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EVALUATION OF REVEGETATION TECHNIQUES USED ON DEGRADED AGRICULTURAL LAND IN THE CENTRAL AVON CATCHMENT, WESTERN AUSTRALIA

Scott James Bartholomew B.Sc. (Env. Mgt.)

Thesis submitted in partial fulfilment of the requirements for the award of Master of Science (Environmental Management)



Centre for Ecosystem Management Edith Cowan University

July 1998

"When land does well for its owner, and the owner does well by his land; when both end up better by reason of their partnership, we have conservation. When one or the other grows poorer, we do not".

Aldo Leopold (1949)



ABSTRACT

The agricultural region in the central Avon catchment is declining in both arable land and native vegetation. This decline has been due to the clearance of large tracts of native vegetation and its subsequent replacement by exotic crops and pasture species. The loss of vegetation has led to regional land degradation in the form of soil erosion and land salinisation. Therefore, changes in land management practices are required to make agricultural production more compatible with land conservation. Revegetation using native plants is the only management solution that integrates both land and ecological conservation with agricultural productivity. The continuation of current agricultural land use will therefore depend on the development of effective methods of revegetation.

The land manager predominantly has a choice between directly sowing seeds and or planting nursery seedlings as methods of revegetation on degraded land. The widespread problem of land degradation in the Avon catchment dictates that the revegetation method chosen must be effective in terms of both seedling establishment and cost on a broad scale. To determine which method is most effective in revegetation, a series of laboratory and field trials were conducted.

A trial was conducted in seed germination incubators to determine the relative importance of genetic and environmental influences on seed viability. The genetics of the seed stock had a significant influence on germination between different biolocalities. Similarly, environmental factors such as different temperature regimes also had a significant influence on seed germination. Consequently, a relationship was established between optimum germination temperature and the average mean winter temperature of the biolocality where the seed was collected. This suggests that temperature may influence seed genetics and subsequent germination, which varied according to the biolocality.

The identification of environmental parameters that influenced seed germination provided a basis for comparison between direct seeding versus planted nursery seedlings in the field. The number of seedlings established in the second and third year at York was greater for direct seeding than from planted nursery seedlings with the exception of two species belonging to the Leguminosae family. However, the failure of seedlings from either method to become established in the field at Tammin emphasised the need to employ a post-sowing and planting management programme. The costs of seedling establishment using the direct seeding method was approximately one sixth the cost of nursery seedlings with the exception of *Kennedia prostrata*.

Abstract

The cost and time invested in revegetation programmes necessitates that the optimum seed biolocality should be selected in order to produce the most effective plant establishment from direct seeding in the field. Biolocality selection based on geographical distance alone did not produce conclusive criteria for effective revegetation. However, the matching of biogeographical regions and climatic conditions with the site of revegetation produced the most effective plant establishment. This predominantly resulted in the selection of a local species but in some situations biolocalities from further afield with a similar climate, also proved effective.

The invasion of weeds at both the York and Tammin trial sites emphasise the potential effects on the establishment of native vegetation. The growth (height & stem width) of nursery seedlings was significantly reduced while the growth height decreased and the mortality of direct seeded seedlings increased when subjected to weed competition.

Reversing the trend in land and ecological degradation within the central Avon catchment can only be achieved through a revegetation programme that integrates nature conservation with agricultural productivity. Direct seeding is a cost effective method of plant establishment provided that adequate attention is given to pre-seed treatment, seed quality testing, selection of the optimum seed biolocality, soil contour and profile preparation and post sowing control of both weed and predators. However, revegetation of degraded land using nursery seedlings can be just as effective when the cost of seed is expensive and difficult to germinate. If the central Avon wheatbelt is to continue to function as the 'bread bowl' of Western Australia then the establishment of native vegetation can only be achieved through the use of direct seeding methods on a broad scale. Current land management practises and the conservation of the remaining remnant vegetation will not maintain a sustainable agricultural based economy or acequately represent existing biodiversity in the central Avon

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I certify that this thesis does not incorporate, without acknowledgment, any material previously submitted for a degree or diploma in any institution of higher education and that, to the best of my knowledge and belief, it does not contain any material previously published or written by another person except where due reference is made in the text

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Acknowledgments

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I would like to sincerely thank all those people responsible for this weighty document and there were many! In particular, Brett O'Brien gave up 'unspare' time late into the night to structure and critique the document. It was Brett's enthusiasm that helped formulate the structural map of the thesis.

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Special thanks go to Peta-Lyn Blyth who endured festering flies in nasal passages and sweltering under the oppressive summer heat whilst counting germinants in the field. She gave me important space and emotional support despite my concurrent love affair with Mrs Apple Macintosh.

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INTRODUCTION

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There is an urgent need to revegetate degraded land within the Avon catchment. The scale of clearing native vegetation for agricultural development is incompatible with characteristics of the region such as soils, landforms & climatic conditions (Boyd *et al.* 1976; Burvill 1979; Pigott 1993). The result of this imbalance is unsustainable land-use practises exacerbated by the time lag between the cause and the subsequent effect of the clearing. The current challenge for land managers is to apply suitable techniques for rapid and effective revegetation. Consequently, research into these methods needs to be developed in order to reverse the trend of land degradation in the southwest of Western Australia.

The type of land degradation and the impact on the economic and social aspects of the community differ within the Avon catchment in a direct relation to the geographical position along the west-east rainfall gradient. In the wetter western areas, waterlogging and water erosion are the most prominent issues (Avon River System Management Strategy 1993). In the central and the eastern sections, the predominant type of land degradation is salinisation and wind erosion, which occurs in shires such as Tammin and York. The need for revegetation has arisen from the simultaneous occurrence of the effect of an unbalanced approach towards land clearing and an increasing environmental awareness in the farming community.

A sustainable approach to land management is dependant on effective revegetation that will promote the continuity of the economic and social structure of agricultural based economies typified by towns such as York and Tammin. Degradation of the land has decreased rural productivity and in some areas has resulted in a cessation of traditional farming practices. A reduction in arable land results in direct economic and indirect social depression in the community (Gemmell 1996). This downturn is causing a shift in attitude away from traditional land management practices (land clearing and cereal cropping) towards one of an increasing environmental awareness (Gemmell 1996).

Page 1

1.1: Revegetation

Revegetation is essential in order to reverse the degradation process (Bell 1989; Davidson & Bell 1989; Dalton 1993). Vegetation provides the primary basis of biodiversity for all other organisms in the food chain, which are dependent on plants for their survival (Davidson & Davidson 1992). Furthermore, vegetation represents the interface between soil (erosion), water (hydrological balance) and air (greenhouse gases). The rate of degradation may be reduced or even prevented by the establishment of perennial vegetation within farming systems (George *et al.* 1993). Establishing vegetation is an effective management tool in the restoration of degraded systems (Munshower 1994). However, a review of the literature indicates that more effective methods of revegetation need to be developed (Malcom 1990; Venning 1991; Dalton 1992; Scheltema 1993; Pigott *et al.* 1994; Yates & Hobbs 1997).

Predominantly, land managers have two choices in the selection of a gene pool for a revegetation programme. One selection involves the planting of 'local' native species and the other is the translocation of 'distant' natives or 'exotic' plant species into the region. These choices involve separating plants along an axis of 'distance of origin' from the revegetation site. Local species are those endemic flora, which originate from the same biolocality as the revegetation site. Distant natives occur within the natural distribution of the species but is geographically distant and climatically different to the site of revegetation. Exotics are those species translocated from within the Australian continent or plants introduced from other continents relative to the site of revegetation.

The genetic integrity of the existing remnant community, proximal to the site of revegetation can be maintained by incorporating species that have a 'similar' deoxy-ribonucleic acid (DNA) 'finger-print' (B. Dixon¹ pers. comm.). However, the ability to discern between DNA finger-prints within a species is an expensive technical science (E. Bunn² pers. comm.) which is impractical for most agricultural land manages and beyond the scope of this thesis. Similarly, provenance selection trials require long term monitoring over space and in time.

¹Robert Dixon: Development Officer, Division of Plant Science at Kings Park ²Dr. Eric Bunn: Research Botanist, Division of Plant Science at Kings Park.

Chapter 1

Provenances are plant populations that have evolved characteristics, which reflect adaptations to a particular set of environmental conditions (Bennett & George 1996). Therefore, land managers require a practical and cost effective method of selecting plant provenances that are pre-adapted to a particular revegetation site. The majority of research on provenance selection of Australian natives has been conducted on the growth of eucalypts for plantation forestry (Awe 1973; Turnbull 1973; Hart 1982; Eldridge & Cromer 1987; Midgley *et al.* 1989; Florence 1996). Therefore, it is mostly likely that the provenance of other species is just as important in determining the selection of characteristics within revegetation species.

The identification of a "biolocality" that has a similar set of environmental conditions to the site of revegetation may assist land managers in the selection process of a particular plant provenance. Biolocalities are confined to an area within the natural distributional range of a particular species. They refer to the locality of the parent plant(s), which provided the seed source. The identification of a particular source of seed is specified by the use of seed lot numbers. Lot numbers are the seed merchant's collection codes, which refers to a systematic method of identifying the closest town or geographical feature proximal to where the seed was collected. The assumption of a biolocality used here is that the seed collected from a particular area contains a specific set of both genotypic and phenotypic characteristics which enable it to survive and reproduce in that locality.

The concept of biogeographical regions (bioregion) provides a useful framework in which to narrow the focus of biolocality selection for effective plant establishment on degraded land. Six bioregions have been recognised by Thackway & Cresswell (1995) in southwestern Australia. Each bioregion is characterised by a high diversity of endemic plants. However, the selection of plants for the revegetation of degraded land has proceeded largely in an ad-hoc fashion (ecoanarchy: Eckersley 1992) with the translocation of distant native and exotic species with little regard to the use of local endemic flora.

The current challenge for land managers is to select the appropriate method of revegetation that would maintain and or increase agricultural productivity and conservation values at patch and landscape scale (Yates & Hobbs 1997).

There are two fundamental methods of plant establishment used in revegetation of degraded land, namely direct seeding and nursery seedlings. Direct seeded seedlings is defined here as field established plants (Plate 1.1) through the process of sowing seeds into prepared ground using agricultural combine or manual sowing methods (Pigott *et al.* 1994). Conversely, a nursery seedling refers to a small plant grown in a soil-filled container under controlled environmental conditions (Plate 1.2) which is then transplanted into the field. Given the large extent of land management problems experienced in the central Avon catchment, the most appropriate method of revegetation is one that is cost effective on a broad scale. The term effectiveness of revegetation is based on the cost per plant established after two to three years in the field. Cost effectiveness is limited to the length of the study period but ideally it should be based upon long term reliability and establishment in the field.

1.1.1: SEED

Seeds are a basic requirement of revegetation and may be used either for direct sowing in the field or in the nursery for seedling production. The components of a seed batch when harvested consist of viable seed, non-viable seed, dead seed and chaff. These terms are explained as follows:

The term seed batch refers to a specific weight and or quantity of seed from a particular species. Viable seed is defined as seed that is capable of germination and development into a normal plant under favourable conditions within a designated time frame. This definition differs from that stated in the literature (Boland *et al.* 1980; Peterson 1987) in that it includes a time constraint. A time constraint was imposed on germination trials due to the incidence of fungal colonisation on seeds, particularly leguminous species, increased over time (see section 4.5.2). Non-viable seed is referred to here as seed that has imbibed water or quiescent seed capable of germination but not within the allocated time-frame. Dead seed is defined as non-fertilised ovules, which are not capable of germination. Chaff consists of impurities such as leaf, twig and fruiting bodies shed after dehiscence.



Plate 1.1: Direct seeded seedlings of *Eucalyptus wandoo* (16 months) after the process of manually sowing the seeds into scalped furrows at York



Plate 1.2: Nursery seedlings of *Eucalyptus* sp. (6 months) grown in a shade house under irrigation at the Chatfield's nursery in Tammin.

Seed purity and quality are terms used in subsequent chapters to describe the germination capacity of a designated seed batch. Purity refers to the number of viable seeds relative to the weight of the seed lot and is expressed in terms of the number of germinants per gram. The weight of the seed lot may consist of the weight of all four seed components (viable seed, non-viable seed, dead seed & chaff particles). Quality refers to the proportion of viable seed to non-viable or dead seed and is expressed in terms of percentage germination per gram.

The purity and quality of different seed lots can be determined by conducting a seed germination test. The germination capacity of a given quantity of seed can be measured prior to sowing. The International Seed Testing Association (1985) has published standard testing procedures, although it does not include a wide range of Australian native species. The small size of the seed, the high proportion of chaff and the similarity in the shape and colour of chaff and viable seed, all make the ISTA methods of seed testing difficult to apply to many Australian native species (Boland *et al.* 1980). Alternatively, other techniques are available for testing the germination capacity of seed lots such as tetrazolium salts, X-ray and embryo excision (Peterson 1987). However, germination tests are more practical, economical and require less expertise in the interpretation and analysis of living seed embryos.

Seeds germinate when prevailing conditions favour the opportunity for seedling establishment (Mott and Groves 1981). Germination is defined in this thesis as the growth of the embryo within the seed until the emergence of the embryonic radicle through the seedcoat (Boland *et al.* 1980). There are two basic factors that inhibit seed germination. Firstly, the environment may not be suitable for germination, and under these circumstances the seed will remain quiescent until such conditions become favourable. Secondly, seeds may fail to germinate even though they have been exposed to suitable environmental conditions.

The most important environmental variables affecting seed germination are moisture (Hegarty & Ross 1981) and temperature (Beardsell & Richards 1987). Water acts as a catalyst in the conversion of complex carbohydrates (starch) stored in endosperm reserves into simple sugars responsible for growth of the embryonic radicle. Furthermore, moisture is an important solvent in the leaching of growth retarding hormones and the synthesis of hormones that initiate germination.

Temperature determines both germination events and seed dormancy responses (Richards & Beardsell 1987). Most seeds of native plants readily absorb available moisture under temperature conditions conducive to germination (Venning 1991). However, extreme ranges of temperatures such as frosts are common in winter in the central Avon wheatbelt (Bureau of Meteorology 1998). These extreme fluctuations in soil temperature may cause seed to become quiescent or dormant which delays germination until conditions become favourable for germinant development.

The failure of seed to germinate under normal environmental conditions is determined by a seed-imposed dormancy mechanism. For example, the permeability barrier of the outer tissue of leguminous seeds has shown to impede the passage and exchange of water and gases with the embryo (Beardsell & Richards 1987). The ecological implications of seed dormancy may be due to the long-term survival of these leguminous species, by ensuring germination occurs at times when seedlings are most likely to establish (eg. following a fire and or heavy rainfall).

1.1.2: NURSERY SEEDLINGS

Nursery seedlings are a common method of revegetation in the central Avon catchment (Runciman 1991). Nursery seedlings are juvenile plants grown under controlled conditions in a container filled with growth media for a limited time period. Container produced seedlings have a high establishment rate in the nursery as the germination phase and the early development of the seedling is protected in a controlled environment. However, for effective revegetation in the field, the most suitable container for a particular seedling crop will depend on both biological and economical factors.

An essential component of nursery production is the use of containers, which can effect the long-term success of a revegetation program. The choice of biologically suitable containers minimise the problem of distorted root systems, such as girdled and circled roots (Londis *et al.* 1990). Distorted roots cause loss of vigour and poor plant establishment in the field. Whitcomb (1981) initially developed an air-root pruning container system, which promotes the development of fibrous roots. The system manipulates the root tip towards the atmospheric environment where it is desiccated and dies, preventing continuous root growth around the inside perimeter of the container.

1.1.3: SOIL RECLAMATION

Soil reclamation involves the process of ripping heavy sub-soils and soil profile construction (Munshower 1994). The purpose of soil reclamation is that it increases the chance of germination and early seedling development in the field (Venning 1991; Scheltema 1993; Stoneman 1993). The attributes of a cultivated soil bed for germination and seedling development include: deep penetration of the seedling root system (C.A.L.M. 1988), increased interface with receptive mineral soil, microsite variation such as shaded depressions (Battaglia & Reid 1993), increase nitrogen mineralisation (Robson & Taylor 1987), increased soil nutrient concentrations (Ponder 1981), hydraulic contact between seed and soil (Bachelard 1985; Gibson & Bachelard 1987) and increased weed control (Pigott *et al.* 1994). Soil preparation provides conditions for germination and subsequent development of native seed when compared with an undisturbed seedbed.

The method of soil preparation for revegetation varies according to environmental factors acting upon the field site and the scale of the revegetation programme (machinery versus hand tools). Most plant establishment methods in the central Avon catchment use machinery because it minimises labour costs, particularly on a broad-scale and the high availability of the equipment.

1.1.4: WEEDS

Soil cultivation and increased fertility as a result of agricultural practices has aided the proliferation of weed species (Croft & Venning 1985). The term weed is a value judgement based upon the perceptions and the context in which the term is used (Burke 1994). Weeds may be undesirable in the landscape from both an economic and ecological perspective (Australian National Parks and Wildlife Service, 1991). The poet Waldo Emerson defined a weed as 'a plant whose virtues have not yet been discovered' (Parsons & Cuthbertson, 1992). The inability to objectively define a 'weed' is contributing to the difficulties confronting land managers in the control of weeds.

Several authors define weeds on the basis of a subjective decision such as, "a plant growing where it is not wanted" (Kerruish & Unger 1985) and species that invade existing remnant communities or ecosystems (Humphries 1992). Similarly, Government Departments define weeds as: "any plant that is objectionable or interferes with the activities or welfare of man" (Bureau of Resource Sciences 1992) or as a plant growing out of place which may occur from within the region, interstate or overseas and may or may not be declared under the Agriculture and related Resources Act 1976 (C.A.L.M. 1993).

The choice of revegetation methods is predominantly between direct seeding and planting out of nursery seedlings. From a practical viewpoint, the most effective revegetation method would not only decrease the rate of degradation in the landscape, but would also provide a maximum return for the cost and time invested. From an ecological perspective, the selection of a biolocality for revegetation would ideally result in a sustainable plant assemblage that would integrate with the existing floral and faunal communities. Therefore, the current focus in the burgeoning field of revegetation is to develop the most effective methods in plant establishment, which incorporate local endemic species that are adapted to the characteristics of the region.

1.2: Aim of Thesis

The overall aim of the thesis is to evaluate two plant establishment methods (direct sowing of seeds versus the planting out of nursery seedlings) on degraded land in the central Avon catchment. Given the contrast of these two methods of revegetation, the criteria for evaluation is divided between pre- and post sowing/planting factors. These include pre-sowing/planting assessment of seed lot quality, purity and biolocality selection. Secondly, post sowing/planting assessment of initial survival, short-term establishment, weed control and relative establishment costs on some key native species. The long-term goal of this study is to provide results to support appropriate techniques for revegetation on a broad scale. It is envisaged that these results will improve the productive capability of agricultural land while also enhancing nature conservation values.

1.3: Structure of Thesis

The thesis is divided into eight chapters: an introduction, literature review, a background, four experiments and a conclusion. Chapter 2 summarises the relevant literature pertaining to the central aim of the thesis. Chapter 3 provides a regional perspective on the study region within the central Avon catchment and includes a site-specific background on the three case studies conducted at both York and Tammin. Chapters 4, 5, 6 and 7 are experiments that relate to the central aim of the thesis. Two of those experiments, 'The Effect of Genetic Variation and Temperature on Seed Germination' (Chapter 4) and 'The Effect of Weed Competition on Native Seedlings' (Chapter 7) were conducted under laboratory conditions. Here, Chapter 4 consists of three separate experiments which are: 'Seed Lot: Purity', 'Biolocality and Temperature: Quality' and 'Frost: Quality'. Chapters 5 and 6 are field experiments, which addressed 'Evaluation of Revegetation Methods' and 'Biolocality Comparisons'.

The 'Seed Lot: Purity' experiment (Chapter 4) was used to determine the capacity of different species to germinate when sown as a basis to the two trial sites (Chapters 5 and 6). Furthermore, the experiment was also used as a pilot study to determine the relative effects of temperature and biolocality selection on seed lot quality for the 'Biolocality and Temperature: Quality' trial. Similarly, 'Frost: Quality' was a preliminary experiment for further research on the effect of sub-zero temperatures on the quality of a range of seed lot species. These experiments (Chapter 4) involve the testing of a theory and explanation of the results using inferential statistics. This deductive method of approach involves the generation of a hypothesis from an existing school of thought followed by data collection and analyses based upon the concept of accepting or rejecting the hypothesis (Haines-Young & Petch 1986).

The deductive method of approach is not always appropriate when applied to certain disciplines conducted in the field (eg. Chapter 5 & 6). The fundamental basis to this thesis involves the process of establishing an environment (revegetation) in which to monitor the development of a synthetic plant community (seed germination, seedling establishment & plant growth) within the constraints of an academic time frame. Furthermore, field environments are subject to a full suite of varying geographical and seasonal climatic variations, which makes replications difficult to factor into an experimental design and therefore, the use of inferential statistics is not appropriate.

Initial trials of vegetation establishment on degraded land were originally designed to provide adequate experimental replications. However, the failure to establish plants at a specific field site (Chapter 5) and high mortality of germinants within a particular revegetation method (Chapter 6) hindered the design of the experiment. The reconstruction of the experiments in order to obtain the required replications and subsequent spatial data was not logistically possible within the designated time frame of the thesis. Therefore, the field trials of 'Direct Seeding Versus Nursery Seedlings' (Chapter 5) and 'Biolocality Comparison in Direct Seeding Establishment' (Chapter 6) involve case studies with descriptive statistics rather than experiments with quantitative analysis.

The examination of the case studies involved a process of experiential education (hypothesis generating) using an inductive method of approach rather than hypothesis testing. Environmental factors that became apparent while conducting the field trials such as germination, temperature and weeds formulated the basis to the topics discussed in the previously mentioned chapters.

LITERATURE REVIEW

2.1: Land Degradation

Land degradation issues in the central Avon catchment has necessitated the need for revegetation within strategic zones in the landscape. This community shift in environmental consciousness is referred to by Hobbs & Saunders (1991) as "re-integration" of a landscape disrupted and injured through historic exploitation and by Eckersley (1992) as a process of "reinhabitation". This process involves the understanding of life supporting systems, which establish ecologically and socially sustainable patterns of existence. The development of an interconnected relationship between people and the environment can be seen with the formation of land conservation district committees (LCDCs) and landcare groups. One of the indirect roles of these organisations is the synthesis of different disciplines from science, industry, conservation and the agricultural community, each addressing the problem of land degradation.

The clearing of native vegetation has exposed surface soils to weathering and erosion, which has resulted in a decline of arable land in the central Avon catchment (ARSMS 1993). The soil becomes desiccated through exposure to the sun, which causes a subsequent increase in soil temperature (Davidson 1983). Dry and denuded soil is more susceptible to wind erosion than land covered by vegetation (Davidson & Davidson 1992). Vegetation stabilises the soil profile by protecting it from the impacts of climatic factors and livestock activity (Plate 2.1).

The shallow and light sandy soils of the upland regions in the central Avon Valley are particularly susceptible to wind erosion (Lefroy 1991). Removal of the topsoil through wind erosion is a cyclic process because it results in a reduction in fertility and further accelerates the decline in vegetation through sand blasting and soil inundation (Copely & Venning 1983).

The impact of salinity and wind erosion on arable land are not obvious in the short term (Bishop & Coakes 1984). Consequently, land management problems in the Avon catchment has been based on a practice of treating the symptoms rather than the cause. The symptoms of vegetation clearance such as a rising water table have either been dealt with on the site where it is expressed in the landscape (Stolte *et al.* 1996) or has been diverted off the property.

For example, the planting of salt tolerant vegetation over water discharge zones (Plate 2.2) and the use of drainage channels to divert excess saline water to road culverts are methods used by landholders in their attempts to deal with the issue at the symptomatic level of the problem. Consequently, both these methods have had minimal impact in reducing salinisation of surface soils in agricultural areas (Bennett & George 1995; Stolte *et al.* 1996).

The salinity management challenges are being caused by a change in the hydrological balance, which results in rising ground water tables (Department of Land and Water Conservation, 1996). The removal of native vegetation and replacing it with shallow rooted annual crops (Plate 2.3) reduces the evapotranspiration of water from the soil, enabling increasing precipitation to percolate through the soil and recharge the water table (George *et al.* 1993; Cox & McFarlane 1995). The rising of the water table mobilises accumulated salt deep within the soil profile, transporting it to the surface (Plate 2.4). In undisturbed landscapes characterised by deep rooted perennial vegetation, capable of transpiring high volumes of water, the ground water is maintained well within the soil profile (McFarlane & Cox 1992).

Salinisation of surface soil and water is not a recent phenomenon. Saltland degradation was first recorded in 1905 at Cranbrook where a railway inspection engineer noted that the water had increased in salinity to such an extent that it caused problems in locomotive boilers. Increased public concern over the salinisation of potable water supplies led to a scientific inquiry into the problem (Wood 1924). It is interesting to note that the study established the first causal link between the clearing of native vegetation and a disruption in the water balance. However, despite the evidence and repeated warnings from agricultural advisers, unplanned land clearing has continued across the state. Most of these areas are prone to salinisation, waterlogging or acidification.

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Plate 2.1: Soil denuded of vegetation through overgrazing showing surface salt and rill erosion (left) compared to a fenced paddock with natural populations of *Halosarcia spp*. (right) at Tammin.



Plate 2.2: Revegetation of a water discharge zone using a salt tolerant species, *Atriplex amnicola*.



Plate 2.3: Mortality of remnant trees within the York Shire caused by ring barking, an historical method used for land clearing.



Plate 2.4: Surface salinisation and gully erosion on upper catchment slopes within the York Shire, likely the result of excessive land clearing.

Salinisation of surface soils in the Western Australian 'wheatbelt' is currently encroaching on productive arable land at the rate of 18,000 hectares per year (Schofield *et al.* 1993). The most degraded lands occur within the shires of Goomalling, Tammin and Dowerin (Fig. 2.1) where 9.26%, 7.93% and 7.06% of the arable land, respectively, has been affected by salt (Select Committee into Land Conservation 1990). The impact of clearing large tracts of native vegetation within agricultural ecosystems has not been fully realised because of the time lag between the cause and effect of land degradation (State Salinity Strategy 1996).

Effective methods in reducing the impact of salinisation are gradually being adopted by land managers (George *et al.* 1993). These methods involve the planting of perennial vegetation over water recharge zones, usually located on productive arable land (Marcar & Crawford 1996). Therefore, given the potential loss of income caused by converting pasture/crop species to perennial plant establishment, it is important that revegetation of such areas effect and rapidly mitigate land degradation. The agricultural community has only just begun to develop sufficient knowledge to practice effective revegetation methods.

2.2: Revegetation

Remnant vegetation within the agricultural regions of Western Australia are inadequate in protecting its biodiversity in the long-term because the size supporting the vegetation area is too small (Hobbs 1991a; Hobbs 1993). However, revegetation using local native species adjacent to remnant flora enhances conservation values by increasing the area of habitat refuge for endemic fauna (Hobbs & Norton 1996). Increasing the area of vegetation can also lead to the establishment of links between disconnected remnant vegetation by forming wildlife corridors (Hobbs 1993) and providing a buffer zone against edge effects from the surrounding agricultural landscape. Revegetation using local native species is particularly important because extensive habitat destruction has occurred in the Avon catchment.

Chapter 2



Figure 2.1: Locality map within the Avon catchment showing the Shires of Goomalling, Tammin and Dowerin, which are most affected by saltland degradation.

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Revegetation often involves the importation and establishment of seeds or seedlings in a reclaimed soil bed. The disturbed land can function as an integral component of the ecosystem provided that adequate attention is given to encouraging biological diversity (Davidson & Davidson 1992) and maintaining the genetic integrity (Eckersley 1992). Biological diversity can be achieved by establishing a diverse plant community with the capacity of regeneration. Such an assemblage of plants creates a community based on interactive cycling of nutrients and flow of energy within the system (Waring 1989).

The overall importance of vegetation is emphasised by defining it as the accumulation of biomass through primary production upon which organisms of both the grazing and detrital food webs ultimately depend on for their survival (Kent & Coker 1992). Therefore, vegetation provides the basis of the habitat within which most organisms complete their life cycle.

Revegetation on degraded lands can have multiple functions. The conservation status of the surrounding region and agricultural productivity can both be simultaneously improved (Lefroy *et al.* 1993). The style of revegetation should therefore be chosen to assist with environmental management and the objectives specific to the site (Plate 2.5 & 2.6). For example, an effective windbreak using diverse native vegetation can also act as a wildlife corridor by establishing links with existing remnant bushlands (Loney & Hobbs 1991).

Native vegetation can host beneficial insectivorous fauna and provide crop windbreaks and stock shelterbelts (Davidson & Davidson 1992). Several studies have shown that many animals colonise native vegetation once established (Landsberg *et al.* 1990). Native shrubs provide nectar for the adults of wasps whose larvae are parasites on defoliating insects of cereal crops (Davidson 1988). The provision of vegetation windbreaks have the potential to increase cropping yields. Bicknell¹ (cited in Runciman 1991) has found a 22% increase in wheat, 47% in oats and 30% in lupin yields due to the presence of vegetative windbreaks. Therefore, native vegetation can indirectly assist in the productivity of agricultural crops and livestock.



Plate 2.5: Revegetation of a saline drainage channel using salt tolerant *Eucalyptus camaldulensis* to help stabilise the hydrological cycle and soil erosion.



Plate 2.6: Landholder and LCDC member John Munkton discussing the merits of an *Acacia saligna* windbreak with environmental management student Peta-Lyn Blyth.

The impacts of translocated seeds or plants into disturbed landscapes can have degrading effects on the local remnant vegetation. Pathogens, pests and weeds (eg. diseases, insects, exotic species) that invade local plant populations can cause high mortality and or genetically pollute remnant species through interbreeding and in doing so, alter the community structure. Translocation should not be viewed in isolation but rather within an ecological context, which alters the relationship between the soil, water and the biotic community (Horwitz 1994).

An example of an introduced species from Western Australia into southern Victoria and South Australia is the invasion of *Acacia saligna* (Whibley 1980) which has increased the cover of the understorey stratum and out-competed local species. This has accelerated vegetation succession and displaced native species within the existing remnant vegetation. The introduction of foreign *Bracychyton* seed can host the seed borer larvae of the Kurrajong weevil *Axionicus insignis* (Kerruish & Unger 1985) which can reduce the number of viable seeds within a particular seed lot. The translocation of genetic material can also pollute the local remnant community. For example, the 'torwood' hybrid initially resulted from the hybridisation between *Eucalyptus torquata* and *E. woodwardii* caused by the translocation of these species (Venning 1988). In their natural habitat these species have different distributions and do not have the opportunity to interbreed. An example of an introduced disease in the Avon catchment is *Phytophthora cinnamomi*, a soil based that causes 'dieback' of remnant vegetation (Landsberg *et al.* 1990).

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The use of local species is not always the optimum choice to achieve the most effective revegetation. This is because the process of land degradation alters the existing landscape, which may no longer be suitable for local species (Yates & Hobbs 1997). In this situation distant natives or exotics may be more successful than local species. These species may outperform local species due to an absence of predators and a pre-adaptation to degraded land, which is similar in conditions to that experienced in its place of origin. For example, pioneer species such as Australian acacia's are used extensively around the world and within Australia to revegetate highly disturbed and or extremely degraded lands (Langkamp & Dalling 1982).

Revegetation of the Avon catchment using local species may be suitable because extreme land degradation accounts for only a small proportion of total degraded land (Naldony 1991). There has been little evidence to suggest that local species are no longer suitable to these altered environment (Naldony 1991). Degraded land has been naturally colonised by native species such as *Eucalyptus camaldulensis* (Chippendale & Wolf 1981) and *Atriplex amnicola* (Malcom *et al.* 1984) within the Avon basin. Therefore, planting local species may contribute to the long-term sustainability of a plant community because local natives are adapted to the surrounding environment (Lefroy *et al.* 1991). Local species have evolved adaptations which enable their long-term survival and hence perpetuation of the species. These survival mechanisms are often expressed in terms of successful reproduction and seedling establishment (Curtis 1990).

2.2.1: SEED DORMANCY MECHANISMS

The range of seeds used for revegetation of degraded land in the Avon Basin contains a large component of leguminous seed (Pigott *et al.* 1994). The Leguminosae group consists of the families Fabaceae (pea), Mimosaceae (Acacia) and Caesalpiniaceae (Cassia). The preponderance of this group is due to their 'opportunistic' (r-selected) colonisation of hostile and highly disturbed environments (Cavanagh 1987). Acacias in particular can colonise land deficient in nitrogen and phosphorus due to the synergistic effect of *Rhizobium* species and the root inhabiting *Mycorrhiza* fungi (Langkamp & Dalling 1982). Legumes have a hard resilient seed coat, which enables a long dormancy period in order to germinate in favourable moisture conditions (Cavanagh 1987). Leguminous seeds rendered dormant by this mechanism require pre-sowing treatment which 'primes' the seed for germination (Richards & Beardsell 1987).

Despite a considerable wealth of scientific knowledge on hard-seeded species and limited research conducted by Fox *et al.* (1987), there is still a lack of understanding on the effects of pre-sowing treatment on germination (Cavanagh 1987). The 'optimum' treatment for many leguminous seeds is not known and both wet and dry treatments have been applied with variable success. Wet treatment involves the use of hot or boiling water, acids, alkalis, organic solvents and alcohols. Conversely, dry treatments use microwave heat energy, manual or machine scarification and impaction. Inappropriate seed treatment in the past has resulted in poor germination and subsequent low numbers of germinants in the field which may have contributed to the perceived reliability of containerised nursery seedlings (Runciman 1991).

2.2.2: CONTAINERISED NURSERY SEEDLINGS

The growth potential and long-term establishment of nursery seedlings is dependent on the propagation medium, methods and container. Seedling growth and survival is dictated by the porosity of the soil medium and the resilience to weathering over time. The long-term establishment of seedlings in the field is determined by the degree of the initial root distortion in order to prevent girdling and wind throw.

Nursery seedlings growth appears to be limited by the availability of root space within a container. The primary limiting resource in a containerised environment is oxygen levels within the growing medium (Whitcomb 1984). The air space in the containerised growing medium is crucial for the relationship of gas exchange and root development. One of the processes of growth involves the respiration and subsequent growth of the plant roots. As the root system grows in a container, the demand for oxygen increases while the demand for carbon dioxide decreases. However, as the oxygen level decreases the carbon dioxide level increases in nonporous media as roots develop into the air spaces and the growing medium decomposes (Whitcomb 1984). Therefore, nursery seedlings may be affected by an oxygen deficit or carbon dioxide toxicity within the growing medium over time.

The length of time seedlings are held in a containerised environment should correspond with the time of transplanting in the field because they can exhaust the essential elements needed for growth in a container. Seed is usually container sown at a particular time in the season, in order to produce a seedling that has been root stabilised in a container by the time of dispatch from the nursery (Y. Toussaint¹ pers. comm.). The time is usually designed to coincide with the start of the winter planting season in June. The soil volume must be proportional to the final size of the seedling (Londis *et al.* 1990) which will be a function of both the species grown and establishment time.

The concept of the air-root pruning container system minimises root distortions in nursery grown seedlings. This has led to complex designs and the development of chemically lined plastic punnets and organic pots. For example, of techniques developed in Western Australia, Ellenby Tree Farm use an air pruning system that is designed in the shape of an inverted stepped pyramid punctured with holes to grow advanced native trees (D. Woodroffe¹ pers. comm.). In the Manjimup Nursery of the Department Conservation and Land Management (C.A.L.M.), copper carbonate paint is sprayed on the inside of the punnets, which effectively kills the root tip of developing seedlings (L. Piggott² pers. comm.). Marrinup Nursery of Alcoa Australia Pty. Ltd. uses a compressed pine bark pot that allows root penetration of seedlings through the fibrous container (Sue Taylor³ per. comm.). A biologically suitable container design can have a dramatic effect on the growth and establishment of native seedlings (Whitcomb 1981).

2.2.3: SOIL RECLAMATION

Plant establishment using native seedlings is usually increased on sites where the soil has been cultivated (Abbott 1984; Abbott & Loneragan 1985; Stoneman 1993). This has been due to several factors such as: improved root penetration into sub-soil (C.A.L.M. 1988), higher capacity of soil for infiltration of precipitation, increased water retention at low suction pressure in soil macropores (Stoneman 1993), reduced predation (invertebrates) through partial burial of seed in porous medium (Stoneman 1993) and a reduction in the density of competing vegetation such as weeds (Scheltema 1993). Soil disturbance increases the effectiveness of a revegetation programme.

¹David Woodroffe: Owner/manager at Ellenby Tree Farm; ²Lindon Piggott: Nursery manager at C.A.L.M., Manjimup; ³Sue Taylor: Nursery manager at Alcoa, Marrinup.
2.2.4: WEEDS

Weeds can be generally categorised as plants that have an efficient metabolism and a successful reproductive strategy. Most weeds metabolise carbon through the process of photosynthesis more efficiently than other species used in revegetation programmes (Kerruish & Unger 1985). Some weeds fix carbon from atmospheric carbon dioxide into simple sugars through a shorter and efficient 'C4' pathway (Kerruish & Unger 1985). Conversely, other plants follow a 'C3' pathway and are not as efficient in utilising carbon. Therefore, C4 weeds have a competitive advantage in establishment relative to C3 plants, particularly in colonising highly disturbed and altered environments (Hobbs 1991b).

Weeds represent the first successional stage in stabilising a disturbed habitat. C4 weed species are 'opportunistic' (r-selected) in colonising newly altered environments through their capacity for rapid reproduction (Recher *et al.* 1990; Hobbs 1991b). Most annual weeds such as capeweed (*Arctotheca calendula*) have a short life cycle so they germinate, reach sexual maturity and reproduce in the one season. Weeds have successfully adapted to colonising degraded agricultural land and reproducing after seasonal rainfall events (Hobbs 1991b). Effective revegetation of degraded land in the Avon catchment may be dependent on weed control as well as biolocality selection and the choice of plant establishment methods (Runciman 1991).

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CENTRAL AVON CATCHMENT: REGIONAL AND SITE DESCRIPTION

3.1: Introduction

This chapter provides a brief summary on the geology, soils, vegetation, land-use and climate within the study region of the central Avon catchment in the southwest of Western Australia (Fig. 3.1). This regional perspective is then focused on the study sites located near the towns of York and Tammin.

The Avon River serves as a major drainage network for the agricultural region and provides both a recreational resource and a link between a series of different vegetation ecotypes. Deterioration of the catchment will naturally result in a perturbation in the characteristics of the river. This has important implications as the river flows through a number of town sites and the city of Perth. Therefore, the Avon catchment and river system has both local and regional significance from an economic, social and ecological perspective (Avon River System Management Strategy 1993).

The Avon River Basin is the largest water catchment (Fig. 3.1) in the southwest drainage division of Western Australia. The river drains 120,000 km² of the wheatbelt (Walker 1986). In doing so, the river acts as a barometer for health of environmental processes within the catchment. Issues such as water quality, siltation (Hansen 1986) and consequent loss of habitat are closely linked to land management problems experienced within the Basin.

Chapter 3



Figure 3.1: Geological features and the trial sites, situated near the towns of York and Tammin within the study region of the greater Avon River catchment, comprising of the Lochardt, Yilgarn and Avon sub-catchments (adapted from the Avon River Management Strategy 1993).

3.1.1: SITE SELECTION OF CASE STUDIES

Two study sites within the Avon River catchment were selected as representative of the environmental variation affecting both direct seeding and nursery seedling establishment on degraded land in the agricultural region. Site 1 (York) is located on private property called 'Woodlands' 97 km due east of Perth and 15 km east of York on the Goldfields Road (Fig. 3.1). The site represents shallow soils exposed to wind erosion. Site 2 (Tammin) is private property located 80 km east-north-east of site 1 and 20 km west of Tammin on Rodger-Kitty Road (Fig. 3.1) and is situated in a landscape depression, adjacent to a saline drainage channel.

3.2: Regional Geology and Landform

The Avon Basin lies east of the Darling Scarp on the Yilgarn Block (Fig. 3.1) which is a shield consisting of Archaean granite and metamorphic rocks (Mulcahy 1967). This geological province is characterised by low relief and shallow drainage channels. The landscape has been subjected to weathering over a very long time producing lateritic soils to great depth.

The basin comprises three main catchments areas, the Yilgarn and Lochardt catchments in the east and the Avon catchment in the west (Fig. 3.1). Despite being similar in formation, the western Avon catchment is distinctively different from the drier eastern catchments. The western part of the Avon catchment is characterised by relatively sharp relief with steep valleys and hilltops. The most prominent geological feature in this region is the Darling Scarp and its relief relative to the Swan Coastal Plain (Seddon 1972). The formation of the scarp is the result of the Darling fault which is caused by the western continental Precambrian block 'riding' over the eastern Perth Basin Cretaceous block (Seddon 1972). This event caused the reactivation of ancient paleo channels in the area (Garden 1979). The erosion forces of these westward flowing river systems has cut into and exposed the parental granite and metasedimentary rocks underlying the plateau to form the current topographical relief (Walker 1986).

East of the Meckering Line (Fig. 3.1), the Avon River flows over a geologically mature landscape such as the Yilgarn and Lochardt catchments. These catchments become less dramatic in relief with broader valleys interspersed with ancient salt and freshwater wetlands (Mulcahy 1967). West of Toodyay (Fig. 3.1) is considered to be the point at which the ancient Avon drainage system ceases and rejuvenation of the river systems begins.

The landscape can be described by three main landforms: the upland sand plains, the valley slopes and the valley floors (Lefroy *et al.* 1991). These landforms are characterised by the following soil types: coarse textured sands and gravels of the uplands, sandy clays and loams of the valley slopes and fine textured clay soils of the valley floors (Fig. 3.2).

3.3: Regional Soils

The landscape in the southwest of Western Australia consists of a Precambrian shield, which has been tectonically stable for a long period of time. The landscape was not rejuvenated in the Pleistocene glaciations (Mulcahy 1967) and as a consequence, the soils are highly weathered and leached resulting in infertile soils (Walker 1986).

Soils within the river basin are characterised according to distances along the river's gradient (Walker 1986). In the developing western catchment of the Avon River, grey alluvial soils are found on the flood plain. Dolerite dykes are evident in this area, responsible for the fertile red brown soils within the valley. As the river's gradient gradually steepens towards the east, the soil type is dominated by white and yellow sand-plain soils and gravel laterite. Fertility tends to decrease as distance from watercourses increases (Walker 1986).

The association of the soil types relative to the position of the landform is often difficult to categorise. Within the various landforms, alluviation and colluviation processes have continually 'worked the parent material to baselevels' resulting in a complex mosaic of different soil types (Mulcahy 1967). The array of different soil types supports a diverse range of native vegetation.

3.4: Regional Vegetation

The region is characterised by a high degree of floral endemism which has been attributed to an explosive speciation during the Quaternary period (Beard 1990) and the isolation from similar climates of southeast Australia by the geographical, climatic and edaphic barrier of the Nullarbor Plain. However, 75% of this native vegetation has been cleared in the catchment since 1945 for agricultural development (Binnie & Partners 1985; Saunders *et al.* 1985; Churchward 1986; Jenkin 1986; Saunders & Hobbs 1989) and has been subsequently replaced with cereal crops and pasture species. As a consequence, the remaining vegetation is fragmented and is represented by nine major vegetation associations (Lefroy *et al.* 1991) listed in Table 3.1.



Figure 3.2: Stylised cross section of the Avon Valley showing the relationships of landform, soil and vegetation types (adapted from Lefroy *et al.* 1991).

The predominant landuse in the Avon catchment is dryland agriculture. The type of agricultural pursuit employed in any one area differs according to the rainfall pattern and soil conditions. A landuse gradient exists across the Avon catchment whereby livestock grazing in the west is replaced with cropping, predominantly wheat, in the east. The western section of the catchment has a relatively high rainfall and fertile soils, which support sheep (meat; wool) and to a lesser extent cattle-based farming activities. The eastern section of the catchment has a relatively low rainfall and infertile soils which is limited to cereal cropping.

Table 3.1:

Typical Vegetation Association with Landforms in the Avon

	В	asın
Landform	Group	Representative species
Sand plain	Wodjil Tamma Grevillea	Acacia neurophylla Allocasuarina acutivalvis Grevillea pritzelii
Valley Slopes	White Gum York Gum Mailee Salmon Gum	Eucalyptus wandoo Eucalyptus loxophleba Eucalyptus erythronema Eucalyptus salmonophloia
Valley Floor	Morrel Salt Lake	Eucalyptus longicornis Atriplex amnicola

Note, Source: Lefroy et al. 1991

3.5: Regional Climate

The Avon Basin experiences a Mediterranean Climate in the western catchment (Bell & Bellairs 1992) and a semi-arid climate in the eastern catchment. Temperature varies according to both season and geographical position within the basin (see section 3.6.3 & 3.7.3 & Fig. 3.5 & 3.7).

Average annual rainfall decreases from 600 mm in the west to less than 300 mm in the eastern range of the basin (Fig. 3.3). Rainfall is highly seasonal with 80% of the annual rainfall falling between the months of May and October.

3.6: Case Study 1, York: Background

(Source: J. & J. Munkton¹)

The York field site is considered to be 'third class' and has a very low crop yield potential. The site was cleared in 1928 by migrants who were lured to the area in search of gold. In 1928 to 1940 the land was cultivated with lupins on a long term cropping rotation alternating with fallow land. Since the 1940's and the advent of fertiliser ('The Green Revolution'), short term cropping rotations have been possible due to the application of macro-elements such as phosphates and trace elements. In this period soil tillage such as mouldboard ploughing was a common practice.

A significant drop in cereal crop yields have been linked to wind blasting from airborne sand and erosion of the relatively fertile topsoil. Effective establishment of a native vegetation windbreak may alleviate desiccation of cereal crops and wind erosion of soil.

3.6.1: CASE STUDY 1, YORK: METHODS OF SOIL ANALYSIS

A soil sample was taken from three sites along the contour (Upper slope, Mid slope & Lower slope), orientated north/south (respectively) within the revegetation area. A hole was excavated to the horizon of the cemented laterite at the three sites using a manual post-hole digger. Soil samples were taken (Approx. 1.5 kg) at each horizon and analysed in the laboratory at Edith Cowan University.

A kilogram of soil from each horizon was agitated through nested sieves into four aggregate fractions ranging from 2mm to <0.02mm. Each fraction was weighed in order to calculate the percentage of the total aggregate and classified into texture according to Handreck and Black (1986).

Distilled water was added to a 50g soil sample that was placed in a 1000ml beaker until it formed a saturated paste with no stratified 'free' water. This sample was mixed using a stainless steel spatula and allowed to stand for an hour. The electrical conductivity and the pH of the paste were measured using an electronic meter with a glass electrode. The meter was calibrated in a buffer of a known electrical conductivity and pH solution.

¹John and Joan Munkton are the property owners of 'Woodlands' at York.

Chapter 3



Figure 3.3: Annual rainfall within the south west of Western Australia (adapted from the Select Committee into Land Conservation 1990).

3.6.2: CASE STUDY 1, YORK: RESULTS & DISCUSSION OF SOIL ANALYSIS

The texture of the soil profile consists of sand over cemented laterite (Table 3.2). The depth of the sand varies as follows: Upper slope: 1280 mm; Mid slope: 680 mm and Lower slope: 100 mm. The shallow nature of the site soil is subjected to both water and wind erosion. This has accelerated since it was cleared of native vegetation for agricultural practices (J. Munkton pers. comm.).

Table 3.2:

Mean Percentage Fractions of Three Soil Samples Showing the Depth of Four Different Aggregate Components and the Composite Texture at York

Depth (mm)	Gravel > 2 mm %	Sand 2.0-0.2 %	Fine sand 0.2-0.02 %	Silt/clay < 0.02 mm %	Texture
		:			
0-10	2	52	32	14	Sand
10-80	1 .	26	38	35	Loam
80-210	7	23	31	39	Loam
210-680	73	13	8	6	Gravel
680	Cemented				Laterite

The topography of the site has a 1:50 fall from south to north and a 1:100 fall running east to west. To the east the land gently rises before sharply breaking away exposing the underlying lateritic caprock which has produced weathered red loam soils.

Mineral salts within the soil are low and decrease slightly with depth. The ionic concentration of the topsoil is 80 micro-Siemens per centimetre (μ S/cm) or 51 parts per million (ppm) of total dissolved solids (T.D.S.). The sub-soil concentrations of ions are 59 μ S/cm or 38 ppm. The soil is moderately acidic (Handreck & Black 1986) with a pH of 5.6 to 5.7 which is within the range to support most types of vegetation communities.

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The association of vegetation and soil types based upon the classification system by Lefroy *et al.* (1991) is described below (Fig. 3.2). The site is situated on the boundary between the upper sand plain (tamma) and the valley slopes (white gum). This is supported by the varying nature of the soils surrounding the site; to the west there is deep yellow sand to a depth of two metres (Wodjil; grevillea) and to the north is found shallow grey sand over deep white kaolinitic clay (white gum), which is indicative of the vegetation type.

3.6.3: CASE STUDY 1, YORK: VEGETATION

In order to re-establish native flora on the York field site, identification of the remaining endemic flora is important. Remnant vegetation is a genetic library of information on some of the species, which may have covered the landscape prior to agricultural development. The predominant species in the remnant vegetation adjacent to the site is: *Acacia colletioides; Allocasuarina campestris; Allocasuarina huegeliana; Banksia attenuata; Dryandra armata; Eucalyptus wandoo* and *Leptospermum erubescens*. According to Lefroy *et al.* (1991), these species represent an ecotone between several vegetation assemblages occuring within different landforms. This may be inferred because of the occurrence of several species typical of both assemblages located at the one site.

3.6.4: CASE STUDY 1, YORK: CLIMATE

The York field site is an elevated escarpment exposed to prevailing winds from the east-north-east. The average mean monthly rainfall for York (Fig. 3.4) is 453 mm per year (Bureau of Meteorology 1998). The daily mean maximum and minimum temperatures for York are expressed in Figure 3.5. The annual mean number of days below zero is 7.4 (Table 3.3).

Table 3.3:

Mean Number of Days Below Zero Degrees Temperature for York

(1001-1350).							
Spring Months	Days < 0°C	Summer Months	Days < 0°C	Autumn Months	Days < 0°C	Winter Months	Days < 0°C
September	0.7	December	0	March	0	June	1.1
October	0.3	January	0	April	0	July	2.0
November	0	February	0	May	0.8	August	2.4

Note. Source Bureau of Meteorology (1998).





Figure 3.4: Mean monthly rainfall data for York (1877-1994) and for each successive year (1995, 1996 and 1997) in which the two field trials were conducted (Source: Bureau of Meteorology, 1998)



Figure 3.5: Mean daily maximum and minimum temperature for York from 1983 to 1997 (Source: Bureau of Meteorology, 1998)

3.7: Case Study 2, Tammin: Background (Source: A. Rodgers¹)

The Tammin district was extensively cleared when European development of agricultural land moved eastward from the alluvial soils of the Swan Valley. The actual site was cleared in 1930 by Allan Rodgers senior predominantly, for cereal (wheat) and sheep (meat & wool). As with site 1, agricultural cropping was made possible through the advent of phosphate and trace element fertilisers in conjunction with the sowing of nitrogenous clover on a crop rotation basis.

The removal of the once dominant woodlands has increased the infiltration rate of rainfall, mobilising existing salt stored within the soil profile down slope to the water channel. This process resulted in the region of Tarimin being ranked the second most affected shire in the Avon catchment (see Fig. 2.1). The current landholder, Allan Rodgers has witnessed an encroachment of salt into once productive arable land (since 1965).

3.7.1: CASE STUDY 2, TAMMIN: METHODS OF SOIL ANALYSIS

A soil sample was taken from three sites along the contour (Upper slope, Mid slope & Lower slope), orientated north/south (respectively) within the revegetation area. A hole was excavated to a depth of 300mm at the three sites using a manual post-hole digger. Soil samples were taken (Approx. 1.5 kg) at each horizon and analysed in the laboratory at Edith Cowan University similar to Case Study 1 (see 3.6.1).

3.7.2: CASE STUDY 2, TAMMIN: RESULTS & DISCUSSION OF SOIL ANALYSIS

The texture of the soil profile consists of grey sand over white sandy clay over white kaolinitic clay (Table 3.4). The depth of the sand varies as follows: Upper slope: 350 mm; Mid slope: 320 mm and Lower slope: 450 mm.

¹Allan Rodgers is the property owner at Tammin.

Table 3.4:

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Depth (mm)	Gravel > 2 mm %	Sand 0.2-2.0 %	Fine sand 0.02-0.2 %	Silt/clay < 0.2 mm %	Texture
0.10			43		Cond
10-120	4 12	47 47	42 20	21	Loam sand
120-300	9	52	20	19	Loam sand
300 +	3	36	21	40	Loam

Mean Percentage Fractions of Three Soil Samples Showing the Depth of Four Different Aggregate Components and the Composite Texture at Tammin.

^a The texture of the soil is classified according to Handreck and Black (1986).

Mineral salts within the soil are relatively high in comparison with the soil at York. The ionic concentration of the topsoil is $320 \ \mu\text{S/cm}$ or $205 \ \text{ppm}$ and the sub-soil is $465 \ \mu\text{S/cm}$ or $298 \ \text{ppm}$. The soil is within the neutral range (Handreck & Black 1986) and has a pH of 6.8.

The topography of the land has a 1:90 fall from north to south and a 1:90 fall running west to east. To the south, the land gently falls to a saline tributary. The creek line is littered with dead tree trunks indicating that the effects of salt expressed on the soil surface is a relatively recent event. Tree species are gradually being replaced with salt-lake species such as *Atriplex amnicola* and *Halosarcia* species.

The site soil is both shallow and permeable, allowing precipitation to infiltrate and dissolve accumulated salts within the upper soil profile. The saline water is intercepted by the impermeable clay subsoil where it is shed down-slope to landscape depressions. The perched saline water table causes secondary salinity and waterlogging of the topsoil, which gradually encroaches into surrounding arable land.

The association of vegetation and soil types based upon Lefroy *et al.* (1991) classification system (Fig. 3.2) is described below. The site is situated on the valley slope and merges from the upper slope (White gum), with the lower slope (York gum & Mallee) landform and vegetation types. This is supported by the varying vegetation communities in and around the field site.

3.7.3: CASE STUDY 2, TAMMIN: VEGETATION

The adjacent remnant vegetation is comprised of a mixture of several vegetation types $\frac{1}{2}$ (Acacia acuminata; Acacia erinacea; Allocasuarina campestris; Calothamnus gilesii; Eucalyptus capillosa; Eucalyptus erythronema; Eucalyptus loxophleba; Eucalyptus salmonophloia). The actual site vegetation is dominated by Eucalyptus species with very few understorey species. The shrubs are declining in vigour with no natural recruitment. This has been due to sheep, which have been permitted to graze on and shelter from the climate extremes within the remnant vegetation.

3.7.4: CASE STUDY 2, TAMMIN: CLIMATE

The annual average rainfall for Tammin (Fig. 3.6) is 344 mm per year (Bureau of Meteorology 1998). This represents a decrease in the rainfall gradient of 109mm relative to the York trial site. The mean daily maximum and minimum temperature for Tammin is expressed in Figure 3.7.





Figure 3.6: Mean monthly rainfall data for Tammin (1911-1995) and for each successive year (1996 &1997) in which the field trial was conducted (Source: Bureau of Meteorology, 1998)



Figure 3.7: Mean daily maximum and minimum temperature for Tammin from 1892 to 1998 (Source: Bureau of Meteorology, 1998).

THE EFFECT OF GENETIC AND TEMPERATURE VARIATION ON SEED GERMINATION

4.1: Introduction

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The process of revegetation has three distinct phases: seed germination, seedling development and plant establishment. The primary phase in direct seedling establishment is germination. Germination represents an important stage in the process of revegetation because germinants are vulnerable to a suite of physical and biological factors (Beardsell & Richards 1987). Seed germination is also influenced by the genetics of the parent stock (Gross 1972; Beardsell & Richards 1987).

The evaluation of the relative importance of environmental and genetic influences on germination, depends on the purity and quality of a given seed lot. A germination trial is one method that can determine the purity and quality of a seed lot because it provides a mechanism to calculate the number (per unit weight) and percentage of seeds that germinates (Scott 1972; Mott & Groves 1981; Beardsell & Richards 1987).

The purity of each seed lot is dependent upon on the provenance, seed treatment, environmental parameters, collection, storage and handling of the seed. The evaluation of seed lot purity enables a calculation of the sowing rate required to produce a given number of plants per hectare (Venning 1991; Scheltema 1993; Pigott *et al.* 1994). The quality of each seed lot is dependent on the same factors mentioned for purity but does not include handling which deals with the removal of chaff. The analysis of seed lot quality assists in the evaluation of seed treatments (scarification, boiling water etc.) and biolocality selection prior to sowing in the field.

The analysis of seed lot quality within different biolocalities is one factor that cannot assume to be constant given the potential interaction between the genetics of the plant and the surrounding environmental variables. The flora of Western Australia has adapted to a wide range of climatic regions and soil types. This has resulted in diverse vegetation assemblages between biolocalities and within a species distribution range (Beard 1990). Consequently, a set of specific environmental parameters may influence the genetic composition of a localised plant population. The potential genetic variation between biolocalities within each species is high. Therefore, the selection of a particular seed lot locality may be an important factor in determining seedling establishment in the field.

The rationale behind biolocality selection is to source the optimum seed lot (Florence 1996) for plant establishment under environmental conditions that prevail at the specific site to be revegetated.

Environmental influences on plant genetics are often expressed as an observable phenotype such as a physiological response or morphological characteristics. Provenance variation has been found in the physiological responses of *Eucalyptus leucoxylon* such as germination rates, frost resistance, seedling growth rates and drought tolerance (Boland 1978). Several researches indicate that the optimum temperature for germination can be hereditary (Young *et al.* 1973; Lodge 1981; Lodge & Whalley 1981). The influence of environmental parameters on the quality of the seed (particularly when plants are subjected to stress) may be necessary for the perpetuation of each species, dependent on inherent germination responses specific to a particular biolocality.

The relationship between genotype and phenotype with regard to seed germination is complex and difficult to discern when applied to the concept of biolocality. For example, species with restricted distributions may contain isolated seed biolocalities that are genetically variable and adapted to specific environmental conditions (ecotypic). In contrast, plant species with a wide geographical distribution may be characterised by a suite of genetic variations that are either ecotypic or clinal (Florence 1996). The climatic influence (temperature) on genetic seed quality can be measured by comparing the relative germination capacity of seed lots for the same species collected from different biolocalities.

A range of temperatures can be used in the laboratory in a germination trial. Ideally, the temperature treatments adopted should reflect those experienced in the seed's biolocality. This allows the evaluation of the influence of an important environmental variable on optimum seed germination.

The occurrence of extreme temperature fluctuations at the soil surface may have increased in the central Avon catchment due to the clearance of native perennial vegetation. The removal of overstorey species increases the diurnal variation in temperature (Stoneman 1993) and can have a considerable impact on the microclimate of the soil bed (Keenan 1986). A vegetation canopy prevents both direct solar radiation during the day and winter frosts at night. This canopy protection may have either a positive or negative effect on seed germination depending on the species and the biolocality of the seed lot. In the central Avon catchment, seeds are sown in winter due to seasonal rainfall events, however this often coincides with the occurrence of sub-zero temperatures.

Effective revegetation in the central Avon catchment may be dependent on the relative interactive effects of temperature, provenance selection and seed treatment on the germination. The importance of seed purity and quality in the revegetation process cannot be underestimated (Beardsell & Richards 1987). The successful establishment of both nursery seedlings and direct seeding methods are dependent on seed germination. The evaluation of seed lot purity and quality is instrumental in effective revegetation.

4.2: Aim

The aim of this experiment is to examine the variability in purity and quality across seed lots from several native species by assessing:

- 1. SEED LOT: PURITY. The purity of several native seed lots under three different alternating temperature regimes.
- 2. BIOLOCALITY: QUALITY. The quality of different seed localities of the same species under four different temperature regimes.
- 3. FROST: QUALITY. The quality of *Eucalyptus wandoo* seed batches subjected to sub-zero temperatures.

4.3: Methods

Approximately 1.2g of fine vermiculite was placed into sterilised 9 cm diameter petri dishes and covered with number 4 Whatman filter paper. Boiling water was applied to each dish as a sanitary measure against fungal colonisation. Distilled water was added to sufficiently wet the vermiculite and filter paper. A set quantity of seed was applied (see Tables 4.1, 4.2 & 4.4) evenly over each petri dish according to the size and weight of the seed. A transparent plastic lid was placed on each petri dish in order to prevent the medium from drying out.

In order to simulate frost, petri dishes were placed in a freezer. The remaining dishes were placed into a refrigerated incubator (Cat N°RI.250/SG). The incubation cabinet was converted to a 'seed germination cabinet' by the installation of low intensity lighting mounted behind the door. Each incubator contained three 30W fluorescent tubes shielded by an acrylic panel to minimise heat transfer into the main cabinet. Most of the surfaces within the cabinet are white in colour for maximum lighting effect. The temperature and light regimes of the refrigerators were set according to each aim stated in 4.2.

In an attempt to break the seed coat imposed dormancy known to occur in the Leguminosae group (Cavanagh 1987), both heat and scarification treatments were employed. *Acacia microbotrya* (seed lot # 424 & 545) was exposed to boiling water for 60 seconds and soaked in cold water for 12 hours. The seed of *Acacia saligna*, *A. microbotrya* (# 900163) and *Kennedia prostrata* (# 960501) seed was scarified for five minutes by shaking the seeds in a glass jar lined with sandpaper. *Kennedia prostrata* (# 642 & 1365) received no seed pre-treatment.

Germination was assessed every second day. Germinants with radicals 2-5 mm long were recorded and then removed and distilled water was added to maintain saturation of the filter paper. A pre-emergent fungicide of Benlate (active constituent: benomyl) was applied at the concentration rate of one gram to one litre to seed experiencing fungal attack.

The experiment was concluded at 28 days after which the filter paper was allowed to dry over a 24 hour period. The remaining plant material was removed and spread over white paper for visual clarity. The germination purity of each sample was calculated by the following descriptive formula:

Germination Purity (per Gram) = <u>Germinated Seed</u> Weight of Seed Lot (grams)

Percentage seed germination is used to compare seed quality between different seed lots, as it is not influenced by variable seed purity (chaff weight) and seed weight (specific gravity) between seed lots. This is particularly important if the weight of the chaff represents a large proportion of seed lots with small seed or if there is variation in the size of seed between seed lot species. Ungerminated seed (non-viable + dead seed) was distinguished from chaff and counted under 10x magnification to determine percentage seed germination. The germination quality percentage was then calculated using the following descriptive formula:

The experimental design consists of germinating seed in a number of petri dishes. The number of individual petri dishes used in this study differs according to the experiment. For statistical purposes the term used to describe each petri dish is a sample or sub-batch of a seed lot and is not considered a repeated measure (replicate). This is because once the seed has germinated, it is no longer investigated. The nomenclature used in the experimental design is:

Samples \rightarrow Batches \rightarrow Seed Lots \rightarrow Species

4.3.1: SEED LOT PURITY

The aim of the purity test was to provide the data to determine the sowing rate of seeds sown in the field during winter at both York and Tammin (Chapters 5 & 6). Germination quality percentages were also used here to eliminate the variables of both chaff and the size of the seed in order to determine the variation between seed lots and within species. This variation is particularly important for use in this pilot study to indicate the relative importance of environmental influences and provenance selection on seed germination.

Available moisture is a seasonally limiting factor in the Avon catchment (Scheltema 1993), and therefore, seeds were sown in winter (Chapter 5 & 6) to coincide with seasonal rainfall (see Fig. 3.4 & 3.6). At the time of seeding, moisture was assumed to be available to stimulate germination and therefore, the major determining environmental influence was temperature. Consequently, moisture was not investigated as an environmental parameter in the seed viability tests.

The ambient incubator temperature used to assess seed lot purity and quality was chosen to reflect winter sowing conditions at both the York and Tammin trial site (see Fig. 3.4 & 3.7). The incubation cabinet was limited to a minimum setting of 8°C and therefore, it could not accurately represent the annual mean daily minimum temperatures experienced in winter at the York (5.7° C) and Tammin (6.2° C) field sites (Bureau of Meteorology 1998). Surface soil temperatures at the field sites were not available at the time of the trial. Surface soils could have a moderating effect on temperature, which would be slightly higher than the ambient temperature figures published by the Bureau of Meteorology (1998).

In order to simulate sowing conditions in the field, seed lots were subjected to fluorescent light each day for 11 hours alternating with 13 hours of darkness. The temperature regime of the incubation cabinet was also adjusted to simulate diurnal and nocturnal fluctuations in temperature for winter sowing times and assumed germination periods representative of the central Avon catchment. Alternating temperatures are expected to be more indicative of field conditions than a daily mean temperature (Mott & Groves 1981). The incubation cabinets were set at the following temperature settings and fluctuated $\pm 1^{\circ}$ C with a 10 minute transition time between alternating temperature regimes:

Cabinet 1: 24°C for 11 hours (day), 12°C for 13 hours (night). Cabinet 2: 19°C for 11 hours (day), 10°C for 13 hours (night). Cabinet 3: 14°C for 11 hours (day), 8°C. for 13 hours (night).

The experimental design consisted of three samples per seed lot for each temperature regime.

The plant species listed in Table 4.1 are representative of the range of species (canopy, shrub & ground cover species) found in undisturbed remnant bushland of the study region. These species are recommended in seed mixes sown on degraded land in the central Avon catchment (Lefroy *et al.* 1991; Scheltema 1993). Morphological descriptions, distribution details and phenology for these species are listed in Appendix 1.

Table 4.1:

The Nine Species Used to Assess Seed Purity Showing Seed Lot	Number,
Weight Sown per Petri Dish and the Supplier.	

5	eed lot	Species	ç∕dish	Supplier
42	4	Acacia microbotrya	1.0	Landcare Services
54	5	Acacia microbotrya	1.0	Landcare Services
90	0163	Acacia microbotrya	1.0	Alcoa of Australia
93	1873	Acacia saligna	1.0	Alcoa of Australia
19	9	Allocasuarina huegeliana	0.1	Landcare Services
52	0	Allocasuarina huegeliana	0.1	Landcare Services
94	0520	Allocasuarina huegeliana	0.1	Alcoa of Australia
14	74	Calothamnus qua drifidus	0.1	Landcare Services
14	90	Calothamnus quadrifidus	0.1	Landcare Services
93	0212	Calothamnus qua drifidus	0.1	Alcoa of Australia
94	0626	Casuarina obesa	0.1	Alcoa of Australia
94	0 5 9 7	Eucalyptus kondininensis	0.1	Alcoa of Australia
14	2 1 [°]	Eucalyptus wandoo	0.1	Landcare Services
14	22	Eucalyptus wandoo	0.1	Landcare Services
95	0053	Eucalyptus wandoo	0.1	Alcoa of Australia
64	2	Kennedia prostrata	10	Landcare Services
13	- 65	Kennedia prostrata	10	Landcare Services
96	0501	Kennedia prostrata	1.0	Alcoa of Australia
93	0122	Melaleuca cuticularis	0.1	Alcoa of Australia

4.3.2: THE EFFECT OF BIOLOCALITY AND TEMPERATURE ON SEED LOT QUALITY

The three plant species that were selected for the biolocality quality trials are listed in Table 4.2. The intention of the selection is to represent the large distributional range of these species within the southwest of Western Australia (Fig. 4.1 & see Appendix 1.1) and the subsequent availability of different seed biolocalities from various seed merchants.

Table 4.2:

Three Species Selected to Evaluate Biolocality Quality in Seed Germination, Showing Seed Lot Number, Seed Weight Sown per Petri Dish and Supplier.

Seed lot	Species	g/dish	Biolocality	Supplier
199	Allocasuarina huegeliana	0.1	Katanning	Landcare se √vices
520	Allocasuarina huegeliana	0.1	Bannister	Landcare services
2603	Allocasuarina huegeliana	0.1	Wickepin	Kimseed
7963	Allocasuarina huegeliana	0.1	Benjaberring	Kimseed
9317	Allocasuarina huegeliana	0.1	Jerramungup	Kimseed
940520	Allocasuarina huegeliana	0.1	Brookton	Alcoa of Australia
147 4	Calothamnus quadrifidus	0.1	Wanneroo	Landcare services
1490	Calothamnus quadrifidus	0.1	Wongan Hills	Landcare services
9784	Calothamnus quadrifidus	0.1	Esperance	Kimseed
9844	Calothamnus quadrifidus	0.1	Esperance	Kimseed
930212	Calothamnus quadrifidus	0.1	Wanneroo	Alcoa of Australia
940995	Calothamnus quadrifidus	0.1	Geraldton	Alcoa of Australia
1421	Eucalyptus wandoo	0.1	Mt. Barker	Landcare services
1422	Eucalyptus wandoo	0.1	Yornaning	Landcare services
4708	Eucalyptus wandoo	0.1	Mt. Barker	Kimseed
7589	Eucalyptus wandoo	0.1	Wickepin	Kimseed
7619	Eucalyptus wandoo	0.1	Brookton	Kimseed
7625	Eucalyptus wandoo	0.1	Moora	Kimseed
950053	Eucalyptus wandoo	0.1	Wongan Hills	Alcoa of Australia

Chapter 4



Figure 4.1: Locality map of the southwest Western Australia showing the biolocality regions where each seed lot was collected.

The principle assumption in this experiment is that the seed quality variation (percentage seed germination) within species collected from different biolocalities was a genetic response to environmental stimuli such as temperature (Table 4.3).

Table 4.3:

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Climate Data for Winter (June, July & August) Showing Daily Mean Temperature and Accumulated Monthly Rainfall for Each Seed Lot

	Biolocality.	
Biolocality	Temperature(°C) ^a	Rainfall(mm)
Bannister	11.1	641
Benjaberring	11.8	154
Brookton	10.8	241
Esperance	13.1	264
Geraldton	14.9	270
Jerramungup	10.6	145
Katanning	10.4	··· 218
Moora	12.5	245
Mt. Barker	11.0	296
Wanneroo	13.9	490
Wickepin	10.7	249
Wongan Hills	11.7	173
Yornaning	10.4	325

^a These figures were calculated by obtaining the mean between the maximum and minimum temperature over three months (source: Bureau of Meteorology 1998).

In order to determine the optimum seed germination within each species, a range of temperatures was trialed, $(12^{\circ}C, 15^{\circ}C, 18^{\circ}C \& 21^{\circ}C)$ which encompassed the upper and lower limits of optimum germination temperatures recorded for these species. Several studies indicate that the optimum germination is 20°C for *Allocasuarina huegeliana* (Fox *et al.* 1987), 20°C for *Calothamnus quadrifidus* (Turnbull & Doran 1987), 15°C - 20°C for *Eucalyptus wandoo* (Turnbull & Doran 1987). The incubators were set at a constant temperature, which fluctuated \pm 1°C. Fluctuation was measured daily using a maximum and minimum thermometer.

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Seed lots were exposed to approximately 10 minutes of fluorescent light every alternate day. The experimental design consisted of five samples per seed lot for each temperature regime.

The percentage germination results derived from the species listed in Table 4.2 were analysed for homogeneity of variance using an F_{max} test (Fowler & Cohen 1993). The data for *Allocasuarina huegeliana* and *Calothamnus quadrifidus* were not homogeneous due to the small number of samples. Non-parametric statistics were therefore employed using the Kruskal-Wallis Test (Fowler & Cohen 1993). Unfortunately, at the time of writing, there was no standard non-parametric test available to conduct inferential statistics on the interaction between two variables (2-way ANOVA equivalent). However, descriptive statistics were used to describe the differences in percentage seed germination between 6 biolocalities and 4 laboratory temperatures with respect to the mean daily winter temperature of the biolocality where the seed was collected.

The estimated effects for *Eucalyptus wandoo* were normally distributed (balanced) and so parametric statistics were employed using a two-way Analysis of Variance (ANOVA). The proportional data were 'normalised' using arcsine transformation prior to statistical analysis (Fowler & Cohen 1993).

4.3.3: THE EFFECT OF FROST ON SEED LOT QUALITY

The experimental design of subjecting *Eucalyptus wandoo* seeds to sub-zero temperatures (Table 4.4) was used here as a preliminary study for further research into the effects of simulated frost on a range of species. *E. wandoo* was selected as a possible indicator species for the effects of extreme temperature on germination as it has a large geographical range which occurs across diverse climate zones including those experienced in the Avon catchment where seasonal frosts are common (see Table 4.5). Furthermore, *E. wandoo* is found in frost susceptible areas (eg. Yornaning) of the lower slopes and inland river systems of the Avon catchment (Bell & Bellairs 1992).

Table 4.4:

Spo Numbo	ecies Subjected to Su er, Seed Weight Sow	ib-Zero T <u>n per Pe</u> t	emperatures tri Dish, Biolo	Showing Seed Lot ocality and Supplier.
Seed lot	Species	g/dish	Biolocality	Supplier
1422	Eucalyptus wandoo	0.1	Yornaning	Landcare services

The seed was collected from the biolocality of Yornaning, some 100 km southsouthwest of the York trial site. The biolocality experiences sub-zero temperatures (Bureau of Meteorology 1998) in the sowing season (Table 4.5). It is presumed that Yornaning is representative of areas which are susceptible to frost in the Avon catchment.

Table 4.5:

Mean Number of Days below Zero Degrees Temperature for the Biolocality of Yornaning (1887-1997).

Spring Months	Days < 0°C	Summer Months	Days < 0℃	Autumn Months	Days < 0°C	Winter Months	Days < 0°C
September	3	December	0	March	0	June	3
October	1	January	0	April	0	July	4
November	0	February	0	Мау	2	August	5

Note. Source Bureau of Meteorology (1998)

Petri dishes containing *Eucalyptus wandoo* (1422) seed were exposed to approximately 10 minutes of fluorescent light every alternate day. The petri dishes were placed in a refrigerated incubator set at 15° C. Five different seed batches of the same seed lot (3 samples per seed batch) were subjected to artificial frosts using a freezer set at -10° C at five different stages during the germination trial. The simulated frost times occurred at 1, 5, 9, 11 and 15 day stage of the trial. Once a seed batch had been exposed to -10° C for 24 hours, it was returned to 15° C for the duration of the experiment.

The control seed batch (3 replicates) was maintained at a constant temperature setting of 15°C.

The seed germination results derived from the species listed in Table 4.4 were not homogeneous and therefore non-parametric statistics were employed using the Kruskal-Wallis Test (Fowler & Cohen 1993).

4.4: Results

4.4.1: SEED LOT PURITY

The number of seeds that germinated per gram varied between each seed lot (Table 4.6). Seed lots of *Eucalyptus kondininensis* (940597), *Calothamnus quadrifidus* (1474) and *Eucalyptus wandoo* (950053) had the highest number of germinants per gram with 2405, 1166 and 1007, respectively. Poor germination occurred in Leguminosae seed lots of *Acacia microbotrya*, *Acacia saligna* and *Kennedia prostrata*.

Table 4.6:

Mean Number and Percentage Seed Germination per gram for Seed Lots of Nine Species, Showing Standard Error Subjected to an Alternating

	lemperatur	<u>e Regime.</u>	% ±S.E. 64 8.2 22 4.6 57 4.5 4 1.4 73 2 75 1.5 75 1.9 98 0.8 97 1.2 92 1.5 45 3.1 99 0.2 96 0.6		
Seed lot	Species	Germinants/g	%	±\$.E.ª	
424	Acacia microbotrya	11	64	8.2	
545	Acacia microbotrya	4	22	4.6	
900163	Acacia microbotrya	3	57	4.5	
931873	Acacia saligna	2	4	1.4	
199	Allocasuarina huegeliana	384	73	2	
520	Allocasuarina huegeliana	405	75	1.5	
940520	Allocasuarina huegeliana	210	75	1.9	
1474	Calothamnus quadrifidus	1166	98	0.8	
1490	Calothamnus quadrifidus	685	97	1.2	
930212	Calothamnus quadrifidus	400	92	1.5	
940626	Casuarina obesa	294	45	3.1	
940597	Eucalyptus kondininensis	2405	99	0.2	
1421	Eucalyptus wandoo	270	96	0.6	
1422	Eucalyptus wandoo	122	87	1.6	
950053	Eucalyptus wandoo	1007	99	0.4	
642	Kennedia prostrata	8	14	2.4	
1365	Kennedia prostrata	1	2	0.6	
960501	Kennedia prostrata	2	41	4.5	
930122	Melaleuca cuticularis	1445	78	2.1	

^a The standard error is calculated from percentage germination.

There was wide variation between species in the overall percentage of seeds that germinated (Table 4.6). Several species for example, *C. quadrifidus* (Av. 95.6%), *E. kondininensis* (99%) and *E. wandoo* (Av. 94%) had a high percentage germination, whereas *A. microbotrya* (Av. 47.6%), *A saligna* (4%), *Casuarina obesa* (45%) and *K. prostrata* (19%) had a low percentage of seeds that germinated.

4.4.2: THE EFFECT OF BIOLOCALITY AND TEMPERATURE ON SEED LOT QUALITY

There was variation in the seed lot quality within each species (Table 4.7 & see also Table 4.6). *Calothamnus quadrifidus* and *Eucalyptus wandoo* recorded a mean germination over ninety percent whereas *Allocasuarina huegeliana* had an average of 71.2 percent for four different temperatures (Table 4.7). These temperatures had very little influence on percentage germination for the majority of seed lots within each species.

Table 4.7:

Mean Percentage Seed Germination for Seed lots of Three Species Subjected to Four Different Temperatures, Showing the Mean and Standard Error.

							·
Seed Lot	Species	12°C	15°C	18°C	21°C	x	±S.E.
199	A. huegeliana	70	75	71	70	72	1.2
520	A. huegeliana	73	76	73	71	73	1.1
2603	A. huegeliana	75	78	83	71	77	1.7
7963	A. huegeliana	49	56	55	44	51	. 1.7
9317	A. huegeliana	82	80	75	77	79	1.6
940520	A. huegeliana	67	81	79	72	75	1.9
1474	C. quadrifidus	99	9 9	98	85	99	1.6
1490	C. quadrifidus	96	98	98	92	98	1.0
9784	C. quadrifidus	100	99	99	65	91	3.7
9844	C. quadrifidus	98	99	97	90	96	0.9
930212	C. quadrifidus	96	96	94	82	92	1.5
940995	C. quadrifidus	99	98	96	93	97	0.6
1421	E. wandco	96	99	99	99	98	0.6
1422	E. wandoo	87	90	95	94	92	1.6
4708	E. wandoo	100	99	100	99	100	0.3
7589	E. wandoo	90	100	100	97	97	1.0
7619	E. wandoo	87	93	99	93	93	1.5
7625	E. wandoo	99	99	99	98	99	1.4
950053	E. wandoo	99	99	99	97	99	0.4

The seed germination results derived from *A. huegeliana* and *C. quadrifidus* listed in Table 4.7 were not homogeneous and therefore non-parametric statistics were employed using the Kruskal-Wallis Test (Fowler & Cohen 1993). However, seed germination results of *E. wandoo* were viewed in isolation as the data was homogeneous and therefore parametric statistics were employed using a Two-Way ANOVA (Fowler & Cohen 1993).

The non-parametric analysis of the variation in percentage seed germination between 6 seed lots within each species was statistically significant for *A*. *huegeliana* (p < 0.0001) and *C. quadrifidus* (p = 0.026) with the probability set at p < 0.05 level of significance (Table 4.8).

The non-parametric analysis of variation in percentage seed germination within different temperatures and within different seed lots for each species was statistically significant for *C. quadrifidus* (p < 0.0001) but not for *A. huegeliana* (p = 0.06) with p < 0.05 level of significance (Table 4.8).

Table 4.8:

Non-Parametric Analysis of Variation in Percentage Seed Germination within 6 Biolocalities and Within 4 Different Temperatures, Using a Kruskal-Wallis Test Showing Species, K value, Degrees of Freedom and Probability.

Species	Variable	к	d.f.	pª
- <u></u>	······	6) - 6 <u>1</u>		
A. huegeliana	Biolocality	55.80	5	0.0001
A. huegeliana	Temperature	7.27	3	0.0600
C. quadrifidus	Biolocality	12.70	5	0.0260
C. quadrifidus	Temperature	61.37	3	0.0001

^a Significance is set at p < 0.05.

The percentage of seeds that germinated for *A. huegeliana* may have been influenced by the interaction between the two environmental variables of temperature and biolocality (Fig. 4.2). The descriptions of these interactions are numerous and repetitive and so only general trends will be mentioned here. The trend lines in Figure 4.2, between seed lot 199 and 9317, diverge with a positive interaction between the upper and lower temperature limits (12°C & 21°C). The extension of the trend lines between seed lots 9317 and 520 both diverge and converge with no parallel lines between germination temperature indicating an interaction between the variables (biolocality / temperature). The continuance of the trend lines between seed lots 520 and 7963 showed minimal interaction between the germination temperatures, with the lines maintaining a similar parallel distance.

The percentage of seeds that germinated for *C. quadrifidus* is unlikely to be been influenced by the interaction between the two environmental variables of temperature and biolocality (Fig. 4.3). The trend lines between the six different seed lots showed minimal interaction between three germination temperatures $(12^{\circ}C, 15^{\circ}C \& 18^{\circ}C)$. These temperature lines maintained a similar trend with a parallel distance, which oscillated within a narrow germination band between 95% and 100%. However, the trend line in Figure 4.3, depicting the germination temperature of 21°C had a distinctive divergence in percentage seed germination with a positive interaction for seed lot 9784 (collected from Esperance) relative to the other temperatures and biolocalities.







Figure 4.3: Mean percentage seed germination showing an improbable interaction in 6 biolocalities of *Calothamnus quadrifidus* (ranked according to increasing mean winter provenance temperatures) between four different temperature regimes.

The variation in percentage seed germination between seven biolocalities (p < 0.00001) and four different temperatures (p = 0.00002) within *E. wandoo* was statistically significant with the probability set at p < 0.05 level of significance (Table 4.9).

Table 4.9:

Parametric Analysis of Variation in Seed Germination of *E. wandoo* Between 7 Biolocalities and 4 Different Temperatures Using a Two-Way ANOVA Showing Variables, Sum of Squares, *F* value, Degrees of Freedom and Probability.

Variables	∑(x-x)	F	d.f.	pª
Biolocality	2745	13.49	6	0.0000
Temperature	934	9.18	3	0.00002
Prov. : Temp.	1721	2.82	18	0.00046
Residuals	3798		112	
	· · · ·			

^a Significance is set at p < 0.05.

The percentage of seeds that germinated for *E. wandoo* were significantly influenced by the interaction (p = 0.00046) in Table 4.9, between the environmental variables of temperature and geographical variable of biolocality. This is also expressed in Figure 4.4. The trend lines in Figure 4.4, between seed lot 1422 and 7589 maintained a distinct difference in percentage seed germination with minimal interaction except for the germination variable of 21°C. However, the extension of the trend lines between seed lots 7589 and 1421 converge with a negative interaction between all four germination temperatures. Furthermore, the continuation of the trend lines between 1421 and 7625 both diverge and converge indicating a positive and a negative interaction between temperature within a narrow germination band of some 4%.



Figure 4.4: Mean percentage seed germination showing a significant interaction (p = 0.0005) in 6 biolocalities of *Eucalyptus wandoo* (ranked according to increasing mean winter provenance temperatures) between four different temperature regimes.

4.4.3: THE EFFECT OF FROST ON SEED LOT QUALITY

Simulated frost at -10°C for 24 hours did not affect the total germination percentage of *Eucalyptus wandoo* when compared to the control set, which germinated at a constant 15°C (K = 0.015 at 1 *d.f.* p > 0.9). The mean percentage germination for seed batches subjected to -10°C varied approximately 10 percent either side of the control (Table 4.10).

Table 4.10:

Mean Percentage Seed Germination for *E. wandoo* After Being Subjected to -10°C for 24 Hours at 1, 4, 9, 11 and 15 day intervals, Relative to the Control, Showing Standard Error

	_	
- 1	100	0
5	89	33.1
9	89	33.1
11	87	3.1
- 15	79	6.4
Control	90	5.3
· .		

Day (-10°C) Mean % \pm S.E.

The rate of germination for seed batches subjected to sub zero temperatures was delayed (Fig. 4.5 & 4.6). Frost simulation arrested seed germination for approximately three days (72 hours), including the 24 hours spent in the freezer. The germination rate curves in Figure 4.5 and 4.6 experience a 'plateau' effect (indicated by the letter D in Figure 4.5 & 4.6) when compared to the relatively continuous curve of the control. The expression of the germination rate of seeds subjected to frost treatment and the control has been divided into two figures (4.5 & 4.6) to improve clarity.

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Figure 4.5: Seed germination of *Eucalyptus wandoo* subjected to simulated frost -10°C for 24 hours at 1, 5 and 9 day intervals which corresponds approximately with a 72 hour delay (D) in germination.



Figure 4.6: Seed germination of *Eucalyptus wandoo* subjected to simulated frost -10°C for 24 hours at 11 and 15 day intervals which corresponds approximately with a 72 hour delay (D) in germination.
4.5: Discussion

4.5.1: SEED LOT PURITY

The germination capacity of seed lots is one factor that cannot be assumed to be constant for individual species. This is supported in other trials of the study species, which show considerable variation in germination capacity within species and between species (Table 4.11). The methods used by other authors for the seed germination tests in Table 4.11 are similar to those employed in this Chapter.

Table 4.11:

Seed Germination showing Seed Lot Purity per Gram and Quality Percentage for the Same Species Trialed in Chapter 3 but Conducted by Different Authors in Other Experiments.

	Species	Purity (g)	Quality (%)	
	Acacia microbotrya	26 ^g	66 °	
	Acacia saligna	30 ^c	54 ^b , 34 ^b	
	Allocasuarina huegeliana	555 ^g	25 ^f 68 ^d	
	Calothamnus quadrifidus	1100 °, 740 °	23 ^d	
	Casuarina obesa	250 °	unavailable	
	Eucalyptus kondininensis	530 °, 313 °	83 ^d	
	Eucalyptus wandoo	303 °	91 ^d	
	Kennedia prostrata	20 °	51 [°]	
	Melaleuca cuticularis	2500 ^g	unavailable	
a. Bell	l <i>et al.</i> (1993)	e. Turnbu	II & Doran (1987)	
b. Cav	anagh (1981)	f. Turnbul	II & Martensz (1982)	
c. Dalton (1993)		g. C.A.L.M. (1996/97)		

d. Enright & Lamont (1989)

In this study, the number of seeds that germinated per gram was highly variable (see Table 4.6) between species and in some cases within species. This is predominantly due to the difference in the physical size and purity of the seed lots. Factors contributing to the variation in the quality of the seed lot such as the proportion of viable seed to non-viable, dormant and dead seed will be discussed later in Seed Lot Quality (4.5.2).

An obvious reason for variation in germination (per gram) between different species is the morphological disparity in the size and shape of the seeds. In particular, the specific gravity of pure seeds will have an effect on the ratio of viable seed relative to the weight of the seed lot. For example, the average number of seeds in an *Acacia microbotrya* seed lot is 26 per gram compared to 300 ± 100 (large error) for *Eucalyptus wandoo* (C.A.L.M. 1996/1997). This questions the relevance of using seed purity as a comparative measure across genera and species. However, the difference in the number of germinants between seed lots per gram within each species will most likely be a factor of seed purity.

The purity of each seed lot is also dependent on seed handling following harvest. Seed handling involves the separation of seed from the reproductive structures borne on the parent plant. The purity of each seed lot depends on the ease with which viable seed can be separated from chaff. Difficulty arises when viable seed resembles chaff and or when the seed is difficult to detect with the naked eye (for example, *Calothamnus quadrifidus* seed). Seed impurity in these species is tolerated due to the high cost involved in purification (Kabay & Lewis 1987).

The purity of each seed lot is dependent on the apparatus employed to separate viable seed from chaff and this varies according to each seed merchant. The ability to clean seed is limited, particularly when the seed is small. Therefore, the proportion of viable seeds to chaff will be variable between different seed lots. This variable purity can have an influence on the selection of a specific seed lot used and the source of the seed when establishing plants on a broad scale using the direct seeding method.

4.5.2: SEED LOT QUALITY

The factors affecting seed quality, such as the proportion of viable seeds to nonviable, dormant and dead seeds, are numerous and difficult to ascertain. However, variables which may have contributed to poor seed quality are seed processing (collection, storage and treatment), genetic (inbreeding depression) and environmental (climatic) on the parent plant. These contributing factors (with the exception of seed dormancy treatment) mostly occurred prior to the purchase of the seed and therefore, were beyond the control of the experimental design. A primary consideration of seed collection is to harvest quality seed. However, poor evaluation of seed maturity and viability within the reproductive structures of the parent plant, will decrease the quality of the seed lot (Kabay & Lewis 1987). This is particularly evident when seed collecting in the southwest of Western Australia where agricultural practices have caused a decrease in the number of viable seeds (Bell & Koch 1980; Bell & Loneragan 1985).

Agricultural development in the central Avon catchment has led to the widespread clearing of native woodland ecosystems (Saunders *et al.* 1985). This practice has caused the fragmentation of native woodlands into isolated networks of remnant vegetation. Seed collected from isolated plants and individuals surrounded by an agricultural ecosystem are likely to be self-pollinated. Information on the size of the remnants from which the seed lots were collected is not available. Isolated plants are susceptible to a genetic inbreeding depression causing low seed viability (Harwood 1990). This is further exacerbated by a decline in the number and diversity of pollinating invertebrate species (Bell *et al.* 1993).

The management of agricultural crops and pasture species is predominantly reliant on the use of chemical insecticides. These chemicals usually do not discriminate between native plant pollinators and perceived agricultural pests, partly because they can be one and the same. Consequently, the application of insecticides causes a reduction in the number of insects available to carry out cross-pollination between different plant populations (Bell & Koch 1980; Bell & Loneragan 1985; Bell *et al.* 1993). Insecticide use, coupled with the destruction or modification of invertebrate habitat (clearing, weed invasion, fire regime etc.) could further reduce genetic diversity and the subsequent production of non-viable seed between native plant communities.

The production of non-viable seed has also been attributed to environmental factors such as seasonal drought, insect predation and poor nutrient availability (Bell *et al.* 1993) which are intrinsic to specific biolocalities. These adverse and seasonally infrequent conditions often restrict the proportion of viable seeds produced in that year. Reproductive events require a large proportion of energy relative to respiration and photosynthesis. Consequently, under environmental stress, reproductive structures (ie. flowers) may not be borne on the parent plant or the fruit may be aborted in order to conserve and redirect energy reserves.

Adverse environmental conditions have influenced the germination response of many species within the Leguminosae group which are dependent on their seed coat imposed dormancy mechanisms. The high degree of variability in seed germination per gram for seed lot species such as *Acacia microbotrya, Acacia saligna* and *Kennedia prostrata* may be associated with the seed treatment employed. The pre-sowing treatment of boiling water and scarification used in this experiment, may have had varying degrees of success in breaking the seed coat imposed dormancy. Clemens *et al.* (1977) states that heat treatment using boiling water can sometimes give erratic results and has been difficult to standardise for various species.

The mechanical hardness of the Leguminosae seed coat has led to an historical practice of hot or boiling water treatment. Many experiments have recorded increased germination percentages following such treatment (eg. Myres 1936; Jones 1963; Farrell & Ashton 1978; Glossop 1980). Bell *et al.* (1993) tested 58 Western Australian legume species and only nine species showed a marked decrease in germination following boiling water treatment. *Kennedia prostrata* was one of the nine species that did not respond to the heat treatment and hence was not heat treated in this experiment. Cavanagh (1980) states that scarification of seed gives the most reliable results but scarification of *K. prostrata* (960501) still recorded low germination. Furthermore, *K. prostrata* (# 642 & 1365) without seed treatment also experienced low germination. The poor germination indicates that either the methods used to break the seed coat imposed dormancy may not have stimulated optimum germination or it has a naturally low viability across its distribution.

A large proportion of the ungerminated seed for *A. microbotrya*, *A. saligna* and *K. prostrata* had imbibed, indicating water permeability through the seed coat. This suggests that the seed coat imposed dormancy may have been broken but the seed still failed to germinate. If the duration of the experiment was extended then a greater number of seeds may have germinated. This situation illustrates a limitation in conducting a seed germination test when compared to other methods that identify viable embryos prior to germinate. The time constraint imposed on both dormant and imbibed seeds to germinate in the laboratory may not be an important factor in the field.

The imbibed seed of both *Acacia* and *Kennedia* were susceptible to fungal attack in the trial and were treated with a pre-emergent fungicide. The employment of hot water treatment for the leguminous species exposed the seeds to fungal colonisation due to contamination of a sanitised medium. Cavanagh (1980) states that fungal colonies can inhibit germination of viable seed.

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Seed viability does decrease over time when held in storage (Harwood 1990). However, storage of seed from most woody perennial species has little effect on the seed lot quality over time provided that certain conditions are maintained (Boland *et al.* 1980). The optimum storage environment for seed is dry, cool, sterile, dark and decreased oxygen levels. For example, the majority of eucalypt seed can remain viable for up to 20 years if stored at $3-5^{\circ}$ C in an air sealed container (Boland *et al.* 1980). It is unlikely that the seed lots donated and purchased for the germination trials in this thesis declined in seed viability due to storage time. This is because Western Australia is a primary resource state which has a high turnover of the stored seed bank held by merchants, predominantly to fulfil contracts for mine-site revegetation (Kabay & Lewis 1987).

4.5.3: THE EFFECT OF BIOLOCALITY AND TEMPERATURE ON SEED LOT QUALITY

Goals for the effective revegetation in the central Avon Valley requires that the optimum germination of a seed lot would be achieved through the selection of a seed biolocality conducive to seedling development at the site of revegetation. Percentage seed germination was significantly different between biolocalities of the same species for *Allocasuarina huegeliana, Calothamnus quadrifidus* (see Table 4.8) and *Eucalyptus wandoo* (see Table 4.9). The explanation for the variation between seed lots can only be hypothesised. However, the differences are likely to be an inherent ecotypic response to biolocality specific environmental parameters such as temperature.

The temperature variable in the laboratory $(12^{\circ}C, 15^{\circ}C, 18^{\circ}C \text{ and } 21^{\circ}C)$ had a significant influence on the germination capacity of seed lots of *C. quadrifidus* (see Table 4.8) and *E. wandoo* (see Table 4.9) but not of *A. huegeliana* (see Table 4.8). However, *A. huegeliana* recorded a difference in the percentage of seeds that germinated between four temperatures but was not statistically different. These findings do not necessarily detract from the known importance of temperature in influencing germination but rather indicates that an increase in the number of samples within *A. huegeliana* may have an influence on the significance of the result.

Percentage seed germination in *A. huegeliana* appeared to have been influenced by interactions between the germination temperatures and seed lots collected from biolocalities with a relatively moderate daily mean winter temperature (ie. # 9317: Jerramungup, # 2603: Wickepin, # 940520: Brookton). This is because the variation between seed lots may be due to their large geographical range (Fig. 4.1) and associated climatic influence on the seeds' physiology in the biolocality.

These biolocalities were more sensitive to the range of laboratory temperatures used in the trial, with relatively large fluctuations in the percentage of seeds that germinated. For example, *A. huegeliana* (2603) recorded a 12% difference in the percentage of seeds that germinated due to the interaction between the laboratory temperatures (21°C & 18°C). This interaction suggests that *A. huegeliana* has an optimum germination temperature of 18°C. These results are difficult to establish definitively, given the minimal data available on climatic statistics for seed biolocalities.

The climatic statistics (see Table 4.3) were recorded from town-site locations obtained from the Bureau of Meteorology and may not have accurately represented the ground temperature in the field where the seed was actually collected. An investigation into the relationship between the ground temperature, water availability in the field and the optimum germination temperature would provide a greater understanding of the seed ecology of direct seeded species used in revegetation.

Percentage seed germination in *C. quadrifidus* appeared not to have been influenced by interactions between germination temperatures and seed lots collected from different biolocalities. A possible explanation is that the variation between seed lots may be minimal, due their similar geographic range (Fig. 4.1) and associated temperature regimes (Table 4.3). The seed lots 9844 and 9784 were both collected from the biolocality of Esperance. Similarly, seed lots 1474 and 930212 were also collected from the same biolocality of Wanneroo. This result does not detract from the potential influence on seed germination from the interaction between biolocality and temperature, but suggests further investigation with increased biolocalities and subsequent temperature treatments.

The percentage of seeds that germinated between each biolocality of *C. quadrifidus* decreased due to the influence of the interaction between the high laboratory temperature of 21°C relative to the other variables. This temperature would be typical of the spring/summer season experienced in the biolocality where the seed was collected, and may trigger a response in the seed to conditions that are not conducive to germinant development in the field. These conditions include desiccation and moisture stress due to high temperatures that could cause a dormancy response in the seed and subsequent lower percentage germination. Therefore, the seeds within a particular biolocality may display an inherent ecotypic dormancy response to temperature conditions. These survival mechanisms are inherent in seeds displaying a recalcitrant response to germination during adverse conditions when temperature conditions are high.

For example, seed germination in *C. quadrifidus* (9784) was 34% lower at 21°C relative to the other temperatures. This clearly demonstrates a response (low germination) to high temperatures from a seed lot with an optimum germination temperature of 12°C. *C. quadrifidus* obviously prefers cooler temperatures for germination and would probably germinate in winter rather than in spring.

Percentage seed germination in *E. wandoo* was significantly influenced by interactions between the germination temperatures and seed lots collected from different biolocalities. In other words, biolocalities respond differently to ambient temperature. A possible explanation for variation in percentage seed germination has already been hypothesised for *A. huegeliana* and may also be relevant for *E. wandoo* seeds. Biolocalities with a relatively lower daily mean winter temperature (# 1422: Yornaning, # 7619: Brookton, # 7589: Wickepin) were more sensitive to the range of laboratory temperatures trialed. These biolocalities had a relatively large difference in the percentage of seeds that germinated compared to the narrow germination band of the remaining seed lots. For example, seed lot number 7619 recorded an 12% difference in the percentage of seeds that germinated (12°C & 18°C) relative to other variables.

The number of seeds that germinated at 12° C in *E. wandoo* was reduced in seed lots collected from cool climates. This temperature may be indicative of winter conditions experienced in biolocalities of seed lots that recorded low seed germination. These conditions may trigger an inherent dormancy response in the seed when temperatures are detrimental to germinant development in the field such as frost. Therefore, the direct seeding of *E. wandoo* collected from a cool climate may increase seed germination when sown in spring due to an increase in soil temperature conditions.

Several authors have alluded to the causal link between the seed germination temperature and the biolocality temperature. In a review of Western Australian species collected from a Mediterranean type climate (Bell *et al.* 1993) determined that optimum germination temperatures corresponded with field temperatures during periods of continuous moisture availability. Bell and Bellairs (1992) found that the optimum temperature for germination of species from the jarrah *(Eucalyptus marginata)* forest was 10 to 15°C where winter temperatures are relatively low. In another study, Bellairs and Bell (1990) found native species of the northern kwongan region (Eneabba), where winter temperatures were relatively high, had an optimum germination of 15 to 20°C. This study showed a similar causal link between the optimum germination temperature and the field temperature of the biolocality where the seed was collected.

4.5.4: THE EFFECT OF FROST ON SEED LOT QUALITY

Seed batches of *Eucalyptus wandoo* (1422) exposed to frost simulation at -10°C in the freezer were not statistically different in percentage germination relative to seed batches that were maintained at a constant 15°C. However, the number of seeds that germinated was dependent on the day in which seed batches were subjected to sub-zero temperatures (see Table 4.10). For example, 100% seed germination occurred when seed batches were subjected to frost treatment on day one compared to 79% germination on day 15.

The trend indicates that sub-zero temperatures may damage the embryo of the seed after the seed has imbibed water. The turgor pressure acting on the internal cell walls may have damaged the embryonic membrane when water molecules expand through freezing (Stoneman 1993). The potential damage to the seed embryo may have contributed to seed mortality by reducing germination.

Abnormal germinants may have been prematurely diagnosed as viable seed upon germination. The term viability can only be confidently determined when the cotyledons are allowed to sufficiently develop from the seed producing a 'normal' germinant (Boland *et al.* 1980). As stated in the section on methods, germinants were recorded after the emergence of a 2-5 mm long radical prior to the emergence of an apical meristem with cotyledons. Premature seed removal may have caused the inclusion of abnormal germinants or non-viable seed in the results (see Table 4.10).

Seed germination in *E. wandoo* is relatively constant (control) for seed exposed to the same temperature. However, seed batches exposed to simulated frost conditions appear to have undergone a secondary dormancy response, resulting in a 'depression' in the germination. The sub-zero temperatures seem to have resulted in a germination 'lag' of some 72 hours or more before warmer temperatures of 15°C broke the frost imposed dormancy response in the seed. Therefore, it appears that *E. wandoo* (1422) is an opportunistic germinator, taking advantage of moisture availability in conjunction with warmer ambient temperatures. Several studies have indicated that seed germination is prevented by extreme high and low temperatures, but for many species, seed germination occurs when incubation temperatures are favourable for the subsequent survival of the seedling (Young *et al.* 1973; Lodge 1981).

Bell and Bellairs (1992) observed that the optimum temperature of 15-20°C for germination of *E. wandoo* was representative of the environmental temperature associated with winter rainfall where the species occurs. *E. wandoo* is found on slopes with shallow, sandy, eroded soils (Boland *et al.* 1980) in the 350-400 mm isohyet where winter precipitation and soil moisture regimes are closely linked to the germination and survival of seedlings.

The seed ecology of some species is adapted to low temperature storage or cold stratification in order to delay germination until late winter or early in spring when frost days are greatly reduced (Baskin & Baskin 1987; Meyer *et al.* 1990). Germination of *Eucalyptus* species in the temperate region of southeastern Australia can be enhanced by cold stratification (Boden 1957; Pryor 1954; Bachelard 1967; Beardsell & Mullett 1984).

The maximum germination recorded (see Table 4.10) was 100 % for the seed batch that was subjected to sub-zero temperatures prior to imbibing water at the start of the experiment. The seed dormancy of *E. wandoo* may be broken by initial cold stratification that primes the seed for germination. The biolocality of Yornaning is susceptible to frosts and therefore cold stratification may enhance germination. This has yet to be definitively established.

The Avon Basin in southwestern Australia experiences a Mediterranean type climate in the west tending semi-arid towards the east. Bell *et al.* (1993) suggests that it is unlikely that cold stratification is used as a dormancy mechanism for native plant species in this region. Delayed germination may reduce the period for active root development, crucial for the survival of plants through the summer months. Bell *et al.* (1993) gives a more likely explanation for the ecological advantage of seed dormancy for Western Australian habitats broken by storage in hot, dry conditions rather than moist, cold stratification. Research into the effects of various storage regimes on seed germination is seriously lacking in the knowledge of germination ecology of Western Australian species.

It is important to note that some seed batches display a delayed effect in germination, irrespective of the frost variable (see Fig. 4.3 & 4.4). In order to substantiate that frost causes secondary dormancy in *E. wandoo* (1422) further samples need to be trialed over a greater number of biolocalities and short successive frosts as would be experienced during winter conditions in the field.

4.6: Summary

A seed germination test enables an effective measure of the potential of a revegetation programme through the analysis of seed lot purity and quality. The evaluation of seed lot purity enables the calculation of the sowing rate (viable seed/kg) required to produce an estimated number of plants per hectare. This is valuable in that it minimises unnecessary time and costs associated with direct seedling establishment. The analysis of seed lot quality enables the evaluation of seed treatments and biolocality selection based on environmental parameters prior to sowing in the field. The evaluation of seed treatment methods, with regard to specific plant families such as Leguminosae, determines whether the optimum technique has been employed. Furthermore, biolocality selection can provide the primary basis for seedling establishment in the field through the assessment of optimum seed germination using temperature as an environmental indicator.

The number of seeds that germinate per gram was highly variable between species and in some case within species. Germination per gram was highly dependent on the variation in the morphological size of seed and the ratio of viable seed to chaff between different seed lots. These variables are especially relevant for the landholder undertaking a large-scale direct seeding programme using a mixture of seed lots, particularly species with small seeds. The variation in these small seeded species was attributed to handling which is dependent on the seed merchant's ability to separate viable seed from chaff.

The variation in seed lot quality may be attributed to the lack of quality assurance associated with the seed purchased from registered merchants. This translates into unpredictable seed germination when sown in the field. Variable germination rates have caused past scepticism in the agricultural community towards direct seeding in preference for nursery seedlings.

Seed lot quality varied in terms of the percentage of seeds that germinated within species and between biolocalities. Percentage seed germination varied in a statistically significant manner for *Allocasuarina huegeliana*, *Calothamnus quadrifidus* and *Eucalyptus wandoo*. This variation is attributed to an inherent ecotypic response to environmental parameters such as temperature.

The environmental variable of temperature under investigation had a significant influence on the germinative capacity of seed lots in *C. quadrifidus* and *E. wandoo* but not for *A. huegeliana*. This variation within and between species may be due to an inherent ecotypic response to the laboratory temperatures, which are indicative of temperature regimes associated with the geographical range of the biolocalities where each seed lot was collected. Unfortunately, this could not be definitively established using inferential statistics because very little climatic information is available on the biolocality where the seed was collected. However, determining the influence of seed germination on the interaction between these two environmental variables could further substantiate the relationship between the biolocality and temperature.

Percentage seed germination in *E. wandoo* was significantly influenced by the interactions between the laboratory germination temperatures and seed lots collected from different biolocalities. This indicates a variation in the optimum germination temperatures between biolocalities. The implication of this finding when direct seeding in the field is that if the mean winter temperature of the revegetation site is equivalent to the optimum germination temperature for a particular seed lot, then climatic matching of that biolocality with the revegetation site may enhance germination and subsequent effective plant establishment.

The optimum temperature for seed germination between seed lots of *E. wandoo* was variable due to the influence of the interaction between the laboratory temperatures $(13^{\circ}C \& 12^{\circ}C)$ with biolocalities that have a relatively low daily mean winter temperature. These temperatures may be indicative of seasonal variations in the biolocalities where the seed was collected that triggers a response to conditions such as high soil temperatures favouring germinant development or sub-zero temperatures causing frost damage.

Therefore, the seeds within a particular biolocality may display an inherent ecotypic response to temperature conditions, which ensures the perpetuation of the species. These survival mechanisms are inherent in seeds displaying both a primed response to conditions conducive to germinant survival and a recalcitrant response to germination during adverse (sub-zero^oC) conditions.

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Sub-zero temperatures did not effect the percentage germination of *E. wandoo* (1422) in a statistically significant manner. However, seed germination did decrease under the influence of simulated frost, the more moisture that was imbibed into the seed over time. This trend was attributed to higher seed mortality due to crystallisation of water molecules through freezing which damaged the seed embryo. However, cold stratification such may enhance germination in seed lots collected from biolocalities prone to frost prior to germination in the field.

Seed germination in *E. wandoo* was relatively constant (control) for seed exposed to the same temperature but seed batches exposed to simulated frost conditions underwent a secondary dormancy response in the seed. The sub-zero temperatures resulted in a delay in germination until warmer ambient temperatures broke the frost imposed dormancy response in seed. Therefore, it appears that *E. wandoo* seed is an opportunistic germinator, taking advantage of conditions conducive to germination as they arise.

EVALUATION OF REVEGETATION METHODS: DIRECT SEEDING VERSUS NURSERY SEEDLINGS

5.1: Introduction

There are three common methods for establishing native vegetation: natural recruitment, direct seeding and the transplanting of nursery seedlings. Effective revegetation depends on choosing the method that is appropriate for the species, site, situation and conditions (Table 5.1).

Method	Revegetation situation	Rationale	
Natural	Adjacent to remnant vegetation	Proximity for seed dispersal Existing seed soil bank	
Direct seeding	Broad hectare application Road side verge Soil stabilisation Wind break Water-table draw-down (salinity)	Economics (less expensive) Natural aesthetics High density planting Multi-tiered canopy Unconfined taproot	
Nursery seedling	Timber production Rare plants (endangered) Genetic control Non-viable seeding plants	Uniform density; arrangement Economics (seed expensive) Limited availability of seed Asexual propagation required	

Table	5.1: Three	Revegetation	Methods	and the	Rationale	in Selecting	the	
		Δι	nnronriat	e Site S	ituation	· .		

Farming communities have traditionally adopted nursery seedling establishment in preference to natural recruitment or sowing of native plant seed (Fig. 5.1).

This has been in part due to the perceived reliability of nursery seedlings (Dalton 1994), the failure of early trials of direct seeding methods (Venning 1991) and the lack of scientifically based information on the methods of revegetation (Dalton 1992). During the late 1980s extensive trials with direct seeding resulted in a greater understanding of successful establishment methods (Venning 1991). Consequently, the demand for native seed has substantially increased with the recognition of the land revegetation merits of many previously unexploited species (Peterson 1987).

The choice of revegetation method to use is often based on value judgements, rather than being empirically derived. Such choices should be based on the establishment reliability, growth rate and relative cost of seedlings. The relative effectiveness of direct seeding versus nursery seedlings has not been determined as a guide to establishing native vegetation in agricultural regions.

5.1.1: PLANT ESTABLISHMENT IN THE FIELD

An important aspect of plant establishment by direct seeding is the timing of germination in relation to the onset of favourable conditions. The timing of germination is largely controlled by the interaction between the physiological state of the seed (dormancy) and the responsiveness to environmental parameters such as the seed soil bed (Beardsell & Richards 1987).

Innate dormancy responses in seeds such as the Leguminosae family may be a reproductive strategy within the species to ensure germination under potentially favourable conditions in successive years (Bell *et al.* 1993). For example, seed remains viable in the seed soil bank due to a thick seed coat that is subjected to weathering over time. The reduction of the mechanical constraint of the testa enables the penetration of water and the exchange of gases to allow the embryo to freely develop (Villiers 1971; Rolston 1978; Mott & Groves 1981).

Pre-sowing treatment of seeds is particularly important in the Leguminosae family in order to increase the effectiveness of directly sown seed in the field. Pre-sowing treatment intervenes in the process of the seed's natural 'biological clock' by breaking the seed coat imposed dormancy. As a consequence the land manager decides when germination should take place according to favourable climatic and soil conditions.

Plant establishment in the field is enhanced by appropriate soil preparation according to site conditions. The selection of a particular method of soil preparation is dependent on soil type, position in the landscape and climate (Lefroy *et al.* 1991). For example, the appropriate soil preparation for sites with porous soils, on upper valley slopes with low rainfall, will aim to redistribute the limited available water by channelling (scalping) it towards the radical/root zone of the developing seed/seedling. In contrast, soil preparation for site conditions with soils that are heavy and subjected to seasonal inundation by ground water will aim to redistribute the water away (mounding) from the seed/seedling to allow aerobic conditions to occur around the root zone.

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Direct seeded seedlings can have establishment advantages in the field relative to nursery seedlings. In 1993, a national questionnaire of 1600 landholders conducted by the Kondinin Research Group (K.R.G.) found that direct seeding method has the lowest mortality rates in seedlings (17%) relative to nursery seedlings (50%) which were planted using a tractor drawn manual tree planter (Fig. 5.2). This may be in part attributed to the ability of direct seeded seedlings to tolerate drought conditions due to the deep penetration of an unconfined root system into the soil profile (Fig. 5.3).

An important factor in nursery seedling establishment is the contact between the interface of the roots with the site soil. Seedlings must rapidly extend roots into the surrounding soil prior to the first summer or they will exhaust the available moisture held within the root system from the nursery environment (Goor & Barney 1968).

Most native plants used in revegetation in Western Australia have a vigorous root system. For example, *Eucalyptus* trees have an intrusive taproot and dominant laterals. Root systems that develop quickly in a containerised environment are prone to distortions such as kinks, circling and girdled roots (Whitcomb 1981). Distorted root structures result in loss of vigour in the aerial portion of the plant and poor establishment once planted out in the field. The loss in seedling vigour is due to the restricted transportation of carbohydrates to the developing apical meristem in the root tip (Whitcomb 1984).

Ninety percent of the root biomass is located adjacent to the walls of the pot in a conventional nursery container (Whitcomb 1984). The seedling root system in a conventional nursery container continues to grow around the perimeter of the pot until being bound by lack of container space causing structural root defects. Conversely, air-pruning pots are biologically designed to train the root tip towards the outside of the pot where it is desiccated and dies (Whitcomb 1984). Air-pruning pots cause good fibrous development, by stimulating and thickening lateral branching of both the primary and secondary root systems.

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Figure 5.1: The proportion of methods used to obtain seed and seedlings by 1600 landholders interviewed in a national questionnaire who were involved in a revegetation programme (K.R.G. 1993).



Figure 5.2: Mortality of seedlings in the field as determined by 1600 landholders in a national questionnaire involved in revegetation using nursery seedlings (N.S): planted mechanically (one person), planted manually and mechanically (two people) and a hand tool versus directly sowing seeds (K.R.G. 1993).



Figure 5.3: Stylised cross section of two root systems showing an unrestricted taproot (direct seeded) and a confined root structure (nursery seedlings) when planted in field soil (adapted from Dalton 1993).

The principle limitation in both conventional and air-pruning pots is that the containers manipulate root development. The majority of containers do not have the volume capacity to accommodate the full extension of a taproot or biomass of roots which often causes restricted seedling development. Consequently, containerised seedlings may have a limited ability to withstand periods of moisture stress relative to direct seeded seedlings in the field due to a distorted root system. Furthermore, nursery seedlings planted in the field which have rapid canopy growth can be susceptible to 'wind throw' due to insufficient root development caused by a distorted root system.

5.1.2: PLANT GROWTH

Nursery seedlings are typically six months old when planted out in the field. As a consequence, the seedlings have a relatively well developed vascular system both above and, to a lesser extent, below the soil surface. A developed seedling may have a height advantage over weeds when competing for sunlight.

Similarly, the structure of a nursery seedling's root system may enable ready uptake of available moisture and nutrients (Venning 1991). Therefore, the developed nursery seedling may have establishment advantages in growth potential in the field relative to direct seeded seedlings.

A two year study conducted by Whitham *et al.* (1986) found that containerised native seedlings grew less than direct seeded plants in height and stem width. Similarly, a comparison in growth rates of Australian natives found smaller tube stock seedlings were higher than 200 mm pot grown seedlings after five years (Clemens & Radford 1986). The disparity in growth and development between seed and nursery seedlings prior to transplanting in the field does not always remain constant and may be reversed over time (Whitham *et al.* 1986).

5.1.3: COST OF REVEGETATION

Land managers have employed nursery seedlings in preference to direct seeding as a method of plant establishment on degraded land. In 1993, the K.R.G. interviewed 1600 farmers nationally who were involved in revegetation on their properties of whom 61 % purchased seedlings and only 5% bought or 27% collected seed (see Fig. 5.1). This is despite the fact that seedling establishment using direct sowing techniques are in the order of one third the cost of a nursery seedling (Dalton 1993; Scheltema 1993). Revegetation of agricultural land using native flora can require a large capital expenditure in the short term given the extent of revegetation required in some areas. The cost of revegetation is the single most prohibitive factor in establishing a rehabilitation programme on degraded land (Venning 1988; K.R.G. 1993). The K.R.G. (1993) found that on average revegetation incurred a total cost of \$1300 per hectare including electric fences, labour and seed/seedlings.

The benefits of revegetation are difficult to quantify in the short term. However, if the initial revegetation programme is effective then it may substantially decrease capital expenditure in the long term by reducing post-planting maintenance.

Capital expenditure on revegetation programmes can be minimised by employing an appropriate method of native plant establishment. The methods listed in Table 5.1 are rated in order of increasing cost. The cheapest method of revegetation is the use of natural recruitment (Barron *et al.* 1996). However, this method is very site specific as the seed soil bank is often absent or minimal on degraded agricultural land. Natural recruitment is therefore limited in its application as a revegetation method. As an alternative, the cost of direct seeding compared to nursery seedlings as a revegetation method is considerably reduced (Dalton 1993).

Direct seeding is a relatively low cost method of plant establishment both in terms of the price of seed versus nursery seedlings and in the labour involved for each respective method. However, there is minimal information available for land managers on the relative short and long-term costs of plant establishment using the two revegetation methods.

5.2: Aim

The aim of this experiment is to compare seedling establishment, plant growth and economics of directly sown seed with planted nursery seedlings on a wind eroded site (Case study 1: York) and a salt affected site (Case study 2: Tammin) both within the central Avon catchment (see Fig. 3.1).

5.3: Methods

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The degradation forces acting upon each site has in part dictated the plant species chosen for the purposes of revegetation. The plant species selected were intended to be representative of native species which have adapted to the environmental and soil conditions of the area. However, due to the limited supply of local seed and seedlings, some species have been collected from distant biolocalities within the catchment. The species and biolocalities sown and planted at both York and Tammin are listed in Table 5.2. These species have been used successfully in revegetation of degraded land (Lefroy *et al.* 1991) in the Western Australian wheatbelt.

In order to maintain consistency with the seed treatment techniques employed in Chapter 3, Leguminosae seed (*Acacia microbotrya, Acacia saligna & Kennedia prostrata*) was scarified for 5 minutes using sandpaper prior to sowing in the field.

Seed lot	Species	Biolocality	Site
900163	Acacia microbotrya	Ravensthorpe	York
931873	Acacia saligna	Jarrahdale	Tammin
940520	Allocasuarina huegeliana	Brookton	York 👳
930212	Calothamnus quadrifidus	Wanneroo	York
940626	Casuarina obesa	Geraldton	Tammin
930122	Eucalyptus kondininensis	Pin erup	Tammin
950053	Eucalyptus wandoo	Wongan Hills	York
960501	Kennedia prostrata	Wanneroo	York
940597	Melaleuca cuticularis	Boddington	Tammin

Table 5.2: Nine Species Sown and Planted for Evaluation in Methods ofPlant Establishment (Direct Seeding/Nursery Seedlings) atYork and Tammin Showing Seed Lot Number and Biolocality.

Establishment in the field is defined for the purpose of this study as survival of plants over two consecutive winter and summer seasons.

The seeding rate sown in the field was calculated on the basis of establishing (after 23 months) an equivalent number of field grown seedlings with planted nursery seedlings for each species using the following formulae:

Seeding Rate = Number of Nursery Seedlings % Field Factor x Number of Viable Seed/gram

At both field sites, 84 nursery seedlings (42 seedlings per tray) were planted at two metre intervals.

The percent field factor in the above formulae is an estimated proportion of seedlings that can be expected to establish from direct seeding in the field. The percentage field factor is partly based upon the size of the individual seed. The percent field factor was calculated at one percent for small seeded species and five percent for large seeded species (compiled from data: Clemens 1984; Venning 1988; Burke 1990; Scheltema 1993).

The number of viable seeds per gram in the seeding rate formulae was calculated from laboratory trials conducted in Chapter 3 and is summarised in Table 5.3. The quantity of seed directly sown at the site was greater in weight than that tested under nursery conditions due to the anticipated high mortality rates in the field.

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Seed Sown (g)	Seed (g)	Germination	% Field Factor	
			<u></u> .	
560	3	1680	5.0	
840	2	1680	5.0	
40	210	8400	1.0	
21	400	8400	1.0	
28.6	294	8400	1.0	
3.5	2405	8400	1.0	
8.34	1007	8400	1.0	
840	2	1680	5.0	
5.8	1445	8400	1.0	
	560 840 40 21 28.6 3.5 8.34 840 5.8	56038402402102140028.62943.524058.34100784025.81445	$\begin{array}{cccccccccccccccccccccccccccccccccccc$	

Table 5.3: Weight of Seed Sown for Direct Seeding at York and Tammin Showing Viability (Chapter 3), Expected Germination and Percentage Field Factor 5.3.1: CASE STUDY 1: YORK

Site 1 consists of a 3128 m² cleared paddock that is fenced from livestock. The preparation for site 1 prior to sowing/transplanting involved the scalping of soil to a furrow depth of 200 mm using a tractor-drawn Kimseed modular seeder in scalping mode. The Kimseeder was the preferred choice of machinery at the York field site due to its proven success in soil profile construction in the district (G. Cockerton¹ pers. comm.).

Scalping is the mechanical displacement of surface soil to form a furrow using a grader blade (Fig. 5.4). This method involves the creation of a receptive seedbed, the removal of the weed seed store and the channelling of rainfall towards the sowing bed. Scalping is an effective method of weed control, particularly when there has been no recorded pre-emergent or post germination herbicide application to the site (Pigott *et al.* 1994).



Figure 5.4: A stylised cross section of a soil scalped profile used as a method of seed niche creation, weed control and water harvesting employed at the York field site (adapted from Lefroy *et al.* 1991).

A total of 20 rows (Fig. 5.5) were scalped to 2.8 metres in width and 68 metres in length with two metres between each row. The rows in which the seeds and nursery seedlings were sown and planted were designated at random. Random selection of the rows attempts to take into consideration site specific variables such as edge effects and soil types. 84 nursery seedlings were planted into two scalped rows (42 per row) for each of the five species.

The nursery seedlings were planted 1.5 metres apart by digging a hole with a hand trowel and backfilled with compression of the overburden. The base of the compressed pine bark pot was removed to allow unimpeded outgrowth of the roots into the soil medium.

¹Geoff Cockerton: Owner/Manager of Landcare Services York.



Figure 5.5: A stylised overview of the field site at York depicting experimental design of planting treatments.

A total of five seed lots were sown into ten rows (2 rows per seed lot) which were selected at random. Each seed lot was directly sown by hand. Seed was distributed over the length of each row by mixing the seed with 7 litres of site soil to act as a buffering medium (Plate 5.1a). The seed was covered with a fine layer of soil by lightly brushing over the soil with the foliage from a tree branch (Plate 5.1b). The rows were identified by the applicable seed lot number and demarcated at the beginning of each row.

5.3.2: CASE STUDY 2: TAMMIN

Site 2 at Tammin consists of a 3400 m² cleared paddock that is fenced from livestock. The site was mounded into linear rows, 3m wide across the contour gradient using a Chatfield seeder. The Chatfield seeder was the preferred piece of machinery due to its availability from the Landcare District Committee in Tammin. Mounding involved the burial of the weed seed to a depth of 200 mm using the overburden from the parallel scalped furrows by elevating the soil surface. Mounding is particularly important when soil is subjected to inundation from seasonal fluctuations in the water-table. The raised mounded bed reduces anaerobic conditions by allowing water to drain away freely from around the developing root zone (Fig. 5.6).



Figure 5.6: A stylised cross section of a soil mounded profile used as a method of niche creation, weed control and water drainage employed at Tammin (adapted from Lefroy *et al.* 1991)

The nursery seedlings were planted using the same method as at the York field site. The furrows between the rows were moist upon sowing due to recent rains. The random designation of rows in which the seed and nursery seedlings were sown and planted attempts to take into consideration site specific variables such as flooding and salinity levels (Fig. 5.7).

The seed was directly broadcasted by hand and sown along the mounded bed into distinct rows for individual seed lots. Each seed lot was mixed with 7 litres of site soil to act as a buffer for even distribution. The seed and soil mix was sown in the next available row. Due to the heavy nature of the soil, a square mouth shovel was dragged across the mounded bed covering the seed with approximately 3 mm of soil overburden (Plate 5.2). The rows were identified with the applicable seed lot number and labelled at the beginning and end of each row.





LEGEND

- 1. M. cuticularis (seedling)
- 2. *M. cuticularis* (seed)
- 3. E. kondininensis (seed)
- 4. E. kondininensis (seedling)

- A. saligna (seedling)
 A. saligna (seed)
 C. obesa (seedling)
- 8. *C. obesa* (seed)

In order to increase the initial success rate, sowing of seeds and transplanting of seedlings corresponded with favourable winter rainfall that occurred in June, July and August for both York and Tammin. The York site was sown/planted on the 6th of July, 1996 (see Fig. 3.4) and the Tammin site on the 1st of August 1996 (see Fig. 3.6). The dates chosen were scheduled around cereal crop planting and the availability of the replanting equipment.

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Plate 5.1a: Manual direct seeding along a scalped furrow at York.

Plate 5.1b: Manual impregnation of seed using foliage from a tree branch at York.



Plate 5.2: Manual impregnation of seed using a square mouth shovel dragged across the mounded bed of heavy soil at Tammin

The sites were monitored approximately every eight weeks by counting all the germinants and nursery seedlings. This method was preferred over random quadrat sampling due to the irregular distribution of germinants from direct seeding. The first 20 seedlings in each row were measured for height using a ruler from the soil level to the active terminal bud. The number of plants and their growth height were recorded at each monitoring.

Special post sowing and planting maintenance of the developing seedlings were carried out at York but not at Tammin. This involved the spraying of a general miticide for the control of red legged earth mites (*Halotydeus destructor*) using (Maldison: active constituent Malathion[®]) and the baiting for European rabbits (*Oryctolagus cuniculus*). The management control of rabbits involved the use of poisonous baits; wheat grains were soaked in $1080^{®}$ (sodium fluoroacetate) and placed around active rabbit warrens. Pest management control took place in September 1996 when pest reproductive cycles were active.

These case studies provide preliminary information on the relative performance of direct seeding and nursery seedlings in the field. The results from the study are difficult to analyse quantitatively because the experimental design of comparing direct seeding with planted nursery seedlings must take into consideration the following factors:

- The germinants and seedlings are in different growth phases and as a consequence grow at different rates.
- The environmental conditions affecting the seeds (predation, burial, dormancy, and moisture) and germinants (desiccation, fungus) are different than those affecting nursery seedlings (predation, competition).
- Seed is broadcast into a myriad of microniches (soil burial depth) resulting in erratic germination and uneven distribution compared with the set planting of nursery seedlings in the field over a demarcated area.

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5.4: Results

5.4.1: CASE STUDY 1: YORK

The number of seeds that germinated at the York field site from direct seeding (Table 5.4) was less than the calculated estimates derived from viability trials in the laboratory (see Table 5.3). In particular, species from the Leguminosae family such as *Acacia microbotrya* (162) and *Kennedia prostrata* (20) recorded low germination. However, with the exception of *K. prostrata* the remaining species produced more germinants than the 84 seedlings that were transplanted.

Table 5.4: Total Plant Establishment in May 1998 at York for Direct Seeding (D.S.) and Nursery Seedling (N.S.) Following Initial Germination and Transplanting Recorded in September 1996 (Percentage Survival of Seedlings).

Species	Method	# Germinants/ Transplants September '96	# Of Plants Established May '98	% Survivalª
A. microbotrya	D.S.	162	63	38.9
A. microbotrya	N.S.	. 84	66	78.6
A. huegeliana	D.S.	1330	621	46.7
A. huegeliana	N.S.	84	77	91.7
C. quadrifidus	D.S.	353	122	36
C. quadrifidus	N.S.	84	31	34.6
E. wandoo	D.S.	647	180	27.8
E. wandoo	N.S.	84	68	81
K. prostrata	D.S.	20	0	0
K. prostrata	N.S.	84	68	81

^a Calculation based on survival of seedlings in May 1998 after seed germination and transplanting recorded in September 1996.

Seed germination in the field was first recorded in the month of September when germinants were visible. The small size of the germinants coupled with weed infestation at the trial site made observations of germinants difficult to detect prior to September. Given that seed germination in the laboratory occurred over a 28 day period (Chapter 4), it is reasonable to assume that seed may have germinated and subsequently died prior to the initial observations in September. Consequently, the number of germinants observed in September may have been underestimated.

At the end of the two year trial period at York, a higher number of small seeded plants were established using the direct seeding method employed at York relative to planted nursery seedlings. Conversely, a low number of large seeded plants were established using the direct seeding method relative to planted nursery seedlings. As expected a large number of the direct seeded germinants at York did not survive the establishment period (Table 5.4). In particular, *K. prostrata*, which had a low number of germinants (20), failed to become established in the field. The highest number of germinants to become established in the field was *A. huegeliana* with 621.

Direct seeded seedlings at York had a lower percentage survival relative to transplanted nursery seedlings with the exception of *Calothamnus quadrifidus* of which 36% survived compared to 34.6% survival for nursery seedlings. Nursery seedlings that performed well were *Allocasuarina huegeliana* (91.7%), *Kennedia prostrata* (81%), *Eucalyptus wandoo* (81%) and *Acacia microbotrya* (78.6%).

5.4.2: CASE STUDY 1: TAMMIN

The number of seeds that germinated at the Tammin field site from direct seeding (Table 5.5) was less than the calculated estimates derived from viability trials in the laboratory (see Table 5.3). In particular, *Acacia saligna* (24%) *Melaleuca cuticularis* (34%) recorded low germination. However, *E. kondininensis* had a relatively high number of germinants with 556.

None of the germinants sown from seed survived the establishment period (Table 5.5) when recorded in April 1997. Similarly, nursery seedlings planted at Tammin also failed to become established in the field resulting in a 100% mortality of seedlings for both revegetation methods. As a consequence, there are no graphs to express the results of the Tammin case study.

Table 5.5: Total Plant Establishment in April 1997 at Tammin for Direct Seeding (D.S.) and Nursery Seedling (N.S.) Following Initial Germination and Transplanting Recorded in September 1996 and <u>Percentage Survival of Seedlings.</u>

Species	Method	# Germinants/ Transplants September '96	# Of Plants Established May '98	% Survivalª
A. saligna	D.S.	24	0	0
A. saligna	, N.S.	84	0	0
C. obesa	D.S.	124	0	0
C. obesa	N.S.	84	0	0
E. kondininensis	D.S.	556	0	0 ⁷²
E. kondininensis	N.S.	84	0	· · · · · 0 · · · ·
M. cuticularis	D.S.	34	0	0
M. cuticularis	N.S.	84	¹ 0	ି 0

^a Calculation based on survival of seedlings in April 1997 after seed germination and transplanting in July 1996.

The following germination Figures were expressed as a percentage, calculated on the basis of seedling survival or mortality between the seasons stated. For example, mortality % =January (# of seeds) / November (# of seeds) x 100.

Acacia microbotrya seed was directly sown at York in July 1996 and recorded 162 germinants in September of that year (Fig. 5.8). From November 1996 to January 1997, seedling mortality was 53.8%. During July to September 1997, there was a 26.1% recruitment of second year germinants. This was followed by a further seedling mortality of 23.2%, occurring between September to November 1997. During the months between November 1997 and May 1998, the number of direct seeded seedlings stabilised with no further mortality.

During November 1996 and January 1997, seedling mortality for *A. microbotrya* nursery seedlings was 20% (Fig. 5.8). Between January 1997 and May 1998 plant numbers stabilised with no further seedling mortality.

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A. microbotrya seedlings that germinated from seeds directly sown in winter (July 1996) had a mean height of 8mm (Fig. 5.9). Seedling growth increased over the first summer (November 1996 to March 1997) to a total mean height of 157mm which slowed during autumn and winter (March to September 1997) to a total mean height of 213mm. A sharp increase in seedling growth occurred during the second spring and autumn (September 1997 to January 1998) to a total mean height of 756mm. Seedling growth over the trial period attained a total mean height of 940mm.

Nursery seedlings of *A. microbotrya* grew from an initial planting mean height of 49mm as at July 1996 to 82mm (Fig. 5.9) during the first spring (November 1996). Seedling growth occurred during the first summer (November 1996 to March 1997) to a total mean height of 463mm, which slowed during the second autumn (September 1997) to a total mean height of 627mm. During autumn (September 1997) and summer (January 1998), seedling growth accelerated to a total mean height of 1051mm. The growth height of nursery seedlings over the trial period averaged 940mm.

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Allocasuarina huegeliana seed was directly sown at York in winter (July 1996) and 1330 germinants (Fig. 5.10) were recorded in the first spring (September 1996). During spring to summer (September 1996 to January 1997), seedling mortality was 53%. During the second winter and spring (July to September 1997), there was a 19% recruitment of second year germinants. This was follo ved by a 16% seedling mortality, occurring during autumn (September to November 1997).

During spring and summer (November 1996 to January 1997), seedling mortality for *Allocasuarina huegeliana* nursery seedlings was highest at 6.1% (Fig. 5.10) which stabilised during summer to autumn (January 1997 to May 1998) with no further seedling mortality.

Allocasuarina huegeliana seedlings which germinated from seeds directly sown in July 1996 had a mean height of 67mm (Fig. 5.11) in spring (November 1996) which increased in the spring to summer period (November 1996 to March 1997) to a mean height of 184mm. Seedling growth slowed to a mean height of 254mm between autumn and the second spring (March to September 1997) which was followed by a sharp increase in seedling growth in the period between September 1997 and January 1998 to a mean height of 900mm. Seedling growth over the trial period attained a total mean height of 1140mm (Fig. 5.11).

Nursery seedlings of *Allocasuarina huegeliana* increased in growth from an initial planting mean height of 277mm as at July 1996 to 82mm (Fig. 5.11) during the first spring (November 1996). Seedling growth occurred between spring and summer (November 1996 to March 1997) to a mean height of 297mm. Seedling growth was reduced from during autumn through till the second spring (March to September 1997) with a mean height of 805mm. Seedling growth accelerated in spring and summer (September 1997 to January 1998) giving a mean height of 1805mm. The growth height of nursery seedlings over the trial period averaged 2290mm.



Figure 5.10: Mortality and survival of *Allocasuarina huegeliana* seedlings comparing two methods of plant establishment at York (1996-1998).



Figure 5.11: Mean growth (height) of *Allocasuarina huegeliana* seedlings comparing two methods of plant establishment at York (1996-1998).

Calothamnus quadrifidus seed was directly sown at York in July and 387 germinants (Fig. 5.12) were recorded in spring (November 1996). Seedling mortality between spring and the first summer (November 1996 to January 1997) was 66.2%. During the second winter to spring (July to September 1997), there was a 144% recruitment of second year germinants. This was followed by a 40% seedling mortality, occurring between spring and the second summer (September 1997 to March 1998) which stabilised in the period between March and May 1998 with no further mortality.

Seedling mortality for *Calothamnus quadrifidus* nursery seedlings during spring and summer (November 1996 and January 1997) was 45.4% (Fig. 5.12). Seedling mortalities declined between the first summer and autumn (January 1997 and May 1998) to only 31%.

Calothamnus quadrifidus seedlings that germinated from seeds directly sown in July 1996 had a mean growth height of 7mm (Fig. 5.13) as at November 1996. Seedling growth in the first spring to summer (November 1996 to March 1997) increased to a total mean height of 114mm. During autumn and the second spring (March to September 1997), seedling growth slowed to a total mean height of 156mm. A sharp increase in seedling growth occurred in the second spring and summer (September 1997 and January 1998) to a total mean height of 620mm. Seedling growth over the trial period attained a total mean height of 810mm.

Nursery seedlings of *Calothamnus quadrifidus* increased in growth from an initial planting mean height of 170mm as at July 1996 to 252mm (Fig. 5.13) in spring (November 1996). Seedling growth occurred between the first spring and summer (November 1996 and March 1997) to a mean height of 598mm. Seedling growth was reduced during autumn to the second spring (March and September 1997) with a total mean height of 796mm. Seedling growth accelerated during the second spring and summer (September 1997 to January 1998) to a mean height of 1123mm. The growth height of nursery seedlings over the trial period averaged 1360mm.



Figure 5.12: Mortality and survival of *Calothamnus quadrifidus* seedlings comparing two methods of plant establishment at York (1996-1998).



Figure 5.13: Mean growth (height) of *Calothamnus quadrifidus* seedlings comparing two methods of plant establishment at York (1996-1998).
Eucalyptus wandoo seed was directly sown at York in July and recorded 647 germinants (Fig. 5.14) in the first spring (September 1996). Seedling mortality (60%) occurred between spring and summer (November 1996 to January 1997). During July to September 1997, there was an 11.4% recruitment of second year germinants. This was followed by a 12.6% seedling mortality occurring between the second spring and summer (September 1997 to January 1998). Seedling numbers stabilised in the period between January and May 1998 with no further mortality.

During November 1996 and January 1997 the seedling mortality of *Eucalyptus wandoo* nursery seedlings was 14.8% (Fig. 5.14). Between January 1997 and May 1998 seedling mortality slowed to 1.5%.

Eucalyptus wandoo seedlings, which germinated from seeds directly sown in July 1996, had a mean growth height of 7mm (Fig. 5.15) as at November 1996. Seedling growth during the first spring and summer (November 1996 to March 1997) increased to a mean height of 115mm which slowed to a mean height of 136mm between autumn and spring (March to September 1997). A sharp increase in seedling growth occurred between the second spring and summer (September 1997 and January 1998) to a mean height of 402mm. Seedling growth over the trial period attained a total mean height of 510mm.

Nursery seedlings of *Eucalyptus wandoo* increased in growth from an initial planting mean height of 205mm as at July 1996 to 208mm in November 1996 (Fig. 5.15). Seedling growth occurred between spring and summer (November 1996 and March 1997) to a mean height of 463mm which slowed during autumn and spring (March to September 1997) resulting in a mean height of 546mm. Seedling growth accelerated to a mean height of 895mm during the second spring and summer (September 1997 and January 1998). The growth height of nursery seedlings over the trial period averaged 970mm.

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Figure 5.14: Mortality and survival of *Eucalyptus wandoo* seedlings comparing two methods of plant establishment at York (1996-1998).





Kennedia prostrata seed was directly sown at York in July and only recorded 20 germinants (Fig. 5.16) in spring (September 1996). Seedling mortality (20%) occurred between spring and summer (November 1996 to January 1997) which increased to 100% during July and September 1997.

Seedling mortality for *Kennedia prostrata* nursery seedlings was 9.5% (Fig. 5.16) between winter and spring (July and November 1996) which reached a peak at 10.5% during spring and summer (November 1996 and January 1997). Seedling mortality slowed to 6.8% in the second year (January to November 1997). Seedling numbers stabilised in the period between November 1997 and May 1998 with no further mortality.

Kennedia prostrata seeds directly sown in July 1996 did not produce seedlings greater than 20mm in width and therefore no growth measurements were recorded (Fig. 5.17).

Nursery seedlings of *Kennedia prostrata* increased in growth from an initial planting mean width of 231mm as at July 1996 to 274mm in November 1996 (Fig. 5.17). Seedling growth occurred between the first spring and summer (November 1996 and March 1997) to a total mean width of 875mm. Seedling growth was reduced during autumn to the second summer (March to December 1997) with a total mean width of 1165mm. Between December 1997 and May 1998, no growth was recorded due to the difficulty in determining the distinction between individual plants.

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5.4.3: CASE STUDY 1: ECONOMICS AT YORK

The total economic cost of materials used in plant establishment at York for both revegetation methods was \$153.10 for seed and \$218.40 for nursery seedlings (Table 5.6). With the exception of *Kennedia prostrata* (\$123.20) the cost of seed used in direct seeding was economical compared to the cost of nursery seedlings. The cost per seedling based on the number of seedlings established in the field was far cheaper relative to nursery seedlings.

Species	Direct \$ Seed	Seeding \$/seedling	Nurser \$ N.S.	y Seedlings \$/seedling
	•	·		······································
A. microbotrya	16.8	0.27	25.2	0.38
A. huegeliana	4.8	0.01	50.4	0.65
C. quadrifidus	6.6	0.05	46.8	1.51
E. wandoo	1.7	0.01	49.8	0.73
K. prostrata	123.2	N/A	46.2	0.68
Total	153.1		218.4	
				an ta' Nganasang tao wa

Table 5.6: Cost of Seed versus Nursery Seedlings (N.S.) Showing the Cost per Seedling Based on the Establishment at York.

5.5: Discussion

5.5.1: SEED GERMINATION

A large proportion of viable seed sown in the field in July 1996 that was expected to germinate (see Table 5.3), but failed to produce germinants (see Fig. 5.7). The estimated number of seeds expected to germinate in the field was low and highly variable with a range of 0.4 percent for *Melaleuca cuticularis* to 15.8 percent for *Allocasuarina huegeliana*. Runciman (1991) and Pigott *et al.* (1994) states that only a small proportion of seeds sown germinate.

Native plant seed is susceptible to a plethora of environmental factors that prevent germination in the field (Beardsell & Richards 1987). The potential contributing factors such as pre-emergent pathogens and seed predation is beyond the realm of this study and will only be briefly discussed. However, the month in which seed germination was observed, seed sowing depth and seed dormancy responses in the Leguminosae group was identified as a probable causal agent for low germination in the field.

The effect of pre-emergent pathogens on the mortality of germinants can be considerable. Mwanza and Kellas (1987) found that the main pathogenic fungi responsible for pre-emergent damping off are *Pythium* and *Fusarium* species. These pathogens accounted for the mortality of 34% of the viable seed in *Eucalyptus radiata* and *Eucalyptus obliqua*. Pathogenic fungi may be responsible for the low germinant survival in the field due to pre-emergent damping off of developing native seed but this was not tested here.

Seed predation by invertebrates has been responsible for reduced germination in the field (Stoneman 1993). Ants have generally been identified as the predominant invertebrates responsible for seed predation (Ashton 1979; Majer 1980; Drake 1981; Andersen 1982; O'Dowd & Gill 1984; Abbott & van Heurck 1985; Andersen 1988; Andersen & Ashton 1988). However, seed predation by ants is not considered a contributing factor to low germination here because of site soil disturbance, seasonal sowing time and seed application quantity involved in the method of direct seeding.

Site preparation for revegetation at York and Tammin involved mass soil disturbance that produces hostile conditions and subsequent displacement of ant colonies. Several studies (Majer 1978; Majer 1980; Majer *et al.* 1984) have recorded that seed removal by ants decreases from the boundary of jarrah forests (*Eucalyptus marginata*) into bauxite pits. The sowing time of seed in the field (July 1996) corresponded with winter rainfall and cooler temperatures (see Fig. 3.6-3.9). Majer and Abbott (1989) observed a decrease in seed predation by ants in winter, relative to the foraging activity in the warm drier months of summer. The artificial broadcasting of a large quantity of native seed in the field (Table 5.3) may have satiated the ant population.

Several studies have reported that mass release of native seed following a fire will satiate ant colonies and allow higher germination than would otherwise occur (O'Dowd & Gill 1984; Wellington & Noble 1985; Andersen 1987; Andersen 1988; Neumann 1992; Yates *et al.* 1996). Therefore, seed predation by ants is unlikely to have been a causal agent in the reduction of seed germination.

Seed germination in the Leguminosae group was low when observed in September relative to the number of seeds sown in July. This may have been due to an inappropriate planting method used to sow endospermic seeds such as acacia's and kennedia's. The hand broadcasting technique employed to sow the seeds and subsequent shallow burial (foliage at York; shovel at Tammin), tended to discriminate against endospermic seeded species. In a direct seeding experiment conducted by Pigott *et al.* (1994), poor germination by large seeded species was attributed to sowing seeds on the soil surface. Sowing of Leguminosae seed at some depth within the soil profile may provide the necessary moisture conditions for germination in order to break dormancy mechanisms.

Environmental conditions acting upon the field sites may have restricted germination in *Kennedia prostrata* (see Fig. 5.1G) and *Acacia saligna* (see Fig. 5.5) due to seed dormancy mechanisms inherent in the Leguminosae group (Cavanagh 1987). Similarly, *Acacia microbotrya* (see Fig. 5.8) had an insufficient number of germinants to establish a high number of seedlings relative to nursery seedlings. This is despite using scarification treatment as a method of breaking the seed coat imposed dormancy (see section 4.3). The problems associated with germination and subsequent seedling survival in the Leguminosae group are similar to those experienced by Pigott *et al.* (1994) but also appear to contradict the results of direct seeding trials conducted by Panetta and Groves (1990) in South Australia.

5.5.2: SEEDLING SURVIVAL

Nursery seedling had a high percentage survival relative to direct seeded seedlings for most species trialed at York (see Table 4.4). Contrary to the hypothesis stated in this introduction, nursery seedling survival was found to be particularly high in periods of limited water availability between September 1996 and January 1997 when compared to direct seeded seedlings (see Fig. 5.8, 5.10, 5.12, 5.14 & 5.16). The advanced vascular system of nursery seedlings upon transplanting in winter enabled successful establishment of seedlings in the field prior to arid conditions experienced in summer. The air pruning peat pots may have encouraged penetration of the nursery seedling root systems into the lower soil horizons where they are protected by a moderating environment which minimises fluctuations in soil moisture and temperature.

Caution should be applied when evaluating nursery seedling survival in the shortterm. The distorted taproot of containerised seedlings may cause 'wind throw' in the long-term if the root system is unable to support the tree canopy. In low rainfall years, nursery seedlings may be susceptible to moisture stress due to the abundance of surface roots encouraged by nursery containers.

The seasonal time in which nursery seedlings were planted may also have contributed to seedling survival at York. Nursery seedlings were planted at York in early July to coincide with seasonal rainfall. Several authors indicate that early transplanting of nursery seedling increases the chance of survival in the field (Lefroy *et al.* 1991; Runciman 1991; Venning 1991).

Environmental factors detrimental to a developing seedling such as water stress, predators and weed competition are minimised under controlled nursery conditions. Nursery seedlings are grown under conditions where the plant's physiological and biological requirements are satisfied. Conversely, direct seeded seedlings are extremely susceptible to environmental factors during the first few weeks after germination in the field (Goor & Barney 1968).

Seedling survival in the field may have been enhanced through a process of phenotypic selection prior to transplanting in the field. In a nursery environment a quantity of seed is directly sown into a soil filled container in order to produce several germinants. Excess germinants are 'pricked out' leaving a single germinant, which is a value judgement based upon vigour and size (phenotypic selection). These phenotypic characteristics may aid survival in the field. Conversely, seed directly sown in the field is subjected to a genotypic selection process influenced by environmental parameters acting upon the site. Consequently, seedling mortality from direct sown seed is high due to the differential sorting of germinants based upon genotypic adaptations to environmental conditions acting upon the field site.

Nursery seedling mortality in York (see Fig. 5.8, 5.10, 5.12, 5.14 & 5.16) was high for direct seeded species over the first summer (November 1996 to January 1997). Similarly, second year recruitment of seed germinants may also have died over their first summer (September to November 1997). These periods corresponded with a reduction in seasonal rainfall (see Fig. 3.4). Furthermore, the failure of seedlings to become established at the Tammin field site are likely to be due to the below average rainfall between December 1996 and May 1997 (see Fig. 3.6). Soil water deficits contributed to high seedling mortality in conjunction with seedling predation observed at the Tammin site. Red-legged earth mites (Halotydeus destructor) have been reported by Venning (1988) to occur in close association with capeweed (Arctotheca calendula). Capeweed was the dominant species to have grown at the trial sites both in York and Tammin in the winter months and as a consequence populations of earth mites were in abundance. Nursery seedlings and germinants are particularly vulnerable to predation by earth mites, which has been known to cause high mortality among developing seedlings (Clemens 1980; Clemens 1984). The symptoms of earth mite damage such as mottled chlorosis and pigmented bronzing were observed on the emergent leaves of seedlings at the Tammin site.

The miticide application used at the York field site appeared to be effective in controlling *H. destructor* as numbers declined rapidly following spraying. However, failure to control earth mite predation may have contributed to the failure of seedling establishment at the Tammin field.

The rabbit (*Oryctolagus cuniculus*) can have a devastating effect on young native seedlings (Scheltema 1993). Rabbit diggings, burrows and fresh scats were observed at both the York and Tammin field sites. Rabbit numbers appeared to have been controlled at York through the use of Sodium fluoroacetate (see Methods) as no fresh markings were observed after baiting. However, the lack of control at Tammin was evident in that nursery seedlings were uprooted and damaged to varying degrees and in some cases the compressed pine bark pots were dug up and eaten.

The major winter weed species recorded at both sites were capeweed (*Arctotheca calendula*) and to a lesser extent wild oats (*Avena fatua*) and Guildford grass (*Romulea rosea*). Blue lupins (*Lupinus cosentinii*) were recorded at the York trial site only. The degree of weed infestation varied according to the type of soil preparation employed at each site.

Weeds tended to colonise the overburden from the mounding technique employed at Tammin where they grew among nurscry seedlings (Plate 5.3) and seeded seedlings. Similarly, weeds infested the overburden from the scalping technique employed at York but did not compete for resources as native species were sown and planted into the adjacent scalped furrow. However, colonisation of the scalped furrows by weeds did occur in the following winter at York. Presumably, the weeds had germinated from seed stored in the soil overburden. Native seedlings at York in the second year were more established and tended to grow through the prostrate habit of capeweed (Plate 5.4) after the weeds had flowered and were receding. The dead capeweed acted as surface mulch suppressing airborne sand and reducing evapotranspiration from the soil.



Plate 5.3: Weed infestation (*Arctotheca calendula* & *Romulea rosea*) at the Tammin trial site that may have contributed to the mortality of (*Eucalyptus kondininensis*) seedlings.



Plate 5.4: A direct seeded seedling *(Calothamnus quadrifidus)* penetrating the prostrate habit of capeweed *(Arctotheca calendula)* in the second year of the trial period.

The failure of the native seedlings at Tammin is in part due to the effects of competition from weed species. Competition from weeds may have retarded the growth development of a deep root system, which enable native seedlings to withstand periods of moisture stress, particularly through the first summer. Weed control is essential in the initial establishment of native seedlings (Bulman & Dalton 1990; Pigott *et al.* 1994). The effect of competition of weeds on native seedlings will be discussed in further detail in Chapter 7.

Extreme fluctuations in ground temperatures, intense light and exposure of direct seeded seedlings to sand blasting by wind could account for the failure of *K. prostrata* to become established at York. In an undisturbed natural community, *K. prostrata* grows as an understorey species where it germinates and grows in low light levels protected from exposure to atmospheric conditions by a vegetation canopy.

5.5.3: SEEDLING ESTABLISHMENT

The number of seedlings that established from seed germinated in the field was greater than that established from nursery seedlings planted at York (see Table 5.4) with the exception of the Leguminosae group. Direct seeding involved the broadcasting of a quantity of seed based on field factor estimates to ensure that a specific proportion of seedlings survive and become established in the field. However, seedling establishment exceeded field factor estimates. Field factor estimates can only be used as general guidelines because they vary between species and according to environmental parameters acting upon the revegetation site. The relatively high direct seedling establishment may be attributed to a combination of interrelated factors such as a receptive soil bed, minimal competition for light and nutrients and the species specific physiological response to environmental conditions.

The number of direct seeded seedlings established in the field varied relative to the number of nursery seedlings that were planted. Calculating the necessary quantity of seed to be sown (see Table 5.3) and factored estimates of seedling mortality demonstrated extremes in variation between species. The difference in the actual number of seedlings established by the direct seeding method recorded in December 1997 (see Table 5.4) relative to the 84 seedlings that were expected to survive ranged from 0 to 506 seedlings. The field factor estimates for seedling establishment of small seeded species (1%) in the field were far higher than expected and for seedling establishment of large seeded species (5%) were lower than expected. The number of seedlings established from seed relative to the number of nursery seedlings that were planted was highly variable. This may be due to inaccurate field factor estimates. Field factor estimates are based upon a general prescription for establishment rates in the field. These estimated do not take into consideration site specific seasonal fluctuations in seasonal and environmental parameters that can influence germination and subsequent seedling survival. Therefore, the field factor estimates need to be adjusted according to the season and the carrying capacity of the site to be revegetated.

The high number of seedlings established from direct seeding relative to planted nursery seedlings may be due to favourable climatic conditions conducive for seedling development. The establishment of directly sown seed is primarily controlled by rainfall patterns that affect soil moisture and subsequent germination (Venning 1988; Buchanan 1989; Venning 1991; Pigott *et al.* 1994). Consequently, the high establishment of small seeded species was in part due to above average rainfall in July and September for 1996 (see Fig. 3.5) and above average rainfall in the critical months of February, March and April for 1997.

Caution should be applied when evaluating seedling establishment in the short term by direct seeding in the field. Uneven distribution of seed resulted in dense clumping (see section 6.4.3) which may cause reduced seedling growth and increased seedling mortality through competition for available resources in the long term.

5.5.4: SEEDLING GROWTH

Nursery seedlings maintained a distinct growth advantage relative to seedlings that were directly seeded at the end of the trial period (see Fig. 5.9, 5.11, 5.13, 5.15 & 5.17). Nursery seedlings are 'semi-established' after developing under relatively 'optimum' conditions for six months where germination and initial survival occurs in a controlled nursery environment prior to transplanting in the field. Nursery seedlings have an advanced vascular system and are therefore more resilient to environmental stress in the field. Furthermore, erect woody annuals established in a nursery environment have distinct height advantages over weeds when competing for sunlight in the field.

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Between July and November 1996, nursery seedlings had a period of acclimatisation (hardening off) with minimal growth while direct seeded seedlings experienced germination and mortality cycles as seedling numbers stabilised. In November 1996 to March 1997, the growth of nursery seedlings increased due to an advanced vascular system that maximised available resources such as moist subsoil and warmer temperatures. Increased growth occurred in seedlings of both revegetation methods following the winter season with rapid growth between September and November 1997 due to warmer ambient temperatures and available moisture.

Acacia microbotrya (see Fig. 5.9), Allocasuarina huegeliana (see Fig. 5.11) and Calothamnus quadrifidus (see Fig. 5.13) which were direct seeded in the field between September and November 1997 experienced rapid growth relative to nursery seedlings. Direct seeded seedlings have the potential to reduce the growth disparity between nursery seedlings over time.

5.5.5: ECONOMICS

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The cost of seed for sowing as a method of revegetation in the field was found to be more economical in comparison to the cost of nursery seedlings in plant establishment (see Table 5.6) with the exception of *Kennedia prostrata*. The cost of sowing leguminous seeds can be prohibitive in the establishment of native seedlings on degraded land, due to the inherent dormancy mechanisms that inhibit germination.

The exclusion of *K. prostrata* from the seeding mix sown at York would have decreased the cost of direct seeding by approximately 80%. The transplanting of nursery seedlings would be a more cost effective method of establishing this species. Alternatively, incorporating *K. prostrata* seed in a direct seeding mix with overstorey species may alleviate poor germination and subsequent establishment problems.

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5.6: Summary

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One of the advantages of nursery seedlings is a high percentage survival (reliability) relative to direct seeded seedlings over the 23 month period of the study. Nursery seedlings complete the most vulnerable phase of their development in a protected environment, so they are planted on site at an advanced stage relative to seeds. In comparison, seedling survival from direct sown seed in the field was low due to environmental parameters adversely affecting less vigorous germinants. This was demonstrated when high rainfall events mobilised existing site soil that caused the burial of developing germinants from direct seeding. Therefore, the basis of comparison in survival between the two methods may not have enough constants to be strictly relevant.

Under adverse conditions there is no difference between direct seeding and nursery seedlings. Both methods of revegetation failed to produce any established plants at the Tammin field site due to a combination of seasonal influences, predation and weed competition. The mean monthly rainfall for Tammin between December 1996 and May 1997 was substantially lower than the average for that period. Failure to control red legged earth mites *(Halotydeus destructor)*, rabbits *(Oryctolagus cuniculus)* and weeds (eg. *Arctotheca calendula)*, emphasises the need to employ a post sowing/planting management programme.

The number of seedlings established from direct seeding was greater for the majority of the species sown when compared with nursery seedlings planted at the York site. Appropriate site preparations such as a receptive soil bed and weed control may have contributed to this result. However, field factor estimates in seedling establishment was higher than expected for species characterised by small seeds and was lower than expected for species with large seeds. Field factor estimates need to be adjusted according to the season and the seedling carrying capacity of the site to be revegetated.

The exception to the findings is that nursery seedlings gave a greater return than direct seeding in the Leguminosae group. The low and highly variable number of seeds that germinated is more a result of seed dormancy treatment and planting depth than a comparison of the relative merits of the two methods of revegetation. Nursery seedlings planted in the field maintained a distinct growth advantage relative to seedlings that were directly seeded. Nursery seedlings are approximately six months old prior to transplanting into the field and have a competitive establishment advantage. Furthermore, nursery seedlings were in a different growth phase due to their maturity relative to direct seeded seedlings. However, some species of direct seeded seedlings experienced rapid growth relative to nursery seedlings in the second year.

The cost of seed for sowing as a method of plant establishment in the field was approximately one sixth the cost of nursery seedlings. Similarly, the cost per seedling based upon establishment in the field was more economical for direct seeding than planted nursery seedlings with the exception of *Kennedia prostrata*.

The direct sowing of seeds at the York trial site in the central Avon catchment is the most effective method of revegetation relative to the transplanting of nursery seedlings. Direct seeding resulted in a higher establishment of seedlings at a lower cost per unit relative to the planting of nursery seedlings.

BIOLOCALITY COMPARISON IN DIRECT SEEDING ESTABLISHMENT

6.1: Introduction

The effectiveness of revegetation is predominantly determined by the survival, establishment and growth of plants in the field (see Chapter 5). The specific characteristics of seed from one biolocality may result in seeds/seedlings having a comparative genotypic advantage over seeds/seedlings from another biolocality. Therefore, effective revegetation may depend on the selection of a seed biolocality that performs under the environmental conditions prevailing at the site of revegetation.

Conditions in the field vary spatially and temporally with a wide range of climatic factors, habitats and soil types occurring in any one locality. As a consequence, most native vegetation has adapted to a wide range of conditions. This presents a challenge to defining the geographical boundaries of plant provenance (Boland *et al.* 1980). The theoretical concept of a provenance boundary within a species distributional range can only be confidently applied when site conditions are homogeneous, distinct and discontinuous from others. However, species with a large geographical distribution over which important environmental variables change gradually (Boland *et al.* 1980) or conversely, where there is considerable heterogeneity in the landscape, are more difficult to separate into distinct provenance zones.

Several studies have shown that the distinction of provenance boundaries can be clearly identified through the difference in morphological (Ladiges 1975; Potts 1985) and physiological (eg. germination response, seedling growth rate, frost resistance and drought tolerance) characteristics (Boland *et al.* 1980) within species. The influence of provenance characteristics has been supported in this study by a laboratory experiment (Chapter 4), which indicated that there was a significant variation in the number of seeds that germinated when derived from different biolocalities. This information may have an influence on revegetation if it could be demonstrated that it also applies to conditions experienced in the field.

The current challenge in the field is to select a provenance from a biolocality which provides the most effective establishment of native vegetation at a site for the time, cost and labour invested. An obvious basis for biolocality selection is the local remnant vegetation, whereby natural bushland relatively close to the revegetation site, which is to be revegetated is targeted for seed collection. However, seed obtained from local remnants is often in short supply through seed merchants, particularly when the remnant is small and degraded. Furthermore, the time available for landholders to collect seed is quite limited. Therefore, the establishment of suitable criteria for the selection of a seed provenance from other distant biolocalitiess should be identified.

Details of the characteristics of the site (soil types, position in landscape, size of remnant and climatic data) would aid in biolocality selection. However, current information available from seed merchants is limited to the date and place at which the seed was collected. Consequently, the criteria for provenance selection is restricted to geographical position (biolocality) and associated generalised climatic data for the area obtained from the Bureau of Meteorology. Boland *et al.* (1980) refer to a process of using climatic data as a basis for identifying biolocalities suitable to the site selected for revegetation. The evaluation of revegetation using local species compared to the same species from a distant biolocality will assist in seed lot selection based on the performance of seeds (germination, establishment and growth) in the field.

The translocation of species from different biolocalities raises an ethical issue of whether the remnant gene pool surrounding the site to be revegetated should be polluted. The relative performance of local biolocality species is promoted in preference to translocated species as a selection criterion guide to plant establishment (Venning 1988; Lefroy *et al.* 1991; Scheltema 1993). The ethical consideration of biolocality selection would be alleviated if it could be shown that the survival and establishment of local species had a higher performance relative to distant species.

6.2: Aim

The aim of this experiment is to determine if any differences exist in the germination, growth and establishment of seeds sown at York, between seed collected from geographically 'local' and those collected from 'distant' biolocalities within the species distribution range.

6.3: Methods

The plant species listed in Table 6.1 have been examined in previous Chapters and are used here for continuity.

	pocifie bood cor itamb		
Seed lot	Species	Biolocality	Locality
424	Acacia microbotrya	Beverley	Distant
545	Acacia microbotrya	York	Local
1 9 9	Allocasuarina huegeliana	Mt. Barker	Distant
520	Allocasuarina huegeliana	Brookton	Local
1474	Calothamnus quadrifidus	Wanneroo	Distant
1490	Calothamnus quadrifidus	Wongan Hills	Local
1421	Eucalyptus wandoo	Mt. Barker	Distant
1422	Eucalyptus wandoo	Yornaning	Local
642	Kennedia prostrata	Collie	Distant
1365	Kennedia prostrata	Wickepin	Local

Table 6.1: The Five Species Used for Biolocality Comparison of Geographically Local Versus Distant Natives Showing Respective Seed Lot Numbers Sown at York

The selection of biolocalities for the experiment was based on a combination of geographical distance, biogeographical region and climatic zones relative to the field site of York. For each species, both a geographically local and distant seed lot were selected relative to the field site of York (Fig. 6.1). 'Local' biolocalities were designated as those seed lots collected from the Avon wheatbelt bioregion whereas 'distant' biolocalities were designated as the Swan Coastal Plain, Jarrah Forest or Warren biogeographical regions. Local climatic zones relative to the site of York were differentiated by precipitation falling within the 400 mm to 600 mm isohyet compared to distant climate zones with rainfall outside this range (see Fig. 3.3 & Appendix 2).

Biolocality selection was also dependent upon the availability of obtaining seed. In some cases 'local' seed lots were not available from seed merchants at the time of the study. For example, seed lots such as *E. wandoo* and *K. prostrata* were collected from a bioregion with a different climate relative to the York site. In these instances the term 'local' is limited to geographical distance alone, relative to the site to be revegetated.

Chapter 6



Figure 6.1: Location of the biolocalities within the different biogeographical regions from which each seed lot was collected, geographically relative to the field site of York (adapted from Thackway and Cresswell 1995).

The seeds were directly sown at York using the same method as those employed in Chapter 5. The York field site consists of a 0.27ha (2720 m²) plot fenced from livestock with the topsoil scalped to a furrow depth of 200 mm using a Kimseeder. A total of 20 rows were scalped at two metre intervals in which 10 seed lots were sown (2 rows per seed lot). Each seed lot was hand sown into two 68m furrows chosen at random (Fig. 6.2). The rows were selected at random to reduce the influence of soil and microclimate variations at the site.





The seeding rate used in the field was calculated with the intention of establishing 5000 seedlings per hectare (Scheltema 1993) or approximately 1360 stems per 0.27ha using the following formula:

Seeding rate = ______ seedlings per hectare

percentage field factor x viable seeds per gram

The amount of seed sown in the field was different for each seed lot within each species due to variations in the purity and quality per gram. The number of viable seeds per gram was calculated from seed germination trials conducted in Chapter 3. In order to produce the same amount of germinants for each seed lot within each species, differential seed weights were sown in the field.

Seed Lot	Species	Weight of Seed Sown (g)	Viable Seed (g)	Estimated Establishment
424	A. microbotrya	60	• 1 1	33
545	A. microbotrya	160	4	32
		بر ا		
199	A. huegeliana	17	405	344
520	A. huegeliana	18	384	347
			·	·
1474	C. quadrifidus	34	685	233
1490	C. quadrifidus	20	1166	233
1421	E. wandoo	45	104	47
1422	E. wandoo	19	244	46
642	K. prostrata	400	1	20
1365	K. prostrata	50	8	20
	Tota	l		1354

Table 6.2:	Weight (gra	ams) of Each	Seed Lot Sow	n in the F	ield and the
	Estimated I	Establishmen	t of Seedlings	Based on	Seed Viability

per Gram Derived from Chapter 4

In a direct seeding experiment which compared early (June-July) with late (August-September) seasonal sowing times, Pigott *et al.* (1994) found that early sowing of seeds was an important factor influencing seedling survival.

In order to increase the effectiveness of revegetation at the York field site, sowing of seeds corresponded with annual seasonal rainfall which occurs in June, July and August (see Fig. 3.5). The seeds were sown early in the season on the 20th of June 1995.

In an attempt to break the known seed coat imposed dormancy in the Leguminosae group (Cavanagh 1987), *Acacia microbotrya* was exposed to boiling water for 60 seconds and then soaked in cold water for 12 hours. *Kennedia prostrata* received no pre-sowing treatment. The seed treatment methods mentioned above were employed on the basis of maintaining consistency with Chapter 4.

The experimental layout of each seed lot sown into two rows at York was random and stratified in order to take into consideration possible site variability factors influencing the results. The intention was to demarcate each row into three replicate blocks in order for statistical analysis. However, quantitative statistics were not used to analyse the differences between seed biolocalities due to an uneven distribution of germinants within each row. Unusually high rainfall events mobilised existing site soil down slope, causing seed burial and or uneven distribution of seed. Therefore, absolute numbers were recorded instead of sampling from quadrats placed within the replicate blocks within each row. Consequently, statistical support was not available for this trial. All the results are expressed in terms of descriptive observations.

6.4: Results and Discussion

6.4.1 SEED GERMINATION

The number of seeds that germinated was variable between biolocalities for each species (Table 6.3). Local seed lot of *Acacia microbotrya* (46 local, 7 distant) and *Calothamnus quadrifidus* (154 local, 96 distant) recorded a far higher number of germinants relative to the distant biolocality seed lot.

One distant biolocality seed lot which had a high number of germinants relative to local biolocality seed lots (Table 6.3) was *A. huegeliana* (221 distant, 184 local). The variability in seed germination is in part due to seed burial and uneven distribution of seed by high rainfall events.

Seed Lot	Species	Locality	Germinants October'95	Final Estab. June'97	% Survival
424	A. microbotrva	Distant	7	. :	71
545	A. microbotrya	Local	46	39	85
199	A. huegeliana	Distant	221	76	34
520	A. huegeliana	Local	184	140	76
1474	C. quadrifidus	Distant	96	10	10
1490	C. quadrifidus	Local	154	11	
1421	E. wandoo	Distant	10	12	120
1422	E. wandoo	Local	4	2	50
642	K prostrata	Distant	12	6	50
1365	K. prostrata	Local	7	0	0
				·	

 Table 6.3: Comparison of Five Geographically Local Versus Distant

 Biolocality Seed Lots at York.

The soil preparation method employed at York involved the scalping of furrows parallel to the contour of the land. The furrows acted as drainage channels for water and water borne soil from high rainfall events. Soil erosion was detected at the field site in York by observing organic debris wrapped around the uphill side of developing seedling stems (Plate 6.1).

The high rainfall events that occurred in July 1995 at York (see Fig. 3.5) that stimulated germination may also have contributed to depressed germinant emergence. With the exception of *Acacia microbotrya* (seed lot # 545), the number of germinants recorded in the field (Table 6.3) was less than the estimated value in Table 6.2. Rainfall events mobilised existing site soil, causing the burial of native seed to varying depths in the soil profile. Several studies indicate that excessive soil burial of small seeded species such as *Eucalyptus loxophleba* (Pigott *et al.* 1994) and *Eucalyptus salmonophloia* (Yates *et al.* 1996) depressed seedling emergence.

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Uneven distribution in seed germination occurred where the seed soil bank was translocated from an area of high topographical relief (mid-slope) to the northern area of low relief at the York field site. Water erosion may account for the mid-slope of the site being denuded of both weeds and native seedlings and the high stand density of seedlings where the seed had accumulated down slope. Uneven distribution of seed germination may have caused competition for resources from developing germinants and a subsequent decrease in seedling survival (Plate 6.2).

Low germination of *Kennedia prostrata* both in the laboratory (see Chapter 4) and in the field at York (also see Chapter 5) appears to be due to several factors such as dormancy mechanisms (see section 4.6.1) and habitat displacement (see section 5.5.1).

6.4.2 SEEDLING SURVIVAL AND ESTABLISHMENT

The majority of biolocalities within each species had a similar seedling survival over time, which fluctuated according to seasonal availability of moisture (Fig. 6.3 to 6.7). Germination in 1995 and subsequent recruitment of new germinants in 1996 (and to a lesser extent in 1997) occurred in the winter months due to high rainfall events. Seedling mortality occurred in the summer months of 1995 and 1996 which is most likely due to desiccation through moisture stress.

The majority of species recorded seedling recruitment in the second year after sowing which indicates that a proportion of the seeds remain viable in the soil following artificial seed dispersal. However, it is reasonable to assume that the majority of seedlings that germinated in the second year after sowing did not become established in the field, given that the number of established seedlings recorded in 1997 was less than that recorded in 1996. Furthermore, weeds tended to dominate the trial site in the second year due to the absence of soil scalping which may have supressed germinant development

Seedling establishment varied according to the biolocality where the seed was collected (Fig. 6.3 to 6.7). Provenance trials of the widely distributed *Eucalyptus camaldulensis* conducted by Turnbull (1973) found variation in performance between species collected from eight different drainage systems and climate zones.



Plate 6.1: Organic debris wrapped around partially buried seedlings (Acacia microbotrya) directly sown at York caused by water erosion of soil down-slpoe.



Plate 6.2: Uneven distribution in seed germination of *Calothamnus quadrifidus* may have caused competiton for resources and a subsequent decrease in seedling survival.

6.4.2.1: Acacia microbotrya

The seed of *A. microbotrya* (# 545) collected from the local biolocality of York had a higher seedling survival (85%) and establishment (39 seedlings) compared to the survival (71%) and establishment (5 seedlings) of the relatively distant seed lot (# 424) collected from Beverley (Table 6.3 & Fig. 6.3). Seed collected from the biolocality of Beverley (# 424) is only marginally distant in terms of geography relative to York (Fig. 6.1).

The climate of Beverley has a minimal decrease in annual rainfall (31mm) and daily mean temperature (maximum & minimum 0.4°C) relative to the climate of York (Appendix 2). However, even slight variations in environmental parameters may translate to differences in establishment and subsequent survival between the local and distant seed lots in the field. A possible explanation for this variation is that the locally selected *A. microbotrya* (# 545: York) may have a physiological specialisation which favours survival in its local environment relative to the distant seed lot (# 424: Beverley) which may not be as well adapted to the site conditions of York.

6.4.2.2: Allocasuarina huegeliana

The local seed lot of *Allocasuarina huegeliana* (# 520) collected from Brookton recorded higher seedling survival (76%) and establishment (140 seedlings) when sown at York, compared to the survival (34%) and establishment (76 seedlings) of the relatively distant seed lot (# 199) from Mount Barker(Table 6.3). This is despite a higher init comminant survival from the distant seed lot in October 1995 (221 seedlings) compared to the local seed lot (184 seedlings) for the same period (Fig. 6.4). However, seedlings from the Mount Barker biolocality experienced a higher mortality during the first two consecutive summers relative to the Brookton biolocality. A possible explanation is that the seed lot collected from Brookton which receives a similar summer rainfall of 36mm (Appendix 2) to the York field site with 34mm, is more acclimatised to reduced availability of moisture relative to the distant seed lot collected from Mount Barker (77mm).

The biolocality of Brookton is located within the same biogeographical region as York (Avon Wheatbelt), and is distinct from the Mount Barker bioregion (Jarrah Forest). The abiotic conditions which delineate biogeographical regions are climate (especially summer rainfall), lithology/geology and landform (Thackway & Cresswell 1995). These conditions may have influenced certain characteristics within the plant seed which enhanced the performance of the Brookton seed lot when sown in the York environs.

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Figure 6.3: Seedling survival and mortality between local (# 545: York) and distant (# 424: Beverley) provenances of direct seeded *Acacia microbotrya* sown at York for June 1995 to 1997.



Figure 6.4: Seedling survival and mortality between local (# 520: Brookton) and distant (# 199: Mt. Barker) provenances of direct seeded *Allocasuarina huegeliana* sown at York for June 1995 to 1997.

6.4.2.3: Calothamnus quadrifidus

The local seed of *Calothamnus quadrifidus* collected from Wongan Hills (# 1490) had a marginally lower survival (7%) and higher establishment (11 seedlings) compared to the higher survival (10%) and lower establishment (10 seedlings) of the distant seed lot (# 1474) collected from the biolocality of Wanneroo (Table 6.3 & Fig. 6.5). The biolocality of Wanneroo is situated on the coast and is subjected to very different climatic pattern (Appendix 2) to the biolocalities of Wongan Hills and York.

The climate of the Wongan Hills biolocality is similar to that of York with only a marginal increase in the annual daily mean temperature (maximum 0° C & minimum 1° C) and a decrease in annual rainfall of 99 mm (Appendix 2). In contrast, the biolocality of Wanneroo has a warmer annual daily mean temperature (maximum 1° C & minimum 3° C) and receives 418mm more rainfall than York. The Wongan Hills seedlings appear to be marginally more acclimatised to the York environs relative to seedlings from Wanneroo.

6.4.2.4: Eucalyptus wandoo

Eucalyptus wandoo (# 1422) collected from the local biolocality of Yornaning had a lower seedling survival (50%) and establishment (2 seedlings) when compared with the seedling survival (120%) and establishment (12 seedlings) of the distant seed lot (# 1421) from Mt. Barker (Table 6.3 & Fig. 6.6). However, the total number of seedling survival was low for both seed lots. Yornaning is situated on the biogeographical boundary between the Avon Wheatbelt and the Jarrah Forest (see Fig. 6.1) while Mt Barker is situated within the Jarrah Forest bioregion. The Jarrah Forest bioregion is characterised by high rainfall (Yornaning: 622mm versus Mt Barker: 743mm) relative to the Avon Wheatbelt region (York: 451 mm). Therefore, the both biolocalities could be considered 'distant', in terms of geography and climate relative to the site of York. Consequently, seedling performance did not provide a clear selection criterion for biolocality based on geographical distance relative to the trial site.





Figure 6.5: Seedling survival and mortality between local (# 1490: Wongan Hills) and distant (# 1474: Wanneroo) provenances of direct seeded *Calothamnus* quadrifidus sown at York for June 1995 to 1997.



Figure 6.6: Seedling survival and mortality between local (# 1422: Yornaning) and distant (# 1421: Mt. Barker) provenances of direct seeded *Eucalyptus* wandoo sown at York for June 1995 to 1997.

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6.4.2.5: Kennedia prostrata

The seed lot of *Kennedia prostrata* (# 1365) collected from the 'local' biolocality of Wickepin had a 100% mortality of seedlings. The seedling survival (50%) and establishment (6 seedlings) of the seed lot (# 642) collected from Collie had higher seedling survival (Fig. 6.7). The seedlings of *K. prostrata* recorded an overall low seedling survival (6 & 0) for both biolocalities (1365 & 642), respectively. The biolocality of Wickepin is geographically closer to the region of the York field site relative to the biolocality of Collie. However, both seed lot biolocalities experience a different climate to that of York (Appendix 2).

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Wickepin receives 506 mm of annual rainfall and lower mean maximum and minimum temperatures (22.9°C & 9.9°C) relative to the field site at York. Similarly, Collie receives more than twice the annual rainfall (954mm) and lower mean maximum and minimum temperatures (24.1°C & 9.4°C) relative to York. The poor performance of *K. prostrata* seedlings may be indicative of the climatic variations within each respective biolocality and suggests a limited ability of the seedlings to acclimatise to the conditions influencing the York field site.

6.4.3: SEEDLING GROWTH

Growth height appears to be one indicator of the species adaptation to site conditions. The local seedlings of *A. microbotrya* (# 545) had a distinct mean height advantage of 269 mm more than distant seed lot (# 424) seedlings (Fig. 6.8). Similarly, local seedlings of *C. quadrifidus* (# 1490) had a marginal mean height advantage of 66 mm more than seed lot 1474 seedlings (Fig. 6.8). In contrast, the distant seedlings of *A. huegeliana* (# 199) and *E. wandoo* (# 1421) had a mean height (32mm & 103mm, respectively) advantage over local seedlings (# 520 & # 1422, respectively). The seedling growth heights of *K. prostrata* were not comparable between seed lots due to the 100% mortality of the local seed lot (# 1365). However, variations in morphological characteristics such as height may exist between the biolocalities and therefore may not be indicative of a species relative performance in the field.



Figure 6.7: Seedling survival and mortality between local (# 1365: Wickepin) and distant (# 642: Collie) provenances of direct seeded *Kennedia prostrata* sown at York for June 1995 to 1997.



Figure 6.8: Mean seedling growth of five species recorded in June 1997 between local (L) and distant (D) provenances direct seeded at York in June 1995 showing standard error bars.

The local seed lots of *Acacia microbotrya*, *Allocasuarina huegeliana* and *Calothamnus quadrifidus* were collected from biolocalities with a similar climate (Appendix 2) and biogeography to the York field site. These local seed lots showed an enhanced seedling performance (in terms of establishment and growth) relative to distant biolocalities. In contrast, the 'local' seed lots of *Eucalyptus wandoo* and *Kennedia prostrata* collected from biolocalities geographically proximal to the York field site but from a different bioregion with different climatic conditions recorded a lower performance relative to distant biolocalities.

6.5: Summary

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The seed lots of *A. microbotrya*, *A. huegeliana* and *C. quadrifidus* collected from biolocality relative to the revegetation site of York had a higher seedling establishment and growth compared to the seed lots collected from distant biolocalities. These local biolocality seed lots that were collected from areas which had a similar climate and biogecgraphy to the York field site, out performed seedlings from distant biolocalities. The growth and establishment of local seed lot species are influenced by their surrounding environment and may have characteristics (inherent ecotypic specialisation) which favour survival at the York trial site. Conversely, seed lots translocated from distant biolocalities may not be as well adapted to the site conditions of York which in turn results in poor seedling performance.

The seed lots of *E. wandoo* and *K. prostrata* collected from 'local' biolocalities relative to the revegetation site of York had a lower seedling establishment and growth compared to the seed lots collected from distant biolocalities. In general, these 'local' seed lots were collected from areas which had a relatively distant geographical distance and different climate and biogeography to the York field site. Similarly, the distant seed lots were collected from areas which had a relatively distant geographical distance, different climate and biogeography to the York field site. Therefore, the term 'local' and 'distant' may be inappropriate to apply to these seed lot biolocalities. The biolocality selection criteria of geographical distance, bioregions and climate were distant, different and dissimilar respectively, relative to the York revegetation site. Consequently, seedling performance in this case (*E. wandoo & K. prostrata*) is only comparable between the 'local' and 'distant' seed lots relative to each other and not to the site of revegetation.

THE EFFECT OF WEED COMPETITION ON NATIVE SEEDLINGS

7.1: Introduction

Weeds are a major economic concern for land managers of annual crops and pastures in the Avon catchment (Dodd *et al.* 1993). There is limited knowledge of the effect of weeds on the establishment of native vegetation (Runciman 1991; Dalton 1992; Pigott *et al.* 1994). The literature indicates that weed control is crucial in minimising competition for resources exerted on native vegetation (Croft & Venning 1985; Venning 1988; Bulman & Dalton 1990) but this has not been definitively established (Runciman 1991; Dalton 1992).

The infestation of weeds at both the York and Tammin trial sites (Chapter 5) highlights the potential effects on native revegetation. A large amount of capital can be expended on chemicals, machinery and labour in the control of weeds on revegetation sites (Copely & Venning 1983; Bulman & Dalton 1990). To determine if the economic effort directed towards the control of weeds is justified, the influence of weeds on native seedlings should be determined. This knowledge will assist in developing more effective methods of revegetation.

Weeds may compete for limited resources such as soil moisture, nutrients and light. If weeds are competitive in relation to other plants then they could retard the development of native seedlings (Clemens & Radford 1986). However, weeds may also provide a benefit to germinants by offering protection from environmental factors such as wind, frost and sun. Therefore, the relation between weeds and native seedlings may be dependent on specific impact assessment criteria. The impact of weeds on the growth and survival of native seedlings can be used as a measure of the benefit of detriment incurred by weeds. Consequently, the issue of the impact of weeds on seedlings is one of an overall interactive effect depending on a range of variables that need to be determined.

7.2: AIM

To improve our understanding of seedling establishment by assessing the effects of weeds on growth and survival of native plants that were direct seeded or planted as nursery seedlings.

7.3: Methods

The experiment was conducted in an irrigated outdoor area at Edith Cowan University, Joondalup (Western Australia) using containerised plants. The irrigation system delivered scheme water through four micro-sprinklers mounted on 1.4 metre high risers. The radius of each wetting zone from the overhead sprinklers extended beyond the base of each riser.

The collective output of the sprinklers was measured by randomly distributing 16 glass beakers (1000ml) across the wetting zone of the site. The beakers were placed in a stratified randomised design to take into consideration the differential watering pattern and other environmental gradients (photoperiod, wind *etc.*). The irrigation system was run for thirty minutes and the volume of water in each beaker was measured. A 26% difference was observed in the range between the highest water volume and the lowest water volume collected in a beaker.

The irrigation regime was set at 6 *am* and 3 *pm* for five minutes at the rate of 15ml per pot every day in the period from April to September and 6 *am* and only for ten minutes at the mean rate of 15ml per pot every third day in the period from October to November. The variation in the watering regime was intended to simulate moisture stress in the field and was compounded by the low natural rainfall events for the months of October and November (Table 7.1).

Month 1997	Mean Minimum (°C)	Mean Maximum (°C)	Monthly Rainfall (mm)
January	18.5	32.4	0.6
February	19	31.3	4
March	15.9	27.5	54
April	14.7	25.7	39
May	10.5	20.7	81
June	9.7	19 .6	105
July	4.4	18.5	138
August	6.8	18.4	119
September	9.3	20.2	109
October	11.8	23.5	42
November	13.2	25.5	8
December	16.9	30	0

Table 7.1: Climate Data for Perth in 1997, Showing the Mean Daily Minimum and Maximum Temperatures and Monthly Rainfall

Note. Source: Bureau of Meteorology (1998).

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The native plant seedlings chosen for the experiment represent common species of various height strata in an undisturbed native vegetation community within the Avon catchment. However, prostrate ground covers were excluded from the trial due to limited availability. The three species of plants used in this experiment are: *Allocasuarina huegeliana* (seed lot # 940520), *Calothamnus quadrifidus* (seed lot # 940995) and *Eucalyptus wandoo* (seed lot # 930329) which have been examined in previous chapters.

The plants were established in the pots by both direct seeding and transplanting nursery seedlings. The seeds sown were obtained from the same biolocalities as those used to establish the nursery seedlings. The seedlings were germinated and grown in compressed pine bark pots by Alcoa Australia Pty. Ltd. and were five months of age when transplanted into the containers.

The nursery seedlings were planted into growing media in high-density polyethylene (HDPE) black plastic pots (140mm). The growing media consisted of crushed pine bark (*Pinus radiata* 37%), sawdust (*Jarrah marginata* 40%) and quartz gravel (23%). The height and stem width of each seedling was recorded at the time of planting.

A quantity of seed was directly sown into identical pots and growing media and placed under glasshouse conditions for a period of two months. Excess germinants were pricked out of each container leaving only one seedling. The mean net weights for both nursery seedling and direct seeded pots after saturation to field capacity was 960 grams after 12 hours of drainage.

The predominant weed found at the trial sites at York and Tammin (Chapter 5 & 6) were capeweed (*Arctotheca calendula*) and to a lesser extent blue lupin (*Lupinus cosentinii*). Both weeds were therefore selected for use in this experiment. *A. calendula* was transplanted bare rooted without soil at the end of June from a property in Joondalup where no red legged earth mites were detected. The weed transplant method was used in preference to seed due to the unsuccessful germination of capeweed seeds in a pilot trial. The average wet weed weight was 17 grams prior to transplanting out into the pots. Two capeweed plants were transplanted around each native seedling. *L. cosentinii* seed was harvested from the York field site and sown directly into the containers. Four lupin seeds were sown in each pot to a depth of 10 millimetres around the native seedlings.

The experimental design consisted of a total of 240 pots of which 120 pots were direct seeded and 120 pots planted with nursery seedlings (Fig. 7.1). Of the 120 containers for each method of plant establishment, 60 pots contained weeds and the other 60 pots were weed excluded. The three native plant species were allocated 20 pots for each treatment (weed & weed excluded). Each species was divided into four blocks (replicates) of five pots. Of the 20 pots containing weeds for each native species, 10 contained transplanted *A. calendula* seedlings while the 10 remaining pots were direct seeded with *L. cosentinii*.

The trial commenced on the 30th of April 1997 and ended on the 30th of November 1997. The seedlings were monitored at the end of each month by measuring the height and stem width of each seedling. The height was measured with a ruler from the soil level to the highest active terminal bud while the stem width was measured at 10mm above the soil level using Vernier Callipers. Stem caliper measurements were not taken for direct seeded seedlings due to the limited expansion of the stern width in the given time frame of the experiment.

The data was analysed for equality of variance for each treatment using an F-test. The data were considered to have equal or similar variances and therefore parametric statistics were employed using an independent t - test (Fowler & Cohen 1993).

7.4: Results

7.4.1: WEED COMPETITION IN NURSERY SEEDLINGS

Competition from weeds had a substantial effect on the growth of native nursery seedlings (Table 7.2). The mean height growth of *Allocasuarina huegeliana*, *Calothamnus quadrifidus* and *Eucalyptus wandoo* seedlings without weeds was greater than those seedlings with weeds (Table 7.2). The difference in height growth was significant between the two treatments (Table 7.2). There was no significant difference in growth between seedlings subjected to *A. calendula* and *L. cosentinii*.
Chapter 7



Figure 7.1: Schematic overview of experimental design (stratified randomised design not shown) showing nursery seedlings and direct seeded seedlings subjected to no weed treatment and weed treatment.

Table 7.2: Comparison of Mean Height Growth Between Weed TreatmentVersus the Weed Excluded Control for Three Species ofNursery Seedlings Showing Standard Error, t value andProbability.

Species	Treatment	Height (mm)	±S.E.	t	p(2-tailed)
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A. huegeliana	Weeds	39.5	9.1	2.9	0.006
A. huegeliana	No weeds	83.7	12.1	• .	
C. quadrifidus	Weeds	20.9	3.0	5.6	0.0001
C. quadrifidus	No weeds	47.0	3.6		
E. wandoo	Weeds	17.3	4.5	3.7	0.001
E. wandoo	No weeds	44.1	5.8	•	· .

The analysis of mean stem width within species revealed that only A huegeliana was significantly different (p < 0.04). C. quadrifidus and E. wandoo were not significantly different in growth stem width (Table 7.3).

Table 7.3: Comparison of Mean Stem Width Between Weed Treatment Versus the Weed Excluded Control for Three Species of Nursery Seedlings Showing Standard Error, t value and Probability.

Species	Treatment	Stem (mm)	±S.E.	t	p (2-tailed)
A huegeliana	Weeds	1.0	0.3	2.2	0.04
Ahuegeliana	No weeds	1.7	0.2		
C. quadrifidus	Weeds	1.7	0.2	1.0	0.3
C. quadrifidus	No weeds	2.0	0.2	.•	···
E. wandoo	Weeds	0.6	0.1	0.7	0.5
E. wandoo	No weeds	0.7	0.1		· .
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7.4.2: WEED COMPETITION IN DIRECT SEEDED SEEDLINGS

Within species analysis revealed that only *C. quadrifidus* was significantly different in height growth between treatments for direct seeded seedlings (Table 6.4). *A. huegeliana* and *E. wandoo* were not significantly different in height growth between treatments for direct seeded seedlings (Table 6.4).

Table 7.4: Comparison of Mean Height Growth Between Weed treatmentVersus the Weed Excluded Control for Three Species ofDirectSeeded Seedling Showing Standard Error, t value andProbability.

Species	Treatment	Height (mm)	±S.E.	t	p (2-tailed)
A huegeliana A huegeliana	Weeds No weeds	22.8 28.0	7.3 5.7	0.6	0.6
C. quadrifidus C. quadrifidus	Weeds No weeds	5.9 17.9	2.1 2.1	4.1	0.0001
E. wandoo E. wandoo	Weeds No weeds	15.4 23.9	2.6 3.7	1.9	0.7

7.4.3: GROWTH OF NURSERY SEEDLINGS

A. huegeliana and C. quadrifidus seedlings with weeds maintained a distinct growth advantage relative to seedlings without weeds between the months of May through to September (Fig. 7.2 & 7.3). However, the seedlings with weeds experienced a dramatic growth reduction in the month of October and continued to decline through November. Conversely, the seedlings of A. huegeliana and C. quadrifidus without weeds accelerated in growth rate from September to November 1997.



Figure 7.2: Comparison of mean height growth in nursery seedlings of *Allocasuarina huegeliana* with weed and without weed treatment.





The growth of both height and stem width for *E. wandoo* seedlings without weeds maintained a distinct growth advantage over seedlings with weeds. *E. wandoo* seedlings showed a rapid increase in growth from October to November (Fig. 7.4). Conversely, the nursery seedlings of *E. wandoo* with weeds experienced a decline in growth during the same period between October and November.

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Seedling height growth for *A. huegeliana* had a similar trend between the two treatments (weed/no weed) for direct seeded seedlings (Fig. 7.5). Direct seeded seedlings of *A. huegeliana* experienced a high growth rate from May to June and tapered off over late winter (July/August) with a sharp decline during spring (September, October & November). However, the *A. huegeliana* seedlings with weeds maintained a slight height advantage over those without weeds until the month of November when the situation was reversed.

Direct seeded seedlings of *C. quadrifidus* with weed and without weed was similar in growth from June to July (Fig. 7.6). Seedlings without weeds had a height advantage and a limited growth rate in August and a rapid growth in height during spring (September, October & November). Conversely, the growth rate of seedlings with weeds declined in August/September increased only gradually during the months of October and November.

Seedling growth (height) of *E. wandoo* without weeds showed a slight height advantage over seedlings with weeds (Fig. 7.7) during the winter period (June, July and August). However, the disparity between growth rates of the two treatments became greater during the months of October and November. Seedlings without weeds experienced a rapid increase in growth while seedlings with weeds receded in growth.

Nursery seedlings of *E. wandoo* without weeds were the only native plants which recorded a higher percentage survival (100%) relative to seedlings with weeds (90%) in Figure 7.8. However, weed competition affected direct seeded seedlings of *A. huegeliana* (45%), *C. quadrifidus* (35%) and *E. wandoo* (70%) recorded a lower seedling survival rate relative to seedlings without weeds (60%, 85% & 75% respectively).

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Figure 7.4:Comparison of mean height growth in nursery seedlings of Eucalyptus
wandoo with weed and without weed treatment.





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Figure 7.6:Comparison of mean growth (height) in direct seeded seedlings of
Calothamnus quadrifidus with weed and without weed treatment.



Figure 7.7: Comparison of mean growth (height) in direct seeded seedlings of *Eucalyptus wandoo* with weed and without weed treatment.



Figure 7.8: Comparison of percentage survival of direct seeded seedlings (D.S.) with nursery seedlings (N.S.) with weed and without weed treatment for *Allocasuarina huegeliana, Calothamnus quadrifidus* and *Eucalyptus wandoo.*

7.5: Discussion

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The growth of nursery seedlings appears to be positively influenced by the presence of weeds when moisture is available but has a detrimental effect on the overall growth of seedlings when soil moisture is limited. Nursery seedlings of *Allocasuarina huegeliana* and *Calothamnus quadrifidus* maintained a distinct height advantage relative to seedlings without weeds between the months of May to September/October. These months represent a period of low temperatures (see Table 7.1) which may be detrimental to a developing seedling. Therefore, the presence of weeds insulating the soil surface may have increased soil temperatures, which improves conditions suited for seedling growth. Weeds may have also assisted the development of a biological layer of organic material, which further increases soil temperatures through microbial decomposition.

Nursery seedlings of *Allocasuarina huegeliana*, *Calothamnus quadrifidus* and *Eucalyptus wandoo* achieved a significantly greater seedling height when weeds were excluded relative to seedlings subjected to weeds. *Allocasuarina huegeliana* was the only species to record a significantly greater stem width when weeds were excluded relative to when seedlings were subjected to weeds. *Arctotheca calendula* and *Lupinus cosentinii* appeared to have had a significant effect on nursery seedling height growth and to a lesser extent on stem width.

Nursery seedlings of *Allocasuarina huegeliana, Calothamnus quadrifidus* and *Eucalyptus wandoo* experienced 'negative' height growth compared to the seedlings which were weed excluded. Negative growth was recorded when the active terminal leader 'died back'. The dieback in nursery seedlings corresponded with the drier period (October/November), during which the overhead irrigation regime was reduced from 15ml per day to 15ml every third day.

The reduction in irrigation watering time coincided with an increase in the ambient temperature. The increase in ambient temperature was due to a transition from the cooler winter season to the warmer spring season, which subsequently caused an increase in evaporation from the growing media and transpiration loss from seedlings. Water stress was observed in a large proportion of the nursery seedlings with weeds where the apical terminal receded (permanent wilt point) and in some cases the leaves defoliated. On the basis of the recorded growth height and the relative incidence of apical recession, it may be concluded that the presence of weeds had a significant effect on the overall performance of native seedlings. This is most likely due to competition for water in the growing medium. Nursery seedlings in the absence of weeds did not fully extend their root systems into the maximum capacity of the growing medium in the container by the end of trial period. The medium was found to be very friable when the nursery seedlings with inverted and the container removed. In contrast, nursery seedling and weeds grown in the same container produced an extensive root system, which formed a firm 'root ball' when the container was removed. In a study conducted by Whitham (1982), both root and shoot development of native seedlings was restricted by perennial ryegrass (*Lolium perenne*). Consequently, root competition from weeds for air space within the medium is a probable causal agent in the suppressed growth of nursery seedlings, with weeds.

The survival of the nursery seedlings did not appear to be affected by competition from weeds with the exception of *E. wandoo*, which was marginally disadvantaged, by weed competition. Mortality of direct seeded seedlings was higher for all seedlings with weeds compared to those without. Direct seeded seedlings appear to be more susceptible to the presence and growth of weeds than the more established nursery seedlings.

The mortality of direct seeded seedlings growing with weeds occurred in spring, which coincided with a reduction in irrigation watering times and seasonal rainfall. A root 'mat' was also observed at the bottom of containers with weeds compared to an open root system in containers without weeds. This may be due to the relatively advanced root systems of weeds that were transplanted *(Arctotheca calendula).* These weeds thrived in those pots which native seed was directly sown, probably because they had had minimal competition for light and root space from the developing native germinants. Therefore, mortality of direct seeded seedlings is suspected to be a result of periodic soil moisture deficits and limited root space caused by the competition by weeds.

The growth height of direct seeded *C. quadrifidus* was significantly different between weed and weed excluded containers. Similarly, both *A. huegeliana* and *E. wandoo* that were weed excluded had a greater mean height than those seedlings with weeds but were not significantly different. It is generally accepted that weeds have a competitive advantage when accessing available resources, which has a detrimental affect on the growth and survival of native seedlings (Messenger 1976; Croft & Venning 1985; Clemens & Radford 1986; Bulman & Dalton 1990; Hobbs 1991b). This is particularly evident when moisture is a limiting factor (Venning 1988), which is compounded in a containerised environment.

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Caution must be applied when extrapolating results from the nursery environment and applying them to field conditions. The limitation of the weed trial and the subsequent inferences drawn from the results must take into consideration the plethora of environmental variables acting upon field conditions relative to the nursery environs. Furthermore, the transplanting of capeweed may not be relevant in the field situation as the planting and seeding of native plants into prepared ground does not occur amongst semi-mature weeds. However, in an attempt to replicate field conditions, the direct seeding of lupin seed demonstrated the effects of weeds on native seedlings.

7.6: Summary

The growth of nursery seedlings appears to be positively influenced by the effect of weeds when moisture is available but has a detrimental effect on the overall growth of seedlings when soil moisture is limited. Nursery seedlings of *Allocasuarina huegeliana* and *Calothamnus quadrifidus* maintained a distinct height advantage relative to seedlings without weeds between the period of May to September/October. Conversely, in the period between October and November, weeds had a negative influence on all three species of nursery seedlings with a significant difference in growth height relative to seedlings that were weed excluded.

Weeds appeared to act as insulators of nursery seedling root systems, shielding them against cold temperatures during the winter months when moisture was available. However, weeds also appeared to contribute to soil moisture deficits in periods of limited water availability, which was attributed to the competition for water within the limited volume of the growing medium. Consequently, weeds appear to have a positive effect on the development of nursery seedlings during cooler months but have a detrimental effect on the growth of seedlings during transitional months when water becomes a limiting factor. It may be assumed that once the weeds die in summer, they provide valuable microclimate advantages to native seedlings.

Nursery seedlings co-planted with weeds produced an extensive root system within the soil filled container, relative to containerised environments that were weed excluded. Growth of the above ground portion of nursery seedlings may have been limited by the availability of air space within a container through the effects of competition from weeds. Consequently, competition for air space from weeds is a probable causal agent in the suppressed growth of nursery seedlings.

The survival of the nursery seedlings did not appear to be affected by competition from weeds. However, mortality of direct seeded seedlings was higher for all three species with weeds compared to those without. Weeds had a greater influence on the survival of direct seeded seedlings because they grew more vigorously in those containers with minimal competition (direct seeded seedlings) than those which contained established nursery seedlings. The mortality of direct seeded seedlings with weeds occurred spring which corresponds with a reduction in irrigation watering times and seasonal rainfall. Therefore, mortality of seedlings may be due to soil moisture deficits and limited root space caused through the competition by weeds.

Direct seeded seedlings free of weeds had a greater mean height than those seedlings, which were subjected to weeds. Weeds have a competitive advantage for available resources, which has a detrimental effect on the growth of direct seeded seedlings when moisture is a limiting facto.

The results indicate that competition for moisture and root space from certain weeds (*A. calendula* and *L. cosentinii*) is a probable causal agent in suppressing growth of nursery seedlings and the growth and survival of direct seeded seedlings. These results emphasise the need for effective weed control in the field. This is particularly critical in the transitional months (spring) when annual weeds are still actively growing, the ambient temperature is increasing and natural rainfall patterns are greatly reduced. Therefore, an effective revegetation programme is to a large degree subject to weed competition, which greatly affects the survival and development of native seedlings.

Conclusion

8.1: Introduction

Landuse practices within the central Avon catchment have not been in balance with the environmental processes relating to the soil type, water budget and climatic characteristics of the region. This imbalance has resulted in the land becoming degraded and the agricultural community being detrimentally affected in ecological, financial and social terms.

The extensive and expanding changes in the landscape, present a challenge to rural communities to develop appropriate methods of using native plants for revegetation. The developing practice of revegetating the landscape requires specific knowledge to direct land managers to adopt the most effective methods available. The success of such methods should be measured in terms of ecological and cost effective establishment of plants on a broad-scale.

The practise of revegetating the landscape needs to focus on the time frame involved in establishing native plants. This period should be viewed within the time it takes for plants to regenerate and mitigate the impact of degradational forces acting upon the site and in the catchment. The most appropriate method may take decades to be evident because it involves the long-term impact on the processes causing land degradation.

The process of revegetation involves seeds germinating, the germinants becoming seedlings and the seedlings developing into sexually mature plants. In context, this thesis has focussed on the developmental phase of seedling establishment. This is a crucial stage in the long-term effectiveness of a revegetation programme because developing seedlings are extremely vulnerable when becoming established in the field. The greater the number of seedlings to become established, the larger the number of plants capable of reaching maturity and continuing the cycle.

This thesis has contributed to the development of effective methods of revegetation by identifying the following key issues that relate to the establishment of native seedlings in the field.

8.2: Seed Lot Purity and Quality

je Z The number of seeds that germinated per weight of seed lot varied considerably between seed lots of the same species. This variation was attributed to the quantity of non-seed material and non-viable seed within each sample. The purity of the sample dictates the number of seeds that have the potential to germinate in the field. The main causal agent of seed impurity between seed lots was found to be in the process of post harvest handling of seed batches.

Seed lot purity is dependent on time, money and the apparatus employed to separate seed from chaff (clean) which varies between each seed handler. From a seed merchant's perspective, the ability to clean seed is dependent on the size of the seed and the nature of the seed relative to non-seed material. The degree of difficulty in separating seed from chaff increases the cost of seed that is in turn transferred onto the landholder.

The factors affecting seed lot viability (viable seed: dormant & dead seed) are also related to the processing of seed (collection, storage & treatment) and environmental influences (inbreeding depressions & adverse conditions) that have affected the parent plants. These contributing influences occur prior to the sale of seed and are not factored into the price, upon purchase. Therefore, a decrease in the viability of any one seed lot is directly passed onto the landholder.

The implications of seed lot quality (impurities and non-viable seed) for landholders is a factor of time and economics. These factors have huge ramifications for direct seeding on a broad scale. The labour vested in sowing non-seeding material will result in a follow up seeding programme due to poor establishment rates in the field. A decrease in the number of stems per/ha will take longer to mitigate the problems associated with land degradation. Therefore, in order to save both labour costs and establishment times, the conducting of a germination test is of paramount importance. However, the cost of plant establishment in the field using direct seeding is unnecessarily increased because farmers still have to pay for non-seeding material.

The practical implication of germination tests for direct seeding are that the seeding rates may have to be adjusted according to the planting densities required in the field. For example, if a landholder requires a certain number of stems per/ha, then a viable seed to weight ratio may have to be employed.

Similarly, when preparing a mixture of seed for sowing with the aim of establishing a plant assemblage, excessive canopy species that have high germination should only comprise a minor proportion of the seed mixture to ensure that it does not have a deleterious effect (eg. competition for resources) on understorey species. A differential sowing rate could be calculated on the assumption that the environmental factors influencing seed lot quality in the field are the same as those in the laboratory after taking field factor estimates into consideration.

The promotion of broad-scale direct seeding in agricultural regions depends on effective germination for the weight of seeds sown in the field. The variable quality between different seed lots, both within and between different seed merchants reinforces scepticism within the agricultural community towards direct seeding as a reliable method of revegetation.

Currently, there is no 'quality assurance' associated with the purchase of native plant seed, such as a certificate depicting seed viability per gram. Consequently, direct seeding as a method of revegetation will continue to produce erratic results in the field until quality assurance is guaranteed with each seed lot purchased from a registered seed merchant. It is recommended that a third party conducts quality assurance in order to remove the vested interest in the results within the industry.

Quality assurance associated with seed purchase would enable a costing structure to be based on a quality to weight ratio rather than a 'blanket' cost per species based on weight alone. A costing structure based on viable seed per gram would also ensure that seed collection and handling are optimised for the time and money invested, resulting in higher purity and quality seed lots. Seed lots of high purity and seed viability provides the basis to an effective revegetation programme in agricultural regions.

8.3: Biolocality and Temperature

Seed germination varied for the same species collected from different biolocalities, indicating that the site where the seed is collected from was important. Temperature had an influence on the germination capacity of different seed lots.

The interaction between biolocality and a range of laboratory temperatures affected seed germination. This indicated that the ambient temperature of the biolocality where the seed lot was collected is an important consideration in biolocality selection. Consequently, seeds collected from a local environment recorded higher plant establishment and growth than species collected form environments distant from the site of revegetation.

The implication of these results for landholders direct seeding in the field, is that if the mean winter temperature of the revegetation site is equivalent to the optimum germination temperature for a particular seed lot, then climatic matching of that biolocality with the revegetation site may be an important factor in biolocality selection and subsequent plant establishment. Consequently the selection of seeds for revegetation from a biolocality of similar climate and proximal to the site of revegetation is important. From an ecological perspective, the selection of a biolocality for revegetation would ideally result in a sustainable plant assemblage that would integrate with the existing floral and faunal communities. Therefore, the selection of local species for a revegetation programme maintains the genetic integrity of the species within the area.

8.4: The Effect of Frost on Seed Lot Viability

The influence of sub-zero temperatures on the number of seeds that germinated was negligible. However, when seeds imbibed moisture, seed mortality decreased marginally over time because of embryo damage. Therefore, the implication of this result for landholders sowing seeds during the winter season should not be dictated by the timing of frost conditions but other environmental factors.

Environmental factors influencing seed dormancy mechanisms in the Avon Basin are likely to be broken by storage in hot, dry conditions rather than moist, cold stratification. However, research into the effects of various storage regimes on seed germination is seriously lacking in the knowledge of germination ecology of Western Australian species.

8.5: Direct Seeding versus Nursery Seedlings

The short-term evaluation of the relative merits of these two methods is indicated by the total number of plants that were established at the end of the trial period. Plant establishment in the field was substantially higher for seedlings derived from direct seeding than from transplanted nursery seedlings. This is provided that seed dormancy mechanisms that operate within a few species must be broken with the application of an optimum treatment prior to sowing. The unit effort and cost of establishing a seedling from seed sown in the field is substantially less than for a transplanted seedling. However, the important phase in evaluation of these two methods is how they contribute to the amelioration of land degradation problems acting upon a revegetated site in the long term.

Land degradation in the Avon basin is extensive. The impact from these degradational forces has yet to be fully realised due to the time lag between the cause (land clearing) and effect (salinisation, water logging etc. From a practical perspective, the most effective revegetation method would decrease the rate of degradation in the landscape on a broad scale and provide a maximum return for the cost and time invested. Consequently, effective long-term amelioration of land degradation problems in the Western Australian wheatbelt is in the broad application of direct seeding methods.

Planting nursery grown seedlings was also a successful method of establishing plants in the field. The survival rate of nursery seedlings was higher and maintained a distinctive growth advantage relative to the number of seeds that germinated and subsequently became established in the field. Nursery seedlings are an important method of application in situations that are small and site specific (steep slopes, salt scolds etc.) provided that adequate attention is given to the development of a structurally sound root system.

A culmination of environmental factors such as low rainfall, predation and weed competition had a detrimental effect on native seedling development at the Tammin field site. Under these extreme conditions both direct seeding and nursery seedlings methods of revegetation were unsuccessful. The failure to control red legged earth mites, rabbits and weeds emphasises the need to employ a post sowing/planting management programme.

8.6: Biolocality Comparison in Direct Seeding Establishment

The seed lots collected from a biolocality near to the revegetation site had higher seedling establishment and growth compared to the seed lots collected from distant biolocalities. These near biolocality seed lots that were collected from areas, which had a similar climate and biogeography to the site of revegetation relative to distant biolocalities. The growth and establishment of local seed lot species are influenced by their surrounding environment and may have characteristics (inherent ecotypic specialisation) which favour their local range. Conversely, seed lots translocated from distant biolocalities may not be as well adapted to the site conditions of York which in turn results in poor seedling performance.

The criteria for biolocality selection based upon seedling performance should not be founded on any one factor in isolation but upon identifying the overall similarities between the environmental parameters of a particular seed biolocality and that of the site to be revegetated. Therefore, the terminology of 'local' and 'distant' should be used to encompass climate, biogeographical regions and soil properties where possible, which will often reflect geographical distance from the site of revegetation. The integration of both bioregions and climatic factors often influences the selection criteria in favour of local biolocalities. The use of local seed maintains the genetic integrity of any remnant vegetation surrounding the revegetation site.

8.7: The Influence of Weeds on Seedling Establishment

Weeds assisted the growth of seedlings in a controlled environment when resources were available. The value of weeds as a method of stabilising the soil and protecting seedlings when they are vulnerable was found to be considerable. However, when resources were limiting, weeds had a detrimental effect on native seedlings. The growth of planted nursery seedlings was reduced when co-planted with weeds. Similarly, weeds reduced the growth and survival of seedlings derived from direct seeding. Therefore, the control of weeds is crucial for an effective revegetation programme at specific times is important. This is particularly important in the initial establishment phase of seedlings when moisture becomes a limiting factor in the transitional months from winter to spring.

8.8: Summary

Direct seeding is the most cost effective method of establishing a large number of seedlings in the field provided that the optimum purity and quality is taken into consideration when selecting a particular seed lot. Germination trials are an important measure of dormancy responses, seed purity and biolocality variation in seed lots. This is crucial for maximising germination in the field. The selection of a local seed lot proximal to a revegetation site increases seedling establishment while maintaining the genetic integrity of the vegetation in the region. Post sowing/planting maintenance is an important component in the process of revegetation in order to ensure that the greatest number of seedlings becomes established in the field. The coexistence of weeds can contribute to the survival and growth of seedlings in a revegetation programme when conditions are favourable. However, weeds compete more effectively for resources than native seedling and therefore should be controlled particularly in periods of limited water availability.

Reversing the trend in land and ecological degradation within the central Avon catchment can only be achieved through a revegetation programme that integrates nature conservation with agricultural productivity. Current land management practises and the conservation of the remaining remnant vegetation will not maintain a sustainable agricultural based economy or adequately represent existing biodiversity in the central Avon catchment. If the central Avon wheatbelt is to continue to function as the 'bread basket' of Western Australia, then the establishment of native vegetation can only be achieved through the use of direct seeding methods on a broad scale.

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MORPHOLOGICAL DESCRIPTIONS, FLOWERING TIME AND DISTRIBUTION DETAILS IN REVEGETATION SPECIES (Adapted from Marchant *et al.* 1987)

1.1: Acacia microbotrya (Manna gum)

DESCRIPTION: A. microbotrya is a small bushy tree to 5 m with smooth grey bark becoming dark with age. Branchlets are glabrous and pendulous in habit. Phyllodes usually falcate, 70-150 x 5-15 mm, glaucous with a central midrib. The inflorescence is a raceme with 3-25 heads per raceme and 3-4 mm across, globular and pale yellow in colour. Pod dark grey to black in colour, linear to narrowly oblong and constricted between the seeds, 70-180 x 6-8 mm with margins red in colour. Seeds are longitudinal in the pod, ellipsoid, 6-7 x 4 mm.

FLOWERING TIME: April to July.

DISTRIBUTION: Extends from the Murchison River through the wheatbelt, southwards to Ravensthorpe.

1.2: Acacia saligna (Western wattle)

DESCRIPTION: A. saligna is a small bushy tree to 5 m with smooth light grey bark. Branchlets are pendulous in habit. Phyllodes usually falcate, $90-170 \times 5-15$ mm, with a central midrib. Inflorescence is a raceme with 5-30 heads per raceme and 3-4 mm across, globular and pale yellow in colour. Pod dark grey to black in colour, linear to narrowly oblong and constricted between the seeds, 70-180 x 6-8 mm. Seeds longitudinal in the pod, black, ellipsoid, 6-7 x 4 mm.

FLOWERING TIME: April to July.

DISTRIBUTION: Extends from Geraldton southwards along the coast through to Esperance, associated with limestone soils.

1.3: Allocasuarina huegeliana (Rock sheoak)

DESCRIPTION: *A. huegeliana* is a tree up to 8m. Branchlets have numerous internodes usually 15-25 mm long. The tree is dioecious, the male spike is distinctly pedunculate with scale leaves (bracts) occurring in whorls of 8-10 with varying width teeth. Alternatively, the female spike is ovoid. The cone is reddishbrown and globular in shape, 15-30 mm x 15-20 mm long. The fertile seed are dark brown with colourless wings, 8-11 mm long.

FLOWERING TIME: May-January

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DISTRIBUTION: Extends from Geraldton to East of the Fraser Range, associated with granitic outcrops

1.4: Calothamnus quadrifidus (One-sided bottlebrush)

DESCRIPTION: *C. quadrifidus* is an erect shrub, 1-2 m high with multiple branching at the lower base of the trunk. Leaves densely crowded, blade terete or slightly compressed, 18-55 x 0.8-1.5 mm. Flowers 4-merous. Floral tube 3-4 mm long and glabrous. Sepals 2mm long with the petals 5-6 mm in length. Stamens are red in colour occurring in bundles. Capsule usually 7-9 mm long, broadest at the centre, usually smooth with 2 prominent terminal sepals.

FLOWERING TIME: August to December

DISTRIBUTION: Extends north from Dwellingup, associated with limestone soils close to the coast and in heavier soils on or near the Darling scarp. Extends from Shark Bay to Israelite Bay and inland to Norseman.

1.5: Casuarina obesa (Swamp sheoak)

DESCRIPTION: *C. obesa* is a tree to 10 m. Branchlets are glaucous with numerous internodes 5-15 mm long. The scale leaves occur in whorls of 12-15 with appressed teeth to the stem. The male spike is pedunculate and cylindrical whilst the female spike is globular. The cone is pale brown and ovoid, 10-22 x 12-20 mm. The seed is straw coloured to grey, 5-7 mm long, glabrous with the wing obtuse and a definite opaque line along one lateral margin.

FLOWERING TIME: Recorded through out the year.

DISTRIBUTION: This species extends from the Murchison River to Israelite Bay and inland to Kalgoorlie, associated with saline flats, rivers and winter-wet depressions.

1.6: *Eucalyptus kondininensis* (Kondinin blackbutt)

DESCRIPTION: The Kondinin blackbutt is a tree up to 15 m high with a bole up to 0.8 m in diameter. The lower bark is rough, black and fibrous to 4 m with smooth, grey bark in the upper branches. It has a spreading habit with youngest branchlets, angular and red. The mature leaves are pale green, glossy, narrowly lanceolate, 8-12 cm long and 1-2 cm wide. Up to 7 creamy white flowers form the umbel with a corrugated, yellow in colour, horn-like projecting bud cap. The fruit is bell-shaped, smooth green when fresh turning wrinkled when dry to a length and width of 5-6 mm. The fertile seeds are dark red-brown, irregularly D-shaped, 2-3 mm long a marked with a net-like pattern. The sterile seeds are dark red-brown, narrowly wedge-shaped to 1-2 mm.

FLOWERING TIME: November to December

DISTRIBUTION: *E. kondininensis* is found from South-East of Quairading to Kondinin and Southward to the area of Newdegate, Lake Grace and Pingrup. It usually occurs in loamy soils near salt lakes.

1.7: *Eucalyptus wandoo* (white gum)

DESCRIPTION: Wandoo is a tree up to 21 m with a bole up to 8 m and a diameter of 0.6-1 m, with a wide spreading canopy. The bark is smooth with a mottled white and grey pattern. The juvenile branchlets are a powdery grey. The mature leaves are alternate, narrowly lanceolate, dull grey-green 8-12 cm long and 1.5-2.5 cm wide. There may be up to 15 horn-shaped flowers, white to creamy in colour with a conical bud cap. The fruit is cylindrical to a length of 0.6-1 cm and 5-8 mm wide. The fertile seeds are light brown, almost round and smooth with a net-like pattern to 1 mm in diameter. The sterile seeds are red-brown, wedge-shaped, about 1 mm long.

FLOWERING TIME: Variable geographically, common from November to April.

DISTRIBUTION: *E. wandoo* occurs from near Three Springs southward to the Kalgan River and in the wheatbelt, extending sparsely to Karalee with prominent stands in the Boddington Toodyay area. Wandoo usually grows in sandy-loam, which may have a gravel component with a clay subsoil.

1.8: Kennedia prostrata (Running postman)

DESCRIPTION: *K. prostrata* is a twining pubescent creeper. Leaflets are broadly ovate, 11-55 x 11-43 mm, glabrous with undulating margins. Flowers occur in pairs on auxiliary peduncles, bracts broadly ovate and deeply incised. Corolla is red in colour, standard reflex, sometimes with a yellow eye, 17-26 mm long and wings 14-24 mm long with an incurved 20-30 mm keel. Pod turgid to slightly compressed and narrowly cylindric, 24-57 x 6-8 mm.

FLOWERING TIME: July to November

DISTRIBUTION: Extends North to Northampton, inland to Southern Cross and South to the Stirling and Porongurup Ranges, stretching East along the Southern coastline to Esperance.

1.9: *Melaleuca cuticularis* (Salt water paperbark)

DESCRIPTION: Tree up to 7 m in height with chartaceous bark. Juvenile branchlets are pubescent. Leaves are opposite, shortly petiolate with the blade narrowly ovate to elliptical, 7-11 x 3mm. Inflorescence is on a terminal head and globulate. Flowers white to cream in colour with a floral tube 3-4 mm long and glabrous. Sepals are triangular with persistent petals 3-4 mm long. Capsule almost globular, usually 6-7 mm long often retaining 5 spreading woody sepals.

FLOWERING TIME: September to November

DISTRIBUTION: Occurs in sands over limestone cliffs along the coastline and bordering rivers. Extends from Dirk Hartog Island to Augusta.
Appendix 2

Biolocality	Climate	Jan	Feb	Mar	Apr	Мау	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Annual	Me
<u> </u>	Rain (mm)	10	13	17	23	56	81	79	59	35	23	14	10	420	
Beverley	Max (°C)	34.1	33.3	30.6	25.7	21.1	17.8	16.7	17.5	20.2	24.2	27.8	32		25
	Min (°C)	16.3	16.7	_14.6	10.9	7.7		5.2	4.9	5.6	7.7	11.2	14.3		់10
	Rain (mm)	11	15	17	26	61	91	87	63	38	26	14	10	459	4
Brookton	Max (°C)	33	32.4	29.2	24.5	20.1	17	16	16.6	19.1	23.3	27.2	31.2		24
	Min (°C)	15.1	15.8	14	10.5	7.2	5.4	4.5	4.4	4.9	7.1	10.6	13.2		9
	Rain (mm)	15.6	15.5	24.3	48.6	128.7	183.3	183.1	140.7	99.9	66.6	32.1	15.9	954	
Collie	Max (°C)	30.5	30.1	27.3	23.2	18.9	16.3	15.5	16.3	18.1	20.7	24.8	28.3		22
	Min (°C)	13.2	13.1	11.5	8.7	6.3	5	4.2	4.5	5.8	7.4	9.7	11.7	분장 <u>실</u> 수 있는	_ 8
	Rain (mm)	24	24	35	56	86	98	107	91	80	71	42	29	743	87
Mt. Barker	Max (°C)	27.1	26.4	24.7	20.9	18.3	15.9	14.7	15.4	16.8	19.5	22.1	24.9		ି <u>2</u> (
	Min (°C)	13.4	13.8	12.7	10.8	9	7.6	6.3	6.1	7	8.3	10.1	12.1		9
	Rain (mm)	9	12	19	46	123	182	173	135	80	55	21	14	869	
Wanneroo	Max (°C)	30	30.5	28.6	24.6	21.1	18.7	17.7	18.3	19.9	21.9	24.8	27.4		2
	. Min (<u>°C)</u>	18.3	18.7	17.3	14.5	<u> 1</u> 1.8	10.4	9.2	9.3	_ 10.3	11.8	14.2	16.5		_1
	Rain (mm)	12	17	20	30	66	91	90	68	47	34	18	13	506	
Wikepin	Max (°C)	30.8	29.9	27.1	22.4	18.3	15.3	14.6	15.1	17.3	21.2	24.9	28.9		2
	Min (°C)	14.6	14.9	13.6	10.9	8.2	7	5.8	5.6	6.3	8.1	10.7	12.8		: · 9
·	Rain (mm)	13	15	18	23	48	65	62	46	24	18	12	8	352	
Wongan Hills	Max (°C)	33.6	33.3	30.3	25.3	20.6	17.1	15.8	16.9	19.8	24	27.8	31.8		2
	Min (°C)	17.3	17.5	16.1	13	9.8	7.4	6.3	6.4	7.3	9.5	12.5	15.2		1
	Rain (mm)	9.5	14.6	16.8	24.1	60	87.9	85.5	65.9	37.4	25.5	12.6	10.1	451	
York	Max (°C)	33.6	32.9	29.9	25.4	20.6	17.3	16.4	17.5	20	23.4	27.9	31.5		2
	Min (°C)	16.6	16.7	14.7	11.2	7.9	6.4	5.2	5.4	6.5	.8.4	12	14.9		1
	Rain (mm)	11	15	20	35	79	117	115	93	61	43	19	14	622	
Yornanina	Max (°C)	32.2	31.3	28.5	23.3	19.3	16.3	15.4	16.2	18.2	21.9	26	30		2
	Min (°C)	14.3	1/1 5	12.4	a 3	64	56	ΛΛ	A 1	<u>л</u> с	63	ολ	12.3		

Mean Climate Data for Seed Biolocalities Sown at York in Chapter 5 (Bureau of Meteorology, 1988).

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