THE DISTRIBUTION, REPRODUCTIVE BIOLOGY, AND MORPHOLOGY OF *Lythrum* SPECIES, HYBRIDS AND CULTIVARS IN MANITOBA

by

Kimberly Alexandra Ottenbreit

Submitted in partial fulfillment
of the requirements for the
degree of
Master of Science

Faculty of Graduate Studies
The University of Manitoba
Winnipeg, Manitoba

September 1991

ISBN 0-315-76868-1

THE DISTRIBUTION, REPRODUCTIVE BIOLOGY, AND MORPHOLOGY OF *Lythrum* SPECIES, HYBRIDS AND CULTIVARS IN MANITOBA

BY

KIMBERLY ALEXANDRA OTTENBREIT

A thesis 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

ABSTRACT

Lythrum salicaria L. (purple loosestrife) is a rapidly spreading, naturalized perennial herb of moist habitats and is considered detrimental to wetland productivity in North America. The first herbarium record for Manitoba was collected from Neepawa in 1896. As of 1991, 38 sites in Manitoba from field work, correspondence and herbarium records were identified. Most of these were in the watersheds of the Red and Assiniboine Rivers. Dispersal is mostly by means of seed. Significantly more seed matures in the capsules in the lower and mid regions of the infructescence compared to those from the top. Seed is shaken by wind from the capsules between late summer and the end of winter. Germinability of seed from 28 cultivated and naturalized populations averaged 92 percent, however cultivars had low seed production (especially 'Morden Pink'). Discriminant analyses identified calyx pubescence, calyx lobe length, and calyx appendage length as significantly reliable features for separation of cultivars from naturalized plants. The cultivars 'Morden Pink', 'Dropmore Purple', and 'Morden Gleam' were self-incompatible, but artificial crosses between these and a naturalized population when legitimate were for the most part fertile. Resulting seeds averaged 98 percent germination and the hybrid plants were highly interfertile.

Thirty-seven percent of the hybrid progeny were classified in discriminant analysis as being indistinguishable from a naturalized population which grows on the Assiniboine River. Controlling the spread of *L. salicaria* will be accomplished by preventing seed production in naturalized populations, and by removing cultivars from locations where they may affect pollen transfer with naturalized populations.

ACKNOWLEDGEMENTS

This study was funded by Ducks Unlimited Canada and the Minnesota Department of Natural Resources.

My thanks are extended to: my advisory committee, Dr. Norman Kenkel, Prof. Louis Lenz, and Dr. John Stewart; Darryl Kroeker of Ducks Unlimited; Lynn Collicutt and Dr. Campbell Davidson of the Morden Research Station for their advice and provision of stock plants; Allen Sturko, formerly of Manitoba Agriculture, for providing me with distributional information on purple loosestrife; T & T Seeds for providing me with 'Dropmore Purple' and 'Morden Pink' cultivars; and Charles Burchill and Alex McIlraith for kindly assisting me with the statistical analyses.

I would further like to thank the Biology Department of the University of Winnipeg for use of their equipment and facilities. I am especially grateful to Karen Jones for enduring my constant presence in the greenhouse and making my time spent there a very enjoyable experience.

I wish to express special thanks and appreciation to my supervisor, Dr. Richard Staniforth, for the assistance and advice given throughout the duration of this study.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER 1

GENERAL INTRODUCTION

Lythrum salicaria L. (purple loosestrife) is an herbaceous perennial of moist or wet environments. It was introduced into North America during the early nineteenth century from Europe. The species is considered an aggressive competitor in wetlands, replacing native vegetation and lowering marshland productivity. As the species is continually expanding its range westward from its beginnings on the eastern seaboard, it now is important to monitor its invasion into new locations as a prelude to implementing methods of control.

Cultivars of *Lythrum* spp. have been developed and are grown as popular ornamentals in many gardens and landscapes eg. *L. virgatum* cv. Morden Pink and *L. salicaria* x *L. virgatum* cv. Dropmore Purple. Some or all of these cultivars are suspected of contributing to the spread of purple loosestrife in the wild.

In recent years the species has been spreading along the major waterways in Winnipeg, Manitoba, the Red and Assiniboine Rivers, and into many ditches, streams and retention ponds. An interest in purple loosestrife's naturalization and controversy surrounding the use of the species as a garden ornamental prompted this investigation.

The objectives of this study were:

1) to determine the distribution of *L. salicaria* in Manitoba.

2) to examine the seed production and germination in several cultivated and naturalized populations.

3) to monitor seed dispersal in a few naturalized populations of purple loosestrife over the winter months.

4) to determine whether floral features may be used to distinguish cultivated and naturalized taxa from each other.

5) to investigate the crossability between three cultivars and one naturalized population of *Lythrum*, and subsequently test the fertility of their offspring.

This research should provide a base for future monitoring of the expansion of purple loosestrife in the province, establish the contribution of cultivars to naturalized populations, and to morphologically distinguish some cultivars and naturalized populations in order to facilitate identification in the nursery trade (providing the sale of these plants continues). Knowledge of the reproductive biology and morphology of *Lythrum* taxa is of use taxonomically and should contribute to the understanding of their heteromorphic incompatibility system.

A couple of terms used throughout this thesis are defined as follows:

1. Wild. The term "wild" refers to introduced or escaped individuals growing outside the garden environment. It does not necessarily mean that one is referring to a native species.

2. Naturalize. To become established as if native.

The following is a list of abbreviations used in the thesis:

CAN	= National Museum of Natural Sciences herbarium
CDA	= Central Experimental Farm herbarium
DBG	= Devonian Botanic Garden
DP	= 'Dropmore Purple'
GEN	= Geneva
LEI	= Leipzig
LL	= Lockport long style/stamens
LM	= Lockport mid style/stamens
LS	= Lockport short style/stamens
MG	= 'Morden Gleam'
MMMN	= Manitoba Museum of Man and Nature herbarium
MP	= 'Morden Pink'
MR	= 'Morden Rose'
MRS	= Morden Research Station, Morden
OSL	= Oslo
TOR	= Toronto
U.of M.	= University of Manitoba, Plant Sciences
UWPG	= University of Winnipeg herbarium
WIN	= University of Manitoba herbarium
Wpg.	= Winnipeg

CHAPTER 2

LITERATURE REVIEW

ORIGINS AND SPREAD

Lythrum salicaria L. (purple loosestrife) was introduced into North America prior to 1814 (Stuckey 1980). A native of Eurasia, this species is nearly cosmopolitan in its distribution (Shamsi and Whitehead 1974a). Freshwater marshes, stream margins, floodplains and ditches are typical habitats of this perennial herb (Thompson *et al.* 1987).

Since its arrival onto the eastern seaboard, purple loosestrife has spread across much of North America. Today it occurs in all ten Canadian provinces (Cody 1978) and is found in all of the contiguous United States north of the 35th parallel, excluding Montana (Thompson 1989).

The impact of this weed on wetland habitats is considered detrimental by many and is a cause for concern (Rawinski 1982, Balogh 1986, Thompson 1989). In many areas, purple loosestrife has displaced more than fifty percent of wetland emergents. Once established many infestations in the Northeast have maintained themselves for over twenty years (Thompson 1989). Colonies studied in Finland were considered young if less than one hundred years old, indicating a potential for a very long lifespan (Halkka and Halkka 1974). It has been observed that in North America small groups of individuals may struggle to survive in an area for 20-40 years and then suddenly proliferate and become troublesome (Stuckey 1980).

Several factors have enabled the species to flourish in North American wetlands: 1) no natural enemies; 2) prolific seed producer, with estimates reaching as high as 2.7 million seeds per individual per year (Thompson *et al.* 1987); and 3) seeds are easily dispersed via wind and water. In addition, accidental spread by waterfowl, people, horticultural planting, and wildflower seed mixes may occur (Henderson 1987, Thompson *et al.* 1987). Purple loosestrife is able to withstand clipping, crushing, and shallow burial or flooding and readily regenerates new growth (Thompson 1989). Mature plants can withstand dry conditions, however moist soil is critical for seed germination and seedling establishment (Henderson 1987).

Lythrum salicaria is reported to replace indigenous vegetation such as *Typha* spp. (cattails), *Juncus* spp. (rushes), and *Carex* spp. (sedges) (Rawinski and Malecki 1984). *Lythrum salicaria* is utilized as nesting sites for red-winged blackbirds (*Agelaius phoeniceus*) and to a small degree by grazing white-tailed deer (*Odocoileus virginianus*). The flowers provide a rich nectar and pollen source for insects such as bees, adult parasitic wasps and syrphid flies (Batra *et al.* 1986), but animals such as marsh wrens (*Cistothorus palustris*) and muskrats (*Ondatra *zibethicus*) use cattails almost exclusively and avoid purple loosestrife (Rawinski 1982). Rawinski (1982) also reports that the loss of open water is another impact which requires consideration. *Potamogeton* spp. (pondweeds), for instance, are unable to survive under the shade cast by purple loosestrife if infestations expand into deeper waters.

Gaudet and Keddy (1988) examined forty-four North American wetland species and discovered a strong relationship between plant traits and competitive ability. These traits were: plant biomass, plant height, canopy diameter, canopy area, and leaf shape. They found that *L. salicaria* had the highest competitive ability, greater than that of other associated marshland genera, such as *Scirpus, Typha, Phalaris*, and *Juncus*.

The monitoring of the spread of purple loosestrife stands, or the detection of new colonies, may be easily determined using remote detection as stands characteristically appear greenish-purple in colour and bumpy textured (Balogh and Bookhout 1989).

Lythrum salicaria is believed to have reached North America via ships ballasts, raw wool, and livestock (Thompson *et al.* 1987). European immigrants also introduced purple loosestrife since the plant was originally valued for its medicinal qualities.

USES

The term "loosestrife" was used by Turner in 1548 for both *Lythrum* spp. and *Lysimachia* spp., and is derived from the greek 'lusis' (deliverance from) and 'makhe' (strife, battle) (Grigson 1974). Pliny (a Roman scholar) described *Lythrum salicaria* as being so potent 'that if placed on the yoke of inharmonious oxen it will restrain their quarrelling.' He also stated that the plant was discovered by Lysimachus, who lived c. 360-281 B.C., and was a companion of Alexander and King of Thrace (Grigson 1974). The generic name *Lythrum* is derived from the greek word 'lythron', meaning blood (Plowden 1970, Gledhill 1985). This refers to the plant's ability to stanch bleeding, to cleanse and heal wounds, ulcers, and sores, and to treat diarrhoea and dysentery (Reader's Digest 1986, Thompson *et al.* 1987). Extracts from *Lythrum* spp. also lower blood sugar levels and have been used as a folk medicine against diabetes mellitus (Lamela *et al.* 1986).

Medicinal uses have been tested in the laboratory for antidiarrheal, antidysenteric, and antispasmodic action. An extract made from the flowers and stems is especially effective as an hypoglycemic agent (Torres and Saurez 1980).

Non-medicinal uses for the plant have also been recognized. Apparently, the high alkaloid content of purple loosestrife made it useful in tanning leather. The young leafy shoots have been recommended for consumption as an emergency vegetable (Klimas and Cunningham 1974, Usher 1974). It has also been used as a hair dye and burned to drive away insects (Reader's Digest 1986).

For many years, *L. salicaria* was highly recommended and valued as a honey plant in the apiculture industry (Mead 1957, Van't Haaff 1968, Bunch 1977a and 1977b, Pellett 1977, Hayes 1979). The plant was known to have a long period of bloom and good yields of nectar (Mead 1957). The honey, though dark in colour and strong in flavour, is considered palatable (Pellett 1944). Thus, it was intentionally sown into many wetlands and streams by beekeepers. Several individuals (Mead 1957, Pellett 1977, Hayes 1979) wrote of deliberately introducing purple loosestrife into their areas. However, beekeepers were warned that *L. salicaria* is an aggressive weed and have been implored not to sow the plant (Hughes 1977).

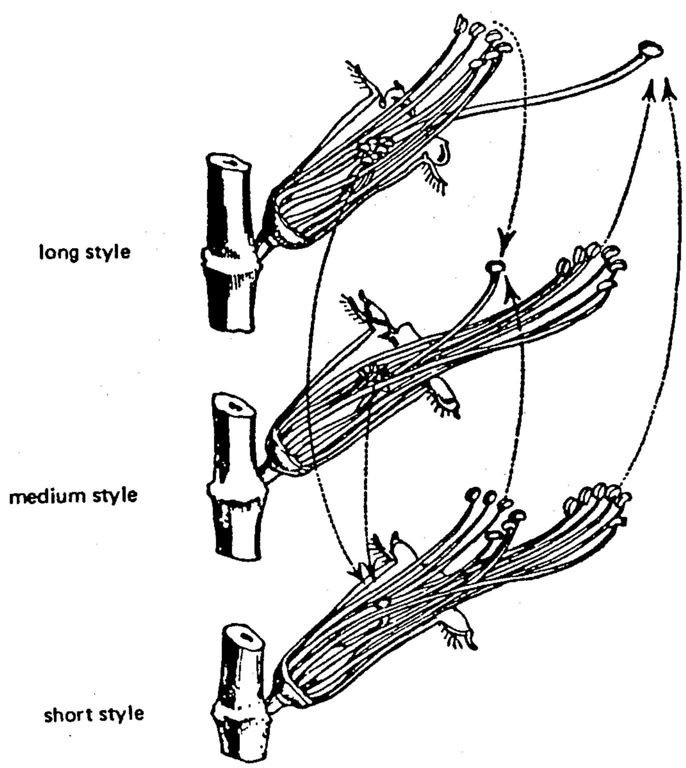
REPRODUCTIVE BIOLOGY

The reproductive biology of *L. salicaria* is complex and is the focus of much research. It is tristylous, a rare breeding system found in only three plant families -- Lythraceae, Oxalidaceae, and Pontederiaceae (Charlesworth 1979, Ganders 1979). Darwin (1865) was the first person to work experimentally with the plant and to recognize its self-incompatibility system. Tristylous species combine different morphological characters with physiological incompatibility (Heuch 1979). A flower of one style length bears two sets of stamens of different lengths. Thus, there are long-styled plants which bear two whorls of stamens at the mid and short levels, mid-styled plants which bear stamens at the long and short levels, and short-styled plants which bear stamens at the long and mid lengths. Legitimate pollinations are restricted to crosses between the style of one length with stamens of the corresponding length. Figure 2.1 illustrates the compatible pollinations that occur between the three style morphs (Darwin 1865).

This heteromorphic, sporophytic, self-incompatibility system means that incompatibility is expressed between the pollen and the style (Brewbaker 1957, Crowe 1964, Vuilleumier 1967) and controlled by the parent (Vuilleumier 1967). Apparently, sporophytic incompatibility factors are derived from the tapetum and the pollen mother cells (Dulberger 1975).

In the genus *Lythrum*, the base chromosome number is 5, with haploid numbers 5, 10, 15, 25, and 30 reported (Dulberger 1968, Tobe *et al.* 1986, Graham *et al.* 1987). *Lythrum salicaria* has been reported to have haploid numbers of n=15 in Israel, and 25 or 30 in Europe (Dulberger 1968). *Lythrum virgatum* has a haploid number of n=15 (Dulberger 1968), and *L. alatum* a haploid number of n=10 (Graham *et al.* 1987). *Lythrum salicaria* and *L. virgatum* are tristylous species, whereas *L. alatum* is distylous, having lost the

Figure 2.1: Tristyly in *Lythrum salicaria*.
Arrows indicate compatible pollinations
(Darwin 1865).


long style
medium style
short style

short-styled form, i.e. the S allele (Ganders 1979, Graham *et al.* 1987). Inheritance of style length in *L. salicaria* is disomic in Australia and tetrasomic in Eurasia (Gilbert and Lee 1980), the latter probably the case in North America. The self-incompatibility system in *L. salicaria*, though effective, is not strict (Darwin 1899, Stout 1923, Mulcahy and Caporello 1970). It is possible that higher ploidy levels cause this increased compatibility, as a diploid species, *L. junceum*, has been shown to have a strict incompatibility system (Dulberger 1970). In distylous species, it appears that pins have a stronger incompatibility system than thrums (Ornduff 1978).

Tristyly is controlled by 2 loci, called S and M, with two alleles at each locus (Ganders 1979, Leach 1983, Real 1983). The S locus is epistatic to the M locus, and S (short style) is dominant to non-short. Plants are mid-styled if they have the dominant allele at the M locus and long-styled if they are homozygous recessive. Thus, ssmm is long, ssM- is mid, and Ss-- is short-styled. These two loci are not linked in *Lythrum* (Frankel and Galun 1977, Ganders 1979).

Equal proportions of the three style forms in *L. salicaria* are generally maintained if colonies consist of twenty individuals or more (Heuch 1980). However, in some stands there tends to be an excess of longs and a deficiency of shorts (Schoch-Bodmer 1938, Heuch 1979). Such an event may occur because of local random effects in nature which result in unequal tendencies for certain forms to disappear (Heuch 1980). It has been proposed that longs are more vigorous (Schoch-Bodmer 1938). Stirling (1933) felt that heterostyly in *L. salicaria* was unstable and at an early stage of evolutionary development. A semi-homomorphic form of *L. salicaria*, in which the pistil and the set of longer stamens are almost of equal length and midway between the standard placement of mids and longs, has been reported and has been considered a reversion to an ancestral form. This form appeared among progeny of self-fertilized mid plants, was self-compatible, and its progeny were of all four style forms (Stout 1925).

The evolution of heterostyly has been explored by several researchers (Crowe 1964, Vuilleumier 1967, Yeo 1975), but it is likely that heterostyly arose by different mechanisms in different plant groups (Solbrig 1970, Ganders 1979). Though the most popular theory, it is uncertain whether physiological incompatibility arose first and was later reinforced by morphological differences, or the reverse (Solbrig 1970). Homomorphic species may have lost alleles and become self-fertile, subsequently lost all their incompatibility alleles, then different alleles may have arisen using a new diallelic system. Stirling (1933) thought that the ancestral form would have been long-styled, but Schoch-Bodmer (1938) suggests that mids with two whorls

1954 (Harp and Collicutt 1983). The parentage of these four cultivars has been well documented. Unfortunately, records of the majority of cultivars have not been kept, and this has led to a lot of confusion and misclassification as to parentages. Such errors are perpetuated in many horticultural publications. Table 2.1 lists cultivated *Lythrum* spp. and their supposed parentages obtained from various horticultural texts. It can be seen that several authors have made errors regarding the correct parentage of some cultivars. This may in turn complicate the consideration of which cultivars are threats in the naturalization of purple loosestrife.

CONTROL

The control of purple loosestrife has been attempted using many methods, but none have yielded completely satisfactory results. Currently, the most effective control is achieved using herbicides, though the most promising option in the future is biological control. This is especially important in Manitoba, where purple loosestrife is recognized as a noxious weed, but no herbicides have been approved for general aquatic use in Canada (Ted Peluk, personal communication). The various methods examined for their effectiveness as a control for purple loosestrife are listed below, and results of research findings reported.

CUTTING-- Rawinski (1982) studied the management of *L.*

Table 2.1: References to parentage of cultivated varieties of *Lythrum* taxa.
[** (A) Giles *et al.* 1980, (B) Buckley 1977, (C) Hebb 1975, (D) Snyder 1983, (E) Beckett 1981, (F) Thomas 1982, (G) Hudak 1976, (H) Still 1982, (I) Allan Bloom, personal communication].

Legend.
♦ indicates known errors in cultivar parentage
+ indicates known correct classifications (according to the Morden Research Station).
* cultivars which may be hybrids with *Lythrum virgatum*.

CULTIVAR	REFERENCE** A	B	C	D	E	F	G	H	I
Lythrum salicaria parentage*									
Atropurpureum									
Brightness	–	–			–	–			
Columbia			–						
Dropmore Purple	–		–					–	
Firecandle			–	–	–	–		–	–
Happy	–		–	–				–	
Lady Sackville		–							–
♦Morden Gleam	–	–	–				–	–	
♦Morden Pink			–			–	–	–	
♦Morden Rose			–				–	–	
♦Pink Spires			–						
Purple Spires			–						
Robert	–	–	–	–	–	–	–	–	–
+Roseum superbum	–								
The Beacon	–	–	–		–	–	–		–
Tom's Choice			–						
Lythrum virgatum parentage									
Dropmore Purple		–		–					
+Morden Pink	–	–		–					
+Rose Queen		–	–		–	–			–
+The Rocket		–		–	–				
Lythrum virgatum x *Lythrum alatum* parentage									
Columbia Pink				–					
+Morden Gleam				–					
+Morden Rose				–					

salicaria and found that cutting shoots in late August was more effective than earlier in the year, as purple loosestrife was less able to replenish carbohydrate reserves. However, effectiveness is determined by plant age and water depth. Cut shoots that were left on moist soil developed adventitious roots and continued growth. McKeon (1959) found that cutting had no effect.

BURNING-- McKeon (1959) found that burning *L. salicaria* had no impact. Rootstocks are likely to survive and regenerate new growth.

WATER LEVEL MANIPULATION-- Under stable water conditions it appears that cattails are able to suppress purple loosestrife (Rawinski and Malecki 1984). Flooding over several years would likely be effective if done before the seedlings were 10 cm tall (Rawinski 1982). Clay (1986) also indicated that long-term flooding reduced plant vigour. Balogh (1986) found that after eight weeks of flooding purple loosestrife at various depths nearly 100 % mortality was obtained. However, Smith (1959) felt that flooding would be unsuccessful with established plants as they were seen to tolerate depths of 60 cm (24 in.), and McKeon (1959) found that water level manipulation had no effect. Rawinski (1982) noticed that carp (*Cyprinus carpio*) appeared to reduce densities of the species when water levels were raised. He found that a flooded marsh with no carp only mildly stressed established purple loosestrife after two years. If problems are less severe, drawdowns in late summer or autumn are best for the control of purple loosestrife. Typical drawdowns commence in the spring in managed marshes. This is followed by a summer or two of mudflat exposure, then a reflooding. Bulrush and cattail seeds can germinate under water if previously exposed to light and oxygen. Purple loosestrife seedlings are intolerant to flooding. Thus, late summer drawdowns may provide native species an opportunity to colonize open flats as any newly germinated purple loosestrife would die once winter arrives (Clay 1986).

HERBICIDES-- Over the past thirty-five years, researchers have attempted to control purple loosestrife using different herbicides with varying degrees of success. McKeon (1959) found that herbicides were either considered ineffective (2-4 Dow, 2,4-D, 2,4,5-T) or not economically feasible (Weedazol). Smith (1959) found that herbicide control with Ammate and Ammate X were successful at 32-40 lbs. per acre from trucks mounted with sprayers at the pre-flowering and flowering stage, achieving 100 % kill. The only problem would be access difficulties into large acreages. Smith (1964) found the best control was attained when using low volatility esters of 2,4,5-T or 2,4-D and 2,4,5-T in mixture, or Silvex, at a concentration of eight pounds acid equivalent per acre when at least 200 gallons of total solution were applied per acre by foliar application.

Rawinski (1982) found that application of the herbicide glyphosate during peak bloom controlled 99 % of purple loosestrife and led to the establishment of native species. Glyphosate is biodegradable and quickly inactivated on exposure to moist soils and, as such, is considered a low hazard to fish and wildlife.

COVER CONTROL SPECIES-- Several plant species have been examined as to their usefulness as a method to control purple loosestrife infestations during a marsh drawdown. Japanese millet (*Echinochloa frumentacea*) was found to out-compete both seeded and naturally established *L. salicaria* seedlings (Rawinski 1982). Balogh (1986) tested two potential plant competitors, nutsedge (*Cyperus ferugmescens*) and Walters millet (*Echinochloa walteri*) and found that they were ineffective. However, pale persicaria (*Polygonum lapathifolium*) did out-compete purple loosestrife for resources in the first year when sown at naturally occurring seed densities.

HAND-PULLING-- Hand-pulling has been accepted as an effective means of control if there are isolated and relatively young plants. This is easiest where water levels are at or slightly above ground level (Smith 1959, Rawinski 1982).

COMBINED CONTROL METHODS-- Clay (1986) recommended that mowing and flooding together as a combined method should be effective. Mowing should be done just prior to flowering, and cuttings removed. Subsequent flooding would eradicate adults. However, this method would not control recolonization. Rawinski (1982) found that the control obtained from cutting and burning in late summer was less effective than cutting alone.

BIOLOGICAL CONTROL-- An effective method of dealing with *L. salicaria* in the future is likely to be by means of biological control. Currently, three insect species are being tested, and release programs are to begin in the summer of 1991 in the United States. The three species are: *Galerucella calmaiensis* and *G. pusilla*, two chrysomelid beetles which attack the leaves and stems; and, *Hylobius transversovittatus*, which is a stem and root boring weevil (Batra *et al.* 1986, Ellen Fuge personal communication). Good control rather than eradication of the species is the goal, without the worry of potentially poisoning or stressing the surrounding environment, as may be the situation with herbicide application. Biological control would be more economically feasible and less labour intensive over the long term, if an effective agent were found.

CHAPTER 3

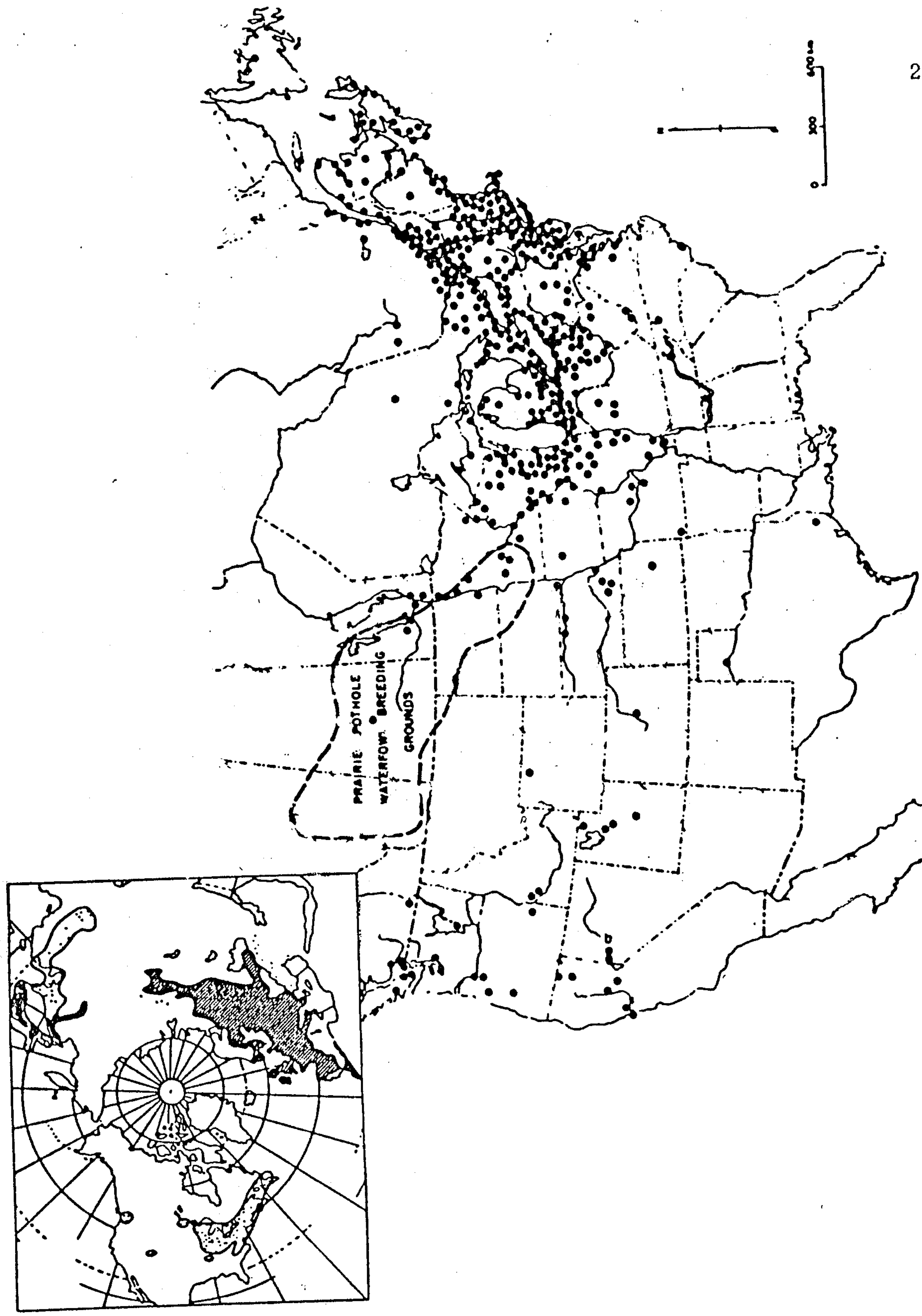
DISTRIBUTION

Lythrum salicaria is a species of nearly cosmopolitan distribution. Within its natural range it is most common in Britain and central and southern Europe, extending as far north as the southern coast of Norway and southeastern Sweden, though it is absent from Denmark and the northwest of Finland. Abundant in central Siberia and along the coastal areas of the USSR, it extends south in Korea, China, and Japan into the Himalayas in northern and western Asia. It is present in the Middle East, the Mediterranean islands and Northern Africa (Shamsi and Whitehead 1974a). Purple loosestrife has invaded other areas beyond its native range and has become established in Ethiopia, Australia, Tasmania, and North America (Thompson *et al.* 1987).

Figure 3.1 shows the distribution of *L. salicaria* in the Northern Hemisphere (inset) and the occurrence of the plant across North America as of 1985 (Thompson *et al.* 1987).

Temperature is believed to be the dominant factor determining its geographic distribution whereas light, nutrient and moisture play significant roles in its local distribution (Shamsi and Whitehead 1977b). *Lythrum*

Figure 3.1: Distribution of *Lythrum salicaria* in North America and across the Northern hemisphere (inset) as of 1985 (Thompson *et al.* 1987).


PRAIRIE POTHOLE WATERFOWL BREEDING GROUNDS

salicaria is restricted to moist or wet habitats such as streams, ditches, and marshes. This is due primarily to the requirements of the seed and seedling during germination and growth (Henderson 1987). *Lythrum salicaria* is a plant of sunny locations, but is able to tolerate up to 50 % shade once established. Purple loosestrife may be found growing in nutrient poor soil with a pH as low as 4 (Shamsi and Whitehead 1974a).

The first reliable report of *L. salicaria* in North America was published in 1814 in Pursh's Flora Americae Septentrionalis (Stuckey 1980). The species initially became established on the eastern seaboard of North America. Canals and the coastal shipping trade aided the spread of *L. salicaria*. Between 1901 and 1941, it colonized the Northwest Pacific coast and the Midwest. Since 1941 the plant has greatly expanded its range with new locations appearing in the Red River Valley along the Minnesota-North Dakota border and into Manitoba and the wetlands of Lakes Winnipeg and Manitoba. All the major watersheds in the U.S. have been colonized except for the Arkansas, Colorado, and Rio Grande river systems. Recently established populations in California, Idaho, Nebraska, Wyoming, and Washington indicate that purple loosestrife may become a problem in the far west in the future (Thompson *et al.* 1987). Most occurrences of *L. salicaria* in North America are within the boundaries of the former Wisconsin glaciation, but west of the Mississippi colonies are more scattered in their distribution (Balogh 1986).

In the eastern United States, where the plant is very well established with an estimated 190,000 hectares of wetlands are invaded each year (Hanna 1989). However, Thompson (1989) estimated the rate of spread between 1940 and 1980 to be 98,721 hectares per year. In Canada the species caused few problems until the 1930's when it became a very problematic weed in the flood plain pastures of the St. Lawrence River (US Fish and Wildlife 1979). However, as recently as 1944 there were no records of *L. salicaria* from Saskatchewan, New Brunswick, Prince Edward Island, or Newfoundland (Groh 1944). At the present time the plant is still most abundant in the St. Lawrence Valley and in Ontario (Stuckey 1980) but it is often seen in wet meadows and along streams in any of the ten Canadian Provinces (Cody 1978).

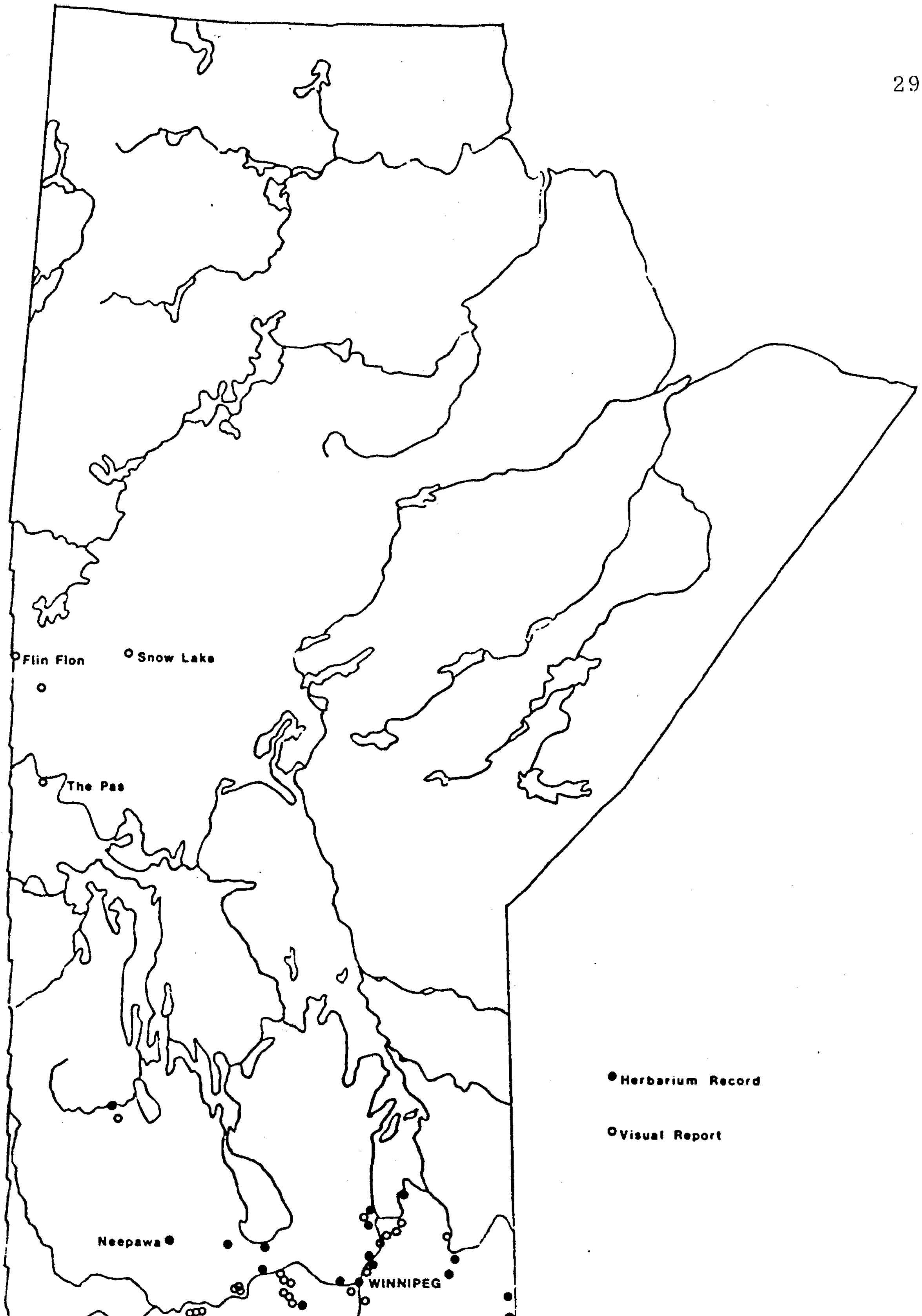
Purple loosestrife is particularly invasive on disturbed sites. Low water levels such as in drawdown or drought situations give the plant an opportunity to establish itself and spread. Isolated potholes are less susceptible to purple loosestrife, as are waterways with steep gradients and frequent scouring. Sites with mild gradients and broad alluvial deposits offer the most suitable colonizing sites (Thompson *et al.* 1987).

The first record of *L. salicaria* in Manitoba was from

Neepawa in 1896 (Scoggan 1957). Almost fifty years passed before a second collection of purple loosestrife was made in the province from Lockport in 1944. The first collection of a specimen from Winnipeg was made in 1951. From this period onward the number of collections has increased. By the end of the 1950's, *L. salicaria* was in Delta, Otterburne, Fannystelle, and Cypress River. Recently it has been reported in communities as far north as The Pas, Cranberry Portage, Flin Flon, and Snow Lake. *Lythrum salicaria* is widely spread along the Red and Assiniboine Rivers, especially in the Winnipeg area. Infestations of the plant seem particularly severe in Cypress River, Fannystelle, and the southern tip of Lake Winnipeg where there is a lot of marshland.

Figure 3.2 is a map of Manitoba indicating known locations of *L. salicaria* as of 1991. Approximately thirty-eight naturalized populations of purple loosestrife are known in the province. This distribution map is based on specimens examined from the following herbaria: University of Winnipeg (UWPG), University of Manitoba (WIN), Manitoba Museum of Man and Nature (MNNN), National Museum of Natural Sciences (CAN), and the Central Experimental Farm (CDA); and from reliable visual reports. The majority of these visual sightings were obtained from a Manitoba Agriculture *Lythrum* survey conducted in 1988 by Weed Supervisors in Manitoba's Weed Districts.

Figure 3.2: Distribution of *Lythrum salicaria* in Manitoba as of 1991. This distribution was established from reliable sightings (o) and from herbarium (UWPG, WIN, MNNN, CAN, CDA) specimens.


Flin Flon
Snow Lake
The Pas
Neepawa
WINNIPEG
Herbarium Record
Visual Report

Purple loosestrife has a strong foothold in Manitoba, but in Saskatchewan and Alberta its presence is minimal. There are three known locations for this species in Saskatchewan: Yorkton, Regina, and Saskatoon. All of these are believed to have been derived from garden escapes (Vernon Harms, personal communication). It is also known from three localities in Alberta: Medicine Hat, Drayton Valley and Calgary (Gary Trottier, personal communication). In British Columbia the plant is more extensively distributed. It may be relatively easy to prevent the spread of *L. salicaria* in the prairies but this would require a careful search for new individuals and a monitoring of colonies, with a follow-up of rapid eradication.

CHAPTER 4

SEED PRODUCTION, DISPERSAL, AND GERMINATION IN NATURALIZED AND CULTIVATED MANITOBAN *Lythrum* POPULATIONS

INTRODUCTION

The perennial herb, *Lythrum salicaria*, is a prolific seed producer, with estimates of annual outputs ranging from 108 000 (Shamsi and Whitehead 1974a) and 300 000 (Batra *et al.* 1986), up to 2.7 million (Thompson *et al.* 1987) seeds per plant. Vegetative propagules are not recorded for the species, though, injured or cut stems and rootstocks show rapid regeneration. It is well known by gardeners that cuttings root readily in water (Radosevich and Holt 1984). Since uprooted plants have been observed in floodwaters (personal observation), it is likely that the species may be spread in this manner also.

The seed is small (averaging 0.9 x 0.4 x 0.3 mm), planoconvex, glossy, light brown, obtriangular in longitudinal section and obliquely elliptic in cross-section (Montgomery 1977), and weighs only ca. 0.06 mg each (Shamsi and Whitehead 1974a). They are dispersed by water and wind (Thompson *et al.* 1987), but may be accidentally transported by humans or animals (Balogh 1986). The majority of seed is not buoyant in water until after germination, at which time it rises to the surface and may be dispersed as seedlings (Teale 1982). Seed maturation is staggered on the plant because of the long blooming season, and the earlier maturation of capsules in the lower part of the infructescence compared to those in the upper region. Killing frosts in the fall terminate the blooming period and the maturation of unripe seed.

The germinability of seed of *L. salicaria* is often very high. Various researchers have found germination levels ranging from 67% to 94% when the seed was provided with light and placed in a variety of temperature and substrate conditions (Anderson 1968, Shamsi and Whitehead 1974a).

Cultivars developed from *L. salicaria, L. virgatum*, and *L. alatum* are usually accepted, and extensively advertised, as sterile by the nursery trade. The cultivar 'Morden Pink', for example, is accepted as male sterile. There is no scientific documentation that establishes this as fact, however. Viable seed output from cultivated-wild or cultivar-cultivar crosses would cause serious problems to wetland and riparian sites. The complexity of the potential crosses of cultivated-cultivated and cultivated-wild populations has probably precluded a comprehensive study along these lines.

The purpose of this study was: 1) to examine the seed production from lower, middle, and upper regions of infructescences; 2) to monitor seed dispersal in a naturalized population over the winter months, and 3) to determine the seed germinability of several cultivated and wild *Lythrum* populations.

MATERIALS AND METHODS

1) SEED PRODUCTION

Infructescences were collected from twenty-three populations of *Lythrum* taxa; six of these were cultivars, eight were seed accessions, and nine were growing in the wild. The cultivated taxa were: 'Dropmore Purple', 'Rose Queen', 'Mr. Robert', 'Lady Sackville', 'Morden Rose', 'Pink Spires'; and seed accessions of *L. virgatum* SHR 80-322, *L. virgatum* GEN 83-122, *L. virgatum* DBG 84-165, *L. virgatum* LEI 83-131 (2 forms), *L. salicaria* TOR 79-018, *L. salicaria* LEI 83-130, and *L. salicaria* OSL 83-029. The naturalized Manitoban populations were from: Lockport (Gunns Creek), Lockport (ditch), Selkirk (Red River), Winnipeg on the Assiniboine River (Assiniboine Park entrance and Assiniboine Park footbridge), Delta, Breezy Point (Netley Marsh), Oak Bluff (ditch), and Portage la Prairie (Crescent Pond).

An infructescent spike was collected from 5 separate plants in each population between August and October 1988, and September 1989. Collections at this time were done at the peak of the fruiting season, though some seed dispersal may have begun. Three additional samples were collected in February 1989 (Delta, Breezy Point, and Portage la Prairie). Such results would reflect seed dispersal during the fall and winter months, and would be lower than those collections made in the fall. Three capsules were randomly chosen from each spike, one from each of the lower, central, and upper regions. Mature seed was removed from each capsule and counted.

2) SEED DISPERSAL

Seed dispersal was monitored over an eight month period from samples collected from a wild population at the Assiniboine Park footbridge on the Assiniboine River in Winnipeg. Sample spikes were collected each month between October 1988 and May 1989, after which time both stems and capsules had decayed. In addition to this, three collections were made from other naturalized populations at Gunns Creek, Lockport (Oct. 16/88, Feb. 2/89, and May 23, 1989) and the bank of the Red River in Selkirk (Oct. 4/88 and Feb. 13/89).

Three randomly selected capsules from each of the five spikes were examined and their seed numbers recorded. In all other respects, procedures were the same as those outlined in the previous section.

Viability of the seed was tested by means of germination trials using 200 seeds (4 replicates each of 50 seeds) from each sample collection. Germination procedures are described below.

3) GERMINABILITY OF CULTIVATED AND NATURALIZED POPULATIONS

Germination characteristics were determined for seed from 18 cultivars and seed accessions and 11 wild populations of *Lythrum* taxa in Manitoba. The selection of cultivars was restricted to those commonly grown as garden plants in Manitoba, or those that are being tested and developed for such purposes in the province. Wild populations were selected based on: 1) their spatial separation from each other, and; 2) their relative ease of access.

The twenty-nine seed sources used in this study were as follows:

CULTIVATED TAXA AND SEED ACCESSIONS--'Rosy Glow', 'Morden Gleam', 'Morden Pink', 'Morden Rose', 'Lady Sackville', 'Mr. Robert', 'Dropmore Purple', and 'Rose Queen' (Morden Research Station); *L. virgatum* cv. Pink Spires DBG 84-161, *L. virgatum* DBG 84-164, *L. virgatum* LEI 83-122, *L. virgatum* SHR 80-322, *L. virgatum* LEI 83-131 (2 forms), *L. salicaria* TOR 74-018, *L. salicaria* OSL 83-029, and *L. salicaria* LEI 83-130 (University of Manitoba Plant Sciences); and an unidentified cultivar from a Winnipeg garden. Two forms of *L. virgatum* LEI 83-131 were specified because there were two distinctly different growth forms present under the same listing indicating possible contamination.

NATURALIZED MANITOBAN POPULATIONS--a ditch on St. Mary's Road near St. Germaine, Lockport (Gunns Creek), Lockport (ditch), Selkirk (Red River), Winnipeg on the Assiniboine River (Assiniboine Park entrance and Assiniboine

Park footbridge), Delta, Breezy Point (Netley Marsh), Oak Bluff (ditch), Portage la Prairie (Crescent Pond), and Cypress River.

Samples of seed from each cultivar were collected in September 1988 from the Morden Research Station (MRS) in Morden, Manitoba, the University of Manitoba Plant Sciences Department, or, in one instance, from a Winnipeg garden. The pollen parents of cultivated samples were unknown. Most of the cultivars and seed accessions were growing in close proximity to each other and so cross-pollination between them was possible. Wild population samples were collected between August 1988 and September 1989.

Capsules were hand-removed from the infructescences and placed in a soil sieve shaker for five minutes to dislodge the seed and separate it from the chaff. Pore diameters of sieves that were used in this experiment were 2.00 mm, 1.80 mm, and 1.19 mm. The seed was then placed in a seed cleaner to remove any remaining chaff fragments. Seed was stored dry at room temperature in brown paper envelopes until required for germination tests. Undeveloped seed was separated by hand from the ripe ones.

Germination conditions were 14h/25°C light and 10h/10°C dark. Two hundred seeds (4 samples of 50 seeds each) from each seed source were placed in petri dishes in an environmental chamber and monitored daily for 19-21 days. Filter paper was placed in the bottom of the petri dishes and these were kept moist with distilled water. Seeds which had germinated were counted and removed. Germination levels of two cultivars, 'Rosy Glow' and 'Rose Queen', were tested using only 160 seeds (4 samples of 40 seeds each), because of the limited amount of seed available for these cultivars. 'Morden Pink' was not included in this test due to low seed output.

RESULTS

1) SEED PRODUCTION

Boxplots were used to illustrate the results of seed production and show the median, the fourth-spread, tail-length, and the outliers for each sample. A boxplot example is labelled and illustrated in Figure 4.1 (Emerson and Strenio 1983). Figures 4.2 and 4.3 show the number of seed produced in samples from each cultivar (Fig. 4.2) or wild (Fig. 4.3) population. Histograms on the x-axes indicate the three regions of the infructescence from which seed capsules were collected; i.e. lower (L), middle (M), and top (T) sections.

It was noted that capsules from the lower and mid regions of most infructescences generally had more seeds than those of the top regions. Another noticeable trend is that there is a lower overall seed output from cultivars compared to wild populations. Median values for cultivated samples ranged from approximately 3-50 seeds per capsule, whereas the median values for naturalized populations ranged from 10-85.

2) SEED DISPERSAL

Figure 4.4 shows the changing seed content of capsules between October 31, 1988 and May 31, 1989. The study population is naturalized and found growing along the Assiniboine River in Winnipeg. Figure 4.5 shows the two

Figure 4.1: Diagram of Boxplot with explanation. The rectangle represents the fourth-spread, with the median indicated by the horizontal line. Fourth-spread is the range of the data defined by the upper fourth and lower fourth, and is closely related to the interquartile range. Tail length is indicated by lines drawn to the most remote points which are not designated as outliers.
Circles and stars represent outliers (from Emerson and Strenio 1983).

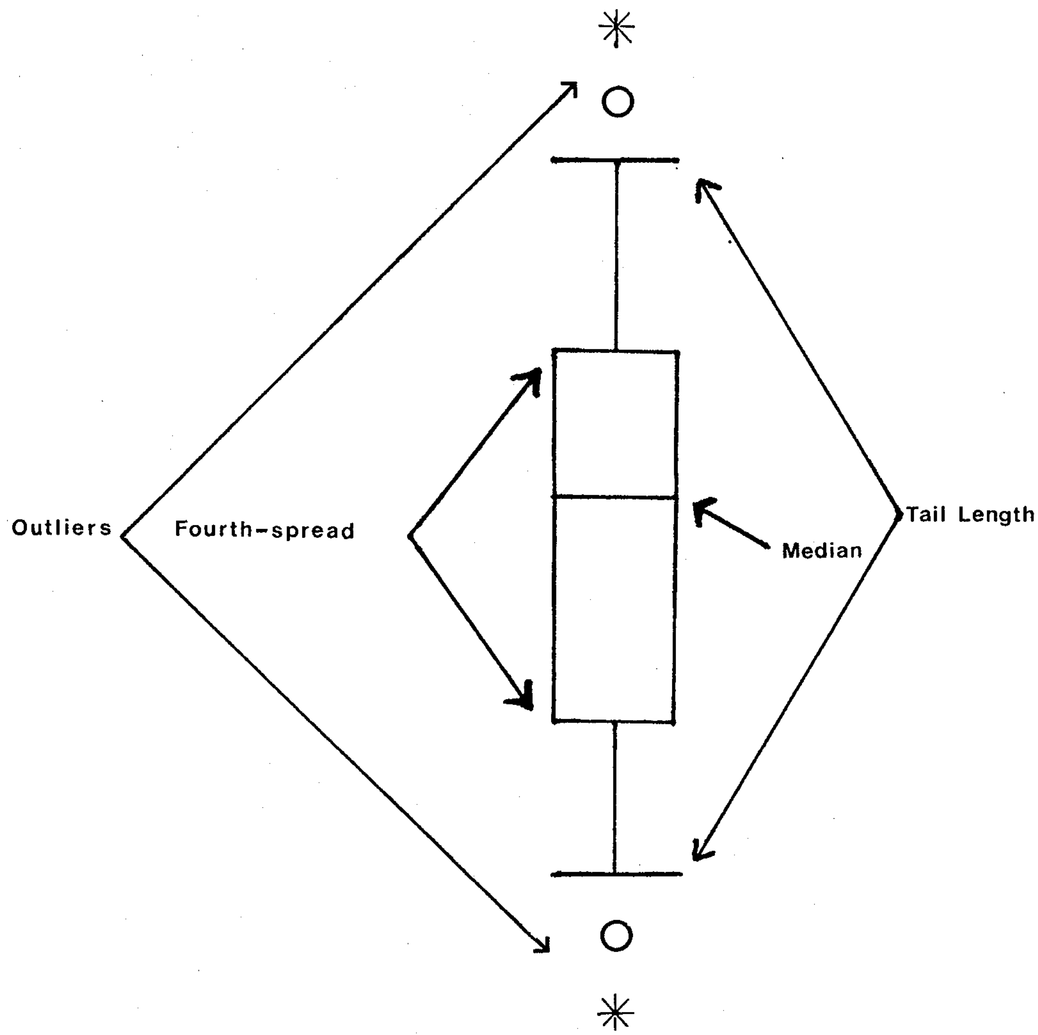
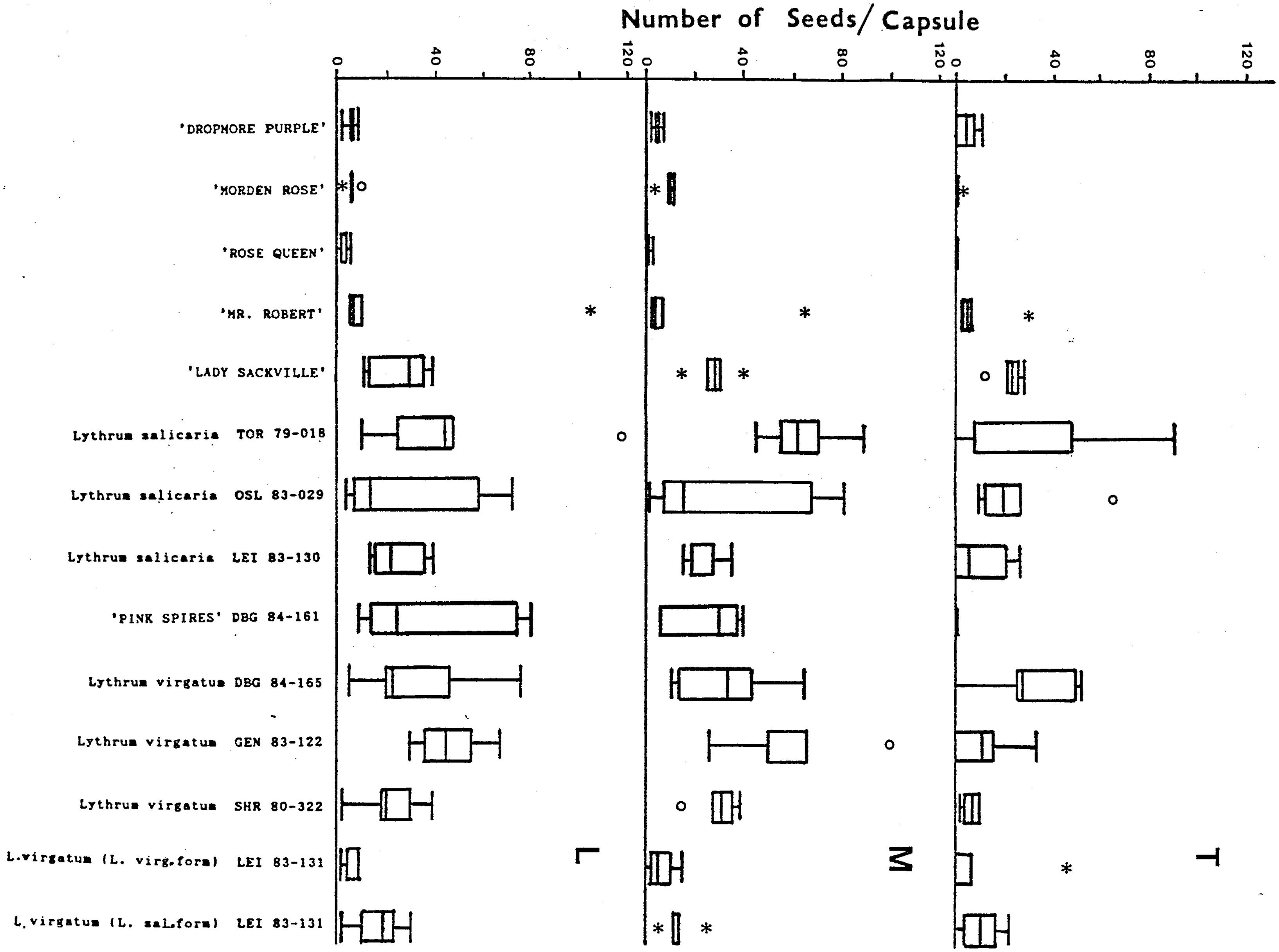

Outliers
Fourth-spread
Median
Tail Length

Figure 4.2: Boxplots illustrating the seed output/capsule for fourteen *Lythrum* cultivars and seed accessions. Capsules were taken from the top one-third (T), middle third (M) and the lower third (L) of the infructescence. (The boxplots show median, fourth-spread, extremes, and outliers, based on a batch of 5 values per sample).



Number of Seeds/ Capsule
0
40
80
120
'DROPMORE PURPLE'
'MORDEN ROSE'
'ROSE QUEEN'
'MR. ROBERT'
'LADY SACKVILLE'
Lythrum salicaria TOR 79-018
Lythrum salicaria OSL 83-029
Lythrum salicaria LEI 83-130
'PINK SPIRES' DBG 84-161
Lythrum virgatum DBG 84-165
Lythrum virgatum GEN 83-122
Lythrum virgatum SHR 80-322
L.virgatum (L. virg.form) LEI 83-131
L.virgatum (L. sal.form) LEI 83-131
L
M
T

Figure 4.3: Boxplots illustrating the seed output/capsule for nine naturalized *Lythrum* populations. Capsules were taken from the top one-third (T), middle third (M), and lower third (L) of the infructescence. (The boxplots show median, fourth-spread, extremes, and outliers, based on a batch of 5 values per sample).

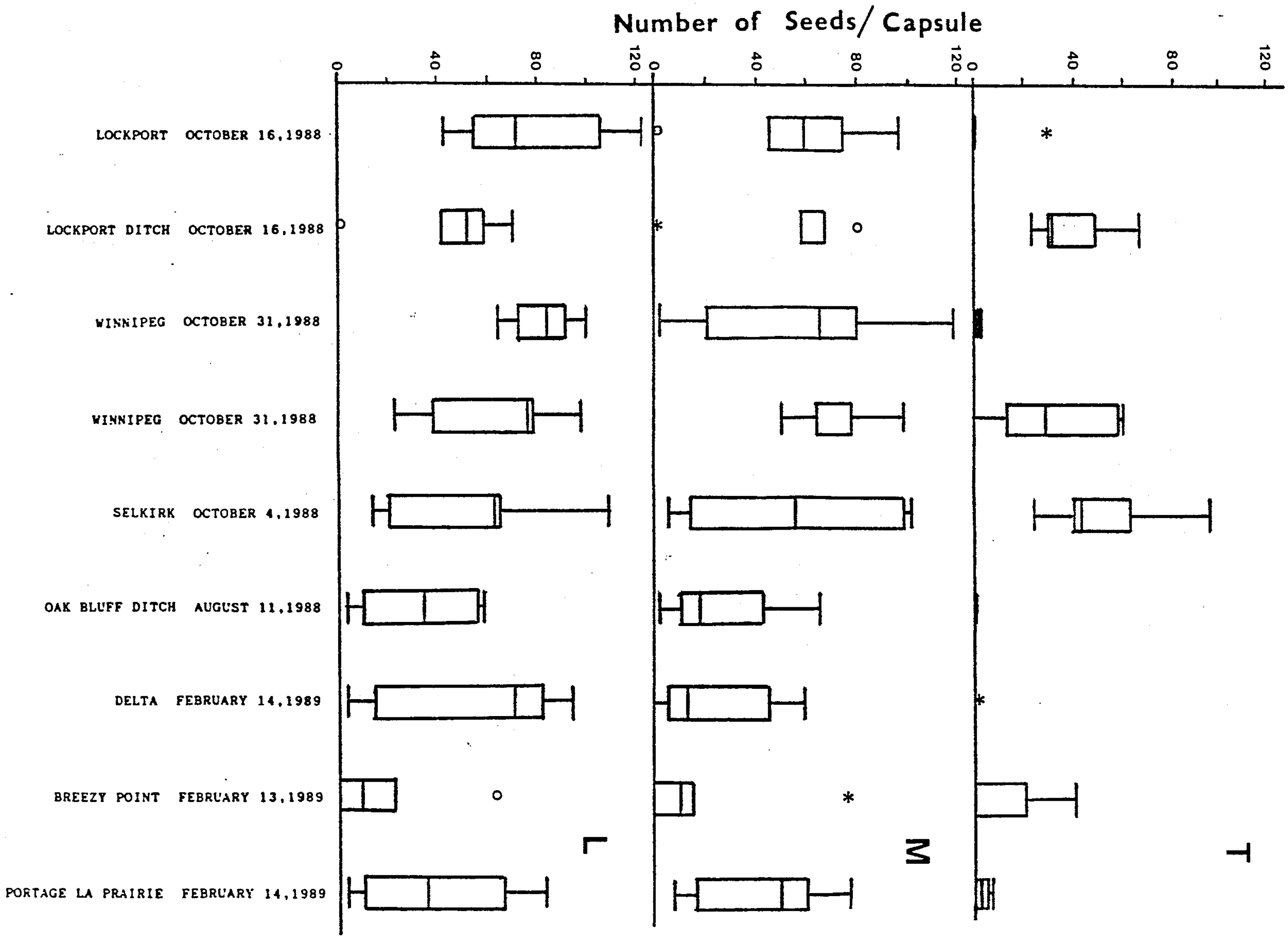

Number of Seeds/ Capsule
0
40
80
120
LOCKPORT OCTOBER 16,1988
LOCKPORT DITCH OCTOBER 16,1988
WINNIPEG OCTOBER 31,1988
WINNIPEG OCTOBER 31,1988
SELKIRK OCTOBER 4,1988
OAK BLUFF DITCH AUGUST 11,1988
DELTA FEBRUARY 14,1989
BREEZY POINT FEBRUARY 13,1989
PORTAGE LA PRAIRIE FEBRUARY 14,1989
L
M
T

Figure 4.4: Boxplots indicating the seed output/capsule from a naturalized population of purple loosestrife growing along the Assiniboine River in Winnipeg over an eight month period. Capsules were taken from the top one-third (T), middle third (M), and lower third (L) of the infructescence. (Boxplots show median, fourth-spread, extremes, and outliers, based on a batch of 5 values per sample).

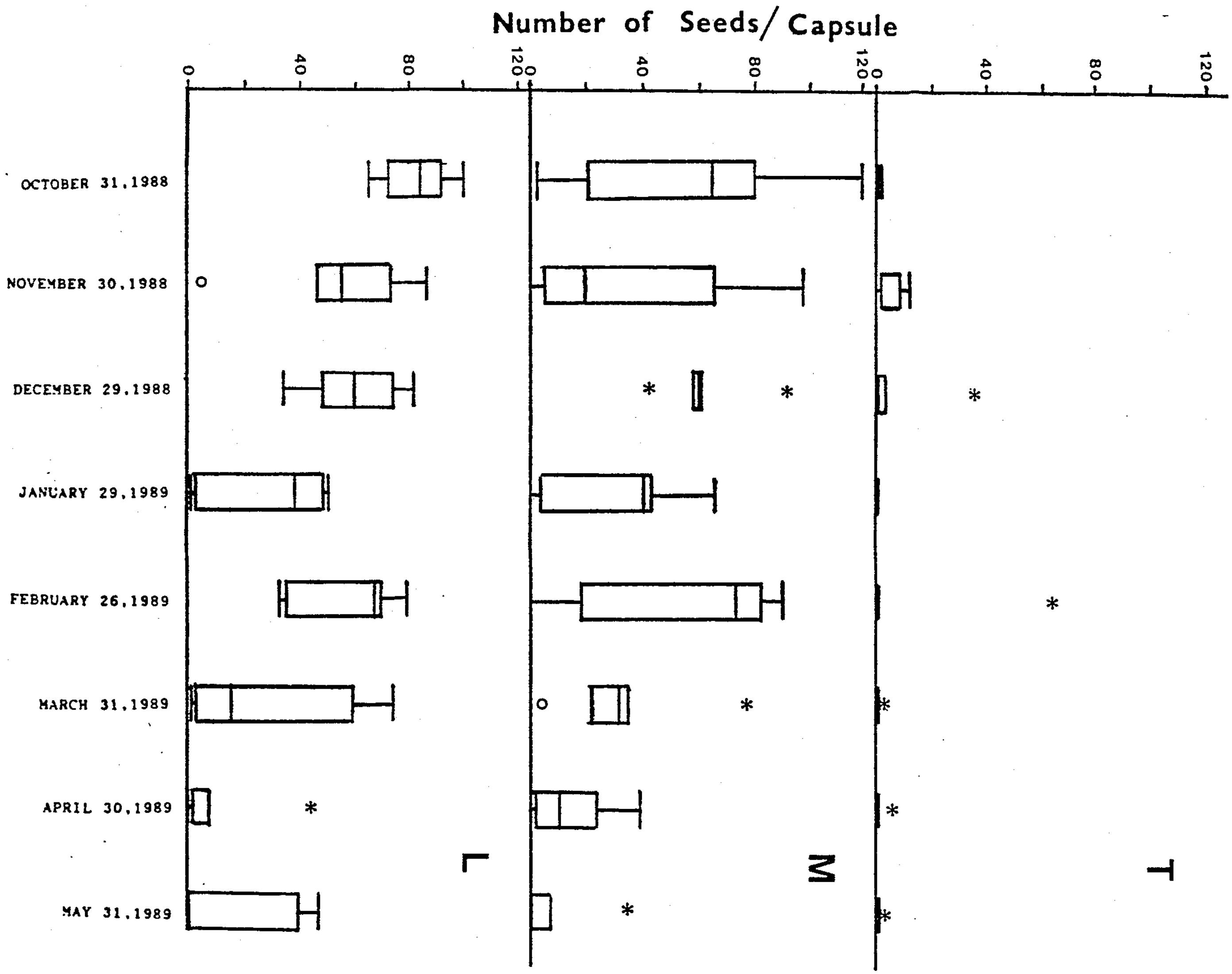
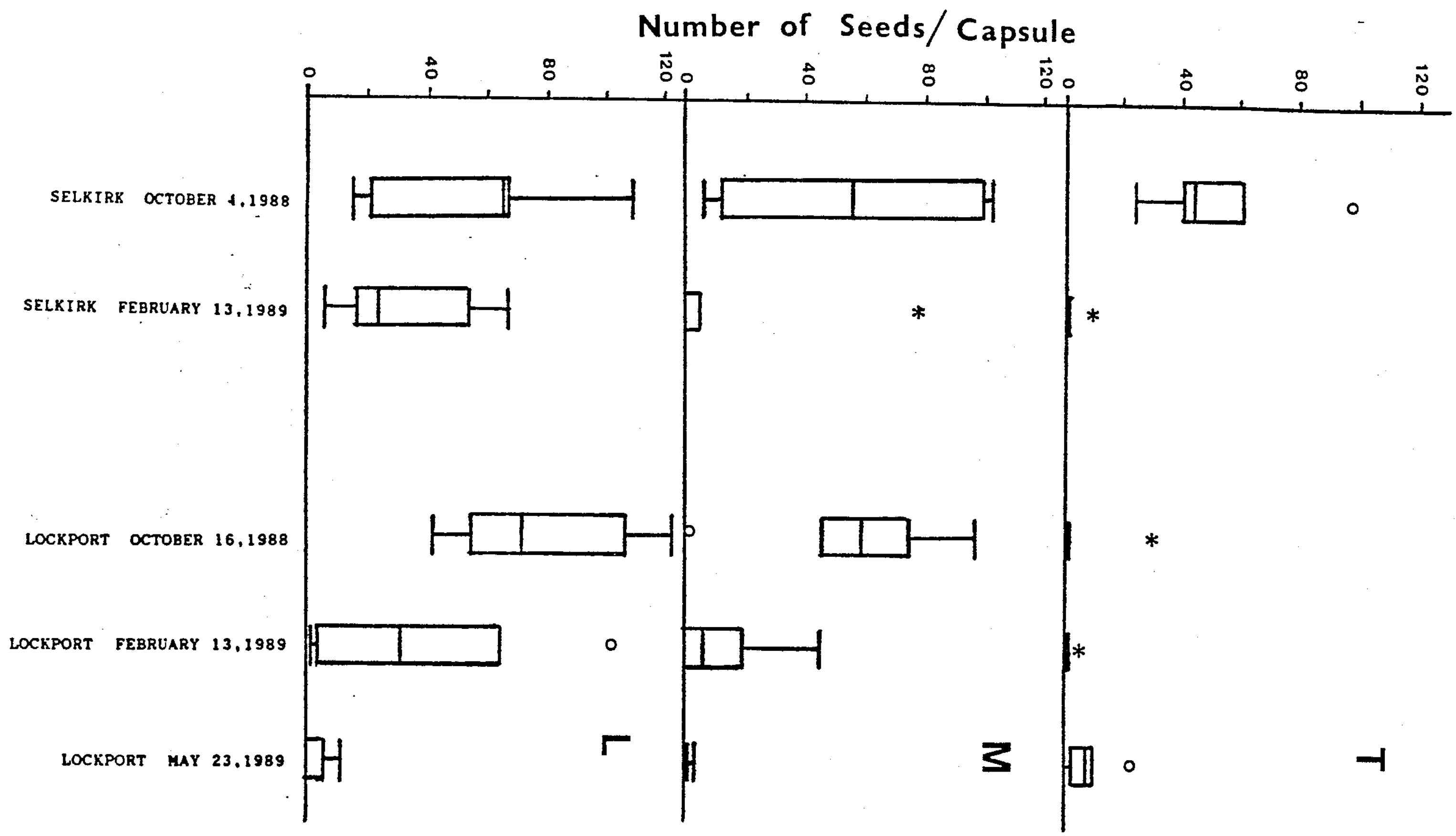

Number of Seeds/ Capsule
0
40
80
120
OCTOBER 31,1988
NOVEMBER 30,1988
DECEMBER 29,1988
JANUARY 29,1989
FEBRUARY 26,1989
MARCH 31,1989
APRIL 30,1989
MAY 31,1989
L
M
T

Figure 4.5: Boxplots illustrating the seed output/capsule for purple loosestrife from Selkirk and Lockport, Manitoba. Capsules were taken from the top one-third (T), middle third (M) and lower third (L) of the infructescence. (Boxplots show median, fourth-spread, extremes, and outliers, based on a batch of 5 values per sample).



Number of Seeds/ Capsule
0
40
80
120
SELKIRK OCTOBER 4,1988
SELKIRK FEBRUARY 13,1989
LOCKPORT OCTOBER 16,1988
LOCKPORT FEBRUARY 13,1989
LOCKPORT MAY 23,1989
L
M
T

collections for the Red River, Selkirk, and the three collections for Gunns Creek, Lockport, respectively.

It can be seen that the seed was dispersed gradually throughout the winter months. Capsules in the lower and mid regions apparently produced more seed than those in the upper region, as found in Figures 4.2 and 4.3. Capsules in the low and middle parts of the infructescence showed decreasing numbers of seed with time. This suggests that seed was dispersed gradually. Those in the capsules in the upper section of the infructescence possessed few seeds at any collection date. It is less likely that this was due to rapid dispersal than to poor seed production in the first place.

Germinability of the seed that had been collected over the duration of this study was tested and found to be at least 94.5 percent for any sample date or population.

3) GERMINABILITY OF CULTIVATED AND NATURALIZED TAXA

Table 4.1 lists the mean percent germination score of each of the twenty-eight seed sources examined. 'Morden Pink', was not included because of its very low seed output, though its seed was 100 % viable. Mean germination was 91.6 percent for the twenty-eight cultivated and wild populations examined, with ranges from 74.4 % to 100.0 %. The majority of seed germinations occurred between the second and eighth days.

Confidence Limits (99% level) were calculated for

Table 4.1: Mean percent germination in seeds of *Lythrum* taxa from naturalized and cultivated populations. (Values are means of 4 samples of 50 seeds each, or 40 seeds each for population indicated by ▼).

Abbreviations for sources are:

MRS = Morden Research Station, Morden
U. of M.= University of Manitoba, Plant Sciences
Wpg. = Winnipeg

DBG = Devonian Botanic Garden
GEN = Geneva
LEI = Leipzig
OSL = Oslo
TOR = Toronto

Populations	Source	Collection Date	Percent Germination
a) CULTIVATED POPULATIONS			
Lythrum virgatum x *L. salicaria* cv. Dropmore Purple	MRS	19/09/88	80.0
Lythrum virgatum cv. Morden Pink	MRS	19/09/88	----
Lythrum alatum x Morden Pink cv. Morden Gleam	MRS	19/09/88	86.5
Lythrum alatum x Morden Pink cv. Morden Rose	MRS	19/09/88	89.5
Lythrum virgatum cv. Rose Queen▼	MRS	19/09/88	74.4*
Lythrum cv. Mr. Robert	MRS	19/09/88	97.0
Lythrum cv. Lady Sackville	MRS	19/09/88	86.0
Lythrum cv. Rosy Glow▼	MRS	19/09/88	90.0
Lythrum salicaria (TOR 79-018)	U. of M.	23/09/88	94.5
Lythrum salicaria (OSL 83-029)	U. of M.	23/09/88	88.5
Lythrum salicaria (LEI 83-130)	U. of M.	23/09/88	92.0
Lythrum virgatum cv. Pink Spires (DBG 84-161)	U. of M.	23/09/88	91.0
Lythrum virgatum (DBG 84-165)	U. of M.	28/09/88	91.0
Lythrum virgatum (GEN 83-122)	U. of M.	23/09/88	93.0
Lythrum virgatum (SHR 80-322)	U. of M.	28/09/88	87.0
Lythrum virgatum (LEI 83-131 L.v.)	U. of M.	23/09/88	75.0*
Lythrum virgatum (LEI 83-131 L.s.	U. of M.	23/09/88	87.0
Lythrum sp.	Wpg.	10/09/88	88.5
b) NATURALIZED POPULATIONS			
Lythrum sp.	Gunns Creek, Lockport	16/10/88	100.0
Lythrum sp.	Lockport ditch	16/10/88	93.0
Lythrum sp.	Assiniboine River, Wpg.	31/10/88	99.5
Lythrum sp.	Assiniboine River, Wpg.	31/10/88	96.0
Lythrum sp.	Red River, Selkirk	04/10/88	99.0
Lythrum sp.	Oak Bluff ditch	11/08/88	100.0
Lythrum sp.	St. Mary's Rd. ditch	--/09/88	97.0
Lythrum sp.	Delta	14/02/89	100.0
Lythrum sp.	Netley Marsh	13/02/89	99.5
Lythrum sp.	Portage la Prairie	14/02/89	99.5
Lythrum sp.	Cypress River	14/09/89	91.5

* germination significantly low (p=.01)

combined germination scores in order to determine if any were outside the expected levels. The formula used was:

$$\hat{p} \pm 2.575\sqrt{\frac{\hat{p} \times \hat{q}}{n}}$$

$\hat{p}$ = proportion germinating
$\hat{q}$ = 1 - p
n = number of populations

The resulting lower and upper limits were 78.17 and 105.11, respectively. Two samples fell below these limits: 'Rose Queen' at 74.4 per cent, and *L. virgatum* (LEI 83-131) at 75.0 per cent. None of the cultivars were sterile.

DISCUSSION

Seed productivity is extremely variable in purple loosestrife, depending on the age of the individual and growing conditions (personal observation). An estimate of annual output may be calculated from mean values; i.e. a plant with an average of 110 seeds/capsule, 5 capsules/whorl, 70 whorls/spike, and 15 spikes/plant would yield approximately 600 000 seeds/year. This estimate (based on the study populations) falls between those of other researchers and is probably a reflection of the average age of plants used in this study. One year-old individuals may have only one to three inflorescences, whereas individuals that are several years old produce many more flowering stems and may resemble shrubs, producing great numbers of seed. Cultivars often have a much reduced seed output when compared to naturalized populations, which would generally be less than half, and as low as a fifth in cultivars such as 'Dropmore Purple', 'Morden Rose', 'Rose Queen', and 'Mr. Robert'.

Seed numbers obtained by examining capsules from the lower, mid, and upper regions of the flowering stem showed that the upper capsules often had less than one-half the seed of the lower and mid regions. Stout (1923) also reported that the average number of seed per capsule was lower toward the top of the branches of the plant. He stated that fewer seeds and smaller capsules, as a rule, are to be expected towards the apex as *Lythrum* spp. have indeterminant inflorescences. A factor that may affect seed output at the top of the infructescences is frost. It was found in the present study that many upper capsules contained immature seed at the time of the first autumn frost. Thus, the main reproductive output of the plant is associated with the lower and middle regions of the inflorescence. Towards late summer, it is likely that more resources are put into storage rather than reproduction in purple loosestrife. Thus, the late developing seeds at the top of the inflorescence are further disadvantaged.

Seed of purple loosestrife is dispersed primarily by wind and water (Shamsi and Whitehead 1974a). Despite the lack of any morphological adaptations for anemochory, their small size with a high surface/volume ratio indicates wind-dispersal is an effective means of spread (Fenner 1985). Long-distance dispersal by wind is unlikely as the seed is not as tiny as fern spores or orchid seed. However, wind would provide a way for short distance dispersal to other appropriate sites. Hydrochory is likely to be a very important means for downstream dispersion of the seed because *L. salicaria* is restricted to moist habitats. Establishment would probably occur if seed was deposited in open sites. The small seed is sensitive to debris accumulation (Keddy *et al.* 1989), and would not do well if dispersed into environments where this was occurring.

In the current studies it was found that seed dispersal in naturalized populations occurred gradually throughout the winter, not just in the late summer and fall. In addition to the kind of wind dispersal described above, wind is capable of moving the seed over the ice of adjacent water bodies both up or downstream, or even inland over snow. In some cases high snow drifts may cover infructescences and prevent dispersal until after the spring thaw. Capsules from the earliest blooming flowers produce ripe seed soonest and disperse their seed first (Thompson *et al.* 1987). However, capsules appressed to the main stem and capsules lower in the infructescence may be more protected from the wind, and may have to decay before dispersing their seeds. Capsules near or at the top of the plant, and those located on the outer periphery of the whorls, are more exposed to the environment and this may dislodge ripe seed more rapidly than those that are more protected.

Germination rates and viability of seed obtained from various naturalized and cultivated *Lythrum* populations in Manitoba were investigated. This was to give an indication of the potential success capabilities of dispersed seed in the wild and the possible contribution of cultivars to the spread of purple loosestrife.

Two important factors which may determine seed germination in *Lythrum* spp. are temperature and light. Shamsi and Whitehead (1974a) found that *L. salicaria* did not germinate below 14°C and determined that the critical minimum temperature was between 15 and 20°C. They found that increased temperatures had no effect on the rate of germination but that maximum percent germination was at 20°C in the light or 30°C in the dark. Shamsi and Whitehead (1977b) also reported that seed of *L. salicaria* required light to induce germination, though very low light intensities were effective. Light eliminates a germination inhibitor in the coats of seeds of *L. salicaria* (Barton 1965). The seed is viable for several years even though they are thin-walled and lack endosperm (Thompson *et al.* 1987).

The germination percentages of twenty-eight seed sources were very high, ranging from 74.4 percent to 100.00 percent. Only two populations had seed germination levels below that of the calculated 99 % confidence limits--*L. virgatum* cv. Rose Queen (74.4), and *L. virgatum* (LEI 83-131) at 75.0 per cent. *L. virgatum* cv. Morden Pink was excluded in this analysis because it had an extremely low seed output, which indicated it was the least fertile of all the samples collected. However, the few seeds that it did produce were viable. Cultivars have lower seed germination levels than naturalized populations, but they are certainly not sterile. *L. virgatum* and cultivars generally have a reduced reproductive capacity then compared to *L. salicaria* or hybrids. Cultivars sold by nurseries have the ability to produce viable seed, if in lower quantities than naturalized purple loosestrife.

Seed viability was very high, the mean germination of the cultivated and wild populations equalling 91.6 per cent. Seed collected over the winter months showed no reduction in germinability, which ranged from 94.5-100 percent. Long term viability of this seed is unknown but Shamsi and Whitehead (1974a) reported that seed is viable for at least three years. Thus, purple loosestrife is able to accrue a soil seed bank with germination occurring when conditions are conducive.

CONCLUSIONS

1. Lower and middle regions of the infructescence produced approximately twice the seed of the upper region. The upper region often had capsules containing immature seed.

2. Seed production studies showed that cultivars have a reduced reproductive capacity when compared to naturalized populations.

3. Seed of purple loosestrife is dispersed gradually over the fall and winter. Thus, seed may be dispersed over ice and snow via wind.

4. Germinability of 28 seed sources (17 cultivated and 11 naturalized) ranged from 74.4 - 100.0 percent. Mean germinability was 91.6 percent. Two samples, *L. virgatum* cv. Rose Queen and *L. virgatum* LEI 83-131 had significantly lower seed germinability at 74.4 and 75.0 percent, respectively, than the other samples. *Lythrum virgatum* cv. Morden Pink had a low seed output, but seeds that were produced were 100.0 percent viable.

CHAPTER 5

CROSSABILITY OF NATURALIZED AND CULTIVATED *Lythrum* TAXA

INTRODUCTION

Lythrum salicaria has a reproductive system designed for outbreeding. The species is tristylous, and individuals are of one of three different flower morphs. Long-styled plants possess two whorls of stamens at the mid and short levels, mid-styled plants possess two whorls of stamens at the long and short levels, and short-styled plants possess stamens at the long and mid levels. Legitimate crosses occur between styles of one length with stamens of the corresponding length (Darwin 1865). However, this self-incompatibility system is not absolutely strict (Darwin 1865, Stout 1923, Mulcahy and Caporello 1970).

Cultivars of *Lythrum* spp. have been developed by the horticulturalists and involve three species: *L. salicaria, L. virgatum,* and *L. alatum*. After the development of a desirable cultivar, propagation is carried out asexually via cuttings (Fehr 1987). Thus, a particular cultivar will exhibit only one style morph and is usually described as sterile by the nursery trade. These cultivars may in fact be capable of contributing viable pollen and producing viable seed-- if cross-pollination occurs with a different cultivar or wild population of a different style length.

Due to the invasive capabilities of purple loosestrife in the wild and the popularity of planting *Lythrum* cultivars as garden ornamentals, it was deemed important to analyze the role of cultivated and wild taxa of *Lythrum* spp. in the establishment of weedy populations. The objectives of this study were:

1. to examine the interfertility within and between three cultivars and one naturalized population of purple loosestrife.

2. to determine whether seed output from successful crosses may contribute to the spread of *Lythrum* spp. in North America.

3. to investigate the fertility of the offspring of these crosses.

MATERIALS AND METHODS

CROSSABILITY BETWEEN CULTIVATED AND NATURALIZED POPULATIONS

In order to test crossability, the three most popular and genetically diverse cultivars sold in Manitoba were selected for study. These were: 1) *L. virgatum* cv. Morden Pink; 2) *L. alatum* x 'Morden Pink' cv. Morden Gleam, and; 3) *L. salicaria* x *L. virgatum* cv. Dropmore Purple. 'Morden Pink' and 'Morden Gleam' are mid-styled, whereas 'Dropmore Purple' is long-styled. These are henceforth in this chapter known as MP, MG, and DP, respectively. The wild population selected for study was from Gunns Creek in Lockport, MB, and assumed to be *L. salicaria*. Since wild populations exhibit all three style morphs, it was necessary for crossing work to subdivide them into short, mid, and long-styled groups. Henceforth, these are known as LS, LM, and LL, respectively.

The pollination work was divided into three areas: 1) self-incompatibility between the long, mid, and short-styled Lockport plants; 2) selfing and crossing within the cultivars, and; 3) crossings between wild and cultivated plants. In total, this required 42 crosses--12 for each set of crossings examining the cultivated and wild population samples, and 18 when examining the interfertility between the cultivated and wild plants. Each cultivar and Lockport style length was represented by a sample of 10 plants, and 5 pollinations on each of the 10 plants were conducted for every cross. Thus, 50 pollinations for each of the 42 crosses totaled 2100 pollinations. (42 x 50= 2100).

This study was conducted in the University of Winnipeg greenhouse between June 1989 and July 1990.

The development of capsules indicated that a pollination had been successful. If not, the carpels withered and fell off. Ripe capsules were harvested from the plants after 6-8 weeks. Seeds were then counted, and germinability tested.

CROSSABILITY BETWEEN OFFSPRING, AND PARENTS AND OFFSPRING

The second part of this study examined the fertility of the offspring obtained from the 42 crosses conducted between naturalized and cultivated populations.

Eleven of the forty-two hybrids were examined. This selection was based on three criteria. First, all hybrids that were recognized as significantly or partially fertile, and of cultivated lineage, were examined. These were: MG x LM, LS x MG, LL x MG, and MP x LM (significantly fertile, RI's ≥ 0.6133); DP x MG, LM x MG, LL x MP, DP x LL, and LS x DP (partially fertile, RI's between 0.1016 and 0.2984). [The Ranging Index (RI) was based on the fertility of each hybrid and is defined later in the Materials and Methods under the Analysis of Data section]. Second, the LL x LL cross's offspring was chosen as it was significantly fertile and would serve as a representative for the wild crosses.

Third, an example of an infertile cross, MG x LL (RI= 0.0352), was included in order to examine the fertility of its offspring.

For each hybrid, there were long, mid, and short-styled representatives. Crossability was tested between the parents and offspring by backcrossing, and crossability was also tested between the offspring themselves.

Backcrosses-- progeny were crossed with pollen from each parent, and each parent received pollen from its offspring. Thus, there were four crosses for each hybrid's three style lengths: F_1 x ♀ parent, F_1 x ♂ parent, ♀ parent x F_1, and ♂ parent x F_1. If the pollen had been obtained from long stamens, DP x LL for example, plants of short or mid-style lengths were used in the crossing study, as they were the morphs that provided the pollen from the long stamens to begin with. If the female parent had originally been long-styled, as in LL x LL, then long-styled plants were used in the crossings.

Interfertility between offspring-- a style length was crossed with each of the three style morphs, and geitonogamously. Thus, there were four crosses for each hybrid's three style lengths: F_1 x itself, F_1 x F_1(short), F_1 x F_1(mid), and F_1 x F_1(long).

In total, for each hybrid there were: 3 style lengths x 8 crosses/style length (backcrosses and offspring interfertility) x 5 pollinations/cross equalling 120 pollinations. (3 x 8 x 5= 120).

In some instances, however, the offspring of a particular hybrid would not include short-styled plants because the dominant S allele was lacking. Only 70 pollinations were done for a hybrid in this situation. This occurred with: LM x MG, LL x MG, LL x MP, and DP x MG offspring, as was expected.

Co-ordination of the flowering of certain individuals was a problem, as some flowered too late and were unable to be included in the study. This occurred when crossing the offspring among themselves. Thus, there are some incomplete results for the following hybrid crosses: MG x LM, MP x LM, LS x DP, and LS x MG. Despite this, however, these data would still provide valuable information as to whether these offspring are sterile or fertile. In total, 1075 pollinations of a potential of 1120 were performed in this portion of the study.

Pollination work was conducted between March 1990 and October 1990.

Mature capsules were counted and removed from the plant, seed output recorded, and germinability tested for each case, as had been done with the parental crosses.

POLLINATION PROCEDURE

All pollination work carried out in this investigation used freshly opened flowers, i.e. less than 24 hours old. According to Stout (1923) fertilization success is best when petals just open and pollen is shed. Emasculation of the stamens was done using scissors where they were longer than the style of the female parent. Short stamens were not removed as they were unlikely to shed pollen on the stigmas, and were difficult to remove without damaging the pistil. Emasculation was done on flowers just prior to or shortly after anthesis. Pollinations were completed by rubbing mature anthers repeatedly over the stigmas. Stigmas were examined after pollination to ensure that there was pollen on them. All equipment (forceps, scissors) was sterilized with 95 % ethanol to prevent contamination between crosses (Davidson 1986).

Self-pollinations were geitonogamous, meaning pollen and stigmas were from the same plant. The flowers were not bagged after pollination because they were too small to be bagged independently. Covering the inflorescence would have further risked the probability of unwanted pollen landing on the stigmas.

Each cross-pollination was marked by a piece of coloured thread tied to the pedicel of the flower (Darwin 1899). In this way, each cross could be clearly marked with a differently coloured thread.

Ripeness of the seed was visually indicated by a browning of the capsules and pedicels. Generally, the fruits were mature and collected 6-8 weeks after pollination.

GERMINATION PROCEDURE

Germinability was tested to obtain a good indication of seed viability. This was done even if the seed production was very low. Only seeds that appeared fully developed were selected. All seed was placed on moist filter paper in petri plates. These were then placed in an incubator under 25°C/14h days and 10°C/10h nights until there was 100 percent germination, or for 19-21 days.

For the forty-two crosses conducted within and between cultivated and wild populations, forty of these yielded some seed. Whenever possible, four plates of fifty seeds per cross were tested. However, fewer seeds were available for use in many cases.

Further germination work was done with seed obtained from six of the F_1 generation crosses (MP x LM, DP x LL, DP x MG, MG x LL, MG x LM, and LL x LL), which included both backcrossings and interfertility tests between the offspring. These six representatives included the lineage of the three cultivars used in the study, and that of a wild cross. Only crosses which produced more than 50 seeds were tested. Two petri plates of 25 seeds per plate were placed in the incubator under the same conditions as above. Eighty-nine crosses were tested. If a particular cross involved using two different morphs, the seed obtained from such crossings were kept apart and germinated separately in the event of any obvious differences in viability.

RESULTS

A t-test was used to compare the fertility index data from a random collection of seed from the Lockport population, and the three greenhouse conducted Lockport crosses expected to have the highest yield. Results of the test showed that there were no significant differences ($t= 1.08$, $\alpha= 0.05$) between the seed output obtained in the greenhouse under artificial conditions and that seen in the wild state.

This allowed the calculation of a 95 % confidence interval using the RI results from the forty-two crosses and three additional seed collections from wild populations in Lockport, Winnipeg, and Selkirk. The formula was as follows:

$$\hat{p} \pm 1.96\sqrt{\frac{\hat{p} \times \hat{q}}{n}}$$

$\hat{p}$= proportion fertile
$\hat{q}$= proportion infertile
n= sample size.

The three naturalized samples were included in the analysis because they were considered indicative of the seed output seen in the wild. They could be used to determine whether any of the results from the cross-pollination work was of a comparable fertility. Three levels of fertility were recognized: significantly fertile (RI $\geq$ the upper limit of 0.2964); infertile (RI $\leq$ 0.0711), and; partially fertile (RI $\leq$ 0.2964, $\geq$ 0.0711).

Table 5.1 lists the forty-two crosses, their capsule yield (%), average seed output/capsule, Fertility Index

Table 5.1: Crosses between forty-two naturalized and cultivated *Lythrum* taxa (50 pollinations/cross).

Abbreviations for *Lythrum* taxa are:
- DP = Dropmore Purple (long-styled)
- MG = Morden Gleam (mid-styled)
- MP = Morden Pink (mid-styled)
- LL = Lockport long style/stamens (♀/♂ parent)
- LM = Lockport mid style/stamens
- LS = Lockport short style/stamens

FI = f(% capsules fertile) x f(average seed output/capsule)

$$RI = \frac{x - \text{minimum FI value}}{\text{maximum} - \text{minimum FI value}}$$

Legend
* significantly fertile
+ partially fertile

Pollen Acceptor	Pollen Donor	Capsule yield (%)	Average seed output/capsule	FI	RI
MG	LM	70	90.0	6300	1.0000*
LS	MG	72	70.0	5040	0.8000*
LL	LL	88	50.0	4400	0.6984*
LL	MG	86	48.0	4128	0.6552*
LM	LM	66	60.0	3960	0.6286*
LS	LS	84	47.0	3948	0.6267*
MP	LM	92	42.0	3864	0.6133*
DP	MG	94	20.0	1880	0.2984+
LM	LL	52	35.0	1820	0.2889+
LM	MG	46	39.5	1817	0.2884+
LL	MP	38	39.0	1492	0.2368+
DP	LL	88	16.0	1408	0.2235+
LS	DP	20	32.0	640	0.1016+
LM (self)		26	20.0	520	0.0825
MG	DP	14	29.0	406	0.0644
LS	MP	22	18.0	396	0.0629
LM	DP	22	17.0	374	0.0594
LM	MP	18	20.0	360	0.0571
LL	LM	26	9.0	234	0.0371
LL	LS	8	29.0	232	0.0363
MG	LS	4	56.0	224	0.0356
MG	LL	12	18.5	222	0.0352
LS	LM	10	22.0	220	0.0349
DP	LM	28	7.0	196	0.0311
MP	DP	8	24.0	192	0.0305
MP	MG	12	14.5	174	0.0276
MP	LS	6	25.0	150	0.0238
LL (self)		10	15.0	150	0.0238
LM	LS	12	12.0	144	0.0229
DP	MP	14	9.0	126	0.0200
MP	LL	10	12.0	120	0.0190
MG (self)		4	27.0	108	0.0171
MP (self)		4	15.0	60	0.0095
MP	MP	4	8.0	32	0.0051
LS (self)		2	10.0	20	0.0032
LL	DP	2	7.0	14	0.0022
LS	LL	2	5.0	10	0.0016
DP (self)		2	5.0	10	0.0016
DP	LS	4	3.0	7	0.0011
DP	DP	2	1.0	2	0.0003
MG	MG	0	0.0	0	0.0000
MG	MP	0	0.0	0	0.0000

values, and Ranging Index values. It also indicates the significantly fertile and partially fertile crosses determined by the confidence interval. These were: MG x LM, LS x MG, LL x LL, LL x MG, LM x LM, LS x LS, and MP x LM (7 crosses significantly fertile); DP x MG, LM x LL, LM x MG, LL x MP, DP x LL, and LS x DP (6 crosses partially fertile). The remaining 29 crosses were ranked as infertile. (The seed output values from the three wild samples included in the analysis of the confidence limits were found to be significantly fertile, as expected).

All three cultivars tested were able to contribute seed and pollen at the partial or significantly fertile levels. If a strict self-incompatibility system is assumed, seven crosses yielded unexpected results. Five of these crosses were classified as infertile when fertile results were anticipated based on the incompatibility system. These were: MP x DP, DP x MP, MG x DP, LS x MP, and LM x DP. Two other crosses were ranked as partially fertile when infertility was expected. These were the LM x MG and LM x LL crosses.

The germinability of the seeds obtained from forty of the forty-two crosses was tested. Mean germination was high, equalling 98.04 per cent (S.D.= 2.03). Germination ranged from 93.18 - 100.00 percent.

Tables 5.2-5.12 list the eleven hybrids used to test offspring fertility. Each table indicates the backcrosses

Tables 5.2-5.12: Backcross and offspring interfertility crosses for eleven hybrids of *Lythrum*. (Pollination work which was not completed because of an insufficient number of flowers to test are indicated by an 'incomplete').

Abbreviations for *Lythrum* taxa:

DP = Dropmore Purple (long-styled)
MG = Morden Gleam (mid-styled)
MP = Morden Pink (mid-styled)
LL = Lockport long style/stamens
LM = Lockport mid style/stamens
LS = Lockport short style/stamens

FI = (%capsules fertile) x (average seed output/capsule)

$$RI = \frac{x - \text{minimum FI value}}{\text{maximum} - \text{minimum FI value}}$$

Legend
* unexpected level of fertility
+ unexpected level of sterility

	Capsule yield (%)	Average seed output/capsule	FI	RI
LL x LL BACKCROSSES				
LL x LL long-styled				
F_1 x LL	0	0	0	0
F_1 x 2 LS, 3 LM	100	52.60	5260	0.3763
LL x F_1	0	0	0	0
2 LS, 3 LM x F_1	80	50.25	4020	0.2876
LL x LL mid-styled				
F_1 x LL	100	66.20	6620	0.4735
F_1 x 2 LS, 3 LM	100	46.80	4680	0.3348*
LL x F_1	100	45.60	4560	0.3262
3 LS, 2 LM x F_1	100	79.80	7980	0.5708*
LL x LL short-styled				
F_1 x LL	100	56.80	5680	0.4063
F_1 x 3 LS, 2 LM	20	55.00	1100	0.0787
LL x F_1	80	80.00	6400	0.4578
2 LS, 3 LM x F_1	80	30.75	2460	0.1750*
LL x LL OFFSPRING INTERFERTILITY				
LL x LL long-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	80	54.50	4360	0.3119
F_1 x F_1(short)	100	48.00	4800	0.3433
F_1 x F_1(long)	20	7.00	140	0.0100
LL x LL mid-styled				
F_1 x itself	100	59.80	5980	0.4278*
F_1 x F_1(short)	100	64.80	6480	0.4635
F_1 x F_1(long)	100	60.60	6060	0.4335
F_1 x F_1(mid)	20	2.00	40	0.0029
LL x LL short-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	80	78.50	6280	0.4492
F_1 x F_1(long)	80	87.75	7020	0.5021
F_1 x F_1(short)	0	0	0	0

	Capsule yield (%)	Average seed output/capsule	FI	RI
MG x LM BACKCROSSES				
MG x LM long-styled				
F_1 x MG	100	41.20	4120	0.2947
F_1 x 3 LS, 2 LL	60	92.33	5540	0.3963
MG x F_1	100	75.80	7580	0.5422
2 LS, 3 LL x F_1	60	57.67	3460	0.2475*
MG x LM mid-styled				
F_1 x MG	40	46.00	1840	0.1316*
F_1 x 2 LS, 3 LL	100	54.00	5400	0.3863
MG x F_1	60	19.33	1160	0.0830*
3 LS, 2 LL x F_1	80	48.25	3860	0.2761
MG x LM short-styled				
F_1 x MG	100	65.00	6500	0.4649
F_1 x 2 LS, 3 LL	100	46.60	4660	0.3333*
MG x F_1	100	89.00	8900	0.6366
3 LS, 2 LL x F_1	40	56.00	2240	0.1602
MG x LM OFFSPRING INTERFERTILITY				
MG x LM long-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	------------------incomplete-------------------			
F_1 x F_1(short)	------------------incomplete-------------------			
F_1 x F_1(long)	------------------incomplete-------------------			
MG x LM mid-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(short)	100	80.80	8080	0.5780
F_1 x F_1(long)	------------------incomplete-------------------			
F_1 x F_1(mid)	0	0	0	0
MG x LM short-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	100	52.20	5220	0.3734
F_1 x F_1(long)	------------------incomplete-------------------			
F_1 x F_1(short)	0	0	0	0

	Capsule yield (%)	Average seed output/capsule	FI	RI
LS x MG BACKCROSSES				
<u>LS x MG long-styled</u>				
F_1 x LS	80	65.00	5200	0.3720
F_1 x MG	100	45.40	4540	0.3247
LS x F_1	100	62.00	6200	0.4435
MG x F_1	100	82.40	8240	0.5894
<u>LS x MG mid-styled</u>				
F_1 x LS	100	107.20	10720	0.7668
F_1 x MG	0	0	0	0
LS x F_1	100	61.40	6140	0.4392
MG x F_1	0	0	0	0
<u>LS x MG short-styled</u>				
F_1 x LS	0	0	0	0
F_1 x MG	100	82.80	8280	0.5923
LS x F_1	40	31.00	1240	0.0887*
MG x F_1	60	62.67	3760	0.2690+
LS x MG OFFSPRING INTERFERTILITY				
<u>LS x MG long-styled</u>				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	100	70.00	7000	0.5007
F_1 x F_1(short)	100	65.20	6520	0.4664
F_1 x F_1(long)	------------------incomplete-------------------			
<u>LS x MG mid-styled</u>				
F_1 x itself	0	0	0	0
F_1 x F_1(short)	100	55.80	5580	0.3991
F_1 x F_1(long)	80	76.60	6128	0.4383
F_1 x F_1(mid)	0	0	0	0
<u>LS x MG short-styled</u>				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	100	74.00	7400	0.5293
F_1 x F_1(long)	100	79.20	7920	0.5665
F_1 x F_1(short)	0	0	0	0

	Capsule yield (%)	Average seed output/capsule	FI	RI
LL x MG BACKCROSSES				
LL x MG long-styled				
F_1 x LL	0	0	0	0
F_1 x MG	80	99.25	7940	0.5680
LL x F_1	0	0	0	0
MG x F_1	100	77.20	7720	0.5522
LL x MG mid-styled				
F_1 x LL	100	118.80	11880	0.8498
F_1 x MG	0	0	0	0
LL x F_1	80	55.75	4460	0.3190
MG x F_1	40	16.00	640	0.0458*
LL x MG short-styled	NO SHORT-STYLED PLANTS			
F_1 x LL				
F_1 x MG				
LL x F_1				
MG x F_1				
LL x MG OFFSPRING INTERFERTILITY				
LL x MG long-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	100	65.80	6580	0.4707
F_1 x F_1(short)	----------	----------	----------	----------
F_1 x F_1(long)	0	0	0	0
LL x MG mid-styled				
F_1 x itself	60	29.67	1780	0.1273*
F_1 x F_1(short)	----------	----------	----------	----------
F_1 x F_1(long)	100	126.20	12620	0.9027
F_1 x F_1(mid)	0	0	0	0
LL x MG short-styled	NO SHORT-STYLED PLANTS			
F_1 x itself				
F_1 x F_1(mid)				
F_1 x F_1(long)				
F_1 x F_1(short)				

	Capsule yield (%)	Average seed output/capsule	FI	RI
MP x LM BACKCROSSES				
MP x LM long-styled				
F_1 x MP	100	30.40	3040	0.2175
F_1 x 2 LS, 3 LL	100	40.80	4080	0.2918*
MP x F_1	100	39.40	3940	0.2818
3 LS, 2 LL x F_1	20	60.00	1200	0.0858+
MP x LM mid-styled				
F_1 x MP	100	22.00	2200	0.1574*
F_1 x 3 LS, 2 LL	100	49.20	4920	0.3519
MP x F_1	0	0	0	0
2 LS, 3 LL x F_1	100	50.60	5060	0.3619
MP x LM short-styled				
F_1 x MP	0	0	0	0+
F_1 x 3 LS, 2 LL	40	86.50	3460	0.2475
MP x F_1	20	12.00	240	0.0172+
2 LS, 3 LL x F_1	60	56.33	3380	0.2418
MP x LM OFFSPRING INTERFERTILITY				
MP x LM long-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	60	29.67	1780	0.1273+
F_1 x F_1(short)	80	20.00	1600	0.1144
F_1 x F_1(long)	0	0	0	0
MP x LM mid-styled				
F_1 x itself	80	41.50	3320	0.2375*
F_1 x F_1(short)	100	43.80	4380	0.3133
F_1 x F_1(long)	100	38.60	3860	0.2761
F_1 x F_1(mid)	-----------------incomplete-------------------			
MP x LM short-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	100	46.20	4620	0.3305
F_1 x F_1(long)	100	44.40	4440	0.3176
F_1 x F_1(short)	-----------------incomplete-------------------			

	Capsule yield (%)	Average seed output/capsule	FI	RI
DP x MG BACKCROSSES				
DP x MG long-styled				
F_1 x DP	0	0	0	0
F_1 x MG	100	93.40	9340	0.6681
DP x F_1	0	0	0	0
MG x F_1	80	75.75	6060	0.4335
DP x MG mid-styled				
F_1 x DP	40	36.50	1460	0.1044+
F_1 x MG	20	32.00	640	0.0458
DP x F_1	0	0	0	0+
MG x F_1	80	27.00	2160	0.1545*
DP x MG short-styled	NO SHORT-STYLED PLANTS			
F_1 x DP				
F_1 x MG				
DP x F_1				
MG x F_1				
DP x MG OFFSPRING INTERFERTILITY				
DP x MG long-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	100	58.60	5860	0.4192
F_1 x F_1(short)	----------	----------	----------	----------
F_1 x F_1(long)	0	0	0	0
DP x MG mid-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(short)	----------	----------	----------	----------
F_1 x F_1(long)	100	75.00	7500	0.5365
F_1 x F_1(mid)	0	0	0	0
DP x MG short-styled	NO SHORT-STYLED PLANTS			
F_1 x itself				
F_1 x F_1(mid)				
F_1 x F_1(long)				
F_1 x F_1(short)				

	Capsule yield (%)	Average seed output/capsule	FI	RI
LM x MG BACKCROSSES				
LM x MG long-styled				
F_1 x LM	100	115.20	11520	0.8240
F_1 x MG	100	73.00	7300	0.5222
LM x F_1	100	44.80	4480	0.3205
MG x F_1	80	73.00	5840	0.4177
LM x MG mid-styled				
F_1 x LM	0	0	0	0
F_1 x MG	0	0	0	0
LM x F_1	100	46.40	4640	0.3319*
MG x F_1	0	0	0	0
LM x MG short-styled	NO SHORT-STYLED PLANTS			
F_1 x LM				
F_1 x MG				
LM x F_1				
MG x F_1				
LM x MG OFFSPRING INTERFERTILITY				
LM x MG long-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	100	115.20	11520	0.8240
F_1 x F_1(short)	----	----	----	----
F_1 x F_1(long)	0	0	0	0
LM x MG mid-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(short)	----	----	----	----
F_1 x F_1(long)	100	139.80	13980	1.0000
F_1 x F_1(mid)	0	0	0	0
LM x MG short-styled	NO SHORT-STYLED PLANTS			
F_1 x itself				
F_1 x F_1(mid)				
F_1 x F_1(long)				
F_1 x F_1(short)				

	Capsule yield (%)	Average seed output/capsule	FI	RI
LL x MP BACKCROSSES				
LL x MP long-styled				
F_1 x LL	0	0	0	0
F_1 x MP	100	58.00	5800	0.4149
LL x F_1	0	0	0	0
MP x F_1	80	53.00	4240	0.3033
LL x MP mid-styled				
F_1 x LL	100	59.20	5920	0.4235
F_1 x MP	0	0	0	0
LL x F_1	100	55.00	5500	0.3934
MP x F_1	0	0	0	0
LL x MP short-styled	NO SHORT-STYLED PLANTS			
F_1 x LL				
F_1 x MP				
LL x F_1				
MP x F_1				
LL x MP OFFSPRING INTERFERTILITY				
LL x MP long-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	100	21.20	2120	0.1516
F_1 x F_1(short)	----------	----------	----------	----------
F_1 x F_1(long)	0	0	0	0
LL x MP mid-styled				
F_1 x itself	100	57.80	5780	0.4134*
F_1 x F_1(short)	----------	----------	----------	----------
F_1 x F_1(long)	100	58.60	5860	0.4192
F_1 x F_1(mid)	60	22.00	1320	0.0944*
LL x MP short-styled	NO SHORT-STYLED PLANTS			
F_1 x itself				
F_1 x F_1(mid)				
F_1 x F_1(long)				
F_1 x F_1(short)				

	Capsule yield (%)	Average seed output/capsule	FI	RI
DP x LL BACKCROSSES				
<u>DP x LL long-styled</u>				
F_1 x DP	0	0	0	0
F_1 x 3 LS, 2 LM	80	41.25	3300	0.2361
DP x F_1	0	0	0	0
3 LM, 2 LS x F_1	80	32.00	2560	0.1831
<u>DP x LL mid-styled</u>				
F_1 x DP	0	0	0	0+
F_1 x 2 LS, 3 LM	80	50.50	4040	0.2890
DP x F_1	100	22.40	2240	0.1606
3 LS, 2 LM x F_1	80	36.00	2880	0.2060
<u>DP x LL short-styled</u>				
F_1 x DP	100	21.60	2160	0.1645
F_1 x 3 LS, 2 LM	60	16.33	980	0.0701
DP x F_1	80	9.75	780	0.0558
2 LS, 3 LM x F_1	40	31.50	1260	0.0901
DP x LL OFFSPRING INTERFERTILITY				
<u>DP x LL long-styled</u>				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	60	29.33	1760	0.1259+
F_1 x F_1(short)	0	0	0	0+
F_1 x F_1(long)	0	0	0	0
<u>DP x LL mid-styled</u>				
F_1 x itself	100	25.00	2500	0.1788*
F_1 x F_1(short)	100	72.00	7200	0.5150
F_1 x F_1(long)	100	48.60	4860	0.3476
F_1 x F_1(mid)	0	0	0	0
<u>DP x LL short-styled</u>				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	80	26.50	2120	0.1516
F_1 x F_1(long)	100	29.60	2960	0.2117
F_1 x F_1(short)	0	0	0	0

	Capsule yield (%)	Average seed output/capsule	FI	RI
LS x DP BACKCROSSES				
<u>LS x DP long-styled</u>				
F_1 x LS	100	39.00	3900	0.2790
F_1 x DP	0	0	0	0
LS x F_1	100	28.80	2880	0.2060
DP x F_1	0	0	0	0
<u>LS x DP mid-styled</u>				
F_1 x LS	100	47.20	4720	0.3376
F_1 x DP	100	50.20	5020	0.3591
LS x F_1	100	44.80	4480	0.3205
DP x F_1	100	40.80	4080	0.2918
<u>LS x DP short-styled</u>				
F_1 x LS	40	17.00	680	0.0486*
F_1 x DP	80	11.75	940	0.0672
LS x F_1	0	0	0	0
DP x F_1	20	37.00	740	0.0529+
LS x DP OFFSPRING INTERFERTILITY				
<u>LS x DP long-styled</u>				
F_1 x itself	40	4.00	160	0.0114*
F_1 x F_1(mid)	100	33.20	3320	0.2375
F_1 x F_1(short)	40	24.50	980	0.0701+
F_1 x F_1(long)	20	16.00	320	0.0229
<u>LS x DP mid-styled</u>				
F_1 x itself	20	6.00	120	0.0086
F_1 x F_1(short)	100	59.80	5980	0.4278
F_1 x F_1(long)	100	43.20	4320	0.3090
F_1 x F_1(mid)	-----------------	incomplete	-------------------	
<u>LS x DP short-styled</u>				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	80	73.75	5900	0.4220
F_1 x F_1(long)	40	54.50	2180	0.1559+
F_1 x F_1(short)	0	0	0	0

	Capsule yield (%)	Average seed output/capsule	FI	RI
MG x LL BACKCROSSES				
MG x LL long-styled				
F_1 x MG	100	89.00	8900	0.6366
F_1 x 3 LS, 2 LM	100	92.20	9220	0.6595
MG x F_1	100	70.80	7080	0.5064
3 LS, 2 LM x F_1	80	36.25	2900	0.2074
MG x LL mid-styled				
F_1 x MG	0	0	0	0
F_1 x 2 LS, 3 LM	20	121.00	2420	0.1731
MG x F_1	0	0	0	0
3 LS, 2 LM x F_1	100	60.80	6080	0.4349*
MG x LL short-styled				
F_1 x MG	100	68.00	6800	0.4864
F_1 x 2 LS, 3 LM	60	128.67	7720	0.5522
MG x F_1	100	83.60	8360	0.5980
3 LS, 2 LM x F_1	40	40.00	1600	0.1144
MG x LL OFFSPRING INTERFERTILITY				
MG x LL long-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	100	87.20	8720	0.6237
F_1 x F_1(short)	100	82.80	8280	0.5923
F_1 x F_1(long)	0	0	0	0
MG x LL mid-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(short)	100	78.40	7840	0.5608
F_1 x F_1(long)	100	89.80	8980	0.6423
F_1 x F_1(mid)	0	0	0	0
MG x LL short-styled				
F_1 x itself	0	0	0	0
F_1 x F_1(mid)	100	103.80	10380	0.7425
F_1 x F_1(long)	100	68.40	6840	0.4893
F_1 x F_1(short)	0	0	0	0

and offspring interfertility crosses conducted, the capsule yield (%), the average seed output/capsule, and the Fertility and Ranging Index values. Pollination work which was not completed due to insufficient flowers for offpsring interfertility tests are indicated with an 'incomplete'. Based on the incompatibility system of the plant, certain crosses were expected to be infertile, whereas others were not. Unexpected results, which were marked by asterisks and plus signs, were most often obtained when crossing mid-styled plants. This was the case in 16 of the 35 anomalous cases, six of these involving mid-style crosses that were selfed, and ten involving crosses between a mid-style and mid-style. The results from two other crosses indicated mid pollen sterility in DP, and three indicated partial sterility of the DP style. Fertility between shorts crossed with short-styles and longs crossed with long-styles (four and three, respectively) is also worth noting. In the results where seed output was obtained unexpectedly, it was usually lower than compatible crosses.

The number of seeds per capsule ranged from 0 in an incompatible cross, to an average of 139.8 in a fertile cross between mid-styled and long-styled LM x MG offspring. This particular cross had been ranked as only partially fertile because of lowered capsule development and seed output. However, the offspring it did produce are extremely fertile. The MG x LL offspring were as fertile as any of the other progeny tested, though such a cross was ranked as infertile. Thus, it may be stated that whether seed output from a particular cross was very high or poor, based on compatiblity, the resultant offspring were generally fully fertile.

The viability of the seed of offspring was tested from six hybrids. These were: MP x LM, DP x LL, DP x MG, MG x LL, MG x LM, and LL x LL. Backcrossings and offspring interfertility pollinations had been conducted on each of these hybrids and seed germinability was tested from eighty-nine crosses. Mean germination was found to be high, equalling 97.35 percent (S.D.= 4.82). Germination values ranged from 72-100 percent. Only four crosses had germination values lower than 90.0 percent. These were at the 72, 74, 84, and 88 percent levels.

DISCUSSION

Lythrum salicaria possesses a heteromorphic, sporophytic self-incompatibility system (Brewbaker 1957, Vuilleumier 1967). The loci controlling self-incompatibility in tristylous species are closely linked with the loci controlling the floral polymorphisms (Ganders 1979). There are two loci (M and S), each with two alleles and epistatic interactions, which control tristyly (Crowe 1964, Real 1983). The S (short-style) locus is epistatic to the M locus. Homozygous recessives are long-styled i.e. ssmm, homozygous recessives for the S locus but a dominant allele at the M locus are mid-styled i.e. ssM-, and all others are short-styled i.e. S---(Ganders 1979). Tristyly serves to enhance compatible pollinations. The incompatibility system permits maximum seed production when pollinations occur between stigmas and anthers of equivalent lengths (Ornduff 1975). The incompatibility reaction comes into effect after pollination has taken place, and governs fertilization. The morphological arrangement is effective at the pollination stage, maximizing compatible style/stamen crosses (Yeo 1975).

This study sought to examine the interfertility within and between a naturalized and three cultivated *Lythrum* populations by analyzing their seed output, and to determine the fertility of the progeny produced. Such an experiment was necessary because the impact of cultivars on the naturalization of purple loosestrife has not been investigated. Many individuals defend the cultivars as sterile, unable to produce viable seed or pollen, and thus, are totally suitable as an ornamental. Others maintain that the cultivars are a problem, and have contaminated wetlands and waterways. A thorough cross-pollination program was required in order to determine the status of these cultivars.

It was found that the cultivars are capable of producing viable seed and pollen. At a partially fertile level all three cultivars studied-- 'Morden Pink', 'Dropmore Purple', and 'Morden Gleam'-- were able to contribute to the production of viable seed in four of the forty-two crosses. These crosses were: DP x MG, LM x MG, LL x MP, DP x LL, and LS x DP. Two of the cultivars, 'Morden Pink' and 'Morden Gleam', were able to contribute to crosses at the significantly fertile level. These four crosses were: MG x LM, MP x LM, LL x MG, and LS x MG. As was expected, the compatible Lockport crosses (LL x LL, LM x LM, and LS x LS) yielded significant fertile results.

A t-test showed that though this cross-pollination work was conducted under artificial conditions, comparisons between the seed output of naturalized Lockport plants was not significantly different ($\alpha=0.05$) from the three compatible Lockport crosses conducted in the greenhouse (LL x LL, LM x LM, and LS x LS). Thus, the results obtained from the crossing work is indicative of the crossability capabilities.

Seed obtained from the crosses was viable, averaging 98 percent germination. An examination of the offspring from eleven hybrids was considered necessary to determine their fertility. If sterile, the impact of the cultivar would be minimized in a naturalized environment as the progeny would be unable to sexually reproduce. If fertile, plants would not only be able to invade an area, but successfully reproduce and spread, even if the original cultivated influence was removed. Results of the capsule and seed output from these hybrids showed that they, too, were fertile. Germination of seed from six of the hybrids in order to ascertain their viability resulted in an average germinability of 97 percent. Though germinability was high from all three morphs, Nicholls (1987) reported that seeds of the mid-styled morph germinate more effectively than those of the long-styled, and seeds of the short-styled morph show very low levels of germination. He also found that mid-styles produce smaller, less fertile pollen but mature more seed than individuals of the other two style lengths. Darwin (1865) also determined that mid-styled plants produced the most seeds/capsule, and short-styles the least. Fisher and Mather (1943) believed that mid-styles showed an overall decrease in fertility in both seed and pollen when the entire plants' output was considered. Schoch-Bodmer (1938) suggested that there may be a difference in the seed viability between the different seeds. However, the current study showed no differences in the seed germinability of the three morphs.

Results from the crossability tests have shown that the self-incompatibility system of purple loosestrife is not strict, as already concluded from other researchers (Darwin 1865, Stout 1923, Mulcahy and Caporello 1970). It was examined again in this study because the flexibility of the incompatibility system may vary depending on the techniques and population samples used. Comparable results were necessary between both wild and cultivated populations in order to recognize fertility levels. The majority of supposedly illegitimate crosses did show reduced seed production. However, some crosses, though considered incompatible, were fecund. A few examples of this were the crosses MP x LM, LM x LL, LM x MG, and several backcross and interfertility tests between offspring.

In the majority of these unexpected fertility or sterility cases (16 of 35) it was the mid-styles which showed either self-fertility with themselves or other mid-styles, though a few incidences of short-short (4 of 35) and long-long (3 of 35) fertility occurred. Darwin (1865), Barlow (1913), Stout (1923), East (1927), and Charlesworth (1979) also found a high degree of self-fertility and interfertility in mid-styles, and much less frequently in long-styles and short-styles. Barrett (1977) reported that another tristylous species, *Pontederia rotundifolia*, also displayed more self-compatibility with mid-styles than the other two forms. Stout (1923) stated that it is the pollen from long stamens which permits compatibility. Dulberger (1970), working with *L. junceum*, found that though mid-styles gave the most fertile crosses, this species maintained its strict incompatibility system. Since *L. junceum* was diploid, and the European and North American *L. salicaria* is tetraploid, it was suggested that the increased ploidy may cause a lessening of the system's effectiveness.

Further unexpected results from the crossability tests indicated mid pollen sterility of 'Dropmore Purple', and partial sterility of its style. Visually, the mid anthers of some flowers never dehisced, or only a couple of the anthers in a flower successfully released pollen. However, many did appear fully fertile.

Stout (1923) stated that there is a lower seed output from capsules towards the apex of the infructescence. When conducting the pollinations, flowers were selected on the sole basis that they were fresh. It is possible that fertility and seed set may vary between those flowers which mature first and those which are differentiated later on in development. Shamsi and Whitehead (1974a) stated that although self-pollinations can be produced artificially, seed production is reduced. This implies that such self-pollinations would not occur in a naturalized or outdoor situation. However, insects are nondiscriminating and are capable of placing pollen from dehisced anthers onto incompatible stigmas. These insects are more efficient at selfing long- and mid-styled plants than shorts (Stout 1923). Thus, illegitimate pollinations involving short pollen are less likely to occur. Nicholls (1987) found that crosses applying illegitimate pollen before legitimate pollen from mid-styled morphs set fewer seeds than mid-style controls. Throughout the duration of the pollination work carried out in the current study, illegitimate pollinations were often made prior to the application of legitimate pollen. For example, if pollen from a cultivar was being used, application was accomplished with the longer stamen length first, then the shorter. If the female parent was short or mid-styled, it was often receiving incompatible pollen prior to compatible (eg. in a LS x MG cross, the short styled ♀ parent received first the long pollen and then the short pollen from the mid-styled ♂ parent). This may have lowered seed output results in some crosses. Pollen grains germinate as well after an illegitimate as after a legitimate pollination, but growth in the former is much slower (Kostoff 1927).

As concern regarding the detrimental effects of naturalized purple loosestrife mounts, it is becoming paramount to prevent its further establishment and spread. Horticultural *Lythrum* taxa are a threat, and this is likely to have ramifications in the nursery trade.

Though one cultivar grown in isolation would have almost a negligible seed output, it has been demonstrated from germinability tests that seed viability is very high, even if production is very low. Heavy rains or strong winds may be able to disperse seeds into an appropriate environment outside the garden. If more than one cultivar is grown together (two style lengths are present), or especially if they are grown near already naturalized purple loosestrife, the likelihood of contributing either pollen or seed is almost ensured. Large insects such as bees are the main pollinators of purple loosestrife (Darwin 1865) and are readily attracted to it for its nectar and ample pollen supply (Mead 1957).

Though the genetic make-up of many cultivars is unknown, development of cultivars has included three known species, *L. alatum, L. salicaria,* and *L. virgatum.* Cultivars with these parentages as well as those of *L. salicaria* are capable of producing viable seed and offspring. Though *L. alatum* is a distylous species, having lost the short form (Graham *et al.* 1987), the horticultural forms obtained from crossing between *L. alatum* and 'Morden Pink' [MG and 'Morden Rose' (MR)] are tristylous. All cultivars appear to be tristylous, regardless of their genetic background. The floral mechanisms of *L. alatum* and *L. salicaria* do not impose mechanical barriers to interspecific pollinations (Levin 1970). However, the viability of such direct crosses has not been commented upon.

There are mix-ups in the nursery trade which further compound the problem, as restricting sales to a couple of relatively hazard-free cultivars may not be adhered to. An example of this was seen regarding the sale of 'Morden Gleam', where long-styled, deep pink flowered plants were being marketed instead of mid-styled individuals. Some nurseries also propagate their stock by seed rather than cuttings. Such a practise is clear evidence that the cultivated *Lythrum* taxa are fertile.

It is important to recognize the ability of the three study cultivars to contribute to the spread and establishment of naturalized populations of purple loosestrife. Other cultivars likely demonstrate similar crossability, seed viability, and offspring fecundity patterns. It is recommended that measures should be taken to minimize the effect of cultivars of purple loosestrife, especially in regions where the plant is not already growing in the naturalized state.

CONCLUSIONS

1. The self-incompatibility system is not strict. Most incompatible crosses yielded greatly reduced seed outputs in comparison to compatible crosses, but some were highly fecund.

Mid-styles showed a high degree of self-fertility with themselves and other mid-styles.

2. Crossability tests between 'Morden Pink', 'Dropmore Purple', 'Morden Gleam' and Lockport individuals has shown that cultivars produce viable seed and pollen.

All three cultivars contributed seed and/or pollen at the partially fertile level (DP x MG, LM x MG, LL x MP, DP x LL, and LS x DP), and 'Morden Pink' and 'Morden Gleam' at the significantly fertile level (MP x LM, MG x LM, LL x MG, and LS x MG).

Compatible Lockport crosses (LL x LL, LM x LM, and LS x LS) also yielded significantly fertile results.

Unexpected fertility was noted in the LM x MG and LM x LL crosses. Sterility was noted in the LS x MP, LM x DP, MG x DP, MP x DP and DP x MP crosses, indicating evidence of some sterility in 'Dropmore Purple' and perhaps 'Morden Pink'.

3. Cross-pollination work with hybrids showed them to be highly fertile.

4. Germinability of seeds from the parental crosses averaged 98 percent. The germinability of the hybrid seeds that were examined averaged 97 percent.

5. Cultivars are capable of contributing viable seed and pollen to the spread of purple loosestrife. The sale of cultivars, regardless of parentage, should be discouraged.

CHAPTER 6

MORPHOLOGICAL CHARACTERISTICS OF *Lythrum* TAXA

INTRODUCTION

The Lythraceae (Loosestrife) family is comprised of approximately twenty-five genera and 550 species (Mitchell 1976). The genus *Lythrum* consists of thirty-five species worldwide. Three of these (*L. salicaria* L., *L. virgatum* L., and *L. alatum* Pursh) were pertinent to this study.

The plants are 4-6-merous and are perigynous. Sepals are short and alternate with appendages (epicalyx) in the sinuses of the calyx. The number of stamens may be equal to or twice as many as the petals in dimorphic and trimorphic species, respectively. The ovary is composed of two fused carpels, and the mature septicidal capsule is enclosed by the persistent hypanthium (Gleason 1974). The plants are herbaceous perennials which perennate by means of rootstocks (Shamsi and Whitehead 1974a).

Characters which have been traditionally used to distinguish the three study species of *Lythrum* are: leaf shape and phyllotaxy; plant pubescence; type of inflorescence; flower styly; and the lengths of calyx appendages and calyx lobes.

Lythrum alatum has linear oblong to ovate lanceolate leaves which are alternate in the median and upper stem leaves which are alternate in the median and upper stem levels. The species is glabrous and distylous, with solitary flowers borne in the inflorescence axils (Scoggan 1979).

Lythrum virgatum has lanceolate leaves narrowed at the base, and like *L. alatum*, is glabrous. This species is tristylous, and the length of the calyx appendages is equal to those of the calyx lobes (Shinners 1953).

Lythrum salicaria has oblong lanceolate to deltoid ovate leaves which are truncate or cordate at the base and sessile on the stem. The leaf arrangement may be opposite, alternate, or whorled, with stems 4-6 sided. The species is typically pubescent and the tallest of the three *Lythrum* species, reaching six to seven feet in height. The species is tristylous and the inflorescence is a spiciform thyrse (Rawinski 1982). The appendages of the calyx are at least twice as long as the calyx lobes (Gleason 1974).

The horticulture industry has developed cultivars from the above-mentioned species or their hybrids. Unfortunately, it is often morphologically very difficult for horticulturalists to distinguish the hybrids from each other or from naturalized populations of purple loosestrife. The invasive, wild-growing species is purportedly *L. salicaria*. However, it is thought by many that horticultural *Lythrum* taxa may be escaping and contributing to the spread of purple loosestrife. Thus, the ability to distinguish the cultivars from each other, and to examine whether cultivated strains may have contributed to wild populations, is important. Such findings may have ramifications in the nursery trade if the continued spread of purple loosestrife in North America is to be prevented.

The purpose of this study was: i/ to determine the best discriminating floral characteristics for the three most common cultivated species and for plants from three naturalized populations; ii/ to determine if the progeny of cross-pollinations between and amongst wild and cultivated plants were morphologically similar to their parents, and; iii/ to construct an identification key for the cultivars based on the best floral discriminators

MATERIALS AND METHODS

BASIC PLANT MAINTENANCE AND GROWTH

Lythrums were grown and maintained in the University of Winnipeg greenhouse, where all cross-pollination work and morphological measurements were conducted. Plants were watered daily and given artificial light conditions of 14 hour days and 10 hour nights. Purple loosestrife will not flower with less than a 13 hour photoperiod (Shamsi and Whitehead 1974b). Also, flowering is retarded or prevented if overcrowded with other individuals in the same pot. Temperature was not controlled, except for a minimum of 17°C. Seedlings were started in flats and then individually repotted into successively larger pots up to 6" in diameter. When plants finished their main flush of blossoming, stems were cut down and rootstocks were placed in 5°C cold storage for no less than 40 days, then returned to the greenhouse. Such a chilling period encouraged very rapid and vigourous growth, with flowering occurring about 3-4 weeks later.

Seed was germinated in petri-plates on filter paper moistened with distilled water and placed in an incubator under a 14h/25°C day and 10h/10°C night photoperiod/temperature regime. When the cotyledons were fully expanded, seedlings were transferred to flats in the greenhouse. They were potted in a 2:1 potting soil:vermiculite mixture. A duration of 8-10 weeks would pass for the plants to reach the flowering stage from the time of seed germination. Shamsi and Whitehead (1974a) found their plants required the same time frame.

Aphids were a problem in the greenhouse and it was necessary to chemically treat the plants with the systemic insecticide Temik (eighth of a teaspoon applied per 6 inch pot on top of the soil). Application was required approximately every three months.

FLORAL MORPHOLOGY OF CULTIVATED AND NATURALIZED POPULATIONS

A) Parent plants.

Three cultivars sold in Manitoba were selected for study because of their popularity in the nursery trade and their genetic differences. These were: *L. virgatum* cv. Morden Pink, *L. alatum* x Morden Pink cv. Morden Gleam, and *L. salicaria* x *L. virgatum* cv. Dropmore Purple. Sample cuttings of 'Morden Pink' and 'Dropmore Purple' were provided by T & T Seeds (Winnipeg), and additional cuttings of 'Morden Pink', 'Dropmore Purple', and 'Morden Gleam' were obtained from the Agriculture Canada Morden Research Station (Morden, MB). Naturalized samples of purple loosestrife were collected from three sites in Manitoba. These were: Selkirk (Red River), Lockport (Gunns Creek off the Red River) and Winnipeg (Assiniboine River).

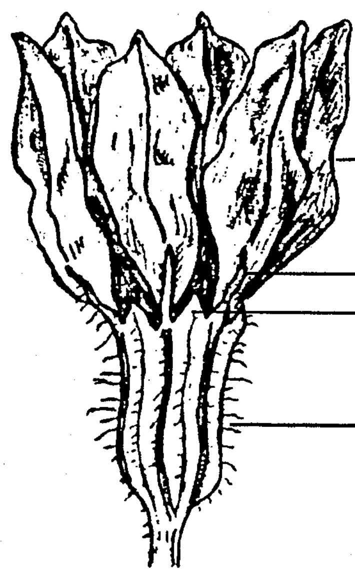
The floral characters selected for measurement were: 1) petal length (mm); 2) petal width (mm); 3) total hypanthium and calyx length (mm); 4) calyx lobe length (mm);

5) calyx appendage length (mm); 6) hypanthium pubescence (length of hairs on the hypanthium ridges (mm); 7) hypanthium hair number (per 0.25 mm^2), and; 8) number of flowers borne in inflorescence axils. Figure 6.1 is an illustration of a purple loosestrife flower, and indicates some of the structures measured in the study (redrawn from Gleason 1974).

Forty flowers, two from each of twenty plants, were measured from each of the six taxa. The one exception was 'Morden Pink', where measurements were obtained from 18 plants (36 flowers, not 40). Measurements were done using a LEITZ dissecting microscope at three magnifications. The 6.4x magnification was used to measure the petal length, petal width, and hypanthium-calyx lengths. The 16x magnification was used for the calyx lobe lengths and calyx appendage lengths. Finally, the 40x magnification was used to measure hypanthium pubescence length and hair number. The number of flowers per inflorescence axil was obtained from an average of ten axil readings per plant. Hypanthium hair number values were based on the average of five measurements per flower over a 0.25 mm^2 area.

Measurements were conducted between the summer of 1989 and summer of 1990 from greenhouse grown plants. The only exception was for the plants from the Selkirk population, whose inflorescences were gathered from the original site and measured the following day.

Figure 6.1: Flower of *Lythrum salicaria*. (Redrawn from Gleason 1974).


petal
calyx appendage
calyx lobe
hypanthium

Sample flowers were chosen randomly from a region of the inflorescence where anthesis had recently occurred.

Data were analyzed using Discriminant Analysis (SAS Program), and Canonical Discriminant Analysis. Canonical discriminant analysis is a multivariate technique which determines whether two or more groups of individuals are significantly different from one another. The method also finds the variables which are the best discriminating variables of the groups, and displays the relationships between groups in an ordination scattergram. The method uses eigenanalysis to obtain linear composites (similar to prinicpal components analysis axes) which maximize group discrimination. Discriminant analysis (eigenvector elements) indicate which variables are the best discriminators for a given discriminant axis. The discriminant analysis using the SAS Program was used to categorize the parent plants into various groupings based on their floral character measurements.

No vegetative characters were measured in this study. It was felt that their variability and susceptibility to modification under various environmental conditions would render them unreliable.

FLORAL MORPHOLOGY OF CULTIVATED AND NATURALIZED POPULATIONS

B) Progeny.

Forty-two cross-pollinations were conducted between and within 'Morden Pink' (MP), 'Morden Gleam' (MG), 'Dropmore Purple' (DP), and the wild population from Lockport. Thirty-seven crosses yielded offspring on which floral characters could be measured. Two of the forty-two crosses, MG x MG and MG x MP, were completely sterile. Three produced a few offspring, but these failed to flower in time for inclusion in this report.

The same floral characters were measured as outlined in the previous section (A). When possible, two flowers from each of ten plants were measured for each cross. In many cases, flowering did not occur within the allotted time limitations, and thus, the data for certain progeny is unavailable. In total, 295 individuals were measured.

These data were analyzed by Discriminant Analysis so that the 295 offspring could be allocated into one of the six populations originally tested. Four of these six taxa were parental stocks ('Morden Pink', 'Morden Gleam', 'Dropmore Purple', and Lockport), and the two others (Winnipeg and Selkirk) were included despite the progeny having no direct genetic affiliation with them. Such comparisons were meant to find the degree of any floral similarity between offspring and their parents. This could be done because the six populations were shown to be distinctly different from one another morphologically in Part (A).

RESULTS

The morphological data collected on the three cultivated and three naturalized population samples were analyzed by Discriminant Analysis to find differences or similarities between the taxa. Table 6.1 lists, for each of the six *Lythrum* populations, the means and standard deviations for the eight floral characteristics measured.

All three wild populations (Winnipeg, Lockport, and Selkirk) had calyx appendages which were longer than calyx lobes. For this character, the wild plants resembled *L. salicaria*. The cultivar 'Morden Pink' had calyx appendages which were equal to or shorter than the calyx lobes, and 'Dropmore Purple' had calyx appendages which were scarcely more than half the lengths of the calyx lobes. However, the calyx appendages of 'Morden Gleam' exceeded the lengths of the calyx lobes.

Secondly, hypanthium pubescence length ranged from 0.12-0.15 mm in the cultivars, whereas in wild populations it ranged between 0.36-0.53 mm. The latter values were more than twice to three times the hair lengths of the cultivars. There was a trend for a greater density of hairs in populations with greater hair length.

Thirdly, the number of flowers borne in inflorescence axils was higher in the wild populations (4.84-6.15) than the cultivated populations (3.34-4.34). Finally, it can be

Table 6.1: Means and standard deviations of floral characteristics for six *Lythrum* populations.

VARIABLE	Morden Pink MEAN	Morden Pink STD.DEV.	Morden Gleam MEAN	Morden Gleam STD.DEV.	Dropmore Purple MEAN	Dropmore Purple STD.DEV.	Winnipeg (A.R.) MEAN	Winnipeg (A.R.) STD.DEV.	Lockport MEAN	Lockport STD.DEV.	Selkirk MEAN	Selkirk STD.DEV.
petal length (mm)	9.72	0.67	9.54	0.71	9.52	1.22	10.12	0.89	8.95	1.40	11.91	1.09
petal width (mm)	4.50	0.49	5.60	0.48	2.99	0.47	3.68	0.94	3.55	0.83	4.46	0.50
calyx appendage length (mm)	1.04	0.18	1.93	0.38	0.68	0.16	1.71	0.42	1.94	0.38	1.98	0.47
calyx lobe length (mm)	1.17	0.10	1.26	0.16	1.12	0.08	0.91	0.12	0.96	0.15	0.99	0.17
hypanthium-calyx length (mm)	7.35	0.35	7.44	0.58	6.50	0.51	6.84	0.74	6.56	0.54	7.44	0.39
hypanthium pubescence (mm)	0.14	0.04	0.15	0.04	0.12	0.02	0.39	0.15	0.36	0.14	0.53	0.13
hypanthium hair number (0.25 mm^2)	0.98	0.42	1.30	0.56	1.93	0.40	2.37	0.72	2.81	0.90	3.29	0.28
flower axil number	4.34	0.54	3.34	0.33	4.00	0.62	4.84	1.45	6.15	1.25	6.08	1.36

observed that there were differences in petal shape between the taxa; for example the 'Dropmore Purple' petal length: width ratio = 3.18, whereas that for the Lockport population was 2.52.

Figure 6.2 is an ordination scattergram of the three cultivars and the three naturalized *Lythrum* populations based on the results of the canonical discriminant analysis. It shows the group centroids for the six populations together with the standard deviation associated with each group on the axis. The first two dicriminant axes accounted for 91 percent of the total discriminating power and therefore the first two discriminant axes were presented. Heck's theta = 0.909 with $p < 0.05$ showed that the groups were in fact significantly different (Morrison 1976).

Eigenvalues are presented in Table 6.2 together with eigenvector elements (discriminant weights). The two main variables on the the first axis were calyx-hypanthium pubescence (0.692) and calyx lobe length (-0.603). The first axis separates cultivars from wild and the second axis distinquishes between the three cultivars based on calyx-hypanthium pubescence (0.697) and calyx appendage length (0.548). The overlap between the three wild populations is considerable and indicates they are not very variable from each other, whereas the cultivars are distinguished from each other on the two axes.

The first axis separates the cultivars from the wilds

Figure 6.2: Ordination scattergram of the three cultivated (MP, MG and DP) and three naturalized (AR, LP, and SK) *Lythrum* populations indicating their centroids and standard deviations (± 1 SD).

Abbreviations for *Lythrum* populations:
AR = Assiniboine River, Winnipeg
LP = Lockport
SK = Selkirk
DP = 'Dropmore Purple'
MG = 'Morden Gleam'
MP = 'Morden Pink'

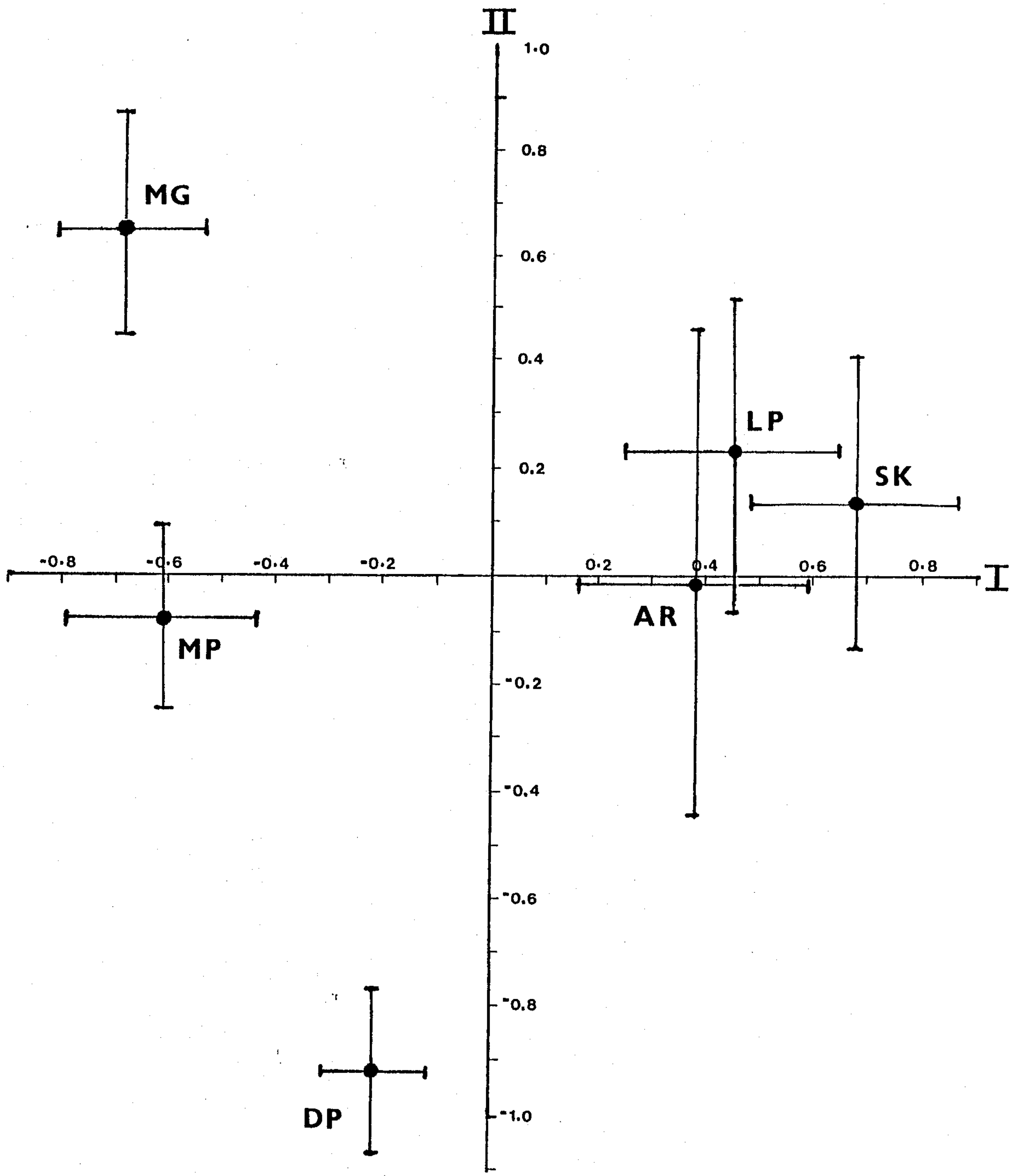

II
1·0
0.8
0.6
0.4
0.2
-0.2
-0.4
-0.6
-0.8
-1.0
-0·8
-0·6
-0·4
-0·2
0·2
0·4
0·6
0·8
I
MG
LP
SK
MP
AR
DP

Table 6.2: Summarization of canonical discriminant analysis results for eight morphological variables measured on three cultivated and three naturalized *Lythrum* populations listing their eigenevalues and eigenvector elements. (Underlined values indicate the variables which were the best discriminators for the two main axes).

		AXIS	
		I	II
	EIGENVALUES	9.962 (69.3%)	3.099 (21.6%)
	VARIABLES		
EIGENVECTORS	petal length (mm)	0.112	-0.142
	petal width	-0.200	0.335
	calyx appendage length	0.272	0.548
	calyx lobe length	-0.603	-0.224
	calyx length	-0.105	-0.040
	calyx pubescence	0.692	0.697
	hair number (0.25 mm^2)	0.128	-0.163
	flower axil number	0.059	0.061

and accounts for 69.3 % of the variance. The main distinguishing features are calyx pubescence in wild and calyx lobe length in cultivated populations. The second axis which is 21.6 %, distinguishes mainly on calyx appendage length and calyx pubescence. Calyx appendage length is greatest in 'Morden Gleam', intermediate 'Morden Pink', and lowest in 'Dropmore Purple'.

The discriminant analysis also performed a classification summary of the membership in each population and reassigned individuals originally in one population into one of the others if its floral characters were more indicative of another group. Table 6.3 lists the number of individuals classified into each population, and their percentages. All fifty-eight individuals measured from the three cultivars remained 100 percent in their appropriate categories, indicating that they are distinctly different from both each other and the naturalized populations. Upon examining the wild populations, however, it was seen that a few individuals were reclassified. These were two from each of the Lockport and Winnipeg populations, and one from the Selkirk population. These five individuals were reclassified into other naturalized populations but not into cultivar groupings. This suggested some similarity between the wild populations but even these on the whole are quite distinct. Individuals were placed into their correct grouping between 90 and 100 percent of the time, showing

Table 6.3: Number and percentage of individuals classified into each of the six *Lythrum* populations.

Abbreviations for the *Lythrum* populations:
AR = Assiniboine River, Winnipeg
LP = Lockport
SK = Selkirk
DP = 'Dropmore Purple'
MG = 'Morden Gleam'
MP = 'Morden Pink'

Number of observations and percents
classified into each of the six taxa.

Population	AR	LP	SK	DP	MG	MP
AR (20)	18 90.00	1 5.00	1 5.00	--	--	--
LP (20)	1 5.00	18 90.00	1 5.00	--	--	--
SK (20)	--	1 5.00	19 95.00	--	--	--
DP (20)	--	--	--	20 100.00	--	--
MG (20)	--	--	--	--	20 100.00	--
MP (18)	--	--	--	--	--	18 100.00

that these plants, based on their measurements, are florally distinct from one another.

FLORAL MORPHOLOGY OF CULTIVATED AND NATURALIZED PROGENY

The floral measurement data collected from the thirty-seven crosses were analyzed using discriminant analysis (SAS Program). This was done in order to classify each individual into one of the six populations that were examined in the initial morphological study.

The results of this classification are shown in Table 6.4, which lists the thirty-seven crosses and shows the placement of the individuals from each cross into one of the six taxa (p=0.05). Table 6.5 summarizes the thirty-seven crosses which were of cultivated, wild, or cultivated and wild parentage; the number of individuals that were measured from each of these three categories; and the percentages and numbers of individuals which were classified with one of their parents. Based on these two tables, it can be seen that of the 295 individuals measured, only 49.8 per cent were classified with one or other of their parents (which were of 'Morden Pink', 'Morden Gleam', 'Dropmore Purple', or Lockport origin, depending on the cross). Progeny were reassigned to Winnipeg's Assiniboine River population, unexpectedly, 36.6 percent (108/295) of the time. This Winnipeg population was of no direct parental link to the individuals measured.

Table 6.4: Classification of the progeny of thirty-seven crosses into the six *Lythrum* taxa using discriminant analysis. (Progeny were obtained from crosses between 'Morden Pink', 'Dropmore Purple', 'Morden Gleam', and Lockport populations).

Abbreviations of the six *Lythrum* populations:

AR = Assiniboine River, Winnipeg
LP = Lockport
  LL = Lockport long style/stamens
  LM = Lockport mid style/stamens
  LS = Lockport short style/stamens
SK = Selkirk
DP = 'Dropmore Purple'
MG = 'Morden Gleam'
MP = 'Morden Pink'

Number of observations and percents classified
into each of the six taxa.

Taxon	AR	LP	SK	DP	MG	MP	total
DP self	0	1	0	0	0	0	1
	0.00	100.00	0.00	0.00	0.00	0.00	100.00
DP x LL	3	5	1	0	0	1	10
	30.00	50.00	10.00	0.00	0.00	10.00	100.00
DP x LM	7	3	0	0	0	0	10
	70.00	30.00	0.00	0.00	0.00	0.00	100.00
DP x LS	3	1	0	0	0	0	4
	75.00	25.00	0.00	0.00	0.00	0.00	100.00
DP x MG	3	5	0	0	2	0	10
	30.00	50.00	0.00	0.00	20.00	0.00	100.00
DP x MP	1	3	0	2	0	3	9
	11.11	33.33	0.00	22.22	0.00	33.33	100.00
LL self	2	6	1	0	0	0	9
	22.22	66.67	11.11	0.00	0.00	0.00	100.00
LL x DP	2	0	0	0	0	0	2
	100.00	0.00	0.00	0.00	0.00	0.00	100.00
LL x LL	6	4	0	0	0	0	10
	60.00	40.00	0.00	0.00	0.00	0.00	100.00
LL x LM	3	6	1	0	0	0	10
	30.00	60.00	10.00	0.00	0.00	0.00	100.00
LL x LS	4	5	1	0	0	0	10
	40.00	50.00	10.00	0.00	0.00	0.00	100.00
LL x MG	8	2	0	0	0	0	10
	80.00	20.00	0.00	0.00	0.00	0.00	100.00
LL x MP	2	4	0	0	3	0	9
	22.22	44.44	0.00	0.00	33.33	0.00	100.00
LM self	2	5	1	0	0	0	8
	25.00	62.50	12.50	0.00	0.00	0.00	100.00
LM x DP	1	8	0	0	1	0	10
	10.00	80.00	0.00	0.00	10.00	0.00	100.00
LM x LL	4	6	0	0	0	0	10
	40.00	60.00	0.00	0.00	0.00	0.00	100.00
LM x LM	1	3	0	0	0	0	4
	25.00	75.00	0.00	0.00	0.00	0.00	100.00
LM x LS	4	5	1	0	0	0	10
	40.00	50.00	10.00	0.00	0.00	0.00	100.00
LM x MG	5	5	0	0	0	0	10
	50.00	50.00	0.00	0.00	0.00	0.00	100.00
LM x MP	2	2	0	0	1	0	5
	40.00	40.00	0.00	0.00	20.00	0.00	100.00
LS x DP	6	3	0	0	0	1	10
	60.00	30.00	0.00	0.00	0.00	10.00	100.00
LS x LL	0	1	0	0	0	0	1
	0.00	100.00	0.00	0.00	0.00	0.00	100.00
LS x LM	2	3	2	0	0	0	7
	28.57	42.86	28.57	0.00	0.00	0.00	100.00
LS x LS	4	6	0	0	0	0	10
	40.00	60.00	0.00	0.00	0.00	0.00	100.00
LS x MG	0	6	0	0	4	0	10
	0.00	60.00	0.00	0.00	40.00	0.00	100.00
LS x MP	3	2	1	0	0	0	6
	50.00	33.33	16.67	0.00	0.00	0.00	100.00
MG self	0	2	0	0	4	0	6
	0.00	33.33	0.00	0.00	66.67	0.00	100.00
MG x DP	1	2	1	1	4	0	9
	11.11	22.22	11.11	11.11	44.44	0.00	100.00
MG x LL	6	4	0	0	0	0	10
	60.00	40.00	0.00	0.00	0.00	0.00	100.00
MG x LM	7	0	1	0	0	0	8
	87.50	0.00	12.50	0.00	0.00	0.00	100.00
MG x LS	3	1	1	0	0	0	5
	60.00	20.00	20.00	0.00	0.00	0.00	100.00
MP x DP	1	1	0	0	0	5	7
	14.29	14.29	0.00	0.00	0.00	71.43	100.00
MP x LL	3	6	0	0	0	1	10
	30.00	60.00	0.00	0.00	0.00	10.00	100.00
MP x LM	3	4	0	0	0	0	7
	42.86	57.14	0.00	0.00	0.00	0.00	100.00
MP x LS	4	3	0	0	0	1	8
	50.00	37.50	0.00	0.00	0.00	12.50	100.00
MP x MG	1	2	0	0	7	0	10
	10.00	20.00	0.00	0.00	70.00	0.00	100.00
MP x MP	1	0	0	1	4	4	10
	10.00	0.00	0.00	10.00	40.00	40.00	100.00
TOTAL	108	125	12	4	30	16	295
PERCENT	36.61	42.37	4.07	1.36	10.17	5.42	100.00

Table 6.5: Number of the thirty-seven *Lythrum* hybrids of cultivated, naturalized, or cultivated and naturalized parentage, and percentage classified with one or other of their parents.

	No. of crosses	Total No.Ind.	% & no. classified with a parent
cultivated	8	62	51.6 (32)
wild	11	89	56.2 (50)
wild & cultivated	18	144	45.1 (65)
	37	295	49.8 (147)

DISCUSSION

Purple loosestrife may vary from only three feet in height up to six or seven feet, and may occur in dense stands depending on its growing conditions. Regardless of stature, its distinctive showy pink-purple thyrses easily distinguish it from almost every other wildflower found growing in North America. Confusion may arise between *Lythrum salicaria* and *Epilobium angustifolium* (fireweed), *Liatris ligulistylis* (meadow blazingstar), and *Agastache foeniculum* (giant hyssop), but only from a distance. Also, the moist or wet habitat requirements of *L. salicaria* are almost never shared by these other species. The plant is also conspicuous throughout the fall and winter, as strong stems support numerous brown seed capsules, creating an easily recognizable profile that is useful in the field for identification purposes (Thompson 1989).

Two other *Lythrum* species, *L. alatum* (winged loosestrife) and *L. virgatum* (wand loosestrife) occur in North America and are of importance to this study. *Lythrum alatum* is a native species of North America that is smaller in stature and less conspicuous, with purple flowers borne one per inflorescence axil. *Lythrum virgatum* is found only in Massachusetts where it was introduced or escaped (Gleason 1974).

Thus, it appears that in North America the three species are easily separable on the basis of morphology or distribution and that *L. salicaria* is the species viewed as the threat to wetlands. However, cultivars of the three species, either hybrids or showy versions of a species, have been developed in the horticulture industry. It has been suspected that these cultivars are contributing to the spread of purple loosestrife via seed and/or pollen. Possibly certain varieties may be a problem whereas others are not, and it was thought important to be able to morphologically distinguish cultivars from each other and from wild *Lythrum*.

A morphological investigation was done to determine if certain floral characteristics could be used to distinguish wild and cultivated *Lythrum* populations. In the first part of the study, three popular cultivars, 'Morden Pink', 'Morden Gleam', and 'Dropmore Purple' were examined, as well as three naturalized populations from Winnipeg, Lockport and Selkirk. Canonical discriminant analysis on these data found that the main discriminating features were calyx pubescence in the naturalized and calyx lobe length in cultivated populations. (Naturalized populations are more pubescent than the cultivars, whereas the cultivars have greater lobe lengths than the naturalized populations). There is a great deal of similarity between the wild populations as there is overlap indicating they are not very variable from each other. The cultivars are distinct from each other based on their calyx appendage length and calyx-hypanthium pubescence. 'Morden Gleam' has the greatest calyx appendage length and pubescence and 'Dropmore Purple' the lowest. 'Morden Pink' is intermediate between the two.

Based on their morphological differences, all naturalized and cultivated plants were very distinct from each other, with individuals being separated confidently into their correct taxon no less than ninety percent of the time ($p=0.05$).

The cultivars are florally very distinct from each other. The nursery trade should have no problems ensuring that there are no mix-ups due to misidentification with these three very popular cultivars. Table 6.6 is an identification key to the 'Morden Pink', 'Morden Gleam', and 'Dropmore Purple' cultivars. The key was based on the following characters: flower styly, petal colour, petal length:petal width ratios, calyx lobes and calyx appendages. The characters selected for this key were derived from the discriminant analysis but included other recognizable characters such as petal colour. The *Lythrum* populations examined were tristylous with whorls of reproductive structures borne at three levels: short, 3-4 mm; mid 7-8 mm, and; long, 9-11 mm. It has been recorded that petal colour may be associated with style length in wild populations (Louis-Marie 1944). Longs were pale, shorts a dark purple colour (though some may be pale), and mids were

Table 6.6: Floral key to three cultivars of *Lythrum*-- 'Dropmore Purple', 'Morden Gleam', and 'Morden Pink'.

FLORAL KEY FOR THREE *Lythrum* CULTIVARS

1. Styles long; petals 3 times as long as wide or more (petals long and narrow); flowers purple . . . . DROPMORE PURPLE

1. Styles mid; petals less than 3 times as long as wide (petals broad); flowers pink to deep pink 2

    2. Calyx appendage length exceeds calyx lobe length . . . . MORDEN GLEAM

    2. Calyx appendage length less than or equal to calyx lobe length . . . . MORDEN PINK

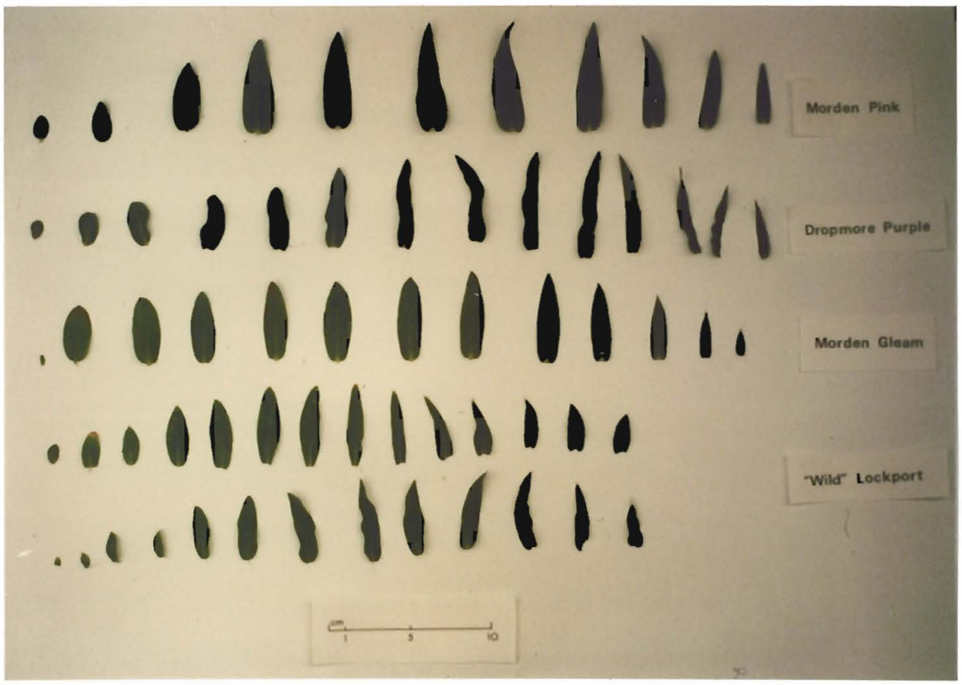
General comments on vegetative characters:
DP - leaves long, tapered; often purplish-red.
MG and MP - leaves broader; ovate-elliptical.
Naturalized - leaves variety of shapes; cordate/truncate sessile base.
MR - leaves elliptical; dark green, glossy.

General comments on floral characters:
<u>Naturalized</u> - flower colour pink to purple.
-calyx appendage length exceeds calyx lobe length.
-hypanthium normally covered in dense, long hairs.
MR - mid-styled, deep pink/rose.

intermediate. These findings were not examined in this study. Fisher and Martin (1947) also determined that petal colour and style length were linked, and that short (S) and purple (P) were the dominant alleles, with recessive alleles of the genes yielding mid- or long-styled plants and pink flowers. Some general observations regarding vegetative and floral characters of both wild and cultivated plants are given as a footnote to this key. Figure 6.3 illustrates the morphological variability of the leaves of populations both within and between individuals. In these studies no quantitative work was done using vegetative characters due to such variability. These differences would be compounded depending on the growing conditions of plants from various greenhouses or wild habitats, as decreased light levels will result in larger leaf sizes and a reduction in the amount of tomentosum and leaf thickness (Shamsi and Whitehead 1974b).

It was anticipated that hybrid progeny of wild origin would be most similar to their parents; i.e. from Gunns Creek, Lockport, Manitoba. Likewise, it was expected hybrid plants of cultivated lineage would most resemble their cultivated parents, and offspring of cultivated/naturalized parentage would be classified with either their cultivated or naturalized parent in the same manner. However, it was found that only fifty percent of the individuals were classified as expected. Surprisingly, thirty-seven percent of the 295 offspring were classified as morphologically

Figure 6.3: Morphological variability of leaves within and between four *Lythrum* taxa: 'Morden Pink', 'Dropmore Purple', 'Morden Gleam', and Lockport.
(Leaves progress from lowest to highest regions of the stem, moving from left to right).


Morden Pink
Dropmore Purple
Morden Gleam
"Wild" Lockport
1
5
10

resembling Winnipeg's Assiniboine River population. This unexpected result suggests that this naturalized population has received some cultivated input. These plants are growing next to Assiniboine Park, where *Lythrum* taxa were once planted as garden ornamentals. Another observation which supports this is the production of red seeds by some of the plants. All other naturalized populations examined produced straw-coloured seeds. The origin of this is unknown, but such dimorphism in the seeds suggests a diverse genetic influence. The only other time red seeds were observed was in seed samples from *L. virgatum.* Input from cultivated plants may have occurred in both the Lockport and Selkirk populations but is not readily apparent.

Generally, it may be stated that many naturalized populations throughout North America have been influenced by cultivated escapes. Dissimilarities between naturalized populations suggests that either they are not pure stands of *L. salicaria* or that the species exhibits a very high degree of plasticity. It would be extremely difficult to detect in the field whether plants found growing in the wild were of cultivated origin or not.

CONCLUSIONS

1. Morphological comparison of floral characters between three cultivars ('Morden Pink', 'Morden Gleam', and 'Dropmore Purple') and three naturalized (Lockport, Winnipeg, and Selkirk) populations showed that calyx-hypanthium pubescence in the wild populations (which were more pubescent) and calyx lobe length in cultivated populations (which were greater in length) were the best distinquishing characters when using canonical discriminant analysis. The cultivars were best distinguished based on their calyx-hypanthium pubescence and calyx appendage length, which was the highest in 'Morden Gleam' and lowest in 'Dropmore Purple'.

2. Thirty-seven percent of the progeny of 'Morden Pink', 'Dropmore Purple', 'Morden Gleam' and Lockport crosses were classified with an Assiniboine River population in Winnipeg. This indicated that cultivars have likely contributed to the naturalization of purple loosestrife, at least in some cases.

3. It is difficult to determine in the field whether there has been cultivated influence. Tomentose plants usually are indicative of *L. salicaria*. In a greenhouse situation this may be useful in preventing the sale of naturalized individuals. However, in the field this character may not

be as useful as different growing conditions occur. Also, interbreeding may have been occurring with cultivars, and glabrous or near-glabrous individuals may no longer be present.

4. The three naturalized populations and three cultivars were morphologically distinct from each other. A floral key was constructed for the three cultivars used in this study. The key utilized the following characters: flower styly, petal colour, petal length:width ratio, and calyx lobe and calyx appendage lengths.

CHAPTER 7

GENERAL CONCLUSIONS

1. *Lythrum salicaria* is distributed widely in Manitoba. There are approximately 38 populations in the province with the plants's range extending as far north as Snow Lake, at almost 55°N latitude.
   It is important to monitor the spread of purple loosestrife in order to implement any method of control.

2. Seed production studies showed that cultivars have a reduced reproductive capacity when compared to naturalized populations. Lower and middle regions of the infructescence produce approximately twice the seed of the upper region. The upper region often has capsules containing immature seed.

3. Seeds of purple loosestrife are dispersed gradually over the fall and winter. Thus, seeds may be dispersed over ice and snow via wind.

4. Germinability of 28 seed sources (17 cultivated and 11 naturalized) ranged from 74.4 - 100.0 percent. Mean

germinability was 91.6 percent. Two samples, *L. virgatum* cv. Rose Queen and *L. virgatum* LEI 83-131 had significantly lower seed germinability at 74.4 and 75.0 percent, respectively, than the other samples. *Lythrum virgatum* cv. Morden Pink had a very low seed output, but seeds that were produced were 100.0 percent viable.

5. Morphological comparison of floral characters between three cultivars ('Morden Pink', 'Morden Gleam', and 'Dropmore Purple') and three naturalized (Lockport, Winnipeg, and Selkirk) populations showed that calyx lobe length, and calyx-hypanthium pubescence were the best discriminators between cultivars and naturalized populations. Calyx appendage length and calyx-hypanthium pubescence were the best variables distinquishing the cultivars from each other using canonical discriminant analysis.

6. The three naturalized populations and three cultivars were morphologically distinct from each other. A floral key was constructed for the three cultivars used in this study. The key utilized the following characters: flower styly, petal colour, petal length:petal width ratio, and calyx lobe and calyx appendage lengths.

7. Thirty-seven percent of the progeny of 'Morden Pink', 'Dropmore Purple', 'Morden Gleam' and Lockport crosses were classified with an Assiniboine River population in Winnipeg.
This indicated that cultivars have likely contributed to the naturalization of purple loosestrife, at least in some cases.
It is difficult to determine in the field whether there has been cultivated influence. Tomentose plants usually are indicative of *L. salicaria*. In a greenhouse situation this may be useful in preventing the sale of naturalized individuals. However, in the field this character may not be as useful if interbreeding has been occurring with cultivars, as glabrous or near-glabrous individuals may no longer be present.

8. The self-incompatibility system is not strict. Most incompatible crosses yielded greatly reduced seed outputs in comparison to compatible crosses, but some were highly fecund.
Mids showed a high degree of self-fertility with themselves and other mids.

9. Crossability tests between 'Morden Pink', 'Dropmore Purple', 'Morden Gleam' and Lockport individuals has

shown that cultivars produce viable seed and pollen. All three cultivars contributed seed and/or pollen at the partially fertile level (DP x MG, LM x MG, LL x MP, DP x LL, and LS x DP), and 'Morden Pink' and 'Morden Gleam' at the significantly fertile level (MP x LM, MG x LM, LL x MG, and LS x MG).
Compatible Lockport crosses (LL x LL, LM x LM, and LS x LS) also yielded significantly fertile results. Unexpected fertility was noted in the LM x MG and LM x LL crosses. Sterility was noted in the LS x MP, LM x DP, MG x DP, MP x DP and DP x MP crosses, indicating evidence of some sterility in 'Dropmore Purple' and perhaps 'Morden Pink'.

10. Cross-pollination work with hybrids showed them to be highly fertile.

11. Germinability of seeds from the parental crosses averaged 98 percent. Germinability of the hybrid seeds that were examined averaged 97 percent.

12. Cultivars are capable of contributing viable seed and pollen to the spread of purple loosestrife, and morphological analyses indicate that this has been occurring. The sale of cultivars, regardless of parentage, should be discouraged.

REFERENCES

Anderson, R.N. 1968. Germination and establishment of weeds for experimental purposes. W.F. Humphrey Press, Inc., Geneva, New York. pp. 236.

Balogh, G.R. 1986. Distribution and seedling ecology of purple loosestrife in Ohio's Lake Erie marshes. Ohio Journal of Science 86(2): 51.

Balogh, G.R. and T.A. Bookhout. 1989. Remote detection and measurement of purple loosestrife stands. Wildlife Society Bulletin 17: 66-67.

Barlow, N. 1913. Preliminary note on heterostylism in *Oxalis* and *Lythrum*. Journal of Genetics 6: 53-65.

Barrett. S.C.H. 1977. The breeding system of *Pontederia rotundifolia* L., a tristylous species. New Phytologist 78: 209-220.

Barton, L.V. 1965. Dormancy in seeds imposed by the seed coat. Handbuchder Pflanzenphysiol 15: 727-745.

Batra, S.W.T., D. Schroeder, P.E. Boldt, and W. Mendl. 1986. Insects associated with purple loosestrife (*Lythrum salicaria* L.) in Europe. Proceedings of the Entomological Society of Washington 88(4): 748-759.

Beckett, K.A. 1981. Hardy perennials. Redwood Burn, Trowbridge, Great Britain. pp. 182.

Brewbaker, J.L. 1957. Pollen cytology and self-incompatibility systems in plants. The Journal of Heredity 48: 271-277.

Buckley, A.R. 1977. Canadian garden perennials. Hancock House Publishers Limited, Saanichton, B.C. pp. 212.

Bunch, Q. 1977a. Purple loosestrife--a honey plant. American Bee Journal 117: 398.

-----1977b. More about purple loosestrife. American Bee Journal 117: 553.

Charlesworth, D. 1979. The evolution and breakdown of tristyly. Evolution 33(1): 486-498.

Clay, R.T. 1986. Purple loosestrife: A literature review and management recommendations. Ducks Unlimited Canada. pp. 29.

Cody, W.J. 1978. The status of *Lythrum alatum* (Lythraceae) in Canada. The Canadian Field-Naturalist 92: 74-75.

Crowe, L.K. 1964. The evolution of outbreeding in plants. Heredity 19:435-457.

Darwin, C. 1865. On the sexual relations of the three forms of *Lythrum salicaria*. Journal of the Linnean Society of London. Botany 8: 169-196.

-----1899. The different forms of flowers on plants of the same species. D. Appleton and Company, New York. pp. 352.

Davidson, C.G. 1986. Experimental taxonomy, propagation and models of inheritance for flower colour and extra petals in *Potentilla fruticosa* L. Ph.D. Thesis, The University of Manitoba. pp. 321.

Dulberger, R. 1968. Chromosome numbers in *Lythrum* in Israel. Israel Journal of Botany 17: 179-183.

-----1970. Tristyly in *Lythrum junceum*. New Phytologist 69: 751-759.

-----1975. S-gene action and the significance of characters in the heterostylous syndrome. Heredity 35(3): 407-415.

East, E.M. 1927. The inheritance of heterostyly in *Lythrum salicaria*. Genetics 12: 393-414.

Emerson, J.D., and J. Strenio. 1983. Boxplots and batch comparison. In Understanding Robust and Exploratory Data Analysis. Eds. D.C. Hoaglin, F. Mastelle, and J.W. Tukey. John Wiley & Sons, Toronto. pp. 447.

Fehr, W.R. 1987. Principles of cultivar development. Volume 1--Theory and Technique. Macmillan Publishing Co., New York. pp. 536.

Fenner, M. 1985. Seed Ecology. Chapman & Hall, New York. pp.151.

Fisher, R.A. and V.C. Martin. 1947. Spontaneous occurrence in *Lythrum salicaria* of plants duplex for the short-style gene. Nature 160: 541.

Fisher, R.A. and K. Mather. 1943. The inheritance of style length in *Lythrum salicaria*. Annals of Eugenics 12: 1-23.

Frankel, R. and E. Galun. 1977. Pollination mechanisms, reproduction and plant breeding. In: Monographs on Theoretical and Applied Genetics 2. Springer-Verlag, Berlin. pp. 281.

Ganders, F.R. 1979. The biology of heterostyly. New Zealand Journal of Botany 17: 607-635.

Gaudet, C.L. and P.A. Keddy. 1988. A comparative approach to predicting competitive ability from plant traits. Nature 334: 242-243.

Gilbert, N. and S.B. Lee. 1980. Two perils of plant population dynamics. Oecologia 46: 283-284.

Giles, F., R. Keith, and D. Staupe. 1980. Herbaceous perennials. Reston Publishing Company, Reston, Virginia. pp. 356.

Gleason, H.A. 1974. The new Britton and Brown illustrated flora of the northeastern United States and adjacent Canada. Volume 2. Hafner Press, New York. pp. 655.

Gledhill, D. 1985. The names of plants. Cambridge University Press, Cambridge. pp. 159.

Graham, A., J.W. Nowicke, J.J. Skvarla, S.A. Graham, V. Patel, and S. Lee. 1987. Palynology and systematics of the Lythraceae. II. Genera *Haitia* through *Peplis*. American Journal of Botany 74(6): 829-850.

Grigson, G. 1974. A dictionary of English plant names (and some products of plants). Allen Lane, London. pp. 239.

Groh, H. 1944. Canadian Weed Survey--1943. Second Annual Report. Department of Agriculture, Canada. pp. 74.

Halkka, O., and L. Halkka. 1974. Polymorphic balance in small island populations of *Lythrum salicaria*. Annales Botanici Fennici 11: 267-270.

Hanna, J. 1989. Purple invader. Seasons - Summer: 20-22,36.

Harp, H.F. and L.M. Collicutt. 1983. Lythrums for home gardens. Agriculture Canada. Publication 1285E. Morden Research Station, Morden, Manitoba.

Hayes, B. 1979. Purple loosestrife--the wetlands honey plant. American Bee Journal 119: 382-383.

Hebb, R.S. 1975. Low maintenance perennials. Arnold Arboretum of Harvard University, Jamaica Plains, Massachusetts. pp. 220.

Henderson, R. 1987. Status and control of purple loosestrife in Wisconsin. WDNR-Research Management Findings No.4.

Heuch, I. 1979. Equilibrium populations of heterostylous plants. Theoretical Population Biology 15: 43-57.

-----1980. Loss of incompatibility types in finite populations of the heterostylous plant *Lythrum salicaria*. Hereditas 92: 53-57.

Hudak, J. 1976. Gardening with perennials. Timber Press, Beaverton, Oregon. pp. 398.

Hughes, R. 1977. Purple loosestrife judged a weed. American Bee Journal 117: 398.

Keddy, P.A., I.C. Wisheu, B. Shipley, and C. Gaudet. 1989. Seed banks and vegetation toward predictive community ecology. In Ecology of Soil Seed Banks. Eds. Leck, M.A. *et al.* Academic Press, Inc., New York. pp. 347-363.

Klimas, J.E. and J.A. Cunningham. 1974. Wildflowers of eastern America. Alfred A. Knopf, New York. pp. 273.

Kostoff, D. 1927. Pollen-tube growth in *Lythrum salicaria*. Genetics 13: 253-255.

Lamela, M., I. Cadavid, and J.M. Calleja. 1986. Effects of *Lythrum salicaria* extracts on hyperglycemic rats and mice. Journal of Ethno-pharmacology 15(2): 153-160.

Leach, C.R. 1983. Fluctuations in heteromorphic self-incompatibility systems. Theoretical and Applied Genetics 66: 307-312.

Levin, D.A. 1970. Assortative pollination in *Lythrum*. American Journal of Botany 57(1): 1-5.

Louis-Marie, P. 1944. La Salicaire dans le Quebec. Institut Agricole d'Oka. Province de Quebec. pp. 46.

McKeon, W.H. 1959. A preliminary report on the use of chemical herbicides to control purple loosestrife (*Lythrum salicaria*) on a small marsh. Proceedings of the Northeast Weed Control Conference 13: 329-331.

Mead, R.M. 1957. The hunt for new honey plants. American Bee Journal 97: 300-301.

Mitchell, A.S. 1976. A summary of the family Lythraceae in the Northern Territory with additional comments on Australian material. Journal of the Adelaide Botanic Gardens 1(1): 55-59.

Montgomery, F.H. 1977. Seeds and fruits of plants of eastern Canada and northeastern United States. University of Toronto Press, Toronto. pp. 232.

Morrison, D.F. 1976. Multivariate Statistical Methods. 2nd Edition. McGraw-Hill, New York. pp. 415.

Mulcahy, D.L. and D. Caporello. 1970. Pollen flow within a tristylous species: *Lythrum salicaria*. American Journal of Botany 57(9): 1027-1030.

Nicholls, M.S. 1987. Pollen flow, self-pollination and gender specialization: factors affecting seed-set in the tristylous species *Lythrum salicaria* (Lythraceae). Plant Systematics and Evolution 156(3-4): 151-157.

Ornduff, R. 1975. Pollen flow in *Lythrum junceum*, a tristylous species. New Phytologist 75: 161-166.

-----1978. Features of pollen flow in dimorphic species of *Lythrum* section Euhyssopifolia. American Journal of Botany 65(10): 1077-1082.

Pellett, M. 1944. Purple loosestrife. American Bee Journal 84: 57.

-----1977. Purple loosestrife spreads down river. American Bee Journal 117: 214-215.

Plowden, C.C. 1970. A manual of plant names. Philosophical Library, New York. pp. 260.

Radosevich, S.R., and J.S. Holt. 1984. Weed Ecology. John Wiley & Sons, New York. pp. 265.

Rawinski, T.J. 1982. The ecology and management of purple loosestrife (*Lythrum salicaria* L.) in central New York. M.Sc. thesis, Cornell University. pp. 88.

Rawinski, T.J. and R.A. Malecki. 1984. Ecological relationships among purple loosestrife, cattail and wildlife at the Montezuma National Wildlife Refuge. New York Fish and Game Journal 31(1): 81-87.

Reader's Digest. 1986. Magic and medicine of plants. The Reader's Digest Assoc., Inc., Pleasantville, NY. pp. 464.

Real, L. 1983. Pollination biology. Academic Press, Inc., Orlando, Florida. pp. 338.

Schoch-Bodmer, H. 1938. The proportion of long-, mid-, and short-styled plants in natural populations of *Lythrum salicaria* L. Journal of Genetics 36: 39-43.

-----1940. The influence of nutrition upon pollen grain size in *Lythrum salicaria*. Journal of Genetics 40: 393-402.

Scoggan, H.J. 1957. Flora of Manitoba. National Museum of Canada, Ottawa. Bulletin No. 140. pp. 619.

-----1979. Flora of Canada. National Museum of Natural Sciences, Ottawa. Publ. bot. No. 7. pp. 1711.

Shamsi, S.R.A. and F.H. Whitehead. 1974a. Comparative eco-physiology of *Epilobium hirsutum* L. and *Lythrum salicaria* L. I. General biology, distribution and germination. Journal of Ecology 62: 279-290.

-----1974b. Comparative eco-physiology of *Epilobium hirsutum* L. and *Lythrum salicaria* L. II. Growth and development in relation to light. Journal of Ecology 62: 631-645.

-----1977b. Comparative eco-physiology of *Epilobium hirsutum* L. and *Lythrum salicaria* L. IV. Effects of temperature and inter-specific competition and concluding discussion. Journal of Ecology 65: 71-84.

Shinners, L.H. 1953. Synopsis of the United States species of *Lythrum* (Lythraceae). Field and Laboratory 21: 80-89.

Smith, L.S. 1959. Some experiences with control of purple loosestrife at the Montezuma National Wildlife Refuge. Proceedings of the Northeast Weed Control Conference 13: 333-336.

Smith, R.H. 1964. Experimental control of purple loosestrife (*Lythrum salicaria*). New York Fish and Game Journal 11(1): 35-46.

Snyder, L.C. 1983. Flowers for northern gardens. University of Minnesota Press, Minneapolis. pp. 385.

Solbrig, O.T. 1970. Principles and methods of plant biosystematics. The MacMillan Company, London. pp. 226.

Still, S.M. 1982. Herbaceous ornamental plants. 2nd Edition. Stipes Publishing Company, Illinois. pp. 303.

Stirling, J. 1933. Studies of flowering in heterostyled and allied species. Part II. The Lythraceae: *Lythrum salicaria* Linn. Publications of the Hartley Botanical Laboratories, The University of Liverpool 10: 1-24.

Stout, A.B. 1923. Studies of *Lythrum salicaria* I. The efficiency of self-pollination. American Journal of Botany 10: 440-449.

-----1925. Studies of *Lythrum salicaria*--II. A new form of flower in this species. Bulletin of the Torrey Botanical Club 52(3): 81-85.

Stuckey, R.L. 1980. Distributional history of *Lythrum salicaria* (purple loosestrife) in North America. Bartonia 47: 3-20.

Teale, E.W. 1982. Stems beyond counting, flowers unnumbered. Audubon 84(4): 38-43.

Thomas, G.S. 1982. Perennial garden plants or The modern florilegium. 2nd Edition. J.M. Dent & Sons Limited, London. pp. 338.

Thompson, D.Q. 1989. Control of purple loosestrife. Fish and Wildlife Leaflet 13.4.11, Fish and Wildlife Service, Washington, D.C. pp. 6.

Thompson, D.Q., R.L. Stuckey, and E.B. Thompson. 1987. Spread, impact, and control of purple loosestrife (*Lythrum salicaria*) in North American wetlands. United States Department of the Interior, Fish and Wildlife Service, Washington, DC. pp. 55.

Tobe, H., P.H. Raven, and S.A. Graham. 1986. Chromosome counts for some Lythraceae sens. str. (Myrtales), and the base number of the family. Taxon 35: 13-20.

Torres, I., and J.C. Suarez. 1980. A preliminary study of hypoglycemic activity of *Lythrum salicaria*. Lloydia 43: 559-564.

U.S. Fish and Wildlife Service. 1979. Another view--purple loosestrife alert. American Bee Journal 119: 383.

Usher. G. 1974. A dictionary of plants used by man. Constable and Company Limited, London. pp. 619.

Van't Haaff, G. 1968. My experience with purple loosestrife. American Bee Journal 108: 244.

Vuilleumier, B.S. 1967. The origin and evolutionary development of heterostyly in the angiosperms. Evolution 21: 210-226.

Yeo, P.F. 1975. Some aspects of heterostyly. New Phytologist 75: 147-153.