of a steep escarpment *ca.* 50 m in elevation. On the north side, the moraine has a gentle slope of *ca.* 1 m per km. Mesisolic organic soils are common in this area, along with gray luvisols and eutric brunisols. Shallow glacial deposits and limestone outcrops occur throughout the area.

Chitek Lake ecodistrict

This ecodistrict occurs south of The Pas moraine. This region is characterized by a north to south trending ridge-swale pattern. Level peatlands consisting of mesisolic soils, and glacial deposits and limestone outcrops, are also common. Gray luvisol soils are common on the ridges, while gleysolic and organic soils have developed in the swales. Eutric brunisols are also found over calcareous glacial till. Mean elevation is 259 m a.s.l., and relief changes are *ca.* 0.6 m per km.

3.1.3 Climate

Climatic data (mean monthly temperature and precipitation, 1966-1990) from the Grand Rapids Hydro meteorological station (53°09'N, 99°17'W) are summarized in Fig. 3.2. Mean annual temperature is 0.5°C, and July is the warmest month (18.8°C). Mean annual precipitation is 48.2 cm (11.6 cm snow, 36.6 cm rain). The highest rainfall occurs in June (7.5 cm), and November the greatest snowfall (2.3 cm) (Environment Canada 1995).

Vegetation sampling took place in June, 1994 when temperature averaged 15.1°C and 10.1 cm of rain fell. Demographic and phenological profile surveys occurred weekly from June to mid-August (1994), and May to September (1995). During these periods mean monthly temperatures were similar to long-term normals, but precipitation values deviated substantially from normal (Fig. 3.3, Fig. 3.4). Precipitation values in June and July of 1994 were above normal (10.1 and 12.8 cm in June and July respectively, compared to long-term normals of 7.5 and 7.5 cm), whereas August 1994 was unusually dry (1.9 cm compared to the normal 6.7 cm). In 1995, the opposite trend was seen. June and July were unusually dry (0.9 and 3.8 cm respectively), whereas August was wet (13.5 cm).

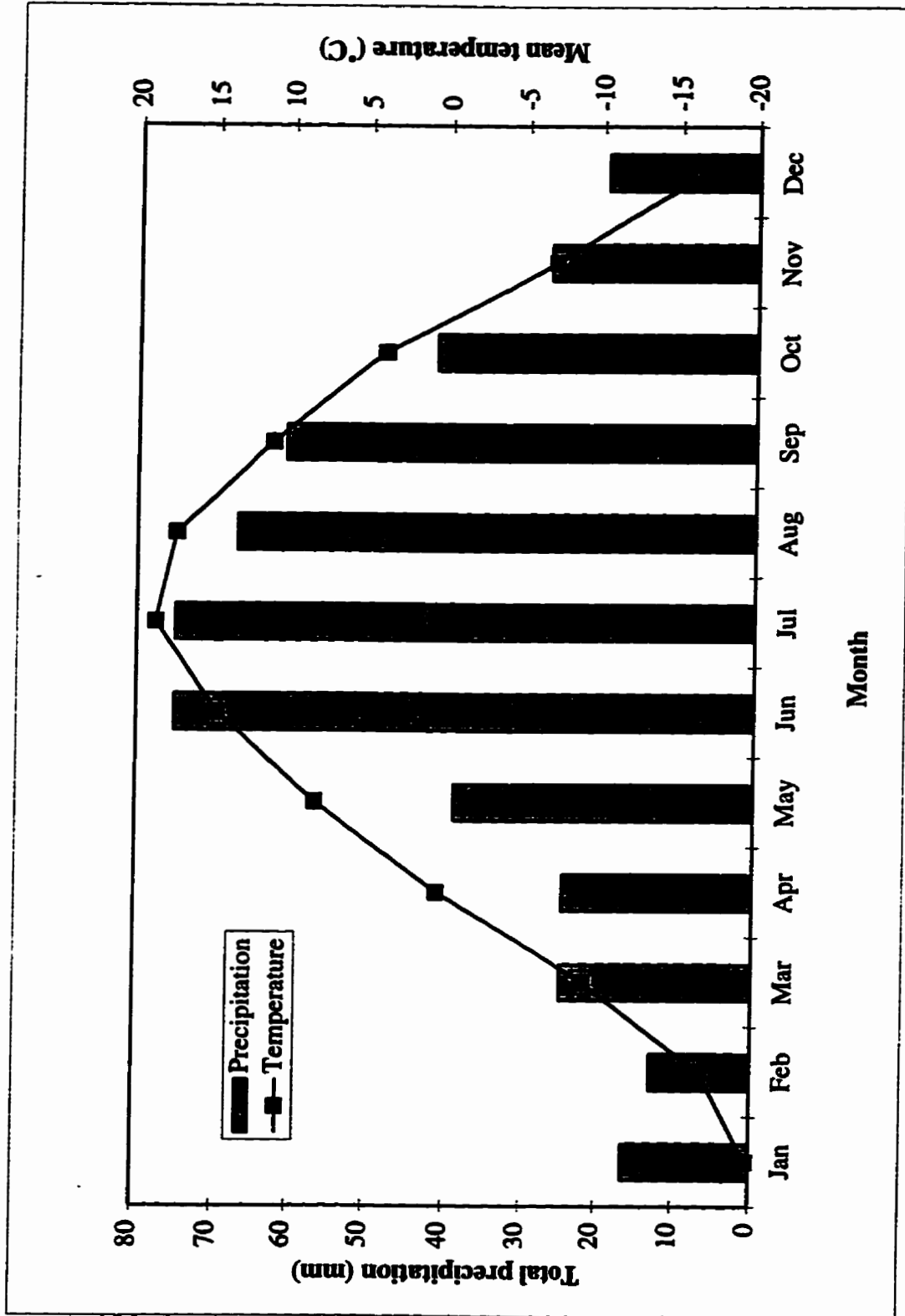


Figure 3.2. Mean temperature and total precipitation data for the Grand Rapids area, 1966-90 (Environment Canada 1995).

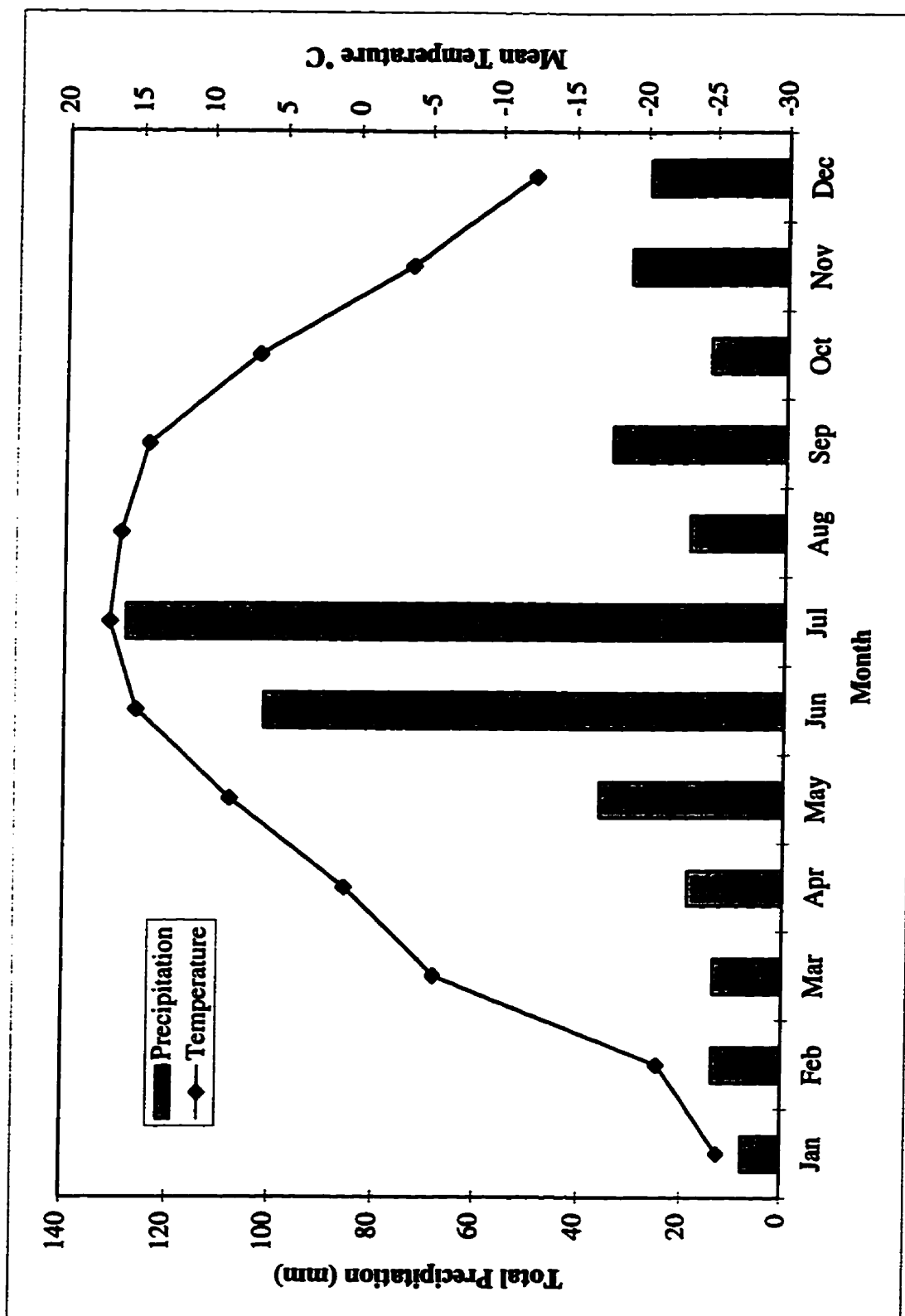


Figure 3.3. Mean temperature and total precipitation data for the Grand Rapids area, 1994 (Environment Canada 1995).

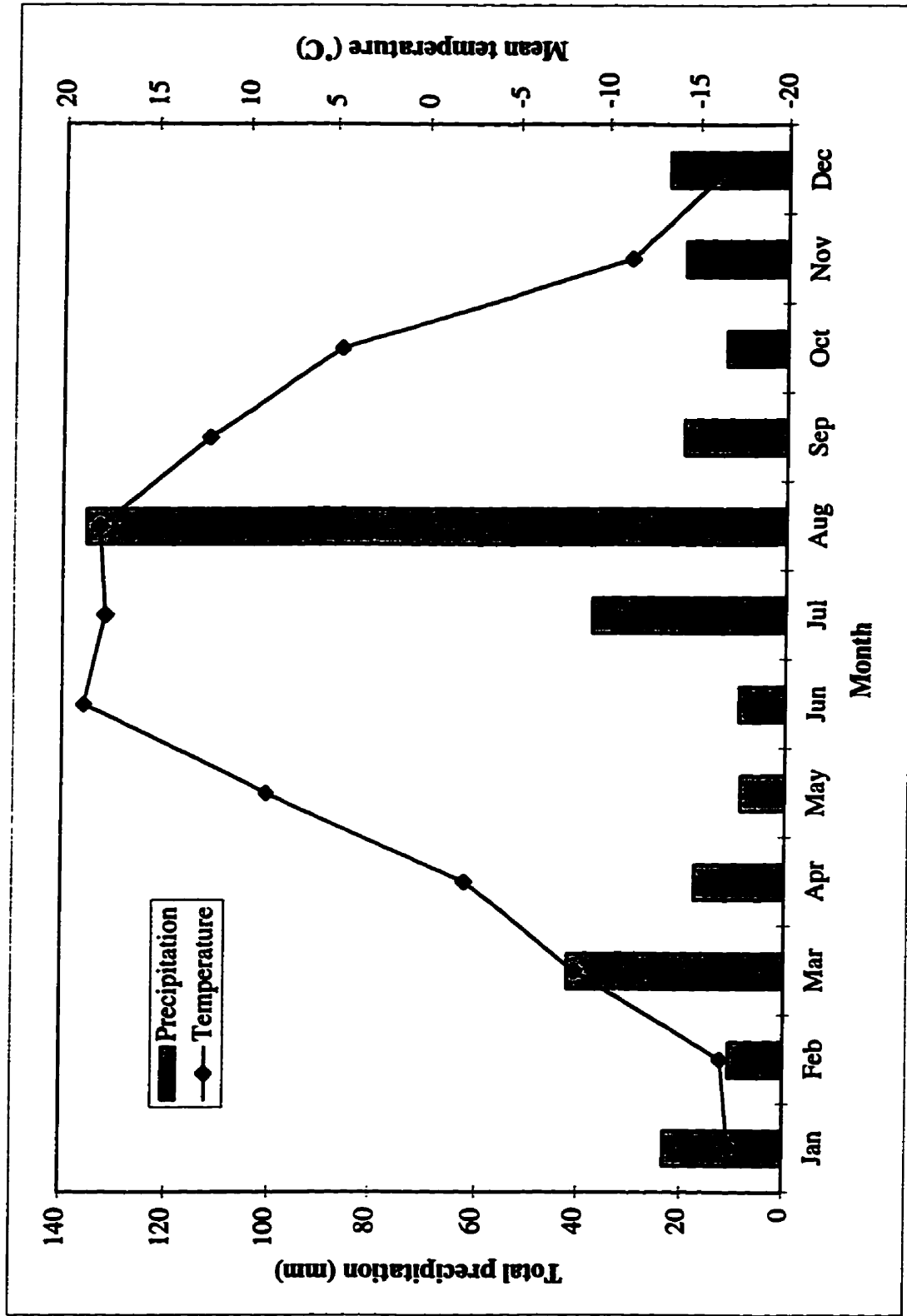


Figure 3.4. Mean temperature and total precipitation data for the Grand Rapids area, 1995 (Environment Canada 1995).

3.1.4 Forest fire history

Forest fire history for each site was obtained from Manitoba Forestry fire-history maps. Large-scale fires occurred in the 1920's and 1930's throughout the study area. More recent fires, which have been smaller and more localized, are detailed in Section 3.1.7.

3.1.5 Vegetation management

Over 70,000 km of electrical transmission and distribution lines are found in Manitoba. Vegetation management along these lines is essential to ensure safety and an uninterrupted supply of power (Manitoba Hydro 1992). Manitoba Hydro currently uses three vegetation management strategies: mechanical removal (tractors, chain saws); herbicides (hand or ground-operated equipment); and biological control (using favorable plant species to outcompete undesirable ones). The first method is currently the most widely used.

3.1.6 Site selection and names

Sites were chosen based on accessibility and the presence of seneca snakeroot. Ten study sites were intensively sampled, nine on the AC line and one on the HVDC line (Fig. 3.5). Seven sites were selected north of The Pas moraine, on the Cedar Lake plain. This area is primarily a dry, flat limestone plain dominated by stands of jack pine (*Pinus banksiana*). Seneca snakeroot is abundant along roadsides and on Manitoba Hydro rights-of-way throughout this area. An additional three sites were located in dry habitats south of The Pas moraine. Seneca snakeroot was rarely encountered on The Pas moraine and in adjacent poorly-drained areas. Individual site names are based on tower number and type closest to the study plots (e.g. site 176 AC refers to tower number 176 of the AC, lower voltage line).

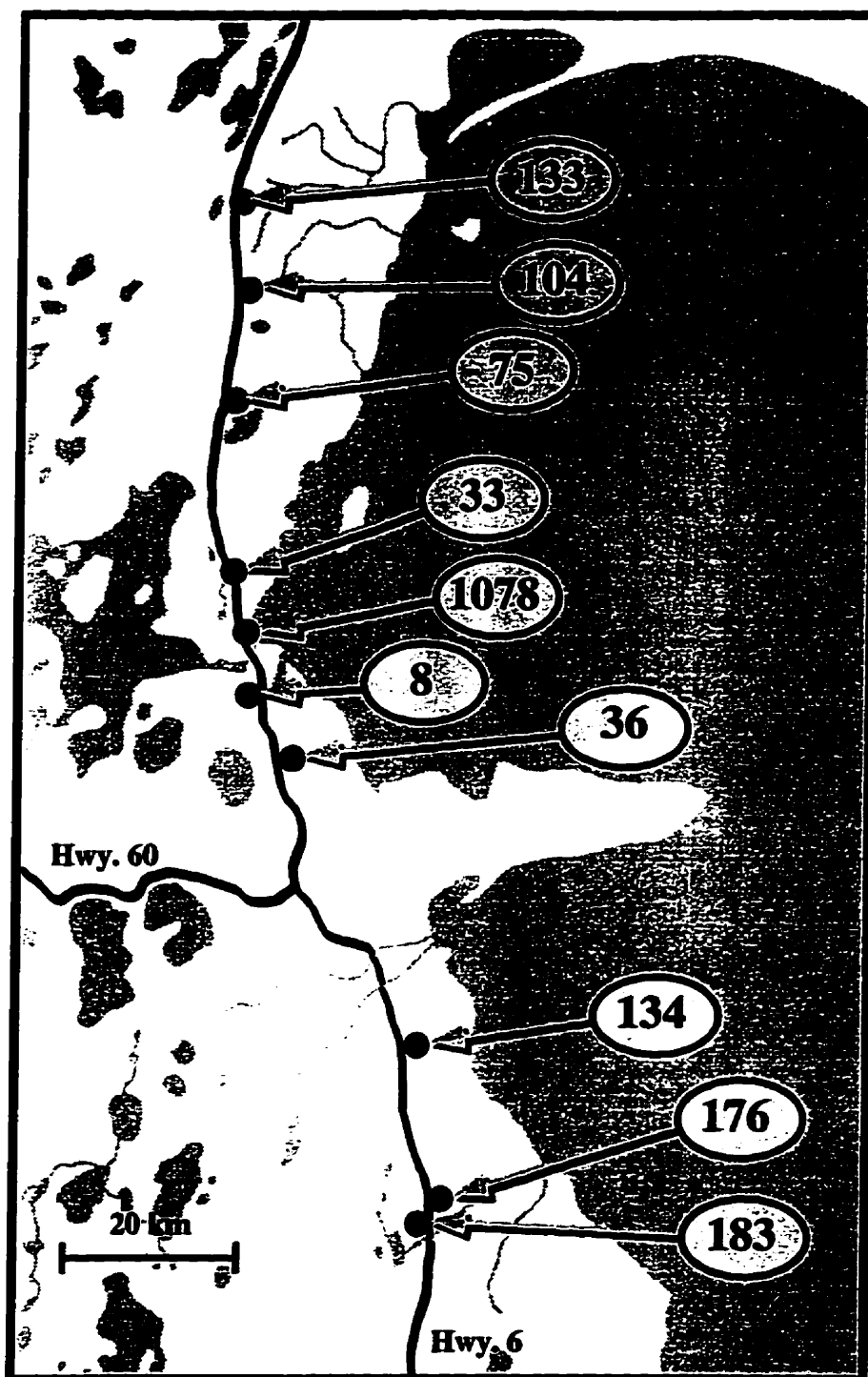


Figure 3.5 Map of study area showing site locations.

3.1.7 Site descriptions

The following site descriptions are based on personal observations, soil survey reports (Fraser *et al.* 1985), and Manitoba Forestry fire-history maps.

133 AC (53°40'N, 99°21'W)

This site is located *ca.* 54 km north of Grand Rapids. The dominant surface texture is loam with patches of bedrock, sand and gravel. The site is rapidly to well-drained and bordered by stands of jack pine on either side. The last recorded burn occurred in 1956. The right-of-way is unshaded and covered with rocky calcareous till.

104 AC (53°34'N, 99°21'W)

This site is located *ca.* 42 km north of Grand Rapids. The site is well to rapidly-drained and dominated by loamy soil, although limestone outcrops are also present. The area was partially burned in 1979 and is bordered by young stands of jack pine. The right-of-way is unshaded and rocky, and slopes slightly towards the north.

75 AC (53°28', 99°21'W)

This site is located *ca.* 30 km north of Grand Rapids near the junction of Highway No. 6 and the Buffalo Lake road. Mature jack pine to the east and a mixture of jack pine and trembling aspen to the west border the site. The last forest fire at this site was in 1937. The dominant soil is a rapidly to well-drained loam-sand-gravel mix. The right-of-way is open and rocky, with some shading from regenerating jack pine and aspen.

33 AC (53°16'N, 99°20'W)

This site, located *ca.* 10 km north of Grand Rapids near a gravel pit road, is bordered by stands of jack pine to the east and a mixed jack pine-black spruce stand to the west. The dominant soil is a rapidly to well-drained loam-sand-gravel mix. This area burned in 1929 and again in 1937.

1078 DC (53°12'N, 99°17'W)

This site, located *ca.* 1 km north of Grand Rapids, is the only one located along the DC line. It is bordered by jack pine and black spruce, with the occasional balsam poplar. A fire occurred nearby in 1976, though there is no evidence of fire in the immediate vicinity of the site. The dominant soil is a rapidly to well-drained loam-sand-gravel mix. The right-of-way itself is rocky and unshaded.

8 AC (53°15'N, 99°20'W)

This site is located *ca.* 5 km south of Grand Rapids and about 1 km down the Wayside Road. It is bordered by stands of jack pine, though black spruce and balsam poplar are also present. The soil is a well to rapidly-drained sand-gravel mix. The area was burned in 1937 and again in 1961. The right-of-way is grass-covered and somewhat shaded by regenerating jack pine and trembling aspen.

36 AC (53°5'N, 99°15'W)

This site is located *ca.* 15 km south of Grand Rapids, in a recent burn area (1989, and previously in 1963). The site is well to imperfectly-drained, with loam and mesic peat

depressions predominating. The right-of-way itself is relatively well-drained, open and unshaded.

134 AC (52°48'N, 98°58'W)

This site is located *ca.* 117 km north of St. Martin Junction. The area is characterized by well to imperfectly-drained clay deposits. The forest bordering the site consists of a mixture of white spruce, jack pine and balsam poplar. The area was burned in 1929, and a smaller fire in 1970 may have also burned the study site. The right-of-way is a grassy, slightly rolling meadow, with some shading from shrubs, tall herbs and grasses.

176 AC (52°36'N, 98°54'W)

This site is located *ca.* 103 km north of St. Martin Junction. The predominant soil is a calcareous, clay-stony till. The area is level to undulating, and imperfectly to well drained. The right-of-way is bordered by stands of jack pine with some white birch. A large fire occurred in 1929, and the site may also have been burned by a smaller fire in 1970.

183 AC (52° 32' N 98° 54' W)

This site is located *ca.* 100 km north of St. Martin Junction. The topography is level to undulating, with imperfectly to well-drained clay-loam to stony till. This site was burned in the huge fire of 1929, and possibly again in 1971. The right-of-way is bordered by stands of jack pine, with young white spruce along the forest edge.

3.2 Field sampling

The ten study sites were sampled to obtain baseline information on the ecology and population biology of seneca snakeroot in Manitoba's Interlake region. Vegetation sampling was undertaken to obtain information on the plant communities in which seneca snakeroot is found, and to quantify seneca snakeroot abundance, demography and biomass. Environmental information (soil and habitat descriptions) were also obtained for each study site.

3.2.1 Habitat descriptions of sites

Descriptions of seneca snakeroot habitats were obtained by recording information on the forest vegetation bordering rights-of-way in which seneca was found. Local topography, shading, and soil moisture conditions were also noted.

3.2.2 Soil sampling and analysis

Three replicate soil cores (8 x 8 x 12 cm in depth) were randomly taken from each of the ten study sites (June 1994) in areas where seneca snakeroot was found. Fresh samples were stored in a cooler in the field and transferred to a cold-room at the University of Manitoba. The soil was then prepared for nutrient, conductivity and pH analysis by Norwest Labs, Winnipeg. Total nitrogen (N) was analyzed by using a CaCl_2 extract and automated colorimetry. An ammonium acetate/acetic fluoride extract and automated molybdate colorimetry was used to determine total phosphorus (P). Potassium (K) was determined using flame photometry and an ammonium acetate/acetic fluoride extract. Sulfur was analyzed by using a CaCl_2 extract and methyl thymol blue automated colorimetry. Soil conductivity and pH were determined using a standard water extract (Norwest Labs 1994).

3.2.3 Vegetation transects

Transects were used to determine the relative abundance of seneca snakeroot, and to provide a summary of associated plant species. At each of the ten sites, two 20 m line transects were randomly placed in areas where seneca snakeroot was present. Percent cover of vegetation was recorded in 1 x 2 m rectangular quadrats placed at every other meter along the transect (20 quadrats/site). Cover estimates were made in late June, 1994.

3.2.4 Demography and phenology

Although it is impossible to determine the age of seneca snakeroot plants, the number of shoots (plant 'size') undoubtedly increases as plants age. Number of shoots per plant was therefore used to obtain demographic profiles of seneca snakeroot populations. At each study site, a 10 x 10 m semi-permanent plot, divided into 100 1 x 1 m grids, was established. Each grid was sampled by recording the location of all seneca snakeroot plants, and counting the number of shoots of each plant. In addition, ten randomly selected plants were permanently marked in each of the 10 semi-permanent plots. All sites were visited weekly from mid-June 1994 to late August in 1994, and again from May to September in 1995, and the condition of marked plants (e.g. flowering, seed production, etc.) was recorded to obtain a phenological profile of the species.

3.2.5 Above and below-ground biomass

Above and below-ground biomass allocation in seneca snakeroot was determined by carefully digging up living plants. A total of 56 plants of various size were harvested in this

way in 1994, from a number of locations. Above and below-ground biomass were separated, dried in an oven at 80° C and massed.

3.2.6 Seed collection and storage

Seed capsules were collected from mid-July to early August (1994 and 1995) from all sites. Capsules were removed by hand, stored in paper bags and brought back to the laboratory, where they were carefully spread out to dry. Drying opened the capsules, releasing the seeds (two per capsule). The seeds were picked out and stored in sealed glass vials at 2°C for later use in germination experiments. The mean weight of 1000 seeds was also determined.

3.3 Laboratory experiments

Laboratory experiments were undertaken to: (1) determine the seed bank present in areas where seneca snakeroot occurs; (2) to investigate seed germination requirements of seneca snakeroot; (3) to investigate methods for vegetative propagation of seneca snakeroot.

These experiments were undertaken in the growth-chamber and greenhouse facilities of the Botany Department, University of Manitoba. A Conviron CMP-3023 growth chamber was used in seed germination and vegetative propagation experiments. The chamber was set at 24°C with 12 hours of light per day. Mean illumination was *ca.* 51 watts/m², from a bank of fluorescent and incandescent bulbs. Two greenhouse areas were used for growing plants. The south-west greenhouse had fluorescent bulb lighting, with a mean illumination of *ca.* 12 watts/m². The south-east greenhouse had similar conditions, but illumination was somewhat higher at *ca.* 17 watts/m². A section of the main greenhouse with high-intensity sodium lights was used for seed bank, seedling and plant propagation experiments. This

greenhouse has a mean illumination of *ca.* 57 watts/m². All the above greenhouse light intensity readings were taken on a clear sunny day in January.

3.3.1 Seed bank

Sampling was undertaken to determine soil seed bank composition, and to determine whether the soil contained germinable seeds of seneca snakeroot. Five soil seed bank samples were taken at each site in July, 1994. Sampling involved taking three scoops of soil, using a garden trowel, from beneath the flowering shoots of mature seneca plants. The scoops were placed in a labelled plastic bag and kept refrigerated. In the laboratory, ten random sub-samples of 70 ml of soil were mixed with 1 liter of water to form a slurry (Shaw 1993). This slurry was carefully poured over a mixture of 2 parts sterilized soil, 1 part peat and 1 part perlite contained in 15 cm wide plastic pots. The 100 pots were then placed in the University of Manitoba greenhouse and watered regularly. Species were recorded and removed upon identification. The experiment ran for eight months.

The above procedure was repeated in September 1995, but half of the soil sample was placed into a 2°C cold-room for two months before potting (cold-stratification treatment). This experiment also ran for eight months.

3.3.2 Seed germination

Numerous experiments (using varied amounts of seeds and repetitions) were undertaken to determine the germination requirements of seneca snakeroot seed (summarized in Table 3.1). Most germination trials took place under light, since preliminary experiments indicated that seeds failed to germinate in the dark. Removal of the aril (elaiosome) from the seed did not appear to increase germination success. A number of other treatment

Table 3.1. Description of techniques used in seed germination trials.

Technique	Description
Anti-fungal treatment	<ul style="list-style-type: none"> •surface sterilization with bleach:water solution (1:20 parts) •surface sterilization with 'No Damp' •alcohol surface sterilization
Aril removal	<ul style="list-style-type: none"> •external appendage (elaiosome) removed from seeds
Cold-stratification	<ul style="list-style-type: none"> •seeds placed between layers of moist sand for 6-8 weeks (2 °C)
Germination medium	<ul style="list-style-type: none"> •petri dishes + filter paper dampened with water •petri dishes + filter paper dampened with gibberellic acid •pots with various combinations of soil mixtures
Growth environment	<ul style="list-style-type: none"> •window sill •growth chamber •greenhouse
Scarification	<ul style="list-style-type: none"> •seed coat slit with scalpel (two slits) •seed coat partially removed with scalpel (cut)
Seed storage	<ul style="list-style-type: none"> •room temperature (fresh seeds) •cold storage (2°C) •freezer storage (-2°C)
Water-soaked	<ul style="list-style-type: none"> •seeds soaked overnight in water to soften seed coat

combinations involving various mixes of germination medium, anti-fungal treatment, scarification, and cold stratification were attempted, as summarized below.

Germination medium

Two germination media were used: (1) damp filter paper in petri dishes; (2) greenhouse soil mixes. The first involved placing seeds on damp filter paper in parafilm-sealed, 9 cm diameter glass petri dishes. These petri dishes were kept in a controlled growth chamber (24°C, 12 hours of light, mean illumination of *ca.* 51 watts/m²). The second method involved sowing seeds on the surface of soil placed in pots. Pots were kept either in the greenhouse, or in a controlled environment chamber.

Anti-fungal treatment

In early experiments, seeds were often attacked by an unidentified fungus before they had a chance to germinate. To overcome this problem, seeds were surface-sterilized. This involved washing the seed in a 20:1 water/bleach solution for 1-2 minutes, and then rinsing with pure water. In most experiments, seeds were soaked in pure water for one day prior to the bleach treatment.

Scarification

Scarification involves scratching or cutting the seed coat to promote germination. Seneca snakeroot seeds were first soaked overnight to soften the seed coat. Under a dissecting microscope, the seed was carefully held with tweezers and the seed coat slit open using a sharp scalpel. Later, as much as possible of the bottom half of the seed coat was removed (see Fig. 3.6), since early experiments demonstrated that this increased germination.

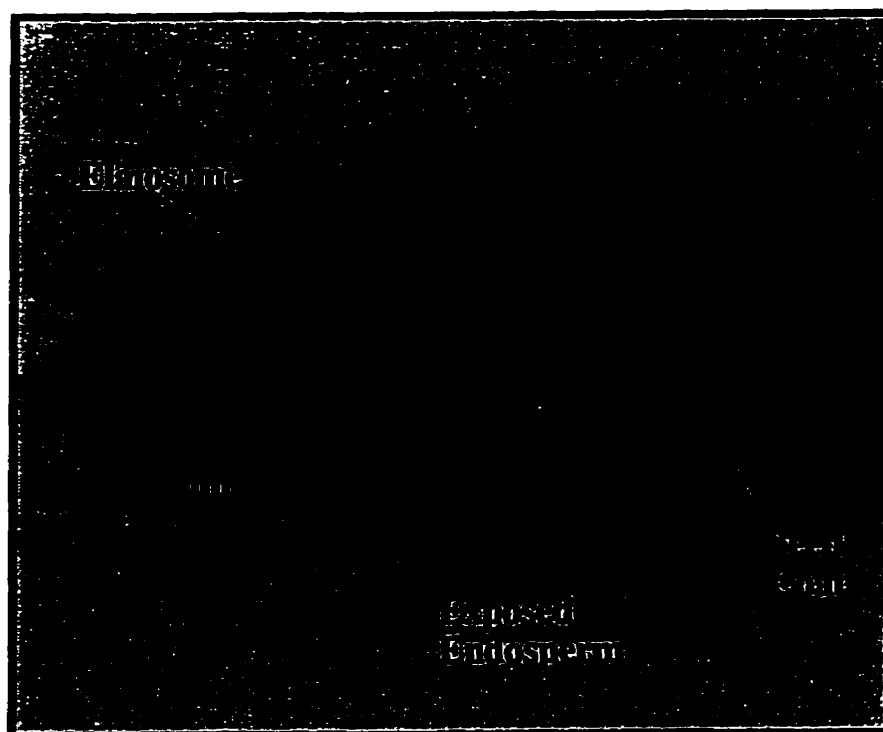


Figure 3.6. Scarified seeds of seneca snakeroot.

Cold-stratification

Cold-stratification involved placing seeds between layers of moist sand, and exposing them to near-freezing temperatures for at least 6-8 weeks. This treatment simulates conditions experienced by seeds in winter. In this experiment, seneca snakeroot seeds were placed between a double layer of cheese cloth covered with moist sand (Fig. 3.7), and kept in a cold-room (mean temperature of 2°C) for periods ranging from two to six months. The sand was kept moist with regular waterings.

Gibberellic acid

Gibberellic acid is a hormone that controls plant growth and development. This hormone is found in actively-growing areas of the plant such as the embryo. Gibberellins promote seed germination, stem and leaf development, flower and fruit development, root growth, and cell differentiation. Seeds that require specific environmental conditions to germinate will often break dormancy when treated with gibberellic acid (Bidwell 1974; Campbell 1993). In this experiment, gibberellic acid was used in varying concentrations (0.1, 1, 10, 25, 50 and 100 mg/L) in an attempt to promote the germination of seneca snakeroot seeds.

3.3.3 Vegetative propagation

Vegetative propagation has two major advantages over seed germination: (1) propagated individuals are identical to their 'parent'; (2) plants are developmentally advanced and will grow more quickly. A number of vegetative propagation methods were attempted, as summarized below (see also Table 3.2).

(1)



(2)



Figure 3.7. (1) Cold-stratification trays of seneca snakeroot seeds. The seeds are layered between moist sand and placed in a cold room (2°C) for two to six months. (2) Diagrammatic illustration of seneca snakeroot seed stratification.

Table 3.2. Summary of vegetative propagation experiments.

<u>Root cuttings</u>	<u>Source</u>	<u>Soil combinations</u>
Cut root into pieces about 5 cm in length Potted entire root from field	Greenhouse (collected from field previous year) Freshly collected from field	2 sand:2 peat:1 soil 1 sand:1 peat greenhouse mix
<u>Shoot cuttings</u>	<u>Source</u>	<u>Soil combinations</u>
Non-flowering shoots (2-5 cm in length) Non-flowering shoots (2-5 cm in length) + root starter Flowering shoots + root starter	Greenhouse (collected from field previous year) Freshly collected from field	1 sand : 1 peat greenhouse mix 1 peat:1 sand:1 soil:1/2 leaf mold
<u>Root division</u>	<u>Source</u>	<u>Soil combinations</u>
Cut entire plant in 1/2 vertically	Freshly collected from field	greenhouse mix 1 peat:1 sand:1 soil:1/2 leaf mold

Root cuttings

Entire plants were dug up, transported to the greenhouse, and potted. After one year of growth, pieces of the taproot from these plants were cut into 2-5 cm long pieces. These root pieces were planted into soil, or placed on the soil surface, in plastic growth trays. The trays were then placed in the greenhouse and watered regularly.

Shoot cuttings

Shoot pieces of varying length were cut from living seneca snakeroot plants, at different phenological stages (e.g. newly elongating shoots, mature (flowering) shoots). These were planted into soil in 15 cm diameter pots. Some shoots were first dipped in commercial root starter in an effort to promote root growth. The root starters used were 'Stim-Root No. 1' (Plant Products Co. Ltd.), which contains 0.1% IBA, and 'Roots' (Wilson Laboratories Ltd.), which contains 0.4% IBA and a fungicide. Pots were placed in the greenhouse and watered regularly.

Whole-plant division

Living seneca snakeroot plants were carefully dug up from the field and immediately transported to the University of Manitoba greenhouse. Whole plants were cut in half vertically and each half repotted. Pots were placed in the greenhouse and watered regularly.

3.4 Field experiments

A series of field experiments was undertaken to investigate some of the factors limiting growth and germination of seneca snakeroot. Manipulative experiments were used to

investigate whether the addition of macronutrients, or the removal of competing vegetation, would lead to an increase in seneca snakeroot growth. A third experiment involved sowing fresh seed into prepared plots in the field to determine *in situ* germination success. These experiments are described in greater detail below.

3.4.1 Nutrient experiment

Macronutrients (nitrogen-potassium-phosphorus) were added to plants growing in the field to determine whether wild populations are nutrient-limited. This experiment was undertaken at three of the study sites (183, 33, and 133). At each of these sites, plants of similar size (total number of shoots) were paired and marked in the field. Paired plants (10 pairs at site 183, 5 pairs at 33, and 15 pairs at site 133) were *ca.* 1 m apart. One plant in each pair was randomly selected for nutrient addition, while the other served as a procedural control. Every two weeks (from June 1- 23, 1995), treated plants were watered with 2.0 L of commercial 20-20-20 (N-P-K) nutrient solution. Control plants received 2.0 L of water without added nutrients. Above-ground biomass of each plant was harvested in mid-summer (July 6, 1995), dried at 80°C, and massed.

3.4.2 Competition experiment

This experiment was undertaken to determine whether removal of competing vegetation resulted in increased growth of seneca snakeroot plants. This experiment was performed at sites 75, 1078 and 8. Ten pairs of size-matched plants were selected at each site and marked. Treated plants had the above-ground biomass of potential competitors (defined as a plant within a 0.5 m radius of the seneca plant) removed every week, beginning in June 1, 1995. Control plants were left untouched. Above-ground biomass of each plant was harvested in mid-summer (July 6, 1995), dried at 80°C, and massed.

3.4.3 Sowing of seed under field conditions

Site 176 was selected for the seed sowing experiment. On June 1, 1995 a grid of six 1 x 1 m plots was marked out with stakes. All plants were removed from within the plots, and the soil was tilled with a hoe to a depth of *ca.* 15 cm. Twenty-five non-scarified seneca seeds were sown into each of three plots, while the remaining three plots were sown with twenty-five scarified (by slicing the seed coat) seeds. All plots were lightly watered immediately after the seeds were sown. Plots were weeded and carefully monitored for evidence of seneca germination and seedling establishment every week until the end of August.

3.5 Data analysis

3.5.1 Competition and nutrient experiments

For both the competition and nutrient experiments, differences in above-ground biomass between the two treatments were tested using two-sided, paired t-tests. The null hypothesis is that mean biomass values for the treatments are not statistically different.

3.5.2 Multivariate analysis of seneca habitats

Correspondence analysis ordination was used to summarize the vegetation composition of the ten study sites. Ordination methods are used to efficiently represent and summarize the major trends present in a complex, multivariable data set. The results are presented in the form of a two-dimensional ordination biplot, in which the sites and species are placed in accordance to their relative communality. The ordination program CANOCO (ter Braak 1987) was used to perform the analysis.

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Ecology of seneca snakeroot

4.1.1 Habitat preferences

Extensive field reconnaissance surveys were undertaken along rights-of-way, highways and in natural areas of the northern Interlake to determine the habitat preferences of seneca snakeroot. The species occurs abundantly throughout the region in dry, unshaded sites. It shows a clear preference for calcareous ($\text{pH} > 7$), limestone-based soils, and is rarely found on the acidic substrates of the Canadian Shield. Seneca snakeroot also shows a strong preference for well-drained soils. While it does occur in moderately-drained sites, it is not found in wet, low-lying, boggy sites. Typically, the species is found adjacent to dry, upland stands of jack pine and trembling aspen. Seneca snakeroot is also found in the native grasslands of southern and western Manitoba.

Seneca snakeroot appears to be quite shade-intolerant. It is abundant in unshaded or lightly shaded areas such as cleared rights-of-way, but becomes infrequent in more shaded habitats such as closed forest stands and unmanaged rights-of-way. The species is common in open areas, which suggests that it is may not be a particularly strong competitor. V-blading (the mechanical management of vegetation under Hydro lines) creates open habitats that are favourable to seneca snakeroot. Dry, recently burned sites are also colonized by seneca snakeroot (e.g. site 36). The abundance of seneca snakeroot in the northern Interlake is attributable to a combination of the calcareous soils, and disturbances that open up habitats for the species.

4.1.2 Soil relations

Soil macronutrient status, conductivity and pH for the ten study sites are summarized in **Table 4.1**. Soils are basic (mean pH = 8.1, range 7.5 - 8.5), indicating calcareous conditions. Mean conductivity is *ca.* 0.3 dS/m, indicating non-saline conditions (values < 1 dS/m are considered non-saline in agricultural soils). Mean nutrient values for nitrate-nitrogen, phosphate, potassium and sulfate were *ca.* 4.2, 2.6, 120.3 and 2.9 ppm respectively. Using agricultural guidelines, these soils would be considered highly deficient in nitrogen and phosphorus (J. Hicks, Norwest Labs, pers. comm.). This suggests that seneca snakeroot can tolerate macronutrient deficiencies.

4.1.3 Associated vegetation

Mean percent cover estimates for the most common species, and for bare ground, are summarized in **Table 4.2** (see **Appendix II** for the complete data set). Total vegetation cover was < 50% in most sites, particularly in the sites north of Grand Rapids (where exposed rock limited plant colonization). Mean cover of seneca snakeroot was 3.15%. Common associates include bearberry (9.78%), sedges (mainly *Carex aurea* and *C. richardsonii*, 9.67%), lichens (mainly *Cladina* spp. and *Cladonia* spp., 7.05%), wild strawberry (5.10%), smooth aster (4.89%), northern bedstraw (4.85%), bryophytes (mostly red-stem moss, 3.32%), American vetch (2.73%), northern reed grass (2.34%), and yarrow (2.20%).

A correspondence analysis ordination biplot summarizing plant community relationships between the ten sites is presented in **Fig. 4.1**. Species common to all sites, such as seneca snakeroot, yarrow, wild strawberry, smooth aster and northern bedstraw, are found near

Table 4.1. Results of soil analysis.

Site no.	Nitrate (ppm)	Phosphate (ppm)	Potassium (ppm)	Sulphate (ppm)	pH	Conductivity (dS/m)
133	7.0	3.0	78.0	3.0	8.1	0.4
176	14.0	3.0	76.0	4.0	7.5	0.2
36	1.0	4.0	191.0	5.0	8.1	0.4
33	5.0	3.0	79.0	4.0	8.0	0.4
183	1.0	3.0	195.0	3.0	7.9	0.2
134	<1	2.0	305.0	2.0	8.2	0.4
75	9.0	2.0	88.0	1.0	8.2	0.4
104	0.5	2.0	59.0	3.0	8.0	0.2
8	1.0	2.0	89.0	3.0	8.1	0.2
1078	3.0	2.0	43.0	1.0	8.5	0.2
Mean	4.6	2.6	120.3	2.9	8.1	0.3
Standard deviation	4.6	0.7	82.9	1.3	0.3	0.1

Table 4.2. Species commonly associated with seneca snakeroot.

Species	Common name	Mean % cover per site										Mean % cover
		133	104	75	33	1078	8	36	134	176	183	(all sites)*
<i>Polygala senega</i>	Snakeroot	3.45	1.63	2.45	4.00	2.75	3.75	0.50	5.75	1.38	5.70	3.15
<i>Achillea millefolium</i>	Yarrow	2.32	0.25	2.38	1.88	1.50	2.75	3.00	3.38	2.38	2.15	2.20
<i>Arctostaphylos uva-ursi</i>	Bearberry	10.22	27.38	0.88	10.50	14.95	14.88	2.63	4.63	6.38	5.13	9.78
<i>Aster laevis</i>	Smooth blue aster	3.73	1.63	2.15	3.00	2.48	4.50	5.88	6.25	8.75	11.13	4.89
<i>Calamagrostis inexpansa</i>	Northern reed grass	0.38	0.25	3.00	0.25	1.50	3.75	8.25	1.75	4.50	0.75	2.34
<i>Carex aurea</i>	Golden sedge	9.83	0.00	5.00	0.00	7.38	0.00	0.00	0.00	0.00	6.80	2.58
<i>Carex richardsonii</i>	Richardson's sedge	0.00	6.75	27.25	14.00	4.50	6.75	1.75	1.88	4.75	0.00	6.44
<i>Fragaria virginiana</i>	Wild strawberry	3.33	1.05	4.25	5.63	8.30	3.25	2.38	5.13	12.75	5.78	5.08
<i>Galium boreale</i>	Northern bedstraw	2.87	1.75	5.60	4.38	2.63	3.50	10.88	2.75	8.25	6.90	4.85
<i>Vicia americana</i>	American vetch	0.45	0.00	0.00	0.00	0.00	0.63	13.38	3.75	2.00	8.25	2.71
<i>Cladina spp, Cladonia spp</i>	Lichens	23.80	4.88	0.25	2.63	7.13	11.13	6.50	0.88	3.63	1.38	7.05
	Bryophytes	1.38	2.75	3.75	6.00	6.88	5.50	7.25	0.00	0.25	0.40	3.32
	Rock	19.85	20.25	50.25	14.00	11.25	14.13	23.88	0.00	6.75	0.00	16.22
	Bare ground	6.18	0.00	0.00	0.00	0.00	0.00	0.00	9.88	0.00	4.35	2.24
	Litter	11.55	17.00	2.75	40.25	24.25	26.25	61.00	49.00	55.25	46.65	32.35
	Dead wood	11.67	0.00	0.25	0.00	0.00	0.00	0.00	0.00	0.00	4.58	2.13

* Mean % cover per site may not add up exactly to mean % cover overall since site 133 is based on 3 transects.

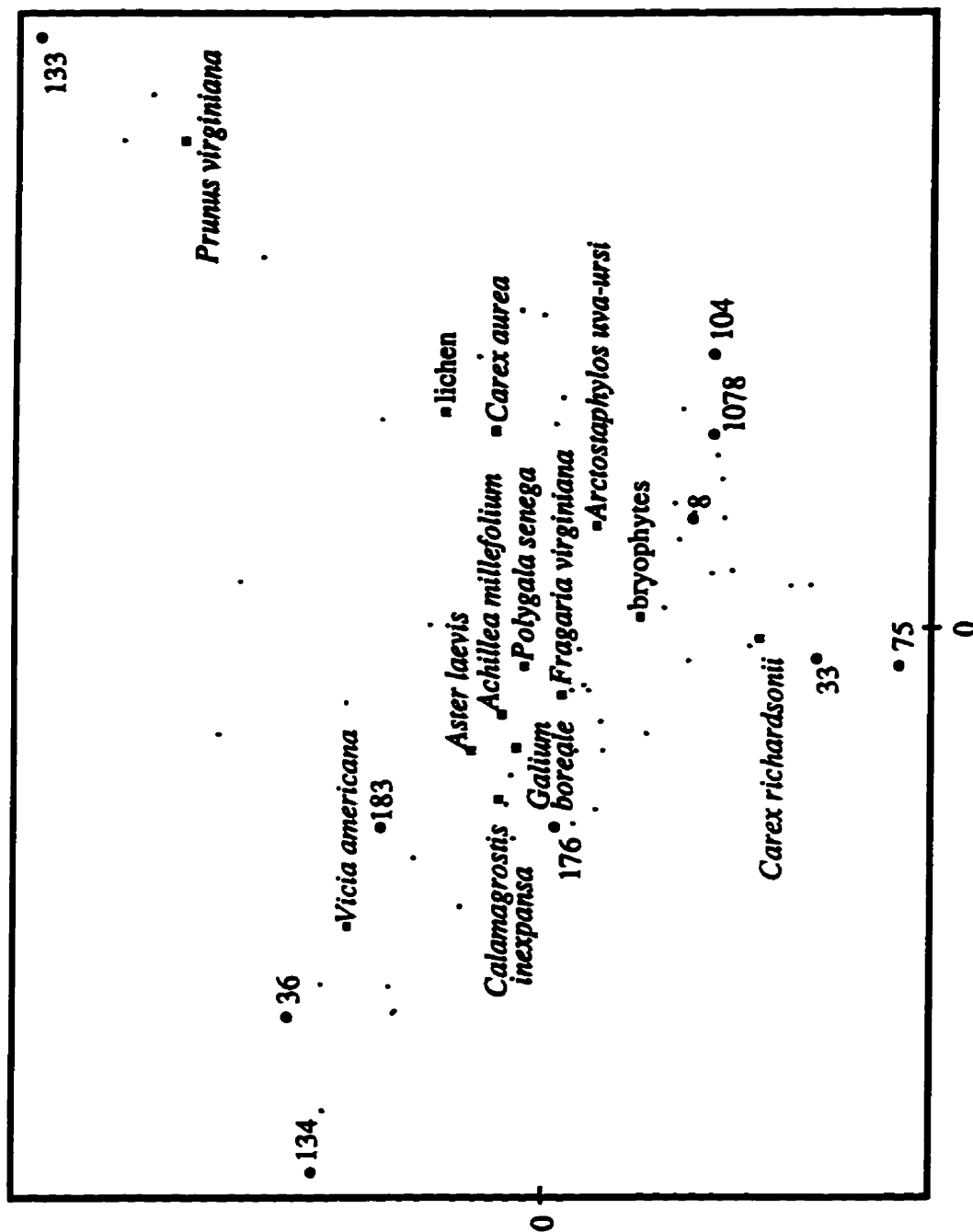


Figure 4.1. Ordination biplot of species and sites with characteristic species labelled.

the center of the ordination diagram. The most northerly site (133) occurs on its own at the top right of the ordination diagram. It has higher lichen cover, and higher shrub abundance, than the other sites. The other northern sites (8, 33, 75, 104 and 1078) occur as a group at the bottom right of the ordination diagram. These sites are characterized by high cover of Richardson's sedge, and bryophyte species. The most southerly sites (183, 176, 134 and 36) occur at the upper left of the ordination diagram. These sites are characterized by higher cover of species characteristic of grasslands, such as smooth aster, northern reed grass, northern bedstraw and American vetch. The results indicate that the southern portion of the right-of-way is dominated by grassland-like vegetation, whereas the northern sites are characterized by boreal species that are tolerant of dry conditions, such as bearberry, bryophytes and lichen species.

4.2 Population biology of seneca snakeroot

4.2.1 Phenology

A phenological profile of seneca snakeroot populations in the northern Interlake region of Manitoba is summarized in **Fig. 4.2** (based on detailed survey data, **Appendix III**). New shoot growth begins in early spring (late April to early May, depending on prevailing soil and air temperatures). These newly emerging shoots are dark purple in colour, with scale-like leaf primordia. Bright green elliptical-shaped leaves form as the shoot elongates. The base of the shoot is scaly and retains its purplish colour. Flowering begins in late May and lasts into early July. The flowering head first appears as a dense, green-coloured, cone-shaped structure at the end of each shoot. The white flowers first appear at the base of the flowering portion of the shoot. The flowering head is cylindrical in shape when the plant is in full bloom, and the sepals turn a pinkish colour. Fruits (two-locular green dehiscent capsules) first appear in early July. Approximately 30 days later, the capsules fall

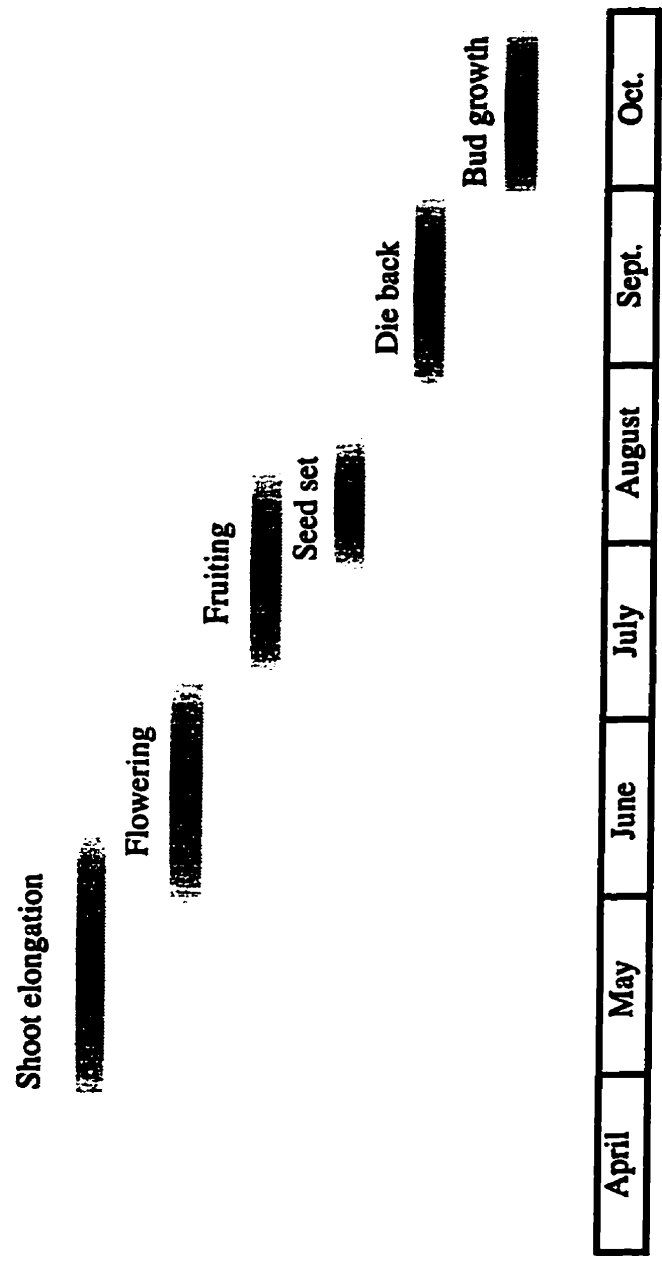


Figure 4.2. Phenological profile of *Polygala senga* L. (1994, 1995 pers. obs.)

to the ground and the shoots are bare of capsules by early to mid-August. The shoots begin to die back in late August to early September. Dead shoots (from previous years) often remain attached to the rootstock until they rot away. New shoot buds develop in late fall (September - October) and over winter until spring.

4.2.2 Population density

Density of seneca snakeroot plants, and the mean number of shoots per plant, are summarized across the ten study sites in **Table 4.3**. Mean density over all sites was *ca.* 2 plants/m² (ranging from 0.47-4.90 plants/m²). The mean number of shoots per plant, over all 10 study sites, was *ca.* 5 shoots/plant (based on a sample of 1863 plants). The smallest plants were found at site 134, which also had the highest plant density. In general, sites with the largest plants (e.g. sites 133 and 1078) were rocky, making the digging of plants difficult. Site 36, which was less well-drained than the other sites, had the fewest plants.

4.2.3 Demography - size and spatial structure

Size-frequency histograms for the ten seneca snakeroot populations are presented in **Fig. 4.3**. The number of shoots/plant showed a strongly L-shaped (positively skewed) distribution at all sites. That is, the populations are characterized by a large number of small plants but comparatively few large plants. This result suggests that: (1) seneca snakeroot populations successfully regenerating by seed, since there are a large number of small plants; (2) older plants are either dying naturally, or are being selectively harvested. The largest plant found, based on a sample size of 1863 plants, had 70 shoots (site 1078). The majority of plants had fewer than 20 shoots, however.

Table 4.3. Population density and mean number of shoots of seneca snakeroot at various sites.

	Site	Density /m ²	Mean No. Shoots
North	133	3.54	5.88
	104	1.01	4.37
	75	2.08	6.23
	33	0.91	5.42
	1078	1.15	8.17
South	8	1.38	4.54
	36	0.47	4.83
	134	4.90	3.32
	176	0.99	6.36
	183	3.20	3.86

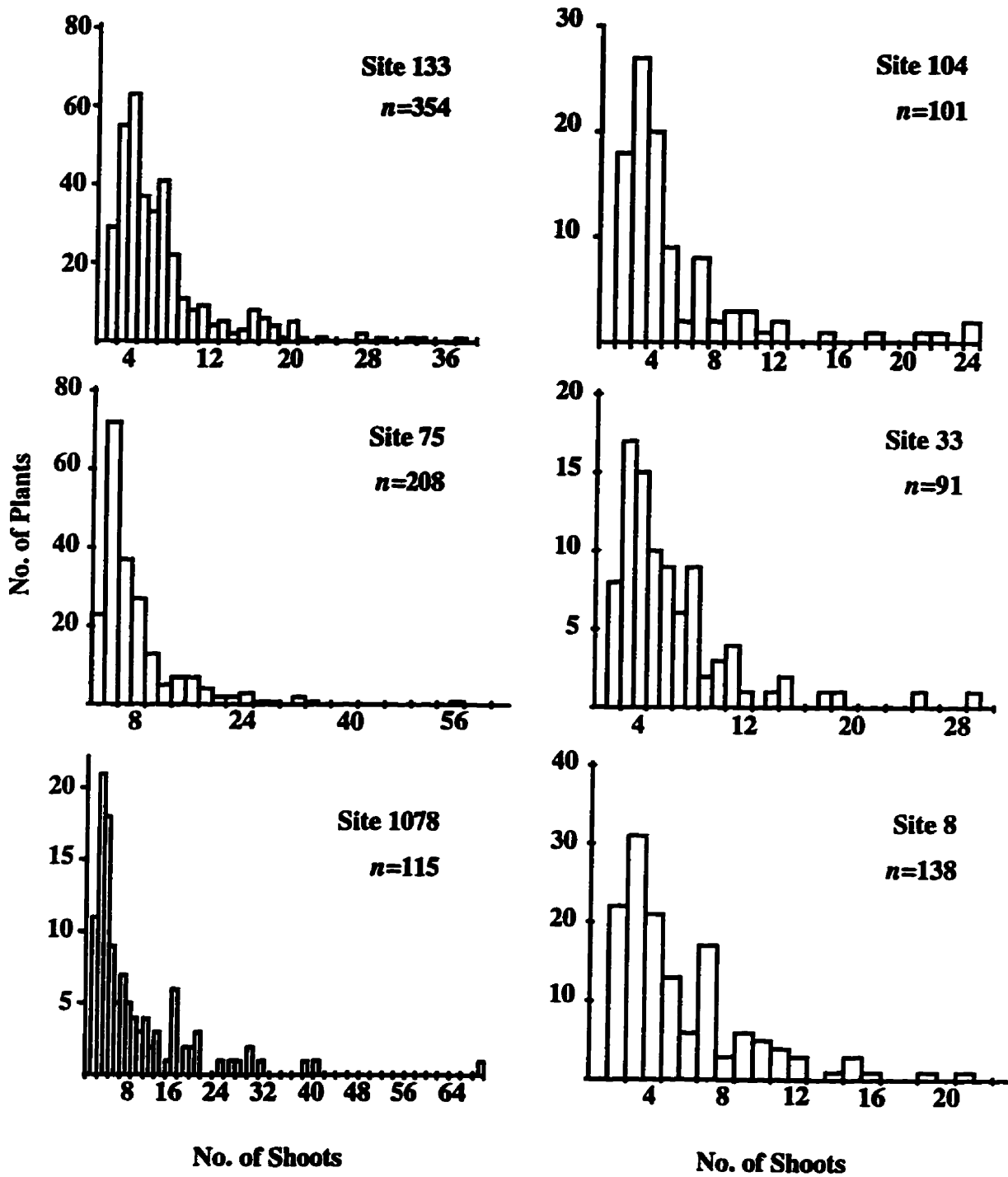


Figure 4.3. Size-frequency histograms for the 10 study sites.

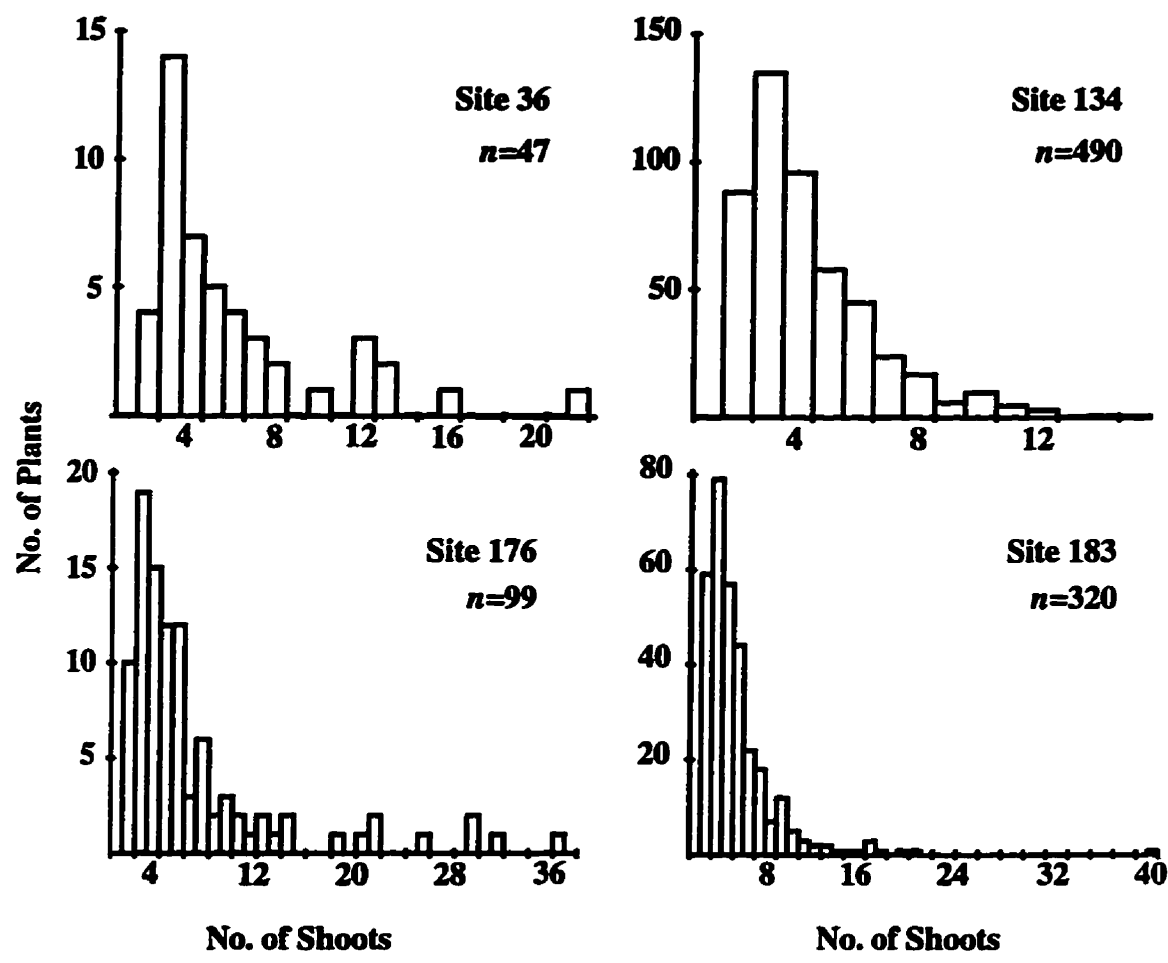


Figure 4.3 continued. Size-frequency histograms for the 10 study sites.

Changes in the number of shoots between 1994 and 1995, for the 100 marked plants (10 per site), are summarized in **Table 4.4**. Thirteen of the plants were lost for various reasons (harvesting, natural mortality, site destruction). Of the remaining 87 plants, 59 showed an increase in the number of shoots (mean of *ca.* 3.7 shoots), 17 showed a decrease (mean of *ca.* 2.9 shoots), and 11 had the same number of shoots. Overall, these results indicate that the number of shoots increases with plant age.

The spatial pattern of individual plants at each site are given in **Appendix III**. While plants at sites 8, 33, 133, 104, and 134 were relatively evenly distributed throughout the study plots, the other sites shows highly clumped (clustered) plant distributions.

4.2.4 Biomass allocation

In the summer of 1994, fifty-six plants were harvested, and dried root and shoot material was massed. The graph of root:shoot mass (**Fig. 4.4**) shows that above and below-ground biomass allocation in seneca snakeroot are approximately equal, over all plant sizes.

4.2.5 Flowering and seed production

In both the field and greenhouse-grown populations, first-year plants (i.e. those with a single shoot) were never observed to flower. However, most plants having two or more shoots produced flowers and fruits. Each shoot produces on average *ca.* 37 flowers (range 15-63, $n = 50$; **Table 4.5**). Since each flower produces a capsule containing two seeds (mean weight per seed was 0.0017 g based on a sample of 1000 seeds), each shoot produces an average of *ca.* 74 seeds per annum. Plants in the study sites had on average 5 shoots, the number of seeds produced is *ca.* 370 seeds/yr. Length of inflorescence per shoot was *ca.* 2.37 cm \pm 1.09 s.d. (range 0 - 4.5 cm, $n = 49$). In both the field and

Table 4.4. Number of shoots per marked seneca plant in 1994 and 1995. A dash (-) refers to a plant that was missing due to harvesting, natural mortality or road construction.

Northern Sites	No. of shoots			Northern Sites	No. of shoots		
	1994	1995	Difference		1994	1995	Difference
133	5	6	1	33	10	18	8
	6	12	6		9	9	0
	15	20	5		6	7	1
	26	-	harvested 1994		3	2	-1
	26	37	11		17	18	1
	9	11	2		3	-	natural mortality
	14	-	harvested 1994		7	3	-4
	15	13	-2		5	5	0
	9	7	-2		4	5	1
	3	4	1		24	24	0
104	20	26	6	1078	15	-	harvested 1994
	2	3	1		4	11	7
	23	16	-7		11	20	9
	2	3	1		26	-	harvested 1994
	11	8	-3		12	18	6
	6	-	natural mortality		19	-	harvested 1994
	2	3	1		11	-	natural mortality
	3	11	8		7	3	-4
	17	20	3		6	-	natural mortality
	11	12	1		7	10	3
75	3	5	2				
	6	-	road constructed				
	20	19	road constructed				
	8	9	1				
	5	6	1				
	8	5	-3				
	14	10	-4				
	5	4	-1				
	1	-	road constructed				
	1	-	natural mortality				

Table 4.4 continued. Number of shoots per marked seneca plant in 1994 and 1995. A dash (-) refers to a plant that was missing due to harvesting, natural mortality or road construction.

Southern Sites	No. of shoots		Difference	Southern Sites	No. of shoots		Difference
1994	1995			1994	1995		
8	5	7	2	176	3	7	4
	2	3	1		5	11	6
	1	2	1		12	18	6
	8	12	4		5	5	0
	8	10	2		4	4	0
	4	7	3		29	-	harvested 1994
	3	3	0		4	4	0
	7	6	-1		21	23	2
	3	3	0		13	17	4
	7	5	-2		10	10	0
36	3	6	3	183	4	6	2
	3	6	3		4	9	5
	2	3	1		7	14	7
	5	9	4		3	9	6
	2	-	natural mortality		15	32	17
	3	3	0		6	8	2
	4	6	2		4	7	3
	11	2	-9		6	7	1
	4	9	5		4	5	1
	15	17	2		11	10	-1
134	5	8	3		4	6	2
	6	6	0				
	3	6	3				
	6	10	4				
	5	8	3				
	3	10	7				
	7	4	-3				
	5	7	2				
	9	7	-2				
	7	13	6				

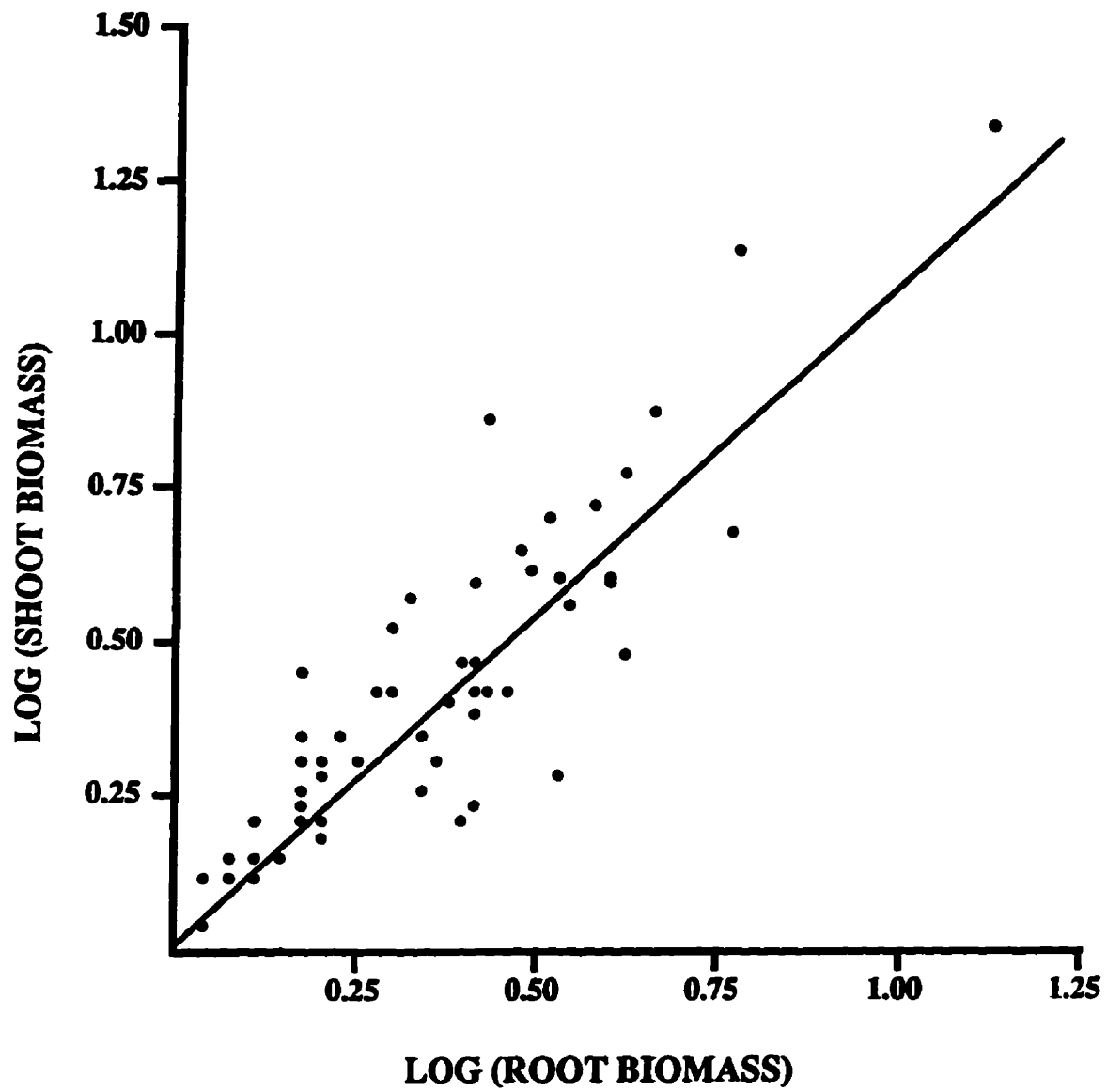


Figure 4.4. Log-log plot of root vs. shoot biomass in grams ($n=56$).
Line indicates a one to one relationship between root and shoot.

Table 4.5. Number of seeds in a random sample of 50 seneca snakeroot shoots.

No.	Number of seeds	No.	Number of seeds
1	26	26	23
2	34	27	24
3	36	28	53
4	54	29	39
5	34	30	31
6	42	31	27
7	26	32	46
8	40	33	43
9	32	34	32
10	51	35	21
11	35	36	25
12	43	37	41
13	27	38	38
14	34	39	36
15	27	40	38
16	35	41	63
17	34	42	55
18	59	43	61
19	35	44	50
20	33	45	43
21	15	46	53
22	36	47	34
23	44	48	41
24	31	49	26
25	33	50	25

Mean 37.3 Standard deviation 10.9

laboratory, it was noticed that flowering shoots that were cut or damaged early in their development bifurcated to produce a pair of flowering shoots.

4.3 Harvesting and mortality in wild populations

Table 4.6 summarizes harvesting and mortality of the 100 permanently-marked plants. Six plants were harvested (based on excavation scars) during the summer of 1994, at sites 133, 1078 and 176. These harvested plants were all relatively large (range of 14-29 shoots), suggesting that larger plants are differentially harvested. Harvesting in the vicinity of sites 75 and 183 was also noted. Five plants could not be relocated in 1995. Since no evidence of excavation was found, it is assumed that these plants died of natural causes. Most of these plants were small (1, 2, 3, 6 and 11 shoots).

4.4 Nutrient experiment

The mean above-ground biomass for the nutrient-treated plants was $1.94 \text{ g} \pm 1.78 \text{ s.d.}$ ($n = 29$), and for control plants $1.66 \text{ g} \pm 1.55 \text{ s.d.}$ ($n = 29$). The paired t-test ($t = 1.957$, $p = 0.060$) suggests that plants treated with nutrients are larger than the control plants, though the results are not quite significant at the $\alpha = 0.05$ level. These results suggest that growth of seneca snakeroot in the Interlake region may be increased by nutrient addition, though more long-term experiments are required. Given the low levels of nitrogen and phosphate in Interlake soils, this result could perhaps have been anticipated. Unfortunately, nutrients in this experiment were only added three times beginning in early June, by which time shoot production and elongation had already taken place and flowering had begun. Future studies should consider adding nutrients earlier in the growing season, before shoot elongation takes place.

Table 4.6. Loss of seneca plants from plots due to harvesting, construction or mortality.

Site	Plants lost in 1994 (includes plant size)	Plants lost in 1995 (includes plant size)
133	harvested, 26 and 14 shoots	no loss
104	no loss	no loss
75	no loss, but evidence of harvesting in area	loss due to construction, 6, 20, 1 shoot(s)
33	no loss	died, 3 shoots
1078	harvested, 15, 26 and 19 shoots	died, 11, 6 shoots
8	no loss	no loss
36	no loss	died, 2 shoots
134	no loss	no loss
176	harvested, 29 shoots	no loss
183	no loss, evidence of harvesting in area	no loss

4.5 Competition experiment

The mean above-ground biomass for plants in weeded plots was $1.09 \text{ g} \pm 1.04 \text{ s.d.}$ ($n = 30$), and for control (unweeded) plots $1.21 \text{ g} \pm 1.37 \text{ s.d.}$ ($n = 30$). The paired t-test ($t = 1.16$, $p = 0.255$) indicates that weeding had no effect on plant biomass. This is perhaps not a surprising result given that the study sites are sparsely vegetated. Seneca snakeroot plants growing in more shaded habitats might be expected to benefit from weeding, but more experiments are required.

4.6 Seed germination - field experiment

Plots sown with scarified and non-scarified seed were monitored throughout the growing season, but no evidence of seneca snakeroot germination was noted. Conditions were unusually hot and dry in June and July of 1995, which might account for these negative results. Further research is clearly required to determine the conditions necessary for seneca snakeroot seed germination under natural conditions.

4.7 Soil seed bank

Results of the seed bank trials are summarized in Table 4.7 a - c. A number of species germinated in these trials, mostly short-lived, 'weedy' species. A single seedling that appeared to be seneca snakeroot came up in the first seed bank trial, but unfortunately it died before it could be definitely identified. No other seneca plants germinated. Seneca snakeroot seeds were undoubtedly in the soil, since collections were purposely taken from beneath existing seneca snakeroot shoots. However, samples were taken in mid-summer,

Table 4.7 (1). Species that germinated from seed bank soil collected in mid-summer 1994.

SCIENTIFIC NAME	COMMON NAME	SITES (NORTH TO SOUTH)									
		133	104	75	33	1078	8	36	134	176	183
<i>Achillea millefolium</i>	Yarrow							x	x		x
<i>Agropyron trachycaulum</i>	Slender wheatgrass							x			
<i>Agrostis scabra</i>	Rough hair grass			x	x	x	x	x	x		x
<i>Anemone multifida</i>	Cut-leaved anemone									x	
<i>Arabis hirsuta</i>	Hirsute rock cress	x	x	x	x		x	x			
<i>Betula sp.</i>	Birch species								x		
<i>Bromus sp.</i>	Brome species										x
<i>Campanula rotundifolia</i>	Harebell			x	x	x				x	x
<i>Cardamine parviflora</i>	Small bitter cress									x	x
<i>Cirsium arvense</i>	Canada thistle		x								
<i>Deschampsia caespitosa</i>	Tufted hair grass			x							
<i>Epilobium ciliatum</i>	Northern willowherb	x	x	x	x	x	x	x	x	x	
<i>Fragaria virginiana</i>	Wild strawberry					x		x		x	x
<i>Galium boreale</i>	Northern bedstraw							x			x
<i>Mentha arvensis</i>	Field mint										
<i>Plantago sp.</i>	Plantain species							x			
<i>Poa compressa</i>	Canada blue grass	x				x		x	x	x	
<i>Poa pratensis</i>	Kentucky blue grass		x			x		x	x	x	
<i>Potentilla norvegica</i>	Rough cinquefoil										x
<i>Sonchus arvensis</i>	Perennial sow-thistle				x		x	x			
<i>Taraxacum officinale</i>	Dandelion	x		x				x	x		
<i>Thlaspi arvense</i>	Stinkweed						x				

Table 4.7 (2). Species that germinated from seed bank soil collected in mid-summer 1995.

SCIENTIFIC NAME	COMMON NAME	SITES (NORTH TO SOUTH)									
		133	104	75	33	1078	8	36	134	176	183
<i>Amaranthus retroflexus</i>	Red-root pigweed	x								x	x
<i>Cardamine parviflora</i>	Small bitter cress									x	
<i>Chenopodium album</i>	Lamb's-quarters	x	x	x	x	x	x	x	x	x	
<i>Cirsium arvense</i>	Canada thistle									x	
<i>Galium boreale</i>	Northern bedstraw	x									
<i>Poa pratensis</i>	Kentucky blue grass							x		x	
<i>Potentilla norvegicus</i>	Rough cinquefoil	x									
<i>Senecio vulgaris</i>	Common groundsel			x		x			x	x	x
<i>Setaria viridis</i>	Green foxtail								x		x
<i>Silene noctiflora</i>	Night-flowering catchfly		x	x	x	x	x		x		x
<i>Sonchus arvensis</i>	Perennial sow-thistle		x								
<i>Taraxacum officinale</i>	Dandelion	x	x	x	x	x	x	x	x	x	x

Table 4.7 (3). Species that germinated from seed bank cold-treated soil collected in mid-summer 1995.

SCIENTIFIC NAME	COMMON NAME	SITES (NORTH TO SOUTH)									
		133	104	75	33	1078	8	36	134	176	183
<i>Agrostis scabra</i>	Rough hair grass								x		
<i>Arabis hirsuta</i>	Hirsute rock cress							x			x
<i>Campanula rotundifolia</i>	Harebell		x		x						x
<i>Chenopodium album</i>	Lamb's-quarters								x		
<i>Cirsium arvense</i>	Canada thistle					x					
<i>Deschampsia caespitosa</i>	Tufted hair grass			x							
<i>Galium boreale</i>	Northern bedstraw			x		x					
<i>Heuchera richardsonii</i>	Alumroot			x							
<i>Viola sp.</i>	Violet species	x					x				
<i>Poa pratensis</i>	Kentucky blue grass		x				x			x	
<i>Potentilla norvegicus</i>	Rough cinquefoil								x	x	

prior to the shedding of the current year's seed. The seneca snakeroot seed collected was apparently either not viable, or their seed coat was not sufficiently broken down to allow germination to occur. The seeds that were viable likely germinated in early spring as evidenced by numerous seedlings that were observed surrounding many of the established plants. Seeds may also have been relocated by ants thereby decreasing the number of seeds present in the seedbank beneath existing seneca snakeroot plants.

4.8 Laboratory seed germination trials

4.8.1 Germination in petri dishes

A complete summary of the petri dish seed germination trials is given in **Table 4.8**. Initial attempts to germinate both freshly collected and cold-stored seed were unsuccessful. Removal of the aril (elaiosome) did not improve germination success. Initial attempts at seed scarification (two slits in the seed coat) resulted in a few seeds germinating. In later experiments, 'scarification' involved complete removal of a portion of the seed coat. This type of scarification greatly increased germination rates. Most experiments were performed in petri dishes placed in a growth chamber, since the highest germination rates were achieved under these conditions. Fungal infection was prevented by soaking the seeds in a 1:20 bleach : water solution (see Methods).

The plant growth hormone gibberellic acid was used in varying concentrations (0.1 mg/L to 100 mg/L) in an attempt to promote germination. In combination with scarification, application of gibberellic acid was found to increase germination rates in most experiments.

Table 4.8. Seed germination trials. In all cases petri dishes were used along with filter paper dampened with water or gibberellic acid (GA), if used. W.S. refers to window sill, G.H. refers to greenhouse and G.C. refers to the growth chamber. All seeds were pre-soaked in water for 24 hours prior to scarification and/or GA treatments except trials in 1994. Germination results with a question mark (?) were only qualitatively noted.

Date (m/d/y)	Anti-Fungal treatment	Seeds used (treatment/age collected)	Scarification	GA (mg/L)	Growth Environment	Germination	Results (% Germination) (overall average)
7/6/94		fresh/1994			W.S.	0/5	0
7/6/94		fresh/1994			darkness	0/5	0
9/21/94		fridge/1994			G. C.	0/100	0
10/24/94		aril removed/1994			G. C.	0/15	0
4/2/95	bleach/water	cold stored/1994	2 slits		G. C.	?/15	few
4/3/95	bleach/water	cold stored/1994	2 slits		G. C.	?/45	few
4/4/95	bleach/water	cold stored/1994	2 slits		W.S.	?/30	few
4/5/95	bleach/water	cold stored/1994	2 slits		W.S.	?/15	few
4/12/95	bleach/water	cold stored/1994	cut		G.C.	?/90	few
4/12/95	bleach/water	cold stored/1994	cut		W.S.	?/30	few
4/12/95	bleach/water	cold stored/1994	cut		G.H.	?/30	few
4/13/95	bleach/water	cold stored/1994	cut		W.S.	?/15	few
4/25/95	bleach/water	cold stored/1994	cut	0.1	G.C.	?/15	few
4/26/95	bleach/water	cold stored/1994	cut	0.1	G.C.	?/15	few
4/26/95	bleach/water	cold stored/1994		0.1	G.C.	?/15	0
4/29/95	no damp	cold stored/1994			G.C.	0/10	0
4/29/95	no damp	cold stored/1994	cut		G.C.	0/15	0
4/29/95	no damp, alcohol	cold stored/1994		0.1	G.C.	0/15	0
4/29/95	no damp, alcohol	cold stored/1994	cut	0.1	G.C.	2/15	13
4/29/95	no damp	cold stored/1994	cut	0.1	G.C.	2/15	13
4/30/95	alcohol	cold stored/1994	cut		G.C.	2/15	13

Table 4.8. continued.

Date (m/d/y)	Anti-Fungal treatment	Seeds used (treatment/age collected)	Scarification	GA (mg/L)	Growth Environment	Germination	Results (% Germination) (overall average)
5/16/95	bleach/water	cold stored/1994	cut		G.C.	5/60	8
5/16/95	bleach/water	cold stored/1994			G.C.	0/15	0
5/24/95	bleach/water	stratified 4 months/1994			G.C.	1/30	6
5/24/95	bleach/water	stratified 4 months/1994	cut		G.C.	52/60	86.7
6/13/95	bleach/water	freezer stored/1994	cut		G.C.	37/75	49.3
6/13/95	bleach/water	freezer stored/1994			G.C.	0/30	0
7/19/95	bleach/water	cold stored/1994	cut		G.C.	3/15	20
7/19/95	bleach/water	cold stored/1994			G.C.	0/15	0
7/19/95	bleach/water	cold stored/1994	cut	1.0	G.C.	8/15	53.3
7/19/95	bleach/water	cold stored/1994		1.0	G.C.	0/15	0
7/19/95	bleach/water	cold stored/1994	cut	10.0	G.C.	13/15	86.7
7/19/95	bleach/water	cold stored/1994		10.0	G.C.	0/15	0
8/1/95	bleach/water	stratified 2 months/1994	cut		G.C.	9/45	20
8/1/95	bleach/water	stratified 2 months/1994			G.C.	1/15	0
8/4/95	bleach/water	fresh/1995	cut		G.C.	4/30	13.3
8/4/95	bleach/water	fresh/1995			G.C.	0/15	0
8/4/95	bleach/water	fresh/1995	cut	10.0	G.C.	10/15	66.7
8/4/95	bleach/water	fresh/1995		10.0	G.C.	0/15	0
9/19/95	bleach/water	fresh/1995	cut	0.1	G.C.	2/10	20
9/19/95	bleach/water	fresh/1995	cut	1.0	G.C.	6/20	30
9/19/95	bleach/water	fresh/1995	cut	10.0	G.C.	11/20	55
9/19/95	bleach/water	fresh/1995	cut	25.0	G.C.	8/20	40
9/19/95	bleach/water	fresh/1995	cut	50.0	G.C.	13/20	65

Table 4.8. continued.

Date (m/d/y)	Anti-Fungal treatment	Seeds used (treatment/age collected)	Scarification	GA (mg/L)	Growth Environment	Germination	Results (% Germination) (overall average)
9/19/95	bleach/water	fresh/1995	cut	100.0	G.C.	12/20	60
9/19/95	bleach/water	fresh/1995	cut		G.C.	2/10	20
9/29/95	bleach/water	fresh/1995	cut		G.C.	8/20	40
9/29/95	bleach/water	fresh/1995	cut	25.0	G.C.	15/20	75
10/3/95	bleach/water	stratified 4 months/1994	cut		G.C.	27/40	67.5
10/3/95	bleach/water	stratified 4 months/1994	cut	25.0	G.C.	18/20	90
10/27/95	bleach/water	stratified 4 months/1994	cut		G.C.	6/10	60
10/27/95	bleach/water	stratified 4 months/1994	cut	25.0	G.C.	18/20	90
10/27/95	bleach/water	stratified 4 months/1994	cut	50.0	G.C.	20/20	100
12/11/95	bleach/water	stratified 6 months/1994	cut		G.C.	24/40	60
12/11/95	bleach/water	stratified 6 months/1994			G.C.	0/10	0
12/11/95	bleach/water	cold stored/1995	cut	100.0	G.C.	10/20	50
12/11/95	bleach/water	cold stored/1995	cut	50.0	G.C.	17/20	85
12/11/95	bleach/water	cold stored/1995	cut	25.0	G.C.	7/10	70
12/11/95	bleach/water	cold stored/1995	cut	10.0	G.C.	12/20	60
12/11/95	bleach/water	cold stored/1995	cut		G.C.	2/10	20
2/2/96	bleach/water	cold stored/1995	cut		G.C.	3/20	15
2/2/96	bleach/water	cold stored/1995	cut	25.0	G.C.	8/10	80
2/2/96	bleach/water	cold stored/1995	cut	50.0	G.C.	11/20	55
2/2/96	bleach/water	cold stored/1995	cut	100.0	G.C.	16/20	80

Seed stratification in moist sand, for periods ranging from two to six months, was also undertaken (in conjunction with seed scarification). A two-month stratification period resulted in *ca.* 20% germination success, but this increased to *ca.* 90% when seeds were stratified for four months. A six month stratification yielded *ca.* 50% germination, suggesting that a four-month stratification period is optimal for seneca snakeroot.

Greatest germination success (85-90%) occurred for scarified seed (removal of at least 25% of the seed coat) that was stratified for four months. The addition of gibberellic acid increased germination further still (to almost 100%), but the resulting seedlings were often weak and somewhat spindly.

Seedlings germinated in petri dishes were successfully transplanted into greenhouse soil once they had developed a strong root and shoot system (generally 2-3 weeks after germination). After a few months of growth, these seedlings were placed in a cold-room (2 °C) for 3 months to simulate over-wintering conditions. All seedlings treated in this way produced numerous shoots when they were removed from the cold-room and placed back in the growth chamber, indicating that they were winter-hardy.

4.8.2 Germination in soil

None of the fresh seeds sown into peat/sand/soil and peat/sand mixes germinated. Seeds that were stratified for four months also failed to germinate when sown into a mixture of soil/sand/perlite. In July of 1995, cold-stored seeds collected in 1994 were soaked in water overnight. Half were scarified prior to sowing into soil, and half were sown without scarification. None of the non-scarified seed germinated, but a germination rate of *ca.* 20% was achieved from scarified seed. The emerging seedlings were weak and soon died,

however. It is therefore recommended that seneca snakeroot seeds be germinated in petri dishes, and the seedlings later transplanted into soil.

4.9 Vegetative propagation

4.9.1 Root cuttings

Results of the root propagation trials are summarized in **Table 4.9**. Overall, the results are not encouraging. A few small lateral roots were produced from small root pieces placed on the soil surface, but otherwise root propagation was unsuccessful.

4.9.2 Shoot cuttings

The results of the shoot cutting experiments are summarized in **Table 4.9**. The first experiments used shoot cuttings taken from plants grown in the greenhouse. The procedure involved placing freshly cut shoot cuttings (*ca.* 2 - 5 cm in length) into greenhouse soil. Only a few roots were observed growing from these shoot cuttings. The next experiment used the same procedure, but the base of the cuttings were dipped in a commercial root starter (Stim-Root No. 1, 0.1% IBA) prior to planting. Again, only a few roots were produced from these shoot cuttings. In a third experiment, shoot cutting ends were dipped in a commercial root starter containing a fungicide (Wilson's Roots, 0.4% IBA). These cuttings showed much considerable root growth after one month.

On May 12, 1995 fresh cuttings were taken from plants in the field. The plants had just begun to grow, and the shoots were short (generally < 5 cm), purplish in colour, and with only a few leaf primordia. These cuttings were taken back to the greenhouse, where they

Table 4.9. Propagation experiments involving root and shoot cuttings and whole plant division.

Date	Procedure (Root cuttings)	Source	Soil mixture	Results
1/13/95	Cut root into 25 pieces (approx. 5cm in length)	Greenhouse	sand:peat:1/2 soil	few
3/16/95	4 root cuttings approx. 4cm in length	Greenhouse	sand:peat	0
3/27/95	3 root cuttings	Greenhouse	sand:peat	0
3/12/95	potted large root	Field	greenhouse mix	0
5/26/95	2 roots in soil	Field	greenhouse mix	0
Date	Procedure (Shoot cuttings)		Soil mixture	Results
3/16/95	4 non-flowering cuttings (2.2, 2.7, 3.0, 4.6 cm)	Greenhouse	sand : peat	few roots
3/27/95	4 cuttings, used Stim-root No. 1	Greenhouse	sand : peat	few roots
4/17/95	7 cuttings, used Wilson gel root starter with fungicide	Greenhouse	sand : peat	good root growth
5/12/95	cuttings, used Stim-root No. 1	Field	greenhouse mix	good root growth
5/18/95	6+5+5=16 cuttings, 3 pots , Wilson root starter	Field	sand:peat	some roots
5/26/95	cuttings, used Wilson root starter	Field	greenhouse mix	some roots
6/1/95	9 cuttings with flowers, used Wilson root starter	Field	soil/sand/peat/1/2leaf mold	soon died
6/17/95	50 cuttings in peat trays (non-flowering), Wilson root starter	Field	soil/sand/peat/1/2leaf mold	some roots
6/17/95	20 cuttings in clay pot (non-flowering), Wilson root starter	Field	greenhouse mix	some roots
Date	Procedure (Whole plant division)		Soil mixture	Results
5/12/95	large plant cut in 1/2	Field	greenhouse mix	took well
5/12/95	med plant cut in 1/2	Field	greenhouse mix	took well
5/26/95	1/2 propagation (6)	Field	greenhouse mix	fair
6/1/95	cut 2 plants in 1/2, cut shoots down	Field	soil/sand/peat/1/2leaf mold	fair
6/24/95	2 plants cut in 1/2, growth chamber	Field	soil/sand/peat/1/2leaf mold	poor

were planted into greenhouse soil after the base was dipped in a commercial root starter (Stim-Root No. 1, 0.1% IBA). These cuttings showed strong above-ground growth, and new shoots were formed after a few months. Shoots collected a week later (May 18, 1995) and treated in the same way did not take as well, however. Shoot collected on June 1, 1995 were flowering, and cuttings did not survive transplanting. Some non-flowering shoots collected on June 17, 1995 survived transplanting, but establishment success was low compared to shoots collected in early May. These results indicate that propagation of seneca snakeroot by shoot cuttings is possible, but only if the shoots are collected in the late fall or early spring (young shoots < 5 cm in length, with leaf primordia only). Older shoot cuttings, particularly if they are flowering, do not root well. Cuttings dipped in a commercial root starter (preferably one with a fungicide) prior to planting produce the best results.

Some of the plants produced from shoot cuttings were grown for six months and then placed in a cold-room for three months to simulate over-wintering. When these plants were placed in the growth chamber, some resumed growth but others did not. Further experiments are required to determine optimal conditions for the establishment of seneca snakeroot shoot cuttings. Success of cuttings taken in late fall should also be determined.

4.9.3 Whole-plant division

In the spring and summer of 1995, individual seneca snakeroot plants were carefully dug from the field, transported to the University of Manitoba greenhouse, and cut in half vertically using a scalpel. The two halves were planted into standard potting soil and grown in the greenhouse. Plants collected in early May survived well, but plants taken later in the growing season (late May and June) did not perform as well (Table 4.9). These results

indicate that whole-plant division should be performed in the early spring (or late fall), while the plants are still dormant or just after they have broken dormancy.

The plants divided in early May were overwintered in a cold-room, and then placed in the growth chamber to resume growth. These plants produced numerous new shoots and even flowered, indicating that whole-plant division is a feasible method for propagating seneca snakeroot.

CHAPTER 5

SUMMARY, SUSTAINABILITY AND RECOMMENDATIONS

5.1 Summary

In order to investigate seneca snakeroot populations in Manitoba, aspects of the biology, ecology, economics and history of *Polygala senega* were considered.

Available literature was reviewed to summarize relevant information on seneca snakeroot. In Manitoba, this perennial herb is typically found on calcareous soils in open, dry habitats. Seneca snakeroot has been used medicinally by many of the aboriginal peoples of North America, for a wide variety of ailments. In the 1700's seneca root was introduced into European medicine, and subsequently became a valuable natural treatment for respiratory disorders. A recent resurgence of interest in natural products has increased the demand for seneca root. This natural product has been, and continues to be, an important part of Manitoba's economy. Increased harvesting and reduced habitat availability (due to land clearing for agriculture) leads to the question of the sustainability of native seneca root populations in Manitoba. Recent attempts to germinate seneca seeds in North America have been unsuccessful, even though a variety of seneca snakeroot is cultivated in Japan.

Baseline information revealed that on Manitoba Hydro rights-of-way in the northern Interlake, seneca snakeroot occurs on dry, calcareous (mean pH of 8.1), nutrient-deficient, silty-clay soil. Common associates include bearberry, sedges, lichens, smooth aster, northern bedstraw, bryophytes and wild strawberry. Seneca shoot buds develop in late fall, overwinter and then continue development in early spring. Flowering occurs from late May to early June. Fruits appear in July and mature within 30 days. Size-frequency diagrams indicate that seneca snakeroot is successfully regenerating at the ten study sites. The species

allocates approximately equal biomass to above and below-ground structures. Loss of larger plants was mainly due to harvesting, and smaller plants due to mortality.

In order to investigate the economics of seneca, persons involved in the seneca snakeroot industry of Manitoba were contacted and interviewed. Diggers may be found throughout the summer along roadsides collecting the root. The dried roots are sold to brokers and exporters throughout the province. Exporters may sell the root directly to pharmaceutical companies, or to international brokers. Pharmaceutical companies either sell the root whole or powder it for incorporation into a variety of products. In Europe and Asia, the root is also sold whole for herbal tea preparations. The majority of the global trade in non-cultivated seneca root appears to be based on material harvested in Manitoba.

The results from germination and propagation experiments indicate that seneca snakeroot can be propagated both vegetatively and from seed. Two seed germination methods proved successful. The first involves stratifying seed in moist sand for four months, and then scarifying the seeds using a fine scalpel. The second method involves scarification and application of gibberellic acid, a plant growth hormone. Vegetative propagation (of material collected in the early spring or late fall) is best achieved by planting shoot cuttings dipped in root starter, or by dividing whole plants.

5.2 Sustainability of seneca snakeroot populations

Seneca snakeroot plants are likely sustainable in the Northern Interlake area, especially in regions north of Grand Rapids. In these regions, outcrops of limestone bedrock and loam-sand-gravel soil mixes commonly occur and make the digging of seneca difficult. Seneca digging is far more efficient and profitable in areas closer to and south of Grand Rapids. The harvesting difficulties due to the rocky terrain combined with the positive results of the

size-frequency histograms (seneca is successfully regenerating at each study site), leads to the assumption that seneca snakeroot plants in the Northern Interlake region can be considered fairly safe from overharvesting - for now. However, Interlake regions in which the soil is characterized by clay or loam mixes, which make seneca snakeroot easier to harvest, could very well be prone to overharvesting. Seneca has been overharvested to near extinction in eastern North America (Coffey 1993) and authors Howarth and Keane (1995a) warn that seneca root is being overharvested in Saskatchewan as they ask readers *'Please do not pick the wild seneca root'*.

In grassland regions, such as parts of Saskatchewan and Riding Mountain National Park (RMNP), seneca snakeroot is present, but not as abundant as in the Interlake region. This is likely a result of overharvesting, competition with other species and loss of suitable habitat. Soil mixes in grassland regions are 'loose' enough to facilitate efficient harvesting. Fortunately, RMNP does not permit harvesting but in similar regions, seneca has no protection. Competition with other species could also be a factor in grasslands. Seneca snakeroot is likely a poor competitor. Seneca thrives in the rocky, dry, nutrient-poor soils of the Interlake where few other species survive, so the competition level is low. In grassland regions, the soil is richer and species diversity is high (compared to northern Interlake rocky areas). Seneca, which is relatively shade-intolerant, is not able to compete as efficiently with the numerous other species and therefore is less abundant than in the Interlake. Seneca snakeroot has also been affected by habitat loss. Agricultural and urban development, especially in southern Manitoba and Saskatchewan, have eliminated appropriate snakeroot habitat. In these regions, remnants of seneca populations may be found in ditches and roadsides.

A complete survey of the seneca snakeroot populations in the province would be useful for determining the abundance of seneca snakeroot and the locations which may be susceptible to overharvesting or habitat loss.

Aside from the pressures on seneca snakeroot populations, it is important to remember that seneca root is a valuable part of Manitoba's economy and traditional life. Rather than suggesting that the harvesting of seneca root be 'banned' in certain areas, further research on the cultivation and propagation of seneca root should be promoted.

The economic value of seneca snakeroot and the stresses on natural populations (such as harvesting and habitat loss) combined with the knowledge gained from the study, have led to the development of four recommendations for the management of seneca snakeroot populations in Manitoba. These are summarized below.

5.3 Recommendations

1. Habitat loss and increased harvesting have placed considerable pressure on the remaining native seneca snakeroot populations in Manitoba. Harvesting pressure on wild populations can be alleviated by promoting the cultivation of seneca snakeroot. It is recommended that a program to propagate and cultivate seneca snakeroot be developed for the northern Interlake (Grand Rapids) region, in cooperation with the aboriginal communities in the area. Research on the propagation and cultivation on seneca snakeroot should be continued. Cultivated seneca root could grow to become an economically viable Manitoba crop. Entrepreneurs from British Columbia (as well as other parts of Canada, including Manitoba) have shown considerable interest in growing seneca snakeroot in their province, where other crops such as ginseng and echinacea, have become economically valuable. Logistically, seneca root should be cultivated in

Manitoba where the environmental conditions are appropriate and where the plant naturally occurs.

2. Seneca snakeroot harvesting and marketing strategies that will bring economic benefit to the aboriginal communities of the Grand Rapids area (and other regions of Manitoba) should be developed. Seneca diggers could be advised that fall harvesting (rather than summer) would allow for the maximum amount of seed to be dispersed and subsequently increased regeneration of plants the following year. Apparently, the concentration of saponins are highest in the fall (Howarth and Keane 1995). A distribution network for the efficient and profitable marketing of cultivated seneca root should be created for the direct benefit of the harvesters and exporters in Manitoba.
3. Manitoba Hydro rights-of-ways in the Interlake region are excellent seneca snakeroot habitats. It is recommended that these populations be maintained and promoted. Regular vegetation maintenance such as v-blading should be continued to control excessive shrub and tree growth, but herbicide use should be avoided.
4. Long-term monitoring of seneca snakeroot populations along Manitoba Hydro rights-of-way should be considered, using the existing 10x10 m semi-permanent plots. The growth rate of seneca plants could be determined (which would be useful in propagation research). Long-term weeding (removal of competitors) and nutrient addition (response to fertilizer) experiments should also be considered. A complete survey of the seneca root population in the province should also be undertaken.

REFERENCES

- Allport, N. L. 1944. The chemistry and pharmacy of vegetable drugs. Chemical Publishing Company, Inc. New York. Pages 106-109.
- American Herbal Pharmacology Delegation. 1975. Herbal pharmacology in the People's Republic of China. A trip report of the American Herbal Pharmacology Delegation. National Academy of Sciences. Washington, D. C. Page 184.
- American Pharmaceutical Association. 1936. The National formulary. Sixth edition. American Pharmaceutical Association. Washington, D. C. Pages 178, 327-328, 357.
- American Pharmaceutical Association. 1943. The pharmaceutical recipe book. Third edition. American Pharmaceutical Association. Washington, D. C.
- American Pharmaceutical Association. 1955. The National Formulary. Tenth edition. American Pharmaceutical Association, Washington, D. C. Pages 511-512.
- Anonymous. 1868. A list of Canadian medicinal plants. Canadian Pharmaceutical Journal. 1(6):83-85.
- Arnason T., R. J. Hebda and T. Johns. 1981. Uses of plants for food and medicine by native peoples of Eastern Canada. Canadian Journal of Botany 59(11):2189-2325.
- Bailey, L. H. 1975. Cyclopedia of American horticulture. Volume 5. Gordon Press. New York. Pages 1390-1391.
- Beattie, A. J. 1983. Distribution of ant-dispersed plants. Sonderbd. naturwiss. Ver. Hamburg. 7:249-270.
- Beattie, A. J. 1985. The evolutionary ecology of ant-plant mutualisms. Cambridge University Press. Cambridge. Pages 73-95, 110-115.
- Begon, M., J. L. Harper and C. R. Townsend. 1990. Ecology. Individuals, populations and communities. Second edition. Blackwell Scientific Publications. Massachusetts. Pages 173-174.
- Berg, R. Y. 1975. Myrmecochorous plants in Australia and their dispersal by ants. Aust. J. Bot. 23:475-508.
- Bidwell, R. G. S. 1974. Plant physiology. Macmillan Publishing Co. Ltd. New York. Pages 502-506, 515.
- Briggs, C. J. 1988. Senega snakeroot. A traditional Canadian herbal medicine. Canadian Pharmaceutical Journal. 121:199-201.
- British Pharmacopoeia Commission. 1993. British pharmacopoeia. Her Majesty's Stationery office. London. Pages 589-590.
- Campbell, N. A. 1993. Biology. Third edition. Benjamin/Cummings Publishing Co. Inc. California. Pages 759, 762-763.
- Canadian Pharmaceutical Association. 1995. Compendium of non-prescription products. 2nd edition. Canadian Pharmaceutical Association. Ottawa. 1995.

- Catling, P. M. and E. Small. 1994. Poorly known economic plants of Canada. 1. Seneca snakeroot (*Polygala senega* L.) Canadian Botanical Association Bulletin. 27(1):10-11.
- Chang H. M. and P. P. But (eds.). 1986. Pharmacology and applications of Chinese materia medica. Volume I. World Scientific Press, Singapore. Pages 551-553.
- Coffey, T. 1993. The history and folklore of North American wildflowers. Facts on file. New York. Pages 145-146.
- Conlon, M. 1995. But why would anyone want to? Globe and Mail, August 26.
- Corner, J. J. , J. B. Harborne, S. G. Humphries and W. D. Ollis. 1962. Plant polyphenols. VII. The Hydroxycinnamoyl esters of *Polygala senega* root. Phytochemistry. Volume 1. pages 73-77.
- Crellin, J. K. and J. Philpott. 1990. Herbal medicine past and present. Volume 2. A reference guide to medicinal plants. Duke University Press. Durham. Pages 387-388.
- Densmore, F. 1913. Chippewa Music - II. Bureau of American Ethnology. Bulletin 53. Pages 64-66.
- Densmore, F. 1928. Uses of Plants by the Chippewa Indians. Bureau of American Ethnology. Forty-fourth Annual Report. 1926-1927. Pages 291, 336-339, 364-365, 376.
- Dugan J. J., P. de Mayo and A. N. Starratt. 1964. Terpenoids. V. Senegenin: Functional groups and part structure. Canadian Journal of Chemistry. 42(3):491-501.
- Duke, J. A. 1992. Handbook of Phytochemical Constituents of Grass Herbs and other economic plants. CRC Press. Ann Arbor. Pages 475-476.
- Dwyer, J. and D. Rattray. (eds.). 1986. Magic and Medicine of Plants. The Reader's Digest Association, Inc. Pleasantville, New York.
- Elsasser, D. 1996. Operation Senega Root. Progress report. Saskatchewan Agriculture and Food Agricultural Development Fund. Unpublished manuscript.
- Environment Canada. 1995. Grand Rapids Hydro climate normals (1966-1990) and monthly meteorological summaries (1994, 1995). Environment Canada. Atmospheric Environment Service. Winnipeg, Manitoba.
- Erichsen-Brown, C. 1979. Use of plants for the past 500 years. Breezy Creek Press, Aurora, Canada. Pages 359-362.
- Eriksen, B. 1993. Floral anatomy and morphology in the Polygalaceae. Pl. Syst. Evol. 186:17-32.
- Fraser, W. R., L. A. Hopkins, R. E. Smith, A. LeSann and G. F. Mills. 1985. Soils of the Waterhen area. Manitoba Department of Agriculture. Winnipeg, Manitoba.
- Fujita M. and H. Itokawa. 1961. Studies on Saponin-bearing drugs. III. Sapogenins of domestic senega and *Polygala*, a Chinese drug. "Yuan Chi". Chem. Pharm. Bull. 9:1006-1008.

- Gillett, J. M. 1968. The Milkworts of Canada. Canada Department of Agriculture, Research Branch. Monograph No. 5. Pages 11-15.
- Great Plains Flora Association. 1986. Flora of the Great Plains. University Press. Kansas. Pages 564-566.
- Grieve, M. 1967. A Modern Herbal. Volume II. Hafner Publishing Co. New York and London. Pages 733-734.
- Grime, J. P. 1979. Plant strategies and vegetation processes. John Wiley and Sons. New York. Page 116.
- Handel, S. N. 1976. Dispersal ecology of *Carex pedunculata* (Cyperaceae), a new North American myrmecochore. American Journal of Botany. 63:1071-1079.
- Hanzawa, F. M. , A. J. Beattie and D. C. Culver. 1988. Directed dispersal: demographic analysis of an ant-seed mutualism. American Naturalist. 131:1-13.
- Hlady, W. M. and B. R. Poston. 1959. A study of the population of Indian ancestry living in Manitoba. Appendix II. The people of Indian ancestry in rural Manitoba. Dept. Agric. Immigr. Winnipeg. Pages 79-86.
- Holldobler B. and E. O. Wilson. 1990. The Ants. The Belknap Press of Harvard University Press. Massachusetts. Pages 549-551.
- Holm, T. 1907. Medicinal plants of North America. 4. *Polygala senega* L. Merck's Report. Volume 16. June 1907. New York.
- Holm, T. 1929. Morphology of North American species of *Polygala*. Bot. Gazette. 88: 167-185.
- Howarth D. and K. Keane. 1995a. Senega root. *Polygala senega* L. David Howarth and Kahlee Keane, Saskatchewan.
- Howarth D. and K. Keane. 1995b. The native garden. Propagating and wildcrafting native plants. Root Woman and Dave, Saskatchewan.
- Huang, K. C. 1993. The pharmacology of Chinese herbs. CRC Press. Boca Raton. Page 224.
- Hughes, L. , M. Westoby and E. Jurado. 1994. Convergence of elaiosomes and insect prey: evidence from ant foraging behavior and fatty acid composition. Functional Ecology 8:358-365.
- Hutchens, A. R. 1992. A handbook of native American herbs. Shambhala Publications. Boston. pages 170-171.
- Jellison, R. M. 1963. Dr. John Tennent and the universal specific. Bulletin of the history of medicine. 37:336-346.
- Johnston, A. 1970. Blackfoot Indian utilization of the flora of the northwestern Great Plains. Economic Botany. 24:301-324.

- Shoji, J., S. Kawanishi and Y. Tsukitani. 1971. On the structure of senegin - II of *Senegae Radix*. *Chem. Pharm. Bull.* 19:1740.
- Shoji, S., S. Kawanishi and Y. Tsukitani. 1972. On the structure of senegin - III of *Senegae Radix*. *Chem. Pharm. Bull.* 20(2):424-426.
- Sievers, A. F. 1948. Production of drug and condiment plants. *Farmers' Bulletin*. No. 1999. U. S. Department of Agriculture. Pages 83-84.
- Silvertown, J. W. 1987. *Introduction to plant population ecology*. Second Edition. Longman Scientific and Technical, Harlow. 229 pages.
- Silvertown, J. W. and J. Lovett Doust. 1993. *Introduction to plant population biology*. Blackwell Scientific Publications. Oxford. 210 pages.
- Smith, R. E., G. F. Mills, R. G. Eilers, H. Veldhuis, C. Selby, M. Santry. 1995. Terrestrial ecozones, ecoregions, and ecodistricts of Manitoba. Unpublished manuscript.
- Tang, W. and G. Eisenbrand. 1992. *Chinese drugs of plant origin. Chemistry, pharmacology, and use in traditional and modern medicine*. Springer-Verlag, New York. Pages 781-786.
- ter Braak, C. J. F. 1987. The analysis of vegetation-environment relationship by canonical correspondence analysis. *Vegetatio*. 69:69-77.
- Thomson, W. ed. 1978. *Healing plants. A modern herbal*. MacMillan London Ltd. London. Page 93.
- Trease, G. E. 1966. *A Textbook of pharmacognosy*. Ninth edition. Bailliere, Tindall and Cassell. London. Pages 482-483.
- Trottier, G. C. 1974. *Range studies, Riding Mountain National Park*. Volumes 1 and 2. Unpublished report, Parks Canada, Riding Mountain National Park.
- Tyler, V. E. 1981. *The honest herbal. A sensible guide to the use of herbs and related remedies*. Pharmaceutical Products Press, New York. Pages 211-212.
- Tyler, V. E. 1993. *The honest herbal. A sensible guide to the use of herbs and related remedies*. Third edition. Pharmaceutical Products Press, New York. Pages 13, 295-296.
- Tyler, V. E. 1994. *Herbs of choice. The therapeutic use of phytomedicinals*. Pharmaceutical Products Press. New York. Pages 97-98.
- Venkatesh, C. H. 1956. The special mode of dehiscence of anthers in *Polygala* and its significance in autogamy. *Bull. Torrey Bot. Club.* 83:19-26.
- Verkerke, W. 1985. Ovules and seeds of the Polygalaceae. *Journal Arnold Arboretum*. 66:353-394.
- Verkerke, W. and F. Bouman. 1980. Ovule ontogeny and its relation to seed coat structure in some species of *Polygala* (Polygalaceae). *Botanical Gazette*. 141:277-282.

APPENDICES

Appendix I. Phytochemical constituents of <i>Polygala senega</i> L. (from Duke 1992)....	114
Appendix II. Mean cover values per species per transect.....	115
Appendix III. Two year phenological profile of seneca snakeroot.....	123
Appendix IV. Spatial patterns of seneca snakeroot.....	127

Appendix I. Phytochemical constituents of *Polygala senega* L. (from Duke 1992).

1,5-Anhydro-(O-Alpha-D-Galactopyranosyl-(1-2)-O-Alpha-D-Galactopyranosyl-(1-2))-D-Glucitol	Polygalic Acid
Arabinose	Polygalitol
Arabinosyl-Alpha-D-Galactosyl-D-Glucose	Presenegin
P-coumaric acid	Raffinose
Cyclosenegin	Rhamnose
3,4-Dimethoxycinnamic acid	Ribose
Ferulic acid	Salicylic-acid methyl-ester-primveroside
Fucose	Saponins
2-O-Alpha-D-Galactopyranosyl-1,5-anhydro-D-Glucitol	Senegenin
Galactose	Senegin
6-O-Beta-D-Glucopyranosyl-1,5-Anhydro-D-Glucitol	Senegenic Acid
Glucose	Sinapic Acid
Hydroxysenegin	Alpha Apinasterol
Manninotriose	Stachyose
Mannose	Tenuifolin
P-Methoxycinnamic Acid	Tenuifolic Acid
Methyl Salicylate	Tenuigenin
Monotropitoside	3,4,5 Trimethoxycinnamic Acid
Pectin	Valerianic Acid
	Xylose

Appendix II. Mean cover values per species per transect (Northern sites).

Transect Species	1078 DC		33 AC		75 AC		104 AC		133 AC		
<i>Achillea millefolium</i>	1.75	1.25	2.25	1.50	2.25	2.50	0.50	1.50	3.55	1.90	
<i>Agropyron smithii</i>					0.75						
<i>Amelanchier alnifolia</i>			1.75	0.50			4.50	11.50	3.75		
<i>Anemone canadensis</i>									1.00		
<i>Anemone multifida</i>	0.25	0.25	3.50	1.75	2.25	0.75	0.50	2.00		0.95	0.60
<i>Antennaria aprica</i>	1.75	1.50	0.75			1.50	0.25	3.25			
<i>Apocynum androsaemifolium</i>					1.00						
<i>Aquilegia brevistyla</i>	0.25			0.25	0.25						
<i>Arabis lyrata</i> var. <i>kamchatica</i>											
<i>Aralia</i> sp.											1.00
<i>Arctostaphylos uva-ursi</i>	10.40	19.50	14.75	6.25	1.75		28.75	26.00	18.55	1.85	10.25
<i>Aster laevis</i>	2.45	2.50	3.25	2.75	1.55	2.75	0.50	2.75	1.98	3.05	6.15
bare ground									9.85	0.45	8.25
<i>Betula</i> sp.											
bryophytes	9.00	4.75	3.75	8.25	7.50		2.50	3.00		1.85	2.30
<i>Calamagrostis inexpansa</i>	2.75	0.25	0.25	0.25	3.00	3.00	0.25	0.25		0.35	0.80
<i>Campanula rotundifolia</i>	0.50	1.25	1.75	2.75	1.25	1.50	0.25	0.75			
<i>Carex aurea</i>	14.75				9.50	0.50			6.98	11.00	11.50
<i>Carex houghtonii</i>											
<i>Carex richardsonii</i>		9.00	9.00	19.00	53.00	1.50	4.75	8.75			
<i>Carex siccata</i>											
<i>Commandra umbellata</i>			1.75	2.00			1.75				
<i>Cornus canadensis</i>									0.25		0.75
<i>Cypripedium calceolus</i>	4.05			0.25							
<i>Danthonia spicata</i>	2.75					1.50					

Appendix II continued. Mean cover values per species per transect (Northern sites).

Transect Species	1078 DC		33 AC		75 AC		104 AC		133 AC		
dead wood					0.50				18.50	9.00	7.50
<i>Deschampsia caespitosa</i>	1.00	0.75	0.25	1.00		0.75					
<i>Elymus innovatus</i>	0.30						0.25	0.25			
<i>Epilobium angustifolium</i>											0.10
<i>Erigeron asper</i>		0.25	0.25								
<i>Fragaria virginiana</i>	9.80	6.80	3.75	7.50	6.00	2.50	0.35	1.75	1.05	5.35	3.60
<i>Gaillardia aristata</i>										0.25	
<i>Galium boreale</i>	1.25	4.00	6.25	2.50	8.45	2.75	1.75	1.75	4.00	2.80	1.80
<i>Habenaria bracteata</i>										0.20	
<i>Heuchera richardsonii</i>			0.25	0.50					0.50		
<i>Hieracium umbellatum</i>					0.25						
<i>Juniperus communis</i>		4.25	0.75	1.75	0.50		24.00	1.50	1.00		
<i>Koeleria cristata</i>	0.25										
<i>Lathyrus ochroleucus</i>			0.25								
<i>Lathyrus venosus</i>					0.25						
lichen	14.25	3.00	2.25	0.50			9.75	20.10	28.40	22.90	
<i>Lilium philadelphicum</i>	1.00	1.25	1.75	0.25	0.25	1.00	1.75			0.20	
<i>Linnaea borealis</i>	0.75	0.50	3.00	0.80		0.50	1.50	2.25	2.20	2.20	
<i>Lithospermum canescens</i>			0.50	0.50						0.90	0.30
Litter	5.50	43.00	41.00	39.50	4.00	1.50	21.00	13.00	8.95	21.20	4.50
<i>Melilotus alba</i>											
<i>Mianthemum canadense</i>		2.25	1.50	1.25	0.11		1.00	5.75	0.50	0.25	0.50
<i>Oryzopsis asperifolia</i>											
<i>Oryzopsis pungens</i>	1.25	2.75	2.25	1.00	0.25		1.25	3.00			
<i>Picea glauca</i>				0.50						0.10	

Appendix II continued. Mean cover values per species per transect (Northern sites).

Transect Species	1078 DC	33 AC	75 AC	104 AC	133 AC
<i>Pinus banksiana seedling</i>	0.10	0.25		0.50	0.50 1.00
<i>Poa pratensis</i>					
<i>Polygala senega</i>	3.25	2.25 5.00	3.40 1.50	3.25	2.35 4.40 3.60
<i>Populus balsamifera</i>	0.30				
<i>Populus seedling</i>					
<i>Potentilla fruticosa</i>	16.25	2.50 1.25 3.25	0.75 0.25	0.75	0.50 1.50 8.50 0.50
<i>Potentilla norvegica</i>					
<i>Prunella vulgaris</i>					
<i>Prunus nigra</i>			0.25	1.50	0.50 13.25 0.50
<i>Prunus virginiana</i>					6.25
<i>Prunus virginiana</i>					
rock					
<i>Rosa sp.</i>	22.50	12.50 15.50	31.50 69.00	30.50 10.00	44.30 8.45 6.80
<i>Rubus idaeus</i>	0.50 1.75	1.00 0.50 1.75	0.75	2.75 0.50	
<i>Salix bebbiana</i>		2.25 0.25			0.25 0.25 0.50
<i>Saxifraga tricuspidata</i>					3.25 0.50 4.00
<i>Schizachne purpurascens</i>					4.10
<i>Senecio canus</i>	0.25		1.25 2.00	1.00 1.00	0.40
<i>Shepherdia canadensis</i>	0.85		0.25	2.00 0.75	1.30
<i>Sisyrinchium montanum</i>	0.25		1.25		
<i>Smilacina stellata</i>					
<i>Solidago hispida</i>	0.75	0.75 7.50 1.25			
<i>Solidago nemoralis</i>					0.50
<i>Sonchus arvensis</i>		0.25			
<i>Stachys palustris</i>					

Appendix II continued. Mean cover values per species per transect (Northern sites).

Transect Species	1078 DC		33 AC		75 AC		104 AC		133 AC	
<i>Symphoricarpos occidentalis</i>					0.70		0.75	0.25	1.45	5.55
<i>Taraxacum officinale</i>	1.00		7.50		1.25	2.50	0.25		0.75	1.55
<i>Vicia americana</i>									0.30	1.05
<i>Viola adunca</i>					0.25		0.25			
<i>Zizia aptera</i>					0.25					
<i>Zygadenus gramineus</i>	1.55	2.25	1.00	2.00	2.90	0.50	0.50	2.50		0.45
Unidentified Herbs			0.50	0.50		0.50			0.25	0.05
Unidentified Grasses		4.00	4.00	2.50	4.75	7.75	1.50	3.75		0.75
Unidentified Shrubs	0.50	0.25		0.50			1.25			

Appendix II. Mean cover values per species per transect (Southern sites).

Transect Species	183 AC		176 AC		134 AC		36 AC		8 AC	
<i>Achillea millefolium</i>	1.45	2.85	2.50	2.25	5.25	1.50	3.50	2.50	4.25	1.25
<i>Agropyron smithii</i>									1.25	0.50
<i>Amelanchier alnifolia</i>			0.25						0.50	1.75
<i>Anemone canadensis</i>	0.20	0.10				0.25	0.50	1.25	0.25	0.25
<i>Anemone multifida</i>			2.50				0.25		0.75	0.50
<i>Antennaria aprica</i>			1.75	2.50					3.00	1.75
<i>Apocynum androsaemifolium</i>			2.50		1.75	1.75			0.50	2.50
<i>Aquilegia brevistyla</i>										
<i>Arabis lyrata</i> var. <i>kamchatica</i>								0.25		
<i>Aralia</i> sp.										
<i>Arctostaphylos uva-ursi</i>	2.00	8.25		12.75	5.50	3.75	0.25	5.00	8.00	21.75
<i>Aster laevis</i>	12.20	10.05	11.25	6.25	6.50	6.00	4.00	7.75	4.50	4.50
bare ground	1.50	7.20			17.00	2.75				
<i>Betula</i> sp.							0.50			
bryophytes		0.80	0.50				5.00	9.50	9.00	2.00
<i>Calamagrostis inexpansa</i>	1.50		8.00	1.00	2.50	1.00	12.00	4.50	4.00	3.50
<i>Campanula rotundifolia</i>		0.45	0.50	2.25		0.50	0.25	0.25	2.50	1.50
<i>Carex aurea</i>	7.00	6.60								
<i>Carex houghtonii</i>							0.25			
<i>Carex richardsonii</i>			1.75	7.75	3.75		1.25	2.25	7.75	5.75
<i>Carex siccata</i>					4.00	4.50				
<i>Commandra umbellata</i>	0.25		2.75	1.25	8.25			3.50	2.50	
<i>Cornus canadensis</i>										
<i>Cypripedium calceolus</i>									0.50	
<i>Danthonia spicata</i>									8.00	

Appendix II continued. Mean cover values per species per transect (Southern sites).

Transect Species	183 AC		176 AC		134 AC		36 AC		8 AC	
<i>Pinus banksiana</i> seedling							2.00	0.75		1.25
<i>Poa pratensis</i>	0.10		4.75		2.00	10.25	4.25	9.00	2.00	0.25
<i>Polygala senega</i>	4.20	7.20	0.50	2.25	7.75	3.75		1.00	3.00	4.50
<i>Populus balsamifera</i>									7.00	
<i>Populus</i> seedling		0.50							0.50	
<i>Potentilla fruticosa</i>		0.25						1.00	5.75	7.00
<i>Potentilla norvegica</i>										
<i>Prunella vulgaris</i>									0.25	
<i>Prunus nigra</i>										
<i>Prunus virginiana</i>									1.50	
<i>Prunus virginiana</i>									0.25	
rock			12.75	0.75			36.75	11.00	8.75	19.50
<i>Rosa</i> sp.	1.25	1.90	2.50	1.50	1.50	2.25		0.50	0.50	2.25
<i>Rubus idaeus</i>										
<i>Salix bebbiana</i>				1.50	0.25				2.00	
<i>Saxifraga tricuspidata</i>										
<i>Schizachne purpurascens</i>	1.05					5.00				
<i>Senecio canus</i>									0.25	
<i>Shepherdia canadensis</i>									0.50	
<i>Sisyrinchium montanum</i>	0.30							0.75	0.50	0.25
<i>Smilacina stellata</i>									0.25	
<i>Solidago hispida</i>								0.50	1.00	0.25
<i>Solidago nemoralis</i>										
<i>Sonchus arvensis</i>			0.50		2.75	1.75	1.00		0.50	0.25
<i>Stachys palustris</i>					0.25					

Appendix II continued. Mean cover values per species per transect (Southern sites).

Transect Species	183 AC		176 AC		134 AC		36 AC		8 AC	
<i>Symphoricarpos occidentalis</i>	1.10	0.45								
<i>Taraxacum officinale</i>	0.40	1.50	2.50	2.00	6.75	1.25	1.50	1.00	1.25	
<i>Vicia americana</i>	15.00	1.50	3.25	0.75	4.75	2.75	12.75	14.00	1.25	
<i>Viola adunca</i>	0.90			0.50				0.25		
<i>Zizia aptera</i>										
<i>Zygadenus gramineus</i>	2.50	1.00	0.50	2.00				0.25	4.50	2.00
Unidentified Herbs	0.60	0.40			1.25	3.00				1.25
Unidentified Grasses	10.20	0.60	5.00	2.75	5.50	5.75	3.75	1.25		
Unidentified Shrubs	0.25	0.25	0.50	0.75					1.50	

Appendix III. Two year phenological profile of seneca snakeroot.

Month	Day(s)	Status
1994		
June	15-17	The plants were just starting to flower in the northern-most sites. Slightly further ahead in the south. Flowers were all white in dense, small cone shaped-heads.
June	22-24	Flowers all white, no evidence of fruits
June	28-29	Flowers mostly white, some evidence of purple colour. Plants were in full bloom. Some shoots have capsules forming on lower flowers(bottom of inflorescence).
July	8,9	Flowers almost all pinkish-white, some white coloured. Capsules formed on some plants but inside seeds are still green.
July	18	Most plants had very dark purple sepals around capsules. Some flowering heads were all white and small as if they had just started flowering. Plants at various stages. Site 8, high elevation, dry, plants furthest ahead.
July	27	Plants almost done fruiting. Half fallen off, to bare ends of shoots.
August	5,6	Fruiting finished for almost all plants. Leaves turning colour and falling off. Some shoots were totally bare of leaves. Lots of evidence of digging. see notes.
August	20	Almost every plant was bare of fruits. Leaves turning purple. Some shoots already dead. Dying back from top down.
Month Day(s) Status		
1995		
May	1	Patches of snow in forest, ice still on small lakes. Temp 12 Impossible to find plants without looking for marked nails. Shoots either not visible or just poking through. Very little evidence of growth of other species, few green areas. Carex sp. growing. Avg length of seneca shoots about .5 - 1 cm. Had to remove litter and top soil to find shoots if present. Shoots purple to purple-green. No leaves yet, just scales.

Appendix III continued. Two year phenological profile of seneca snakeroot.

Month	Day(s)	Status
1995		
May	11	<p>Very cold. Temp 2 ° C . Freezing rain & snowing. Snow on ground at site 183 forming a hard crust. Not much difference from last week, shoots may be a bit longer 1 - 1.5 cm. Many marked plants still not visible. Quite a difference in other species from last week. <i>Carex richardsonii</i> , prairie crocus, willows flowering. Quite a bit more green, but patchy. Yarrow, strawberry, <i>Taraxacum</i> , <i>Antennaria</i> just starting.</p>
	25	<p>Seneca plants various sizes. Some not visible, some just popping up. The majority were 2-4 cm long (shoots). Very few with small green, dense flowering heads. (i.e. just beginning to form flowers.)</p>
	30	<p>Plants about 6-10 cm tall. Many have green flowering heads. Small, tight, with no white showing. Noticed some seedlings similar to ones in greenhouse.</p>
June	5	<p>Shoots 15+ cm tall. Flowering heads green. Some of the bottom flowers open on some plants.</p>
	16	<p>Plants in full flower! Noticed green capsules on bottom of some flowering heads. Some flowers dropping off at bottom, (probably not pollinated). May have something to do with the very warm weather. Some pink colour noticed on some flowers. Noticed ants crawling all over plants at some locations.</p>
	23	<p>Capsules formed on all plants. Almost entirely all full of capsules. Top few flowers may still be present. If not pollinated, flowers fallen off. Shoots partly bare. Didn't notice the pink/purple colour and full flowering stage as did last year. The hot weather must have accelerated things. Last week only a couple of capsules were formed on each shoot, now they were almost entirely full of capsules.</p>
June	28	<p>133 - Plants full of capsules (green). Some capsules noticed on ground. Noticed the pink/purple colour on some. 75 - some full of capsules, or partly to fully bare 176 - no capsules noticed at all. Very few with small white flowers on tip, rest bare. Either not pollinated, or capsules fallen off already, didn't notice any capsules on the ground. Perhaps too hot and dry?</p>

Appendix III continued. Two year phenological profile of seneca snakeroot.

Month	Day(s)	Status
1995		
July	6	133 - Full of capsules and sepals have dark purple colour or bare to partly bare of capsules. Leaves turning purple. Capsules that have fallen to ground have dried up and opened. Last week it was easy to see green capsules on ground, this week very difficult because capsules yellow - totally dried up. Some black seeds noticed on ground. Site 8 - almost all capsules gone, didn't notice anything on ground. 183 - almost all capsules gone, impossible to collect seed here.
	10	Site 183 - Shoots bare, rarely saw a shoot with a couple of capsules left on. Found what looks like an ant refuse heap, collected a sample. 75 - Some capsules still on (1/2 on) but had to look hard to find them. 133 - best site. Most have capsules still on (1/2 to full). Will be bare in a week or two. Sepals/capsules very purple, a few still green. Many have bare shoots as well. Most leaves are a red/purple colour.
	26	No capsules found on plants at any site. Lots of purple coloured leaves. Some yellow colour noticed, some leaves falling off. Sites seem very dry. Noticed shoots of some plants had split near the ends once or twice.
August	14	Met senega digger Henry Chartier digging on side of road. Plants hard to find, because no flowers/capsules etc. Collected seed bank soil. Sites fairly damp due to recent heavy rain. Some plants doing good, some entirely purple.
	30	183: Site very wet, leaves green, plants o.k. 133: very wet, many plants under water. Some with stems and leaves very dark purple, some plants green and healthy looking. Perhaps due to recent heavy rains. Few had leaves turning yellow.
Month	Day(s)	Status
1996		
October	10	Plants collected in the Ashern region in late fall resembled plants collected in early spring - i.e. dark purple shoots with scale-like leaves emerging from the ground - suggesting that the shoots emerge (begin new growth) in the fall rather than early spring as previously thought.

Appendix IV. Spatial patterns of seneca snakeroot plants in study plots. Site number is indicated at top right of each plot. Scaling on the axes is in meters.

