# Ecological, genetic and cultural status of *Solanum aviculare*, poroporo (Solanaceae)

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# Abstract

*Solanum aviculare*, endemic to Australasia, is an opportunist pioneer secondary successional plant occupying disturbed and open lowland habitats, and was an important medicinal and cultural species to Māori known as poroporo. It is currently in 'decline', the ecological decline appearing to correspond to a decline in knowledge and cultural use of the species. To gain understanding of the reasons for the decline, enhance ecological knowledge, assist conservation and cultural restoration of *Solanum aviculare* this research documented the successional role and cycle of regeneration dynamics and tactics, established morphological characteristics, investigated the genetic diversity and recorded cultural and conservation information.

The successional status and role was identified. Growth data identified cohort development and inter-site differences, metadata found height and crown spread growth to be significantly correlated and likely part of an early reproduction and dispersal strategy. Germination of soil cores from differing depths confirmed a viable seed bank exists sufficient for species maintenance. Viable seed spread via animal gut passage was determined by germination and chemical tests, results showed rats passed higher rates of viable seed than birds. Seed germination trials with stratified and fresh seed confirmed temporal and depth behaviour, flowering observation documented temporal differences with the closely allied species *Solanum laciniatum*, indicating a relationship to stasis induction. Leaf morphology studies documented differences between the two allied species and proposed further nomenclature.

Genetic diversity was investigated through the use of PCR/ISSR techniques. Chloroplast DNA was extracted by CTAB and DNA kit protocols. CTAB extraction was unable to effectively remove RNA, although use of DNA samples with high quantities of RNA confirmed that RNA was not an inhibiting factor in PCR production. The production of consistent reliable ISSR bands proved difficult, with no technical explanation found. ISSR findings indicate that *Solanum aviculare* is highly monomorphic, consisting of predominant invariant monomorphic loci. Twenty primers were tested with no polymorphic loci

identified and no intra species variation documented. Indications were also that *Solanum aviculare* and *Solanum laciniatum* are inter species invariant on monomorphic loci. Monomorphic loci may possibly be the evolutionary markers of generic differentiation within *Solanum*.

Surveys identified *Solanum aviculare* as uncommon and rare, existing mainly as single plants or small groups in the majority of areas surveyed. The Threatened and Uncommon Plant listing of *Solanum aviculare* as an 'at risk declining' species is confirmed and a further Recommendation category proposed. Ecological decline and corresponding decline in cultural use and knowledge of *Solanum aviculare* was identified through specialist interviews; appearing to be related to removal from Māori of control over their land. The name poroporo being now associated mainly with non indigenous *Solanum* species. Māori cultural concepts in practice were highlighted as fundamentally important to reversing the decline, with an example of a successful traditional practice based (tikanga) integrative collaborative restoration program being documented.

This research forwards that optimal collaborative solutions, programs integrating scientific and tikanga knowledge and practices, provide the best opportunity of reversing the declining trend and for increasing and maintaining knowledge associated with the traditional role of poroporo.

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# CHAPTER 1
# INTRODUCTION

The ecological research presented within this thesis documents the dynamics, systematics, ecology, genetic diversity, conservation and associated cultural status of the indigenous lowland shrub *Solanum aviculare* Forst. within the Eastern Bay of Plenty and locations in the Waikato region of the North Island, New Zealand. *Solanum aviculare*, commonly known as poroporo, was a valuable and significant species to Māori, culturally and medicinally; at one time being grown commercially for medicinal alkaloid extraction in New Zealand and continuously grown for these purposes in Europe and Asia. Research into the autecology, genetics, and conservation and cultural status is considered pertinent due to a documented decline in populations of *S. aviculare* (de Lange et al. 2008), the unknown underlying causes of the decline, coupled with an associated reduction in knowledge of, and use of poroporo by Māori.

Colonisation by humans in New Zealand has led to major destruction and permanent clearances of what were contiguous natural ecosystems, in particular of lowland and coastal forest and wetland systems, for farming and settlement, leaving a landscape of fragmented habitats invaded by introduced animal and plant pests. The majority of remaining natural habitats, managed for conservation purposes, are located in upland and mountainous areas, and a major challenge is to halt and reverse the decline in indigenous biodiversity in lowland and coastal ecosystems that are heavily modified, and in particular those areas not protected for conservation (New Zealand Biodiversity Strategy (NZBS) 2000). New techniques for conservation in species management and pest control have not halted species decline, and integrated management solutions are proposed that preclude ad hoc decision making, reduce a paucity of knowledge, and move toward management programs with a community and ecosystem focus rather than singular species. The successful implementation of ecosystem based conservation strategies however still requires a need to understand single species, linking the mechanisms of, and how they interrelate within their ecological setting, in an interdisciplinary manner (Beyschlag & Ryel 2007).

The cultural decline in knowledge and use of poroporo appears to be correlated with the ecological decline; therefore integrated interdisciplinary and collaborative programs may play an important role in implementing a reduction in the decline, and reviving subsequent cultural use of *S. aviculare*. Scientific programs can supply precise quantifiable results, and traditional knowledge qualitative information; although, while neither system separately is always effective in resource protection, combined can produce sustainable co-management (Moller et al. 2004) of at risk species. Restoration is determined by the long term survival of, and ecosystem services supplied by, the species being planted, therefore successful restoration requires information on regenerative and reproductive strategies employed by the planted species. Knowledge gained by undertaking detailed studies of a species differing ecological aspects, including the dynamics and functions can be vital to a restoration projects success (Beyschlag & Ryel 2007). Hence this thesis undertakes ecological, plant regeneration dynamics, genetic, conservation and cultural research to attempt to integrate the understanding of, and contribute solutions to the ecological and cultural decline of *S. aviculare.*

## 1.1 Research background and rationale

William Colenso (1868) documented that *S. aviculare* in conjunction with *S. nigrum* L., (he considered to be a European introduction prior to colonisation circa 1850), was common and widespread over the whole of the North Island from sea-level to 1500ft (± 450 m). Cheeseman (1925) recorded the abundance of *S. aviculare* in lowland districts as far south as Foveaux Strait, (although he appears to include *S. laciniatum* as *S. aviculare* to Foveaux Strait). Salmon (1967) described *S. aviculare* as common throughout the North Island and upper South Island. Webb et al. (1988), and Burrows (1999) noted that *S. aviculare* commonly occupied scrub, coastal shrub areas, forest margins, gaps and clearings from natural destructive events, and sometimes more modified habitats including forest areas destroyed by human activity. Crowe (2004) observed *S. aviculare* as being very common in places along lowland forest edges and scrub from the North Island, south to Dunedin, yet in 2008 *S. aviculare* was classified as ‘At Risk’, ‘Declining’, ‘sparse’ and ‘data poor’ by the New Zealand vascular

plant panel (de Lange et al. 2008), suggesting a fairly rapid decline from being common and abundant, especially in areas outside of managed reserves.

My personal interest in poroporo came after being given poroporo (*S. laciniatum*) plants by an ecologist friend for my garden. While growing poroporo I was unable to locate any plants in my travels around the Bay of Plenty despite searching for them. Tangata whenua (Indigenous New Zealanders) who had grown up in the area noticed the plants growing at my residence and commented that those types of plants had not been seen in the local area for >50 years (P. King Kaumatua pers. comm. 2002). I had not readily observed the frequent occurrence of *S. laciniatum* or *S. aviculare* in the Eastern Bay of Plenty as described by Colenso (1868), Cheeseman (1925), Salmon (1967) Webb et al. (1988), Burrows (1999) or Crowe (2004) until I found two populations of *S. aviculare* on slips, within the Tarawera Scenic Reserve, developed following large rain events between November 2005 and February 2006. The discovery of these slips, raised questions as to why and how *S. aviculare* had appeared in such numbers, when I had not observed them growing in the area previously. Aston (1913) undertaking a post eruption vegetation survey on the Western face of Mt. Tarawera, documented *S. aviculare* growing in a gully above Kanehapa (Kanaehapa) Beach between lake level at 1040 ft (± 300m) and 1500 ft (±450m). I speculated that the populations of the Tarawera slips may have originated via seed banks, from plants descended from eruption survivors such as noted by Aston (1913).

My interest in enhancement and restoration of scarce native species, and plant succession includes why and how plants appear and develop in particular places. These interests developed in the course of my Bachelor of Environmental Studies at Te Whare Wānanga o Awanuiārangi, and the opportunity to further my understanding of plant succession and restoration processes continued in the studies toward my Masters degree at the University of Waikato (Te Whare Wānanga o Waikato). Interest in restoration of species stimulated interest in genetics, to gather enhanced information on how population connectivity and the existence of eco-types may aid restoration programs.

Research and discussions with tangata whenua in the Eastern Bay of Plenty, determined that *S. aviculare* or *S. laciniatum* were generally not currently recognised as poroporo and information regarding their uses was lacking. Further research found there to be a lack of published studies that document 1) the seral role of *S. aviculare* in lowland forest disturbance and succession regimes, including the dynamics in regard to seed banks via bird dispersal, and dispersal agents, 2) leaf morphology, 3) inter and intra species genetic diversity data, 4) reasons for the species decline, and 5) reasons for the reduction in cultural knowledge of the uses of the species, despite many publications documenting medicinal uses relating to alkaloid content.

Rowarth et al. (2007) in reviewing native seed germination research established that information on individual species requirements and their ecology was lacking, and information on storage and germination techniques was incomplete. New Zealand studies on *S. aviculare* have centred on chromosome numbers (Baylis 1954), cytogenetic and breeding work (Baylis 1963), day length and flowering (Baylis 1968), documentation of characterisation of developmental phases of shoot and leaf growth (James & Mantell 1994), seed germination behaviour (Burrows 1996, 1999) and incidental bird predation (McEwen 1978; Dijkgraaf 2002; Stanley & Lill 2002; Campbell 2006). International studies have centred on steroidal alkaloid extraction applications and phylogenetic work on the taxonomic status of the *Archaesolanum* sub-genus, of which *S. aviculare* belongs (Gerasimenko 1970, 1971; Panina et al. 1977; Cham & Daunter 1990; Cham et al.1991; Bohs & Olmstead 1997; Bohs 2005; Weese & Bohs 2007; Pozcai et al. 2008).

## 1.2 Thesis aims, objectives and outline

The aim of this research was to gain, in an integrated holistic manner, an overall understanding of the systematics, ecology, conservation status and related cultural aspects of *S. aviculare*. These may suggest possible reasons for the decline, and provide some potential sustainable long term management, enhancement and restoration solutions. Māori words (nga kupu) are integrated into the text with translations immediately following inclusion. Additionally a glossary is included (Appendix 1) to enhance translation and meanings.

The research is presented in six chapters including overall conclusions and recommendations in the final chapter. Outlined below are the objectives for each chapter.

**Chapter 2: Biological review**

This chapter describes the biology of *S. aviculare* in the form of a comprehensive literature review, and comprises systematics, taxonomy, cytology, morphology, as well as ecology and utility. The *Archaesolanum S. aviculare* allied complex is described, along with introduced *Solanum* species occurring in New Zealand, also known as poroporo.

**Chapter 3: Plant dynamics and ecology**

This chapter is presented in four parts; the dynamics being separated into different topics for enhanced clarity, with the research methodologies and results of each topic being presented separately and discussed. The separate topics are then combined in conclusions and recommendations. Part A analyses and discusses the successional structure, population dynamics, regeneration strategies and tactics of *S. aviculare* relating to disturbance regimes, including population structure, reproductive status, lifespan estimation, cohort development and metapopulation data. Part B investigates and discusses the presence of a viable seed bank, the dispersal agents and mechanisms involved in viable seed spread. The viability of seed recovered from dispersal agent's gut passage deposited faeces was endeavoured through chemical and germination trials, and the dispersal tactics employed by *S. aviculare* considered. Part C undertakes and discusses seed germination experiments to consider preferential germination times, seasonal differences relating to day-length and temperature, any species differences, colonisation and germination tactics relating to stasis or fresh seed and depth reduction responses. Part D undertakes and discusses leaf morphology studies documenting differences between *S. aviculare* and the closely related species *S. laciniatum*, determining heteroblastic development, influences on juvenile and adult leaf development, documenting flowering timing and undertaking insect association and damage monitoring.

**Chapter 4: Conservation genetics of *Solanum aviculare***

This chapter documents the genetic diversity of *S. aviculare;* attempting to identify polymorphic loci, investigate and document genetic variation between and within spatially distinct populations of *S. aviculare*, documenting any monomorphic status of *S. aviculare* and any invariance between *S. aviculare* and *S. laciniatum*. The documentation entailed leaf collection, extraction of DNA and the use of Polymerase Chain Reaction (PCR) based Inter Simple Sequence Repeat (ISSR) genetic analyses to estimate the genetic variation within and between populations. A consideration of conservation genetics and restoration planning for *S. aviculare* is undertaken based on the information gained from the genetic work.

**Chapter 5: Conservation and cultural status**

This chapter is presented in two parts, part A Conservation status and part B Cultural status; the research methodologies and results of the conservation status and cultural aspects are presented separately, discussed, and integrative collaborative practices discussed in a final conclusion. Part A considers the current conservation status of *S. aviculare* as a declining species defined in de Lange et al. (2008), and attempts to provide further information on the 'declining', 'sparse' and 'data poor' status, suggest reasons for the increased uncommon situation in the North Island, provide an initial generalised nationwide assessment, and increase confidence in the current classification by providing increased quality data on *S. aviculare*, and includes an estimation of the status of *S. laciniatum.* This is undertaken through electronic surveys with Department of Conservation field staff, private ecologists and individuals. Part B attempts to understand the cultural aspects and the current cultural status of *S. aviculare* as poroporo and a Māori taonga species, through interviews and electronic surveys with tangata whenua. An attempt is made to explain the conservation decline and current cultural status of poroporo from the mātauranga koiora Māori perspective (Māori biological knowledge) by utilising the information gained with associated published information on mātauranga Māori (Māori traditional knowledge base) and mātauranga koiora Māori.

Conclusions and recommendations from Part A and B are combined. The tangata whenua information is collated with the conservation information to consider any similarities and/or differences between how the species is viewed and documented from conservation and cultural perspectives. An attempt is made to consider species enhancement and conservation from, and with an integrated view and framework, combining and clarifying mātauranga Māori concepts, practices and values along with modern science that may address and provide for both conservation and cultural concerns, and the future management of this species.

**Chapter 6 : Summary: Conclusion and Recommendations**

This concluding chapter summarises the findings of the previous chapters, including referral to the research aims and objectives. Recommendations are made on all issues discovered, including conservation and restoration planning and strategies for poroporo.

Photo Moana Kapoor

***Solanum aviculare* flower with Lake Tarawera background: TAR 3**

# CHAPTER 2

# BIOLOGICAL REVIEW

## 2.1 INTRODUCTION

This chapter presents a review of information regarding the bio-systematics, ecology and utility of *Solanum aviculare* Forst. (Solanaceae, *Archaesolanum*), that is available through published and unpublished sources. *Solanum aviculare*, commonly known as poroporo, is a New Zealand native nightshade (Wall & Cranwell 1936) associated with disturbed lowland forest, and with bird dispersed fruit (Webb et al. 1988; Wardle 1991; Burrows 1999; Winterbourne et al. 2008). It can be characterised as an opportunist seral (ecological successional community) pioneer providing initial facilitation for many regenerating native canopy species. It is one of two closely allied indigenous species both known as poroporo belonging to the same sub-genus *Archaesolanum*, the other being *S. laciniatum* Aiton. (Martin 1961; Wardle 1991). Three other extant less closely related species, *S. nodiflorum* Jacq. (*Solanum*), *S. americanum* Mill. (*Solanum*) and *S. nigrum* L. (*Solanum*) are also known as poroporo, but considered either pre-European Polynesian transports, or post-European introductions (Allan 1961; Webb et al.1988). To remove any confusion with other species also being known as poroporo, in this study only the researched taxon will be referred to as poroporo when not using scientific nomenclature.

Cockayne (1967) described the terms indigenous/native as applying to a plant species being wild and present without human aid. If plants arrived through humans either intentionally or unintentionally they are considered introduced. The processes and arrival time in New Zealand of *Solanum* species known as poroporo are unresolved (Allan 1961; Webb et al. 1990; Leach 2005), however *S. aviculare* and *S. laciniatum* are indigenous being present prior to human arrival, and *S. nodiflorum* and *S. americanum* are doubtfully indigenous being likely introduced by Pacific voyagers either prior to or with the ca.1350 AD migration. *Solanum nodiflorum* var., *S. ni*grum var., *S. americanum* var., and other species were introduced post-European arrival (refer Colenso 1868).

Poroporo leaves, fruit and stems have traditionally been utilised and highly valued by Māori in many rohe (traditional tribal areas) for rongoā (medicinal), kai (food) when the berries were ripe, and cultural purposes, but its use has currently declined (M. Home Ngai Tahu environmental liaison pers. comm. 2009; D. Heke-Kaiawha traditional rongoā practitioner pers. comm. 2009). European settlers also utilised the leaves and fruit of *S. aviculare* for food and medicine (Riley 1997). Scientific recognition of the medicinal qualities of *S. aviculare* led to isolation of an active alkaloid component, solasodine, with *S. aviculare* and *S. laciniatum* commercially cultivated for solasodine extraction in New Zealand, Europe and Asia (Cooper & Cambie 1991). The majority of research on *S. aviculare* has focussed on the alkaloid component, associated uses for steroidal contraception and rheumatoid arthritis anti-inflammatory applications rather than ecology.

*Solanum aviculare* is documented as being 'At Risk, declining, sparse and data poor' in New Zealand (de Lange et al. 2009), and while relevant information is available it is spread widely throughout literature, and information on the ecology of *S. aviculare* is deficient. A review is considered pertinent due in part to the data deficiency status, the increased awareness of the slow ecological decline of *S. aviculare* and subsequent loss to Māori of a taonga (highly valued) species and associated mātauranga koiora (Māori biological knowledge). There is increased popularity in replicating natural regenerative and successional processes in restoration programmes that encourage biodiversity, retain genetic variation and reduce replanting of climax species (Atkinson 1994, 2001; Clewell et al. 2004), and interest in *S. aviculare* as an early successional species in forest restoration is increasingly suggested (D. Gosling nurseryman/ecologist pers. comm. 2008). Restoration may also provide for traditional use requirements by making poroporo more readily accessible for harvest.

This review brings together information, previous and current research, on the taxonomy, nomenclature, cytology, morphology, ecology, utility, conservation and cultural references of *S. aviculare*, as well as other publications referring to *S. aviculare*.

## 2.2 Origin and development of New Zealand Flora

The origins of New Zealand's flora lie in the important role played by New Zealand's geographic history. New Zealand geomorphology has ancient elements of pre-Cambrian (>510 million years Before Present (my BP), Devonian (410-360 my BP) granites and Jurassic (200-145 my BP) Gondwanan relic flood basalts, that along with Carboniferous (360-300 my BP) schists, greywacke and recent volcanics dominate the basement rock and mountainous regions (Haase 1990; Campbell & Hutching 2007). Prior to approximately 83 my BP (Late Cretaceous), New Zealand formed part of South-Eastern Gondwana, the super-Continent that included Antarctica, Australia, South America, Africa and India. A land mass, Zealandia, that was precursor to New Zealand, broke from Gondwana, was substantially larger than modern-day New Zealand, and largely submerged by 25 my BP (Late Paleogene/Miocene) (Figure 2.1). The shape of modern-day New Zealand has been formed through plate collision, up-lift and volcanics over the previous approximately 20 my BP (Miocene), and particularly since approximately 5 my BP (Pliocene & Pleistocene) (Stevens et al. 1988; Campbell & Hutching 2007; Wallis & Trewick 2009).


**Fig. 2.1 Modern New Zealand and nearby islands showing in shade the outline of the now largely submerged continent of Zealandia.** (Wallis & Trewick 2009)

New Zealand biota can be considered to have evolved from two origins; vicariance and long distance dispersal (Thorsen et al. in press). Vicariance relates to the separation of Gondwana, forming other continental land masses including Zealandia; any original Gondwanan biota potentially then ceasing to influence Zealandia, apart from transoceanic dispersal that then took place from the other land masses, especially during periods of low sea level. The biota of Zealandia initially then would have been identical to Gondwana and the other land masses, but evolved from those remnants, and from dispersed arrivals, to form the distinctive biota which can be considered to be directly related to New Zealand's modern flora and fauna. There is on-going discussion over whether Zealandia was completely submerged through the Oligocene and Miocene (37 my BP – 20 my BP), the length of inundation and if New Zealand's flora was formed entirely from and evolved since dispersal events, or whether islands of land remained as refuges for remnant Gondwanan species that continued evolving in isolation, with the dispersed arrivals (Wardle 1968; Raven 1973; Stevens et al.1988; Pole 1994; McGlone et al. 2001; Campbell & Hutching 2007; Wallis & Trewick 2009). Recent phylogenetic reviews suggest that much of New Zealand's flora derives from dispersal, with <10% of species studied (n=ca.100) plausibly being of Gondwanan origin (Wallis & Trewick 2009).

New Zealand's flora potentially is a combination of vicariance and dispersal, considered unique having evolved and developed in isolation from other landmasses since the separation from Gondwana (NZBS 2000; Thorsen et al. in press). There is a high degree of endemism evolved from within a diverse range of climatic and glacial conditions, and the subsequently formed habitats ranging from subtropical to subantarctic and temperate forests, swamp, tussock and shrub lands. Many common northern temperate species are absent or under-represented in the New Zealand flora, which evolved without the presence of large grazing mammals, whose niche appeared to be filled by birds such as the moa (*Dinornithidae* spp.), kererū (*Hemiphaga novaeseelandiae*) and kokako (*Callaeas cinerea*) (Cockayne 1947; Clout & Hay 1989). Birds appear also to be important as seed dispersers. Fleshy-fruit bearing species are conspicuous in New Zealand forests (Lord 1999); approximately 48 % of woody plants and 12% of all plant species have fleshy 'fruits' (Wotton et al. 2008). The dispersal mode of many species appearing to be adapted to bird predation, due in part to their

fleshy pericarps, fruit size and colouration, with 32.6% of total flora species, and 13.6% of frugivorous flora species utilising endozoochory (dispersal via animal gut transmission) (Clout & Hay 1989; Haase 1990; Wardle 1991; Burrows 1994; Wotton et al. 2008; Thorsen et al. in press).

Ancestors of the sub-genus *Archaesolanum* complex are suggested to have arrived in Australasia in the early evolutionary movement of *Solanum* as it has little affinity to other *Solanum* groups (Bohs 2005; Poczai et al. 2008). The arrival of *S. aviculare* in the New Zealand landmass can be considered pre-human, however initial arrival timing is unresolved and debatable, ranging from an origin pre-dating the break-up of Gondwana and vicariance, to long distance dispersal methods via sea passage and bird epizoochory (seeds stuck to body) and endozoochory during periods of low sea levels (Wardle 1968; Raven 1973; Pole 1994; Symon 1994; McPhail 1997; McLoughlin 2001, McGlone et al. 2001; Wallis & Trewick 2009). The original existence of *S. aviculare* on Lord Howe and Norfolk Island (Symon 1994), and the possibility of complete submergence of New Zealand in ancient times (Campbell & Hutching 2007) contributing to the distance dispersal postulation.

## 2.3 Morphology

### 2.3.1 Vegetative description

*Solanum aviculare* is an unarmed perennial heteroblastic (progressive change in leaf form with maturity) woody shrub, growing up to 3-4 m in height (Baylis 1963). The branching behaviour is dichasial cymose, of sympodial, acrotonic, and scorpioid nature following from new shoots that emerge from the axils of top leaves which subtend a terminal inflorescence (Figures 2.2 & 2.4). Later branching orders may tend to monochasium (James & Mantell 1994). Stems are initially green, strongly tending to purple, and glabrous (lacking hairs) except for the hypocotyl, with apical buds developing in each axil (Salmon 1980; Webb et al. 1988; Eagle 2006) (Figures 2.2 & 2.3). The axillary shoots produce specific numbers of leaves (12-14) until terminated by an inflorescence, with further anthoclades (single shoot generation) continuously occurring (Brimble 1960; James & Mantell 1994).

The sympodial growth form of *S. aviculare* promotes a readily spreading canopy providing a wide area of protection to regenerating seedlings of canopy species when *S. aviculare* inhabits a disturbed site as a prisere (pioneer initial community in a succession). This horizontal growth habitat is often persistently observed in nursery seedlings, where lateral growth becomes dominant, requiring apical buds on lateral shoots to be pinched out to encourage vertical growth (D. Gosling pers. comm. 2008).

Photo G. Weavers

**Figure 2.2: Dichasial growth habit of juvenile *in situ S. aviculare* (ca. 11 months) with glabrous purple stem.**

Photo Moana Kapoor

**Figure 2.3: Hypocotyl showing hairs, with glabrous cotyledon.**

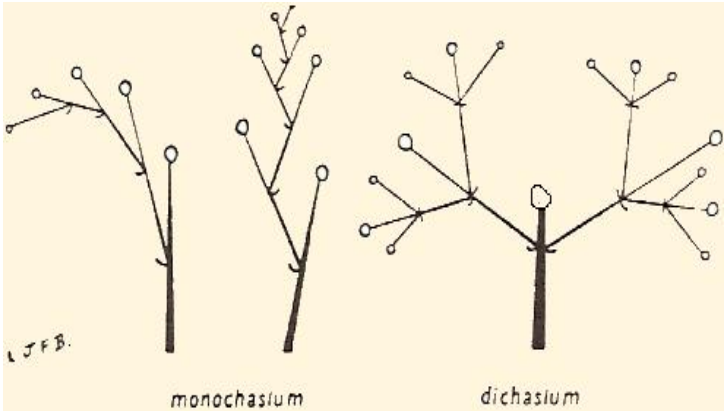


**Figure 2.4: Representation of dichasial and monochasial branching showing acrotony, sympodial and scorpiod behaviour (**adapted from Brimble 1960**).**

2.3.1.1 Leaf description

*Solanum aviculare* leaves are alternate, simple, and entire, having the petioles extending decurrently from the laminar to the base and fused to the stem (Allan 1961; Symon 1981; Webb et al. 1988). Adult and juvenile leaves are dark green in colour, relatively soft, membranous and glabrous, similarly the cotyledons are glabrous. Juvenile first and occasional second initial leaves are ovoid and have minute epidermal and mainly peripheral hairs (trichomes) (Figure 2.5) (Symon 1981). The leaves vary in length from 10-30cm, and width from 30-45mm, with juvenile leaves often being longer than adult (Symon 1981). Baylis (1963) documented the variation in shape; juvenile stages exhibiting pinnately lobed leaves, with adult leaves formed in leaf axils (axillary) exhibiting an entire linear-lanceolate, lanceolate leaf shape with a pinnatifid (pinnasect) shape irregularly re-appearing at maturity (Figure 2.6). Linear and pinnatifid leaves commonly occur on the same adult plant, with variable pinnatifid irregular re-occurrence in adult's possibly either associated with vigour and environmental conditions or genetic based, as some plants display the occurrence more than neighbouring plants of the same group (D. Gosling pers. comm. 2008).

Photos Moana Kapoor

**Figure 2.5: Initial juvenile leaves showing hairs (trichomes), and glabrous cotyledons.**

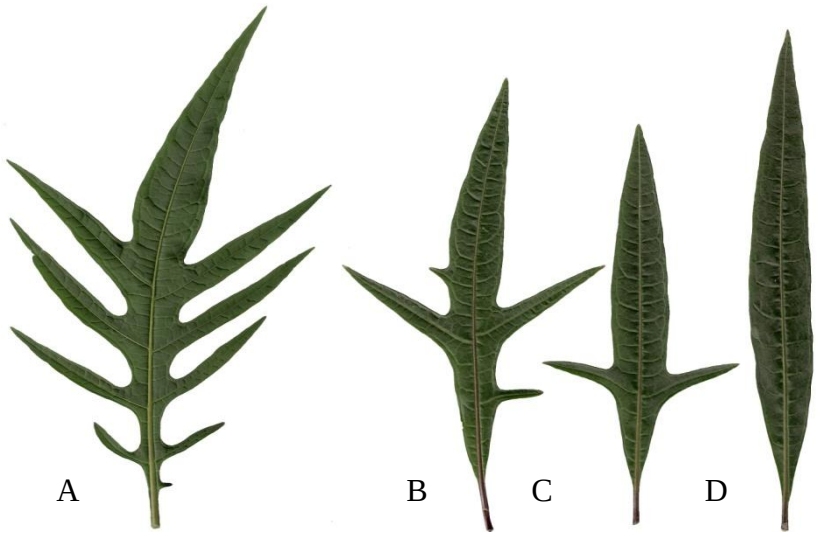


**Figure 2.6: *S. aviculare:* Left to right: A) juvenile pinnately lobed leaf, B) & C) adult pinnatafid leaves and D) adult linear leaf.** (Scans G. Weavers)

2.3.2 Reproductive characters

*Solanum aviculare* exhibits reproductive features common to many *Solanum* species, with cymose inflorescences, lobed corollas, anthers open via slits or pores, fruit a berry and ovary superior (Allan 1961; Webb et al. 1988). Reproduction is sexual and readily self-fertilising without pollinating agents, although they appear highly attractive to insects which alight to the flower immediately on opening (G. Weavers pers. obs. 2008). Pollen grains are 25-31 μ, not being exposed until 12-24 hours after flower opening, with germination occurring within 2 hours of placement on the stigma (Baylis 1963). Cross pollination appears common with hybrids formed, Symon (1994) related that Gerasimenko (1970) reported ready crossing between collections within both *S. aviculare* and *S. laciniatum*, although Baylis (1963) documented the *S. aviculare* complex is not readily cross compatible, as the production of sterile seed occurs when crossed between species, except for partial fertility where *S. vescum* is the female parent. *Solanum aviculare* flowers within 12 weeks of germination at dark photoperiods of 16 + hours (James & Mantell 1994). Baylis (1968) determined the flowers demonstrate a long-day adaption of generally >8+ hours for the inflorescences to fully develop.

Stamen filaments are 3-4mm long, either long or longer than the anthers, which are 3-5mm long, erect, oblong, spreading and open at the tips with transverse

slits (Cheeseman 1925; Allan 1961; Baylis 1963; Symon 1981; Webb et al. 1988) (Figure 2.7). The stigma protrudes above the anthers, and is receptive at flower opening. The calyx is 3-7mm long, and accrescent (growth continued after flowering), with a 2-3.5 cm diameter stellate fused corolla. The corolla colouration is variable ranging from pale bluish-lavender to violet, with five white tipped bluntly pointed lobes, and a predominant five pointed slightly deeper violet and white central star pattern (Baylis 1954; Allan 1961; Symon 1981; Webb et al. 1988). Flower shape and colour are the main points of distinction in identifying *S. aviculare* from *S. laciniatum* (Figures 2.9-2.12). The corolla persists for 3-6 days, displaying nictonasty by closing at night and opening in the morning.

Axillary inflorescences flower continuously through November to April, with fruit often ripening into winter. Inflorescences form in a scorpiod cyme of up to twelve flowers, some flowers can appear as pedicellate in the stem fork (Allan 1961; Symon 1985; Webb et al.1988). *Solanum aviculare* fruits are ovoid succulent bilocular pendent berries 1.5-2.5cm long and 1.5-2.0cm wide. They are first green turning to yellow and orange/red when fully ripe. The green fruits contain toxic glycoalkaloids, remaining poisonous until fully ripe (Connor 1977). They contain 200-600 obovate seeds with a rugose ridge pattern, 1-2mm long, and 1.2-1.5mm in width at maturity, with inconspicuous stone cells that accumulate as granules (Figure 2.8) (Allan 1961; Baylis 1963; Gerasimenko 1971; Webb et al. 1988; Burrows 1999).

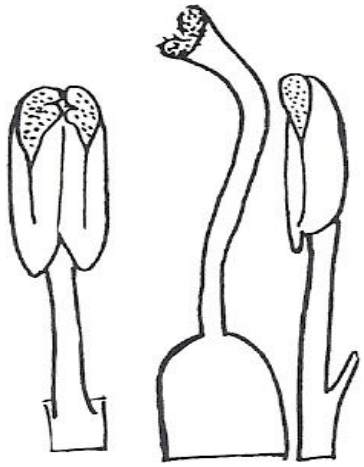

**Figure 2.7: Sketch showing *S. aviculare* stigma, anther and filament (**Symon 1981**)**.

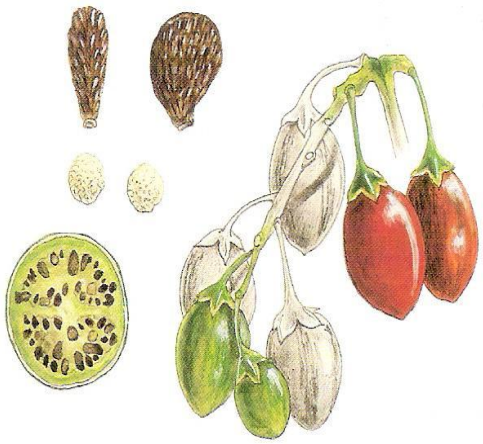

**Figure 2.8: *S. aviculare* fruits, seeds and stone cell accumulations (**adapted from Dashorst, Symon 1994**)**


**Figures 2.9 & 2.10: Left: *S. aviculare* fully open. Right: *S. laciniatum* fully open.**


Photos Moana Kapoor

**Figures 2.11 & 2.12: Left: *S. aviculare* partially open. Right *S. laciniatum* partially open. Note colour variation.**

It has been noted that over several kilometres of Miocene Limestone in the Otway Ranges, Victoria, Australia that *S. aviculare* always produces white flowers, whereas elsewhere in the Otway ranges the plants produce the normal purple flowers. No studies have been undertaken to confirm phenotypic or genetic origins (D. Symon & G. Carr pers. comm. 2009).

## 2.4 Systematics

2.4.1 Taxonomy and relationships

*Solanum aviculare* Forst. (Pl. Esc. 42. 1786. Type: New Zealand. "Queen Charlotte's Sound", *Forster 107* (lectotype, BM, designated by Baylis, 1954; isolectotype, M) is of the order Solanales, a member of the diverse and wide spread Solanaceae Juss., residing within the sub-family Solanoideae Schltsl.,

tribe Solaneae Miers., sub-tribe Solaninae Hunz. and genus *Solanum* L. Within *Solanum*, *S. aviculare* is a member of the sub-genus *Archaesolanum* Bitt.ex Marz. and informally recognised *Solanum* clade I and cluster VI (Bohs & Olmstead 1997; Olmstead & Palmer 1997; Bohs 2005; Poczai et al. 2008). *Solanum aviculare* was first identified by Europeans in New Zealand at Queen Charlotte Sound by Johann and George Forster on Cook's second voyage in 1773, and subsequently confirmed as an esculent (edible) plant by George Forster (1786) in his doctoral thesis, *Dissertatio inauguralis botanico-medica de plantis esculentis insularum oceani australis*. This thesis with its clear description of *S. aviculare* is considered the first valid publication of *S. aviculare.* The *Flora insularum australium prodromus* (Forster 1786) describing *S. aviculare* in abbreviation is often cited as the first place of publication but was published later in October 1786.

*Solanum* L. is a large genus (ca.1500 spp.) with at least seven recognised sub-genera (Weese & Bohs 2007; Pozcai et al. 2008), many of which contain large numbers of species and show distinct Continental radiation patterns. The majority of sub-genera are polyphyletic and widely dispersed globally, however *Archaesolanum* is a small monophyletic sub-genus (Bohs & Olmstead 1997) that is morphologically and cytologically distinctive, differing in leaf shape and chromosome numbers. Members of this sub-genus are endemic to the Australasian region being found only in New Guinea, Australia, New Zealand and off-shore islands (Baylis 1963; Randell & Symon 1976; Symon 1981, 1985, 1994; Bohs 2005).

The sub-genus *Archaesolanum* consists of eight recognised species and two varieties. Gerasimenko (1970) proposed three series or subdivisions of *Archaesolanum*, but they have not been verified by formal botanical recognition. There were six species as recognised by Baylis (1963): *S. aviculare* (with two varieties), *S. capsiciforme* Baylis, *S. laciniatum*, *S. simile* Muell., *S. symonii* Eichl., and *S. vescum* Muell. The two varieties are *S. aviculare* var. *albiflorum* (*S. cheesemanii* Gerasim.), recognised by Cheeseman (1920), which is a white flowered variant of a recessive single gene (Baylis 1963), and *S. aviculare* var. *latifolium* (*S. baylisii* Gerasim.), recognised by Baylis (1963), a broad-leafed variant of a partially dominant, single gene mutation (Baylis 1963). Baylis (1963)

also confirmed that these two varieties of *S. aviculare* exist solely in New Zealand, confined mainly to northern New Zealand and northern off-shore islands.

A further two species were recognised by Symon (1981, 1985). *Solanum linearfolium* Gerasim. was named after being found in plants grown near Moscow with seed from Canberra, Australia. *Solanum multivenosum* Symon was a new high altitude species found only in New Guinea. In addition Symon (1994) noted five named variants of *S. aviculare* and *S. laciniatum* documented during variety trials for drug production in Russia by Korneva et al. (1972), but as no nomenclatural types were indicated, nor holotypes or lectotypes nominated, they have not been formally validated botanically. Gerasimenko (1971) also documented and named five forms of *S. laciniatum*. While the features were practically indistinguishable in the forms, as also noted by Baylis (1968), her observations noted the individual morphological features were apparently heritable, and considered to be genotypic, influenced through phenotypic causation by geographical and ecological factors, rather than directly from their complex genotype (sub-genus *Archaesolanum*), as the differentiation only affected specific individual features. They were also not formally recognised botanically. Gerasimenko (1973) noted and named two varieties of *S. vescum*, but again were not formally validated botanically (Table 2.1).

The eight recognised species extend in overall range from Southern New Zealand, through mainly Eastern and Southern Australia, from Tasmania to Queensland, to the New Guinea Highlands, forming a complex or uniform group. *Solanum aviculare* is the only member found in all locations, covering a latitudinal area from $6^{o}$ S to $44^{o}$ S. (Baylis 1963; Baylis 1968; Olmstead & Palmer 1997; Symon 1981, 1985; Bohs 2005). Only *S. aviculare* and *S. laciniatum* are found naturally in New Zealand. These two taxa overlap in distribution, with *S. laciniatum* generally found in lower latitudes than *S. aviculare*, ranging from $35^{o}$ S to $46^{o}$ 30’ S (Baylis 1954, 1968).

**Table 2.1: Distribution of the named species, varieties and forms in the *Archaesolanum* sub-genus.**

| TAXON | New Zealand | Australia | New Guinea |
|---|---|---|---|
| *S. aviculare* Forst. (1786) | X | X | X |
| *S. aviculare* var .*albiflorum* Chees. (1920) | X | | |
| *S. aviculare* var *latifolium* Baylis (1963) | X | | |
| *S .aviculare* var. *grandifolium* Korneva (1972) | | X | |
| *S. aviculare* var. *grandiflorum* Korneva (1972) | | X | |
| *S. aviculare* var. *patulum* Korneva (1972) | | X | |
| *S. aviculare* var. *acutifolium* Korneva (1972) | | X | |
| *S. aviculare* var. *hybridum* Korneva (1972) | | X | |
| *S. capsiforme* Baylis (1963) | | X | |
| *S. laciniatum* Aiton. (1789) | X | X | |
| *S. laciniatum* forma *tasmanicum* Gerasim. (1971) | | X | |
| *S. laciniatum* forma *novozeylandicum* Gerasim. (1971) | X | | |
| *S. laciniatum* forma *australiense* Gerasim. (1971) | | X | |
| *S. laciniatum* forma *cultum* Gerasim. (1971) | | X | |
| *S. laciniatum* forma *viridicaule* Gerasim. (1971) | | X | |
| *S. linearfolium* Gerasim. (1965) | | X | |
| *S. multivenosum* Symon (1985) | | | X |
| *S. simile* Muell. (1855) | | X | |
| *S. symonii* Eichl. (1963) | | X | |
| *S. vescum* Muell. (1928) | | X | |
| *S. vescum* var. *kibalczecii* Gerasim. (1973) | | X | |
| *S. vescum* var. *davidii* Gerasim. (1973) | | X | |

### 2.4.2 Cytology

*Solanum* is a diploid group, 2n=12, however the *Archaesolanum* complex is of a secondary polyploid nature, which is the first found for the genus with no fossil or ancestral species of chromosomal numbers n=12 or n=24 found or proposed (Baylis 1954; Symon 1981, 1994). *Solanum aviculare* has the chromosome number n=23, considered an aneuploid reduction from n=24 (Bohs 2005; Weese & Bohs 2007; Pozcai et al. 2008). *Solanum capsiforme, S. linearfolium, S. simile,* and *S. vescum* are also n=23, with *S. laciniatum, S. multivenosum* and *S. symonii* tetraploids at n=46. It is considered possible that polyploidy in New Zealand

flora may have stemmed from Pleistocene glacial isolation where the original diploids failed to survive through an apparent lack of ability to diversify (Hair 1966), although that may not fully explain the n=23 status of *S. aviculare* in Australia and New Guinea as well.

### 2.4.3 Phylogeny and Nomenclature

The sub-genus *Archaesolanum* is distinct within *Solanum*, however the position of *Archaesolanum* is still unclear in relation to the other *Solanum* lineages, and the complete evolution of sub-genera within *Solanum* is still not fully understood (Bohs & Olmstead 1997; Bohs 2005). The phylogenetic distinctiveness of the *S. aviculare* complex was highlighted by Olmstead & Palmer (1997) using *cp*DNA restriction site variation analysis and Bohs & Olmstead (1997) utilising chloroplast gene *ndhF* (dehydrogenase complex) sequence analysis where *Archaesolanum* (species *S. aviculare* & *S. laciniatum*) were joined with little support to other *Solanum* clades. However Weese & Bohs (2007) combined chloroplast *ndhF*, *trnT-F* and nuclear *waxy* DNA analyses found >80 % support to placing the *Normania* clade (species *S. trisectum* & *S. herculeum*) and the African non-spiny *Solanum* clade (species *S. aggregatum*) as sister taxa to *Archaesolanum* (species *S. aviculare* & *S. laciniatum*).

The *Normania* and African non-spiny clades, similarly to *Archaesolanum*, are other isolated groups without close relatives, being endemic to north-west Africa, the Macaronesian Islands of Madeira and Canaries (*Normania*), and extreme Southern Africa (African non-spiny). Unusually they have no close macro-morphological similarities to *S. aviculare* or *S. laciniatum* despite the support values connecting them. The analyses indicate these isolated groups without obvious close relatives, may be part of an early *Solanum* radiation and dispersal event, or vicariance (Bohs & Olmstead 1997, 2001), possibly being Tertiary or Pliocene relics (Bohs & Olmstead 2001).

Through the recent work by Weese & Bohs (2007), deeper relationships between *Solanum* clades, and *Archaesolanum*, are beginning to be resolved and confirmed. Most phylogenetic molecular marker studies include only one or two *Archaesolanum* species to investigate relationships among *Solanum* (Bohs &

Olmstead 1997; Bohs 2005), and until current research by Pozcai et al. (unpublished), no studies had utilised molecular markers to investigate relationships among all eight *Archaesolanum* species (Pozcai pers. comm. 2009).

Species of the *Archaesolanum S. aviculare* complex are closely related, particularly *S. aviculare* and *S. laciniatum* (Symon 1994; Pozcai pers. comm. 2009), with differences between species in flowers, leaves, pubescence and fruit (Symon 1981, 1985, 1994). Baylis (1954) documented clear distinction between the sister taxa *S. aviculare* and *S. laciniatum* in chromosomal number, flower shape and colour, pollen size, fruit size, seed size and number, however they are still often confused with each other vegetatively (Baylis 1963; Symon 1981). Bohs & Olmstead (1997) utilising chloroplast *ndhF* sequence variation found 100% bootstrap value uniting *S. aviculare* and *S. laciniatum*. Poczai et al. (2008) using accessions from *S. aviculare, S. laciniatum* and *S. simile* found that *S. aviculare* and *S. laciniatum* share specific RAPD (Random Amplified Polymorphic DNA) bands restricted to them, but mostly lacking in *S. simile*. In addition Pozcai (pers. comm. 2009) considered *S. aviculare* may be the parent of *S. laciniatum*, which appears to be a hybrid, as noted by Baylis (1963), involving *S. aviculare* as the male parent and *S. vescum* as the female parent.

New Zealand had two indigenous species and two doubtfully indigenous species variously known historically and currently as poroporo (Clarke 2007). Two purple berried species, one of which was *S. nodiflorum*, identified and documented on Cook's first voyage by Joseph Banks and Daniel Solander as *S. nodiflorum* but logged in Solander's journal as *S. nigrum*, and the other *S. americanum*, both without the name poroporo. Two orange berried, one of which is *S. aviculare*. The other being *S. laciniatum*, found also on Cook's first voyage by Banks and Solander but logged in Solander's journal as *S. lanceum*, also being the first recorded use by Europeans of the name poroporo in New Zealand. At present it is considered that there are only three extant indigenous or doubtfully indigenous taxa known as poroporo, *S. aviculare, S. laciniatum*, and *S. americanum* (refer Allan 1961; Webb et al. 1988).

The form of *Solanum nodiflorum* collected by Banks and Solander, as noted by Baylis (1958) is considered extinct and now represented by introduced

naturalised *S. nodiflorum*, S. *nigrum*, *S. americanum* and others (B. Snedden pers. comm. Te Papa 2009). Allan (1961) treated *S. nodiflorum* as a presumed indigenous species, though Webb et al. (1988) considered *S. nodiflorum* to be indistinguishable from the widespread *S. americanum* considered indigenous to America, Australia and the Pacific regions, and presumed as either indigenous to New Zealand or an aboriginal pre-European introduction. Leach (2005) also considered *S. americanum* was likely to have been a pre-European Polynesian import due to its presence as a possible introduced 'weed' in the Pacific based on Solander's observations of 'weeds' in the Society Islands in 1769.

Edmonds & Chweya (1997) recognised *S. nodiflorum* as a subspecies of *S. americanum*. However AFLP (Amplified Fragment Length Polymorphism) marker analysis by Manoko et al. (2007) concludes that the two are different species. They differentiate *S. nodiflorum* Jacq. from their recommended nomenclature of *S. americanum* sensu stricto, with no support for the existence of subspecies within *S. nodiflorum*. *Solanum nodiflorum* is an autogamous (self fertilising & associated with colonising ability) species spread worldwide, additionally suggesting the movement of *S. nodiflorum* to New Zealand via early Polynesian voyaging transport.

The name poroporo though originally applied to *S. aviculare* and *S. laciniatum* in New Zealand by Māori has with linguistic variation, widespread use in the Pacific, but associated only with purple berried species (Clarke 2007). *Solanum americanum* is listed as found in the Pacific Islands and known as poro, poro puaka and poroporo in the Cook Islands, oupoo in the Marquesas Islands, polo fua and polo kai in Niue, magalo, magālo and polo in Samoan, 'oporo in Tahitian and polo kai in Tongan (Institute of Pacific Islands Forestry 2009). Both *S. nodiflorum* and *S. americanum* appear in Hawaii and are called polopolo, 'olohua, pōpolo, pōpolohua (Niihau) (Bishop Museum 2009). It is noted that Solander logged *S. laciniatum* as polopolo as well as poroporo, most likely due to the presence of the Ra'iatean tohunga, rangatira and interpreter Tupaia accompanying the voyage.

#### 2.4.3.1 Common Māori Names

*Solanum aviculare* commonly had a variety of names associated with growth stages; Tūhoe also calling it pōporo before it fruits, and kaoho and kahoho when fruiting (Best 1903; Best 1907; Best 1942). Symon (1994) documented that Hooker (1853) described poroporo as a North Island name, and kohoho as a South Island name, with Colenso (1880) reversing it by describing kohoho as used in the North, and poporo or poropora in the South. Allan (1961) listed poroporo, hareto (fruit), horeto, houto and poporo as *Solanum* species; kahoho, kohoho, kaoho, peoi, and poroporo tanguru as *S. aviculare* and *S. laciniatum*. Beever (1991) named *S. aviculare* and *S. laciniatum* as poroporo, poroporo tanguru, and pōpopo, with pōporo and peoi before fruit bearing, hāreo when fruiting, hōreto and hōtou bearing ripe fruit, kahoho and kaoho when the plant is in fruit and kohoho after fruiting. Clarke (2007) adds the names kohokoho, turunui and koheuheu. The name poroporo was referred to by kaumatua of Ohiwa Harbour, Bay of Plenty as relating to poroporoake or farewell to the dead, apparently relating what would happen to anyone who ate the unripe green fruit (P. King pers. comm. 2003).

#### 2.4.3.2 Common English Names

Early European colonists called poroporo Māori gooseberries, pronouncing the word poroporo as bulli-bulli, probably due to the misunderstanding of local dialect and phonetics. The p becoming b, and the r heard as i or d. It was further corrupted by becoming bulli-bull, bulla-bull or bullibul, most likely due to the last vowel in a reduplicated Māori syllable being faintly pronounced (Laing & Blackwell 1927; Martin 1961; Beever 1997; Riley 1994). Senior Pākehā (60+ years New Zealander of European descent) hunters related they knew *S. aviculare* as “the staghorn plant” (anonymous pers. comm. 2009).

## 2.5 Ecology

### 2.5.1 Distribution in New Zealand

*Solanum aviculare* inhabits temperate lowland and coastal forest, generally from sea-level to ± 450 m, although plants in Whirinaki Forest Park have been found

at 566 m above sea level (G. Boyt DOC pers. comm. 2008). *Solanum aviculare* is noted as tolerating a variety of substrate and climatic conditions from the Kermedec Islands, through the North Island (Te Ika a Maui) and off-shore Islands, the Chatham Islands (Rekohu/Wharekauri), and the South Island (Te Wai Pounamu) as far south as Banks Peninsula, the Karamea River and Cascade River in South Westland (Cheeseman 1925; Baylis 1954; Cockayne 1958; Salmon 1980; Webb et al. 1988; Poole & Adams 1994; Burrows 1999; Eagle 2006).

The southerly extent of *S. aviculare* appears to be related to day-length and frost susceptibility. Evidence for this was documented by Baylis (1968), who established that, although *S. aviculare* growth was not inhibited by short days or lowered temperatures (5$^{o}$C), a latitudinal pattern of long-day length in inflorescence development suggested that the day length adaption may prevent southerly spread. Winterbourn et al. (2008) noted that poroporo (*S. aviculare*) is likely eliminated by cold in Canterbury. *Solanum aviculare* is tolerant of medium levels of irregular frost, but can succumb to regular heavy frosts (D. Gosling pers. comm. 2008). Plants were defoliated by heavy frosts in Otago growth trials and Waitara, Taranaki, when grown in a commercial operation (Martin 1999).

Currently *S. aviculare* is in 'decline' (de Lange et al. 2008) and in the Eastern Bay of Plenty appears locally extinct within the Rangitaiki and Galatea Plains, and only found more commonly within Te rohe o Tūhoe (Te Urewera) in inland and mountainous areas such as Maungapohatu, Waikare and Ruatahuna (DOC field staff pers. comm. 2008/2009; K. Tamiana Tohunga ngahere pers. comm. 2009).

### 2.5.2 Habitat and Ecological Role

*Solanum aviculare* appears as an opportunist pioneer coloniser of mainly small scale disturbed sites such as earth slips, canopy gaps, stream and forest margins, within and surrounding lowland forest and re-growth areas; surviving through the regular occurrence of such disturbances and opened areas (Horn 1974; Connell & Slatyer 1977; Veblen 1992). The ecological role of *S. aviculare* as a

prisere appears to provide soil stability, shade, humidity and moisture regulation for a degree of initial facilitation for seedlings of other secondary successive and later canopy species. *Solanum aviculare* is not a nitrogen-fixing plant such as *Coriaria arborea* (Walker et al. 2003), and is considered only to facilitate favourable environmental factors.

*Solanum aviculare* is not a long-lived species, surviving up to seven to ten years in natural environments (Gerasimenko 1971; Wardle 1991; Symon 1994). At times *S. aviculare* can be considered almost ephemeral as it has been noted as appearing, flowering and senescing within two seasons (D. Gosling pers. comm. 2008).

### 2.5.3 Faunal Associations

#### 2.5.3.1 Avifauna

Bird in Latin is *avis*, and *aviculare* means small bird relating to leaf shape (Eagle 2006). Fruits of around 70% of New Zealand trees, and 48% of woody plants including *S. aviculare*, are attractive to, eaten and assumed spread by birds (Clout & Hay 1989; Burrows 1994; Wotton et al. 2008). Consumption of *S. aviculare* fruits by birds was first documented by Forster (1786), though the bird species were not named, and there is little information on exactly which native birds are involved in predation and viable seed dispersal. However native pigeons or kererū (*Hemiphaga novaeseelandiae*) are documented by McEwen (1978), Dijkgraaf (2002), and Campbell (2006) as consuming *S. aviculare* fruits and two *Solanum* genera and three species are noted by Thorsen et al. (in press) to be dispersed by frugivorous vertebrates in New Zealand.

Native bird species such as kiwi (*Apteryx australis mantelli*) (Reid et al. 1982), and parakeets (kakariki) (*Cyanoramphus novaezelandiae* & *C. auriceps auriceps*) (Greene 1998) have been recorded as consuming *Solanum* species, and frugivorous species such as weka (*Gallirallus australis*), tui (*Prosthemadera novaeseelandiae*), bellbird (*Anthornis melanura*), and kokako are known to disperse seed in New Zealand forests (Clout & Hay 1989). It can be assumed that all these species feed on *S. aviculare* fruit and disperse the seed when available.

Adventive bird species starlings (*Sturnus vulgaris*) and blackbird (*Turdus merula*), and naturalised silvereye (*Zosterops lateralis*) were observed disseminating *S. aviculare* and *S. laciniatum* seed (McEwen 1978; Fergusson & Drake 1999). Silvereye is also documented as an endozoochory species consuming and disseminating viable seed of *S. aviculare* in Australia (Stanley & Lill 2003). Senior Pākehā hunters stated, while observing *S. aviculare* on the side of the Humphries Bay, Tarawera trail, that "...the other one..." (*S. laciniatum*) was sometimes seen growing epiphytically (anonymous pers. comm. 2009), presumably deposited by birds roosting. *Solanum aviculare* is considered by Symon (1979) to be bird dispersed in Australia.

2.5.3.2 Mammals

Brushtail possums (*Trichosurus vulpecular*) eat fruits and pass viable seed of *S. aviculare*, with the potential to be important dispersers of native seeds (Williams et al. 2000). Dungan et al. (2002) documented the same result, including enhanced germination, for *S. laciniatum*. Goats (*Capra hircus*) grazed *S. aviculare* in Raukumara Ranges (East Coast North Island) and deer (*Cervus* and *Dama* spp.) in Whirinaki Forest Park (G. Boyt pers. comm. 2008). Cockayne (1908) documented cattle in Waipoua Forest; Northland thrived on eating *S. aviculare*, amongst other shrubs. *Solanum laciniatum* was grazed but not killed in a horse paddock at Onuku, Akaroa (G. Weavers pers. obs. 2008).

*Solanum aviculare* fruits were predated and abundant viable seed excreted by ship rats (*Rattus rattus*) (Williams et al. 2000). Kiore (*Rattus exulans*) and mice (*Mus musculus*) in the same trials interestingly did not predate *S. aviculare*, although both species were observed caching some fruit. Predation and dispersal by Norway rats (*R. norvegicus*) and pigs (*Sus scrofa* spp.) is unknown, but considered possible and likely where *S. aviculare* is growing in association with these introduced mammals (Beveridge 1964; Williams et al. 2000). Wallabies (*Macropus* spp.) were not observed browsing *S. aviculare* foliage at Tarawera, even though adjacent shrubs such as *Coprosma robusta* were heavily grazed (G. Weavers pers. obs. 2009). Symon (1986) noted a lack of herbivore dietary knowledge in Australia, and that *S. simile* foliage was a minor diet item in quokka wallaby (*Setonix brachyurus*), although fruit were eaten when ripe.

Symon (1986) also suggested that the presence of high quantities of solasodine alkaloids found in *Archaesolanum* species may inhibit the grazing by wallabies.

#### 2.5.3.3 Insects and Reptiles

Insect associations and predation are apparent in the wild (Burrows 1999). Forty seven insect species from twenty three families and seven orders were found on *S. aviculare* and *S. laciniatum* plants during commercial alkaloid extraction, with the majority being herbivorous pests of *S. aviculare* and *S. laciniatum*, the remainder mainly being predators of certain of those pest species (Martin 1999). Duthie et al. (2006) documented that tree weta (*Hemidenina crassidens*) consumed native fleshy fruits, dispersed the seed over their range and passed viable seed, which had higher germination rates than control seeds manually extracted from pulp, therefore may also be a dispersal agent for *S. aviculare*

In New Zealand reptilian (*Saurochory* spp.) predation can be important in frugivorous species that are low shrubs associated with small fruit and exposed habitats, although such predation is considered to be complimentary to bird dispersal (Thorsen et al. in press). Lizards were noted by Burrows (1994) and Wotton et al. (2008) and Wotton (2002) as having involvement in consuming and dispersing seed of small fleshy-fruited species. Symon (1979) reported in Australia that reptiles, as fruit predators, are associated with fruit characters of colour, smell and fruit drop at maturity. The Australian lizard (*Trachydosaurus rugosus*) ate tomatoes (*Solanum* spp.), and coupled with the lizard (*Tiliqua scinoides*) was found in the vicinity of *S. simile* which has aromatic, succulent fruit that drop to the ground in the manner of *S. aviculare* (Symon 1979).

## 2.6 Utility

### 2.6.1 Māori

Historically legend has related that poroporo (*S. aviculare*) was found in Hawaiki (Māori ancestral homeland), where the berries were reserved for the use of priests (tohunga). The practice appears to have continued when poroporo was found in Aotearoa (New Zealand) after the arrival of early settlers from Hawaiki

(Gordon no date). Gordon (no date) and Grey (1855) relate that poroporo (poporo) fruit stolen by Tama Te Kapua and his brother from around a priest's house in Hawaiki, ultimately led to the migration of Te Arawa (Rotorua district Tribe) waka (vessel/canoe) to Aotearoa. Gordon (no date) and Riley (1997) state that the breaking of tapu (ceremonial restriction) by eating the ripe berries of poroporo by a Rotorua tohunga prior to undertaking the birth rites of the Rangatira (chiefly) child Tūtānekai, led to his being put to death by drowning.

Traditionally Māori used poroporo for kai, rongoā and cultural purposes. Poroporo plants were spread to suitable sites (Holland 2000), and cultivated around kainga (home living areas & buildings), especially as kai for children (in difference to legend) (Colenso 1880; Best 1925; Best 1942; Symon 1994). Fruits were eaten raw, with the leaves being used in umu (earth oven) to flavour food (Riley 1997).

Medicinally the leaves, including juice, inner skin and the underside were pulped and used as a poultice for skin irritations, ulcers, established sores, eruptions and scabies (Goldie 1903; Salmon 1967; Salmon 1980; Brooker et al. 1987; Riley 1997; Moon 2005). When heated in water with ngaio (*Myoporum laetum*), kawakawa (*Macropiper excelsum*), and tātarāmoa (*Rubus cissoides*) the leaves were utilised as a vapour bath for joint pain (Riley 1997). Pulped berries were also used for scabies and joint pain (Riley 1997; D. Heke-Kaiawha pers. comm. 2009). The leaves, inner bark and pith were also used for bruises and established itch sores (Salmon 1980; Brooker at al. 1987; Moon 2005). Leaves were boiled and liquid used as a shampoo, noted as giving hair a shiny gloss, and as a dandruff treatment (Riley 1997; Moon 2005). Poroporo leaves were taken as a decoction just before menstruation for contraceptive purposes, even in to the mid 20th century (Riley 1997; W. Te Pairi pers. comm. 2008; D. Heke-Kaiawha pers. comm. 2009; K. Tamiana pers. comm. 2009).

Juice from the leaves was used to size the wood of waka, and to wash whare (house) prior to kōkōwai (red ochre) application to help colour retention in kowhaiwhai (painted panels). When the juice was mixed with kōkōwai, soot (un-named tree spp.) and / or shark oil it was used to colour facial tattoos (moko) (Salmon 1967; Brooker et al. 1987; Riley 1997; Auckland Museum 1998; Clarke

2007). Clarke (2007) notes also that flutes were made from the woody stems of poroporo. Riley (1997) noted Best described the dried leaves being used as a maro poroporo or a type of 'under garment' worn beneath maro kuta (small skirt) by young women of rank.

In the modern era use of *S. aviculare*, as poroporo by tangata whenua for rongoā, kai and cultural purposes has declined or ceased. Tangata whenua interviewees (T. Te Pairi Rangatira pers. comm. 2008; anonymous pers. comm. 2008/2009; K. Tamiana pers. comm. 2009; D. Tupe rongoā practitioner pers. comm. 2009) in the Eastern Bay of Plenty and Hawkes Bay, in their kōrero (speaking/interviews) state that the name poroporo is now associated mainly with the purple berried species *S. nigrum*, *S. americanum*, and others. Conservation decline indicates poroporo may not have been common in the Bay of Plenty and Hawkes Bay for ± 70 years. Many others only know *S. aviculare* as poroporo by story, though some memories of its rongoā use remain.

The use of *S. aviculare* as a rongoā, and oral knowledge (mātauranga) relating the name and use of poroporo to *S. aviculare* is not completely lost, but appears scarcely, appearing to be correlated to the ecological decline that along with an associated decline in dispersal agents such as kererū, is attributed to altered land use with land extensively cleared for urban development, and pasture with stock continuously grazed (anonymous pers. comm. 2008/2009; M. Home pers. comm. 2009; Lyver et al. 2008; M. Tamiana ngahere expert pers. comm. 2009). Many interviewees consider that the land being no longer in tangata whenua ownership (raupatu) (Lyver et al. 2008) is a causal factor in the altered land use that has led to poroporo becoming scarce along with knowledge of its use.

Areas where *S. aviculare* is known to be currently growing are often within public conservation managed forests, reserves and parks where access is difficult and collection cannot take place without a permit. Many Māori have also moved away from their rohe (traditional home areas) where kai and rongoā were collected as part of daily routine, losing connections to traditional kōrero associated with plants and their uses. A combination of decline and reduction of poroporo in readily accessible areas, harvest sites having restricted access and located a distance from traditional rohe, no or limited harvest sites available in e

noho kainga (urban areas where they are currently living) all contribute to the weakening use of traditional resources and related mātauranga, leading to eventual loss of cultural use and knowledge of *S. aviculare* as poroporo.

### 2.6.2 European settlers

Consumption and use of the berries was first documented by Forster (1786) describing the taste as "acid, a little sweet, almost sickening" (Symon 1994). Ripe berries were used by early colonists for making into pies and jam, as well as presumably eaten raw, as the flavour and consistency were described as delicious at times, and at others not unpleasant, sweet but slightly acrid when ripe, tasting like a date and tomato, and the pulp sweet and resembling a fig (Harvey & Godley 1969; Riley 1997; Crowe 2004). Europeans also used *S. aviculare* leaves for general healing, mixing it with lard and using it as a salve, and for eczema, dermatitis, itching and festering itch sores as a decoction and bath. It was also considered effective for scab (undescribed condition) in sheep (Riley 1994).

### 2.6.3 Contemporary use and economic importance

Wool home-spinners have used the leaves and twigs to make dyes, and a mild flavoured honey has also been produced (Crowe 1999). Modern medicinal uses of the active ingredient solasodine have followed traditional ones; for rheumatoid arthritis anti-inflammatory applications, skin disorders including sarcoma and melanoma, and a naturally occurring contraceptive, as well as having indications of cardiac effects and ecdysone (hormonal interference) insecticidal properties (Krayer & Briggs 1950; Schreiber & Manske 1968; Weston 1976; Cambie et al. 1981; Cham & Daunter 1990; Cham et al. 1991).

Solasodine was first identified and isolated as solasonine from *S. aviculare* at Auckland in 1942 by Professor Lindsay Briggs (Bradley et al. 1978; Cooper & Cambie 1991). Solasodine, is found in many *Solanum* species, but in high quantities in *Archaesolanum* species, the leaves, berries and young shoots all containing the product. Green unripe berries and young shoots contain a higher quantity than the leaves, with the alkaloid content of the berries reducing as fruit

ripens (Bradley et al. 1978; Bradley et al. 1979; Cooper & Cambie 1991). *Solanum aviculare* yields the highest solasodine values of the *Archaesolanum* complex (Symon 1994), Cooper & Cambie (1991) reporting that *S. aviculare* in New Zealand cultivation yielded twice the quantity of solasodine than *S. laciniatum*, depending upon climate and substrate.

Cropping trials utilising *S. aviculare* and *S. laciniatum* were initiated to test commercial extraction in 1964 by Ivan Watkins-Dow in New Plymouth, Taranaki and continued by the DSIR in 1969-1970 at Canterbury (Cooper & Cambie 1991; Symon 1994). Trials started in Otago ended in 1977 as a result of frosts and unseasonable wet weather (Riley 1994). Commercial extraction of solasodine from *S. aviculare* including a cultivar NA38 (Symon 1994), commenced in 1976 at Waitara, Taranaki in a joint venture, *Solanum* Extraction Industries Ltd, with Fletcher Holdings and Diosynth Netherlands BV, a subsidiary of Akzopharma the pharmaceutical arm of the Dutch company AKZO NV. Production ceased in 1981 mainly due to cheaper production costs from worldwide competition with initial mono-cultural production difficulties caused by insect pests, virus and potato blight. Both *S. aviculare* and *S. laciniatum* have, from the 1960's, been continuously cultivated widely in Holland, Hungary, Eastern Europe, Russia and China for contraception and rheumatoid arthritis. Trials have been carried out in Pakistan, Egypt, Israel, Japan, Cuba, England and Indonesia (Bradley et al. 1979; Cooper & Cambie 1991; Symon 1994).

*Solanum aviculare* has not received a great deal of attention as an ornamental plant, despite the attractive combination of the green foliage with purple flowers, and the ease of propagation from cuttings and seed, possibly due to the toxic nature of its green unripe fruit. The constant pruning needed to maintain a satisfactory size and shape in the highly suitable growing conditions found in urban gardens may also reduce its ornamental appeal. *Solanum aviculare* is not widely used in restoration projects despite its apparent role as a prisere in natural communities, though it is available through specialist nurseries (D. Gosling pers. comm. 2008).

## 2.7 Conclusions

*Solanum aviculare* is an ancient flowering shrub, potentially of Gondwanan antiquity, with arrival in New Zealand being pre-human. *Solanum aviculare* is a unique plant that occupies a wide range of lowland habitats through a broad latitudinal range, inhabiting disturbed and open environments from the coastal zone to inland hill country; playing an important seral role in those habitats, including vegetation succession and erosion control, providing regular food for bird species that act as dispersal agents. As a short lived plant it survives through scattered deposition, and a continuous cycle of small scale disturbances providing the favourable mechanisms for growth and maturation.

*Solanum aviculare* was commonly used traditionally for food and medicinal purposes by Māori and European settlers, and commercially exploited for solasodine production in the modern era. Currently *S. aviculare* is in decline; the ecological decline, coupled with the introduction of alternative food and medicinal sources, appears to have contributed to a decline in the cultural knowledge of, and use of *S. aviculare* as poroporo for traditional purposes. The name and uses of poroporo is now mainly associated with the purple berried species *S. nigrum*, *S. americanum*, and others.

The decline of *S. aviculare* and methods required for reversing the decline, coupled with the valuable roles played in seral dynamics and vegetation succession makes continued research necessary, and imperative. *Solanum aviculare* research should include a full documentation of the seral dynamics and a full genetic appraisal of variation and diversity that may influence eco-sourcing for restoration, as Baylis (1968) noted *S. aviculare* exhibited eco-types in regard photoperiod adaptation to latitude and temperature.

Bio-prospecting possibilities exist for exploitation of New Zealand generated material with documented medicinal properties and it is important to monitor species such as *S. aviculare*, as any publicised ecological research undertaken can increase the awareness of such species potential for bio-prospecting exploitation.

Promoting the use of *S. aviculare* and *S. laciniatum* in ecological restoration and enhancement will potentially provide mitigation against further decline, by ensuring seed dispersal for future germination and colonisation. An increase in availability of poroporo through such programs can enhance and make provision for traditional Māori values.

Photo Moana Kapoor

***Solanum aviculare*, flowering and fruiting in Tarawera slip site: TAR 3**

# CHAPTER 3

# PLANT DYNAMICS AND ECOLOGY

# PART A: GROWTH STRUCTURE AND SUCCESSIONAL STATUS

## 3.1 INTRODUCTION

Plant vegetative dynamics as stated by White (1979) are initiated partly by disturbance, which produce conditions an early successional species requires, and upon which colonisation can be dependant (Platt & Connell 2003). Exogenous disturbance in the form of tree fall gaps (gap-phase regeneration) and earth slips (catastrophic regeneration) are an accepted common feature of secondary succession in the regeneration of New Zealand lowland forests, and many species exhibit establishment responses to gaps (Ogden 1985; Lusk & Ogden 1992). Disturbance of a habitat, in particular substrate perturbation, plays a role in maintaining diversity and ecological processes by opening space for secondary vegetative regeneration, creating a series of patches which determine composition and structure (Horn 1974; Ogden et al. 1991; Lusk & Ogden 1992; Lloyd et al. 2003).

Secondary succession is a sequential process of vegetation change, often beginning from a soil seed bank (Crawley 1986) in the re-establishment of a community following an allogenic short term or temporary disturbance that opens a space (Horn 1974; Connell & Slatyer 1977; Huston & Smith 1987). Mature forest areas are characterised by closed canopies and low light conditions (Denslow 1987) that are not generally conducive for an early (pioneer) succession species to survive and reproduce locally. New openings that provide conditions conducive to growth of pioneer species are generally ephemeral, and pioneer species must utilise strategies to allow them to colonise and complete a reproductive cycle before these ephemeral openings disappear (Horn 1974). Within a vegetative community individual species will exhibit adaptive strategies allowing establishment and reproduction within a gap or disturbance patch (Burrows 1990; Platt & Connell 2003). Connell & Slatyer (1977) and Platt & Connell (2003) describe conceptual models of mechanisms (adaptive plant

strategies/ life history traits) (Model 1-III, Model 3b) that promote secondary successional change after a disturbance, and consider how an early species may persist, including resting stages, until the next disturbance. Adaptive diversification or the ability of a species to vary strategies allows it to succeed under differing conditions (Denslow 1987; Huston & Smith 1987) with Platt & Connell (2003) noting that disturbance sites can exhibit varying levels of resource availability, and pioneer species can develop specialisations to enhance colonisation and reproduction.

Adaptive successional strategies displayed by species, for invasive success must include processes for rapid plant growth rate to flowering and fruit set, successful wide seed distribution, seed morphology and behaviour that allow survival until the right conditions present appropriate germination cues (Connell & Slatyer 1977). Early secondary successional species therefore tend to be characterised by temporal flexibility of seed germination in relation to germination cues, fast initial growth rates, expansive vegetative growth habits, rapid onset of flowering and fruit set, short life span, efficient seed distribution processes for wide dispersal, and seed characteristics including tactics for longevity until germination requirements are met (Huston & Smith 1987; Smith & Smith 2001).

Increased soil and air temperatures via increased direct sunlight intensity and duration are important effects of gap opening, leading to rapid early growth to maximum size and providing ideal conditions for secondary regeneration processes to begin, as closed canopies are deficient in photosynthetically active light wavelengths (Denslow 1987). Huston & Smith (1987) documented early successional species are characterised by small size at maturity (compared to later successional species), low structural strength, rapid early growth, low later stages growth, and a short life span. Brokaw (1985) found that pioneer species in gap-phase regeneration exhibited rapid height growth, significantly greater than canopy species, following germination of the pioneer species in a soil disturbance containing light or heat sensitive seeds. This rapid growth in height declined rapidly with age, and early recruitment into a gap afforded the greatest chance of survival and reproduction for the secondary successional species.

*Solanum aviculare* is a pioneer secondary successional species appearing in New Zealand lowland forest after disturbance events. In particular *S. aviculare* is strongly associated with tree fall gaps, earth slips, forest, stream edges and bird roosting sites. As a prisere species (pioneer initial community in a succession) *S. aviculare* is considered to exhibit the characteristics and adaptive strategies required for rapid invasion of ephemeral disturbances, quickly and successfully completing a cycle of regenerative processes. These involve fast initial growth rates with expansive vegetative growth, rapid on-set of flowering and fruit set, wide seed dispersal mechanisms via fleshy-fruit for bird consumption (Burrows 1999), seed bank dynamics including stasis and flexible germination patterns (Burrows 1994; Rowarth et al. 2007).

This research aimed to answer the questions 1) what growth strategies and tactics (mechanisms) are employed by *S. aviculare* to successfully colonise and complete its life cycle and 2) do these tactics conform to the description and conditions of a pioneer successional species in earlier studies. Therefore the aims of this research were to determine and quantify the cycle of regeneration tactics including growth dynamics and successional processes of *S. aviculare* occurring within a secondary disturbance regime in lowland podocarp-broadleaved forest of the Eastern Bay of Plenty and Waikato, New Zealand by recording *in situ* growth data from single plants in *S. aviculare* populations and groups.

## 3.2 Methodology

### 3.2.1 Study sites

Two slip sites, (TAR 1 & 2) containing populations >30 plants were initially located below and above Tarawera Falls, Eastern Bay of Plenty, New Zealand. Following discussions with Department of Conservation field staff (J. Willcocks pers. comm. 2008) a further slip site (TAR 3) with a population of >30 plants was located on the Humphries Bay track on the northern side of Lake Tarawera. The Tarawera slip sites were documented as occurring between November 2005 and February 2006, twenty eight months prior to data collection in March 2008 (J. Willcocks pers. comm. 2008). Field searches located 18 plants at Lake Rotoma (ROT), six plants at Lake Okataina (OKA) (Rotorua), which were combined as one site consisting of 24 plants, and populations >30 plants located

at Maungatautari north and south enclosures (Waikato). Five plants were located at Pirongia sites (Waikato) by Elizabeth Overdyk (pers. comm. 2009). Enquiries with a private ecologist located >30 plants at the Ohutu Stream, Te Urewera (URE) (W. Shaw pers. comm. 2008) and enquiries with tangata whenua located 13 plants at O Tane Wainuku, Tauranga (TAU) (D. Heke-Kaiawha pers. comm. 2008) (Plates 3.1-3.5). Full site descriptions are contained in Appendix 2. All sites were coded for identification (Table 3.1).

**Table 3.1: Locations, site codes, habitat and numbers of *S. aviculare* plants used in the research.**

| Location | Site code | Location type & plant number |
|---|---|---|
| Tarawera | TAR 1-3 | Slip sites within reserve > 30 each |
| Rotoma | ROT 1 & 3 | Road edge Individual plants |
| Rotoma | ROT 2 | Tree fall gap 15 plants |
| Rotoma | ROT 4 | Lake edge Individual plant |
| Okataina | OKA 1 | Tree fall gap 3 plants |
| Okataina | OKA 2 | Track edge Individual plant |
| Okataina | OKA 3 & 4 | Road edge Individual plants |
| Tauranga | TAU 1-13 | Forest edges 13 plants |
| Te Urewera | URE 1 | Slip site 4 plants |
| Te Urewera | URE 2 | Stream edge 2 plants |
| Te Urewera | URE 3 | Slip site >24 plants |
| Maungatautari North | MAU N 1 -3 | Tree fall gaps 1, >3 & >30 plants |
| Maungatautari South | MAU S 1 | Track edge 5 plants |
| Maungatautari South | MAU S 2 & 3 | Tree fall gaps 2 & 5 plants |
| Maungatautari South | MAU S 4 | Bird roosting site >20 plants |
| Pirongia Corcoran Rd | PIR 1-2 | Track edge 2 plants |
| Pirongia Kaniwhaniwha | PIR 3-5 | Track edge 3 plants |

All of the sites located consisted of similar lowland podocarp (totara *Podocarpus totara*, rimu *Dacrydium cupressinum*) – broadleaved (tawa *Beilschmedia tawa*, tawhero *Weinmannia racemosa*) dominant forests. The Tarawera sites had the addition of dominant pohutukawa (*Metrosideros excelsa*) at TAR 1, and rewa-rewa (*Knightia excelsia*) at TAR 2. Locations of the nine study sites were considered widespread and broad enough to enable a comparison of population connectivity and cohort development and variation (Figure 3.1). Low Impact, Research and Collection permits were obtained through the Department of Conservation, National Permit Numbers BP-23541-FLO & WK-24608-FLO. In addition permission to collect at Maungatautari was obtained through Waipa

District Council and the Maungatautari Ecological Island Trust, and at Okataina through the Okataina Scenic Reserve Board. Entry permits for the Tarawera forest for the duration of the research were supplied by Māori Investments Ltd. Herbarium voucher samples, one per population, were collected from all sites except Pirongia, and lodged in the University of Waikato Herbarium (WAIK).

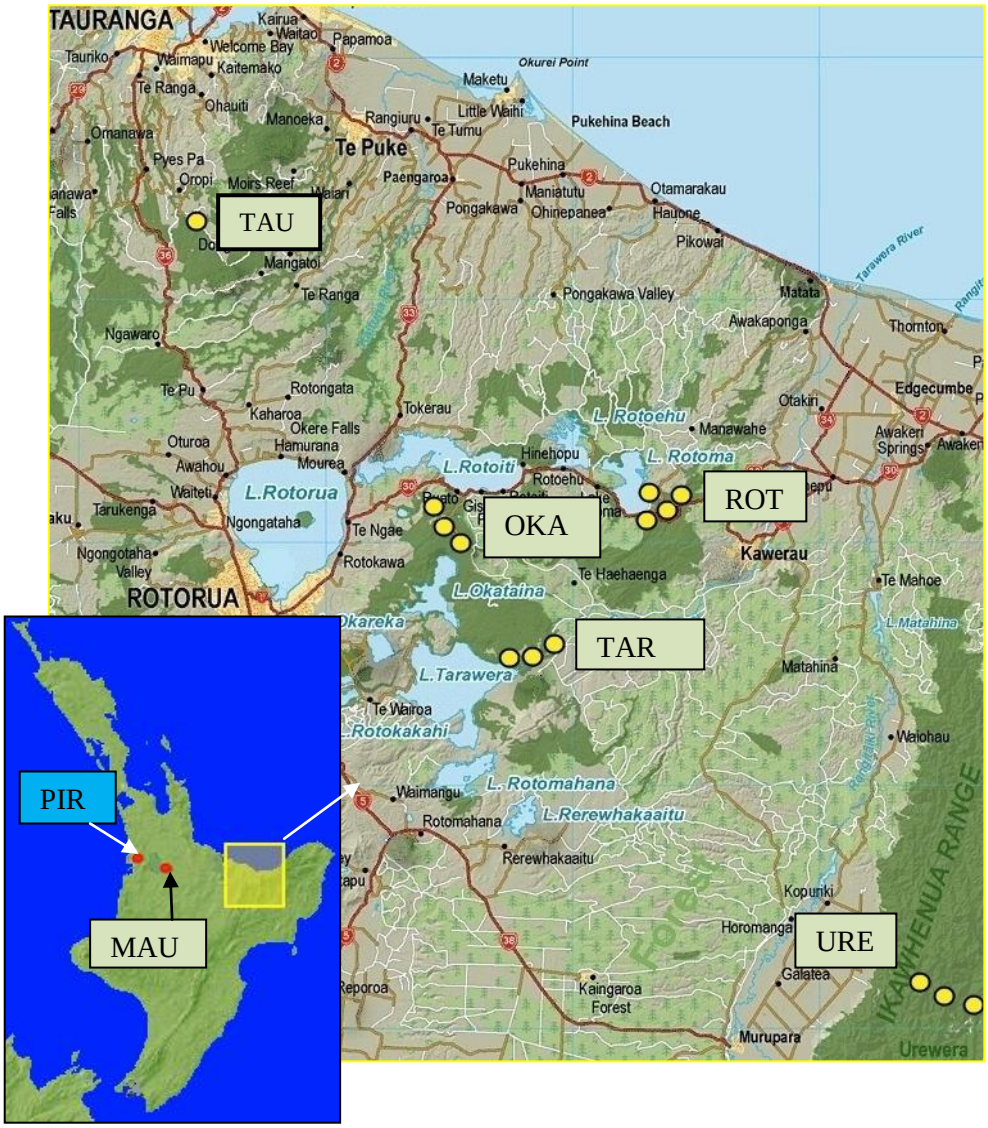


**Figure 3.1: Main location map showing relative positions of Bay of Plenty and Waikato sites. Key: yellow = Bay of Plenty & red = Waikato (Maps adapted from EBOP and Landcare NZ)**

Photo Moana Kapoor

**Plate 3.1: Rotoma site 1 (ROT 1) showing plant location on road side.**

Photo G. Weavers

**Plate 3.2: Tarawera site 3 (TAR 3) showing steep terrain and location of poroporo plants (straight arrow) and Recce vegetation plots (dotted arrow).**

Photo G. Weavers

**Plate 3.3: Rotoma site 2 (ROT 2) showing plant locations within tree fall gap.**

Photo D. Heke-Kaiawha

**Plate 3.4: Typical poroporo plant location on forest edge, Tauranga (TAU 13).**

Photo G. Boyt

**Plate 3.5: Ohutu site 2 (URE 2) poroporo plant on Ohutu stream edge in association with *Buddleia.***

3.2.2 Growth Structure- research design

Where >30 plants were present at a site, an entry point was selected randomly, and the first 30 plants encountered measured. In sites with <30 plants, all plants were measured. Height and crown spread were measured, flowering or non-flowering status recorded as juvenile or adult by the obvious presence/absence or evidence of fruit or flowers. Tarawera sites were continuously monitored to follow growth patterns and any changes over the study period. A stem sample for ring analysis was taken in March 2009 from the recovered trunk of a large dead adult plant at Tarawera site TAR 2. This was senesced in September 2008, six months after the growth data was collected.

3.2.3 Data analysis and statistics

Cohort development was indicated by utilising data analysis of all sites with >20 plants present. Metapopulation data analysis was determined from all sites combined. The data was entered into Microsoft Excel 2007 spreadsheets, and histograms produced. The data was then re-entered into Statistica 9.0.725.0 (StatSoft 2009) to confirm results by producing distribution, box and scatter plots, and multiple regression analyses. Significance tests and non-parametric correlations were produced by Spearman Rank Correlations ($p=<0.05$).

## 3.3 Results

Growth data was collected from a total of 192 plants, consisting of 115 adult and 77 juvenile plants. Ninety plants were measured at Tarawera sites TAR 1, TAR 2 and TAR 3, which combined contained >150 plants when the data was recorded in March 2008. Rechecking of the sites in September 2008 showed a large senescence of plants and by March 2009 only 15 plants in total remained. Tarawera site TAR 1 had no survivors. Tarawera site TAR 2 contained 11 survivors, with six only having one branch containing leaves. Tarawera site TAR 3 had five remaining plants. Ring analysis of the recovered stem from TAR 2 indicated the plant was approximately three years of age (±1 year) at senescence. The diameter of the trunk was 7cm at .3m from the ground. Recorded height and

crown spread were 1.9m and 6.72m$^2$. The majority of other senesced plants at TAR 2 were of similar height, crown spread and apparent age.

All sites with > 20 plants (n= 6) were consistent with being cohorts, appearing to have developed at the same time at each site (Figures 3.2 & 3.3). Cohort growth development is indicated, with recruitment still occurring in all sites. Tarawera TAR 1 shows a population where recruitment is occurring by the appearance of juvenile (non flowering) plants in the 0-2.0m height and 0-1.0m$^2$ crown spread range, the population appears to be stabilising with juveniles decreasing and adults increasing in number in higher height and crown spread classes. Tarawera TAR 2 indicates active recruitment is occurring with juvenile plants appearing in the 0-2.0m height and 0-4.0m$^2$ crown spread range, the population is also stabilising with juvenile plants decreasing and adult plants increasing in the higher height and crown spread classes. Tarawera TAR 3 indicates that active recruitment is strongly occurring with juveniles dominating in the 0-2.0m height and 0-4.0m$^2$ crown spread range, with the population beginning to stabilise, but with a juvenile outlier still occurring in the highest height and crown spread range.

Okataina/Rotoma OKA/ROT sites indicate a mature reproductive population that has stabilised, very little recruitment is still happening and adult plants dominate in all height and crown spread classes. Ohutu URE indicates an actively developing population with high recruitment of juvenile plants and low appearance of adult plants in both height and crown spread, but especially in the higher height classes. Maungatautari MAU indicates a population that is stabilising, with recruitment still taking place but not in the lowest height range. Adult plants dominate, increasing in number while juvenile plants decrease in the higher height and crown spread ranges.

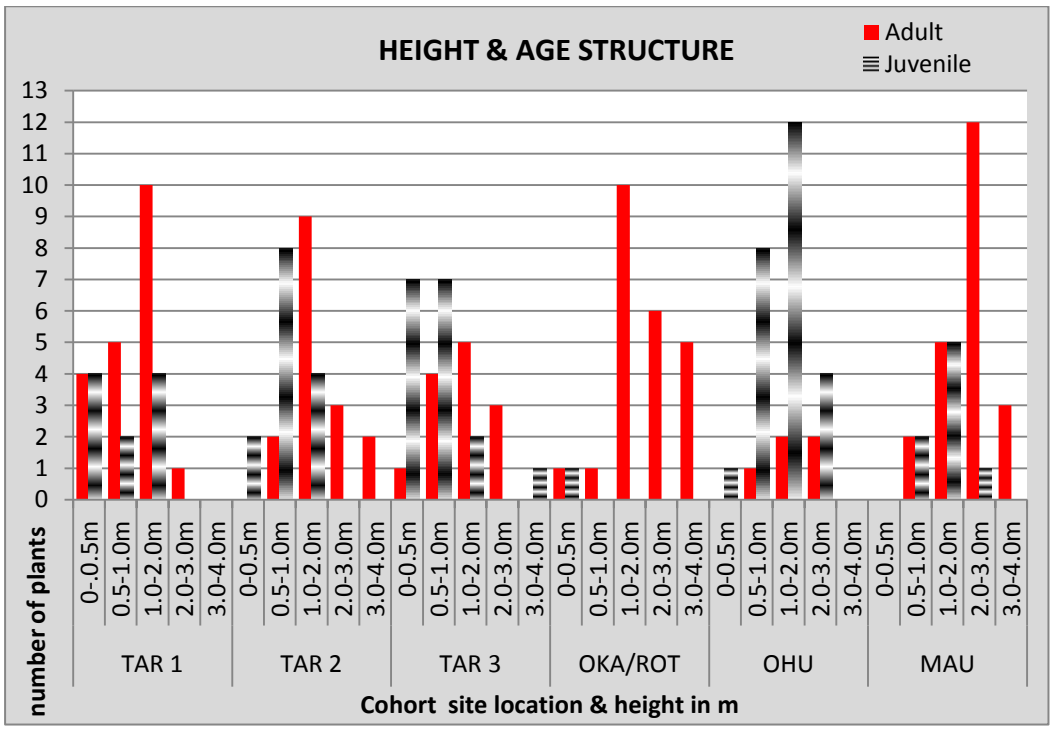


**Figure 3.2: Height of *S. aviculare* in all sites >20 plants, indicating cohort development.**

Overall cohort height structure shows adult and juvenile plants appear together at the height range up to 2m, with adult plants predominant over 2m. The exceptions are TAR 3 where one juvenile plant appears as an outlier, MAU with one plant up to 3.0m, and Ohutu where juvenile plants dominate. Crown spread structure follows height structure with similar appearance of adult and juvenile plants $<3m^2$ and adult plants dominating $>3m^2$, the juvenile outlier in TAR 3 appears also, but with no juvenile appearance in MAU over $1.0m^2$ and the juvenile plants also dominant at Ohutu. Spearman Rank Order Correlations show no significant correlation between height and crown spread between the sites, height p=1.00 and crown spread p=.10. There is significance between site and height with p=.03 at R=.853, but not site and crown spread p=.20. Apart from Ohutu and outliers the data shows that as height and crown spread increase, apparently the population is stabilising, juvenile plants decrease and adult plants dominate.

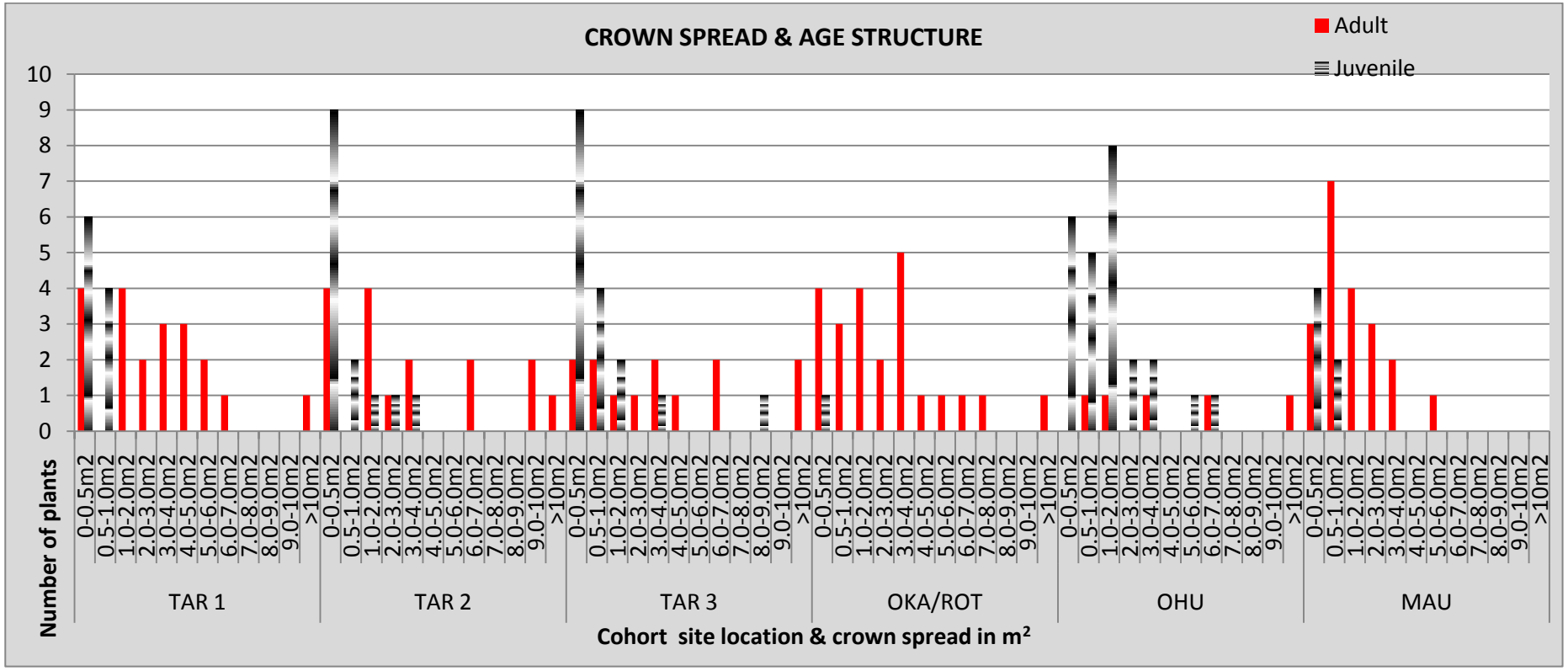


**Figure 3.3: Crown spread of *S. aviculare* in all sites >20 plants, indicating cohort development**

This is reflected in the metadata (n= 8) in that as plant height and crown spread increases, the frequency of adult reproductive plants increases. The frequency of adult and juvenile plants < 2 m in height was evenly spread; n = 73 adult, and n = 71 juvenile, with juvenile plants dominating <1.0m. There is a sharp decline in the frequency of juveniles as the height increases > 2m; n = 6 juvenile and n = 42 adult (Figure 3.4). The frequency of adult and juvenile plants < $3m^2$ crown spread was also evenly spread; n = 64 adult and n = 71 juvenile, with a sharp decrease in juvenile plants > $3m^2$; n = 6 juvenile and n = 51 adult (Figure 3.5). The highest frequency of total plants is in the < 2m height (n=144/192) and < $3m^2$ crown spread range (n=135/192), large plants >2 m height and >$3m^2$ spread reducing in frequency, the total numbers showing sharp decline; n = 48/192 height and n = 57/196 crown spread (Table 3.2). The metapopulation data summary of all 192 plants from all sites (n=8), Spearman Rank Order Correlation showed significant correlation between height and crown spread (p <.00), with a strength of R=.7723. The eight sites overall average height was 1.53 m and overall average crown spread was 2.41 $m^2$.

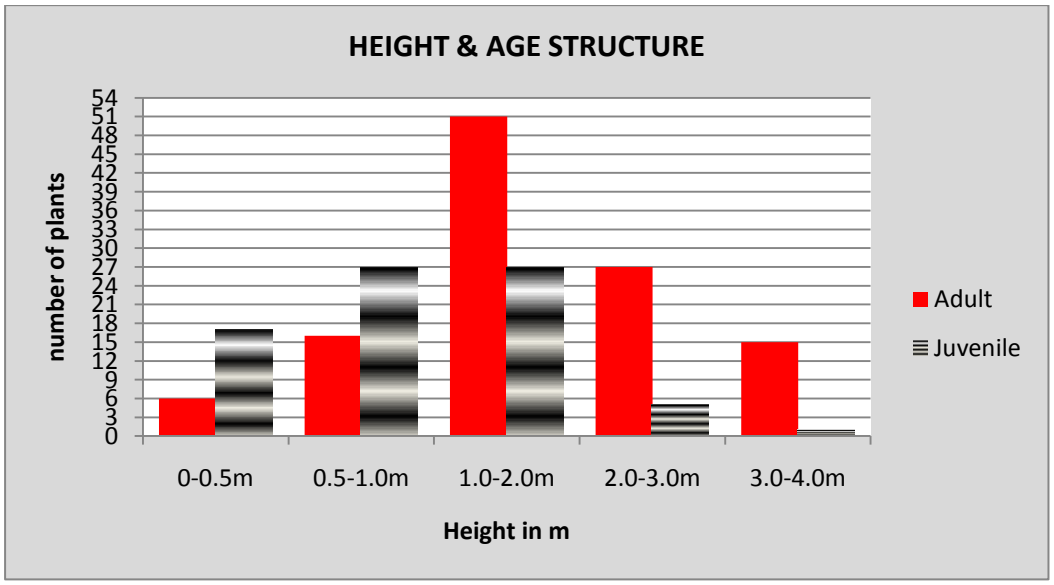


**Figure 3.4: Metapopulation height and numbers of plants (n=192) in relation to age of *S. aviculare* plants.**

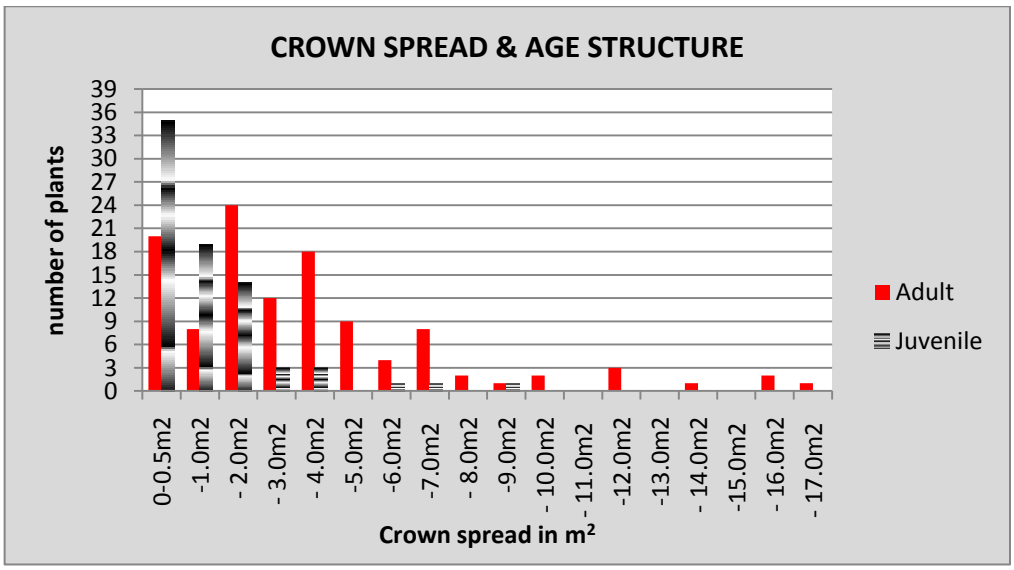


**Figure 3.5: Metapopulation crown spread and numbers of plants (n=192) in relation to age of *S. aviculare* plants.**

**Table 3.2: Metapopulation data summary showing numbers of plants below and above approximate 2m average height and 3m$^2$ average crown spread (n=192).**

| | HEIGHT | | CROWN SPREAD | |
|---|---|---|---|---|
| | **< 2.0m** | **> 2.0m** | **< 3.0m$^2$** | **>3.0m$^2$** |
| **Juvenile** | 71 | 6 | 71 | 6 |
| **Adult** | 73 | 42 | 64 | 51 |
| **Totals** | 144 | 48 | 135 | 57 |
| | 192 | | 192 | |

## 3.4 Discussion

*Solanum aviculare* appears to conform to the characteristics and described attributes of a light demanding pioneer (prisere) seral species as described in Brokaw (1985), Denslow (1987) and Huston & Smith (1987). The cohort data shows the pattern of population development in sites, and coupled with the metapopulation data, the inter site variation and pattern of height and crown spread differences may indicate a relationship of cohort development to site resource availability, prevailing environmental conditions and site precariousness similarly as described in Platt & Connell (2003) (refer Appendix 2).

Site TAR 1consisted of a shallow slip with low resource availability, a scoria sub-strate predominating and on-going instability and scouring caused by rain. The site was not directly exposed to wind, being sheltered to some extent by intact forest on all sides. Site TAR 2 was a steeper slip consisting of no scoria but deep low fertility ash, the middle of the slip was strongly scoured by rain with further small slips happening at the upper reaches. The site was exposed to wind funnelling up the valley leading up to and over the Tarawera Falls. Site TAR 3 was a very steep slip consisting of deep low fertility ash, some scoria and large obsidian rocks. The middle was very unstable with strong scouring by rain and regular sub-strate movement. The majority of *S. aviculare* plants were to the sides of the slip where the ground was more stable. The whole site was exposed directly to the south and the south west at the bottom, with some shelter to the middle and upper reaches from the south west by intact forest on the prevailing side. The OKA/ROT sites consisted of areas of higher resources, either forest edges or tree fall gaps consisting of mature higher fertility sub-strate material and soil, which in the main were sheltered from prevailing winds by surrounding intact forest. The URE sites consisted of two unstable slip sites and one stream edge site. Both slips were unstable with one (URE 1) actively moving when the data was being collected. The other slip was highly shaded as it had formed through the centre of a grove of predominant Tawa (*Beilschmedia tawa*). All of the URE sites were sheltered from prevailing winds by intact surrounding forest, although they were directly exposed to rain events. The MAU sites were in higher resource areas consisting of either forest track edges or Tawa tree fall gaps within mature canopies. They were also protected from prevailing winds by intact surrounding mature forest.

The two sites with <20 plants were TAU which was a high resource area consisting of plants on the edge of mature intact and actively managed forest, and the PIR sites consisted of track edge or tree fall gaps of medium to high resource within an intact forest reserve. The aberrant appearance an individual juvenile plant at TAR 3 at maximum height and grown spread and the predominance of juvenile plants at Ohutu are interesting outliers in the data and not fully explained. Those plants exhibited no sign of flower or fruit productions at the time the data was collected, were growing in the same sub-strate and environmental conditions as the adult plants and may relate to the site being

younger than the other sites, specific site differences or genetic causes. It was not possible within the scope and time frame of this research to explore these differences in more detail.

Both the cohort and metapopulation data demonstrate the frequency of plants at maximum height and crown spread (later growth stage) declines with maturity. Senescence occurred in >83% of plants at Tarawera sites that were recorded at ± three years of age. The height and crown spread of the plant in TAR 2 from which the trunk sample was recovered indicates plant growth was at maximum and late growth stage. Platt & Connell (2003) noted that disturbance sites can exhibit varying levels of resource availability, and pioneer species can develop specialisations to enhance colonisation and reproduction. *Solanum aviculare* is documented as living for seven to ten years, therefore senescence of these plants at three years, apparently reaching maximum growth and reproduction at an early age, may relate to low resource availability, stresses such as adverse climatic conditions and a precarious life style position in the slip sites, that a flexible specialised strategy of early rapid growth and reproduction can overcome to ensure seed spread before conditions may change (refer Huston & Smith 1987; Platt & Connell 2003). The plants observed at Tarawera sites all dying at approximate age 3 years did not unfortunately allow planned continuous growth monitoring and leaf morphology to be undertaken. This situation is not an uncommon phenomenon in plant studies, as Crawley (1990) noted many populations go extinct during the course of a study.

The metapopulation data of height and crown spread show growth to maximum size is rapid and significantly correlated, $p = .00$ (Table 3.2, Figure 3.5 & 3.6), and are most likely in response to the increased active photosynthetic sunlight levels available in the disturbance gap (Denslow 1987). This growth data (refer Table 3.2, Figure 3.5 & 3.6) coupled with the seed bank germination results (refer Part B Figure 3.6, Tables 3.4 & 3.5) further suggest that it is gap openings and soil disturbance that are a necessary requirement for supplying the light and subsequent heat requirements *S. aviculare* requires for growth and survival. The metapopulation and cohort growth data also shows the highest frequency of *S. aviculare* plants occurs at the approximate average height of <2m and the approximate crown spread average of $<3m^2$, at ± 1-2 years of age. The rapid

vegetative growth is in conjunction with early reproduction; adult reproductive plants appearing in similar frequency with juvenile plants at early growth stages. Following further growth the plants achieve maximum crown spread and height, with the frequency of plants then declining rapidly, conforming to the attributes of a prisere species as described by Brokaw (1985).

The rapid growth rates appear as a strategy providing an advantage to *S. aviculare* as well as facilitation to other early succession species, the advantage and facilitation increasing with height and crown spread size (Huston & Smith 1987). This rapid growth in height and crown spread, coupled with the sympodial acrotonic inflorescence development, and the number of adult reproductive plants appearing in early growth stages ensures that *S. aviculare* places fruit in a positive position for birds to predate and subsequently disperse seed early in establishment time. The early rapid growth, high seed numbers coupled with fruit production readily available for dispersal, appears as a specialised tactic or strategy to ensure that *S. aviculare* has produced and dispersed propagules, while maintaining a dominant light advantage over later species, which in the event of changing environmental or habitat conditions will ensure *S. aviculare* persistence (refer Horn 1974; Huston & Smith 1987; Platt & Connell 2003).

The definition of a species as early or late succession is relative to, and dependent upon the life history characteristics of the species (Platt & Connell 2003). The growth structure results of this study, while appearing as flexible early succession survival strategies for *S. aviculare*, also provide a degree of facilitation within the areas of secondary disturbance sites for the growth and survival of later canopy and other early successive species. In this research facilitation is considered 'somewhat consistent' between the classic model I and model II descriptions of Connell & Slatyer (1977), and model 3b of Platt & Connell (2003); in that *S. aviculare* appears to modify the environment making it somewhat more suitable for establishment of not only later successional species which will eventually replace *S. aviculare* (model I & 3b), but also for other early successional species, all of which are generally able to recruit and survive regardless (model II); but does not appear to make conditions suitable for recruitment of its own seedlings (model II) (Connell & Slatyer 1977; Platt &

Connell 2003). No apparent *S. aviculare* seedling growth was observed over a two year period under the canopies of any of the 192 plants documented, despite fruits senescing on the plants and apparently dropping to the ground. No further seedling establishment was observed over 18 months after senescence of the Tarawera plants, additionally suggesting light availability was not a limiting factor in the lack of seedling growth beneath canopies (refer Denslow 1987).

The expression of facilitation is related as 'somewhat consistent' with model I in determination of replacement, in that while growth and establishment of later canopy species such as *Weinmannia racemosa* and *Metrosideros excelsa* was enhanced, early successional plants *Coriaria arborea, Hebe stricta* and *Kunzea ericoides* in the slip sites were not actively inhibited. Those species were observed growing outside of *S. aviculare* canopies, but they grew in greater numbers beneath *S. aviculare* canopies, in conjunction with ferns and mosses which were generally growing more readily beneath *S. aviculare* (Plates 3.6 & 3.7).

Photo Moana Kapoor

**Plate 3.6: Photograph highlighting shading and early and late successional species growing beneath mature *S. aviculare* canopy TAR 2.**

Photo Moana Kapoor

**Plate 3.7: Close up photograph of Plate 3.6.**

Senescence of the plants in the slip sites did not allow for continued observance, documentation and analysis of this phenomenon. Facilitation is also not considered to be classic model I in the sense of *S. aviculare* providing essential modifications required for recruitment without which establishment would not proceed, such as *C. arborea* nitrogen fixation in primary succession (Walker et al. 2003). The slip sites are areas of secondary succession (model II & 3b), with the other secondary species as noted also being documented as being present and germinating from the soil seed bank (refer Part B Table 3.5).

*Solanum aviculare* is determined to be an 'opportunist' pioneer species that is able to sequentially occupy a space first and grow to maturity rapidly as described in Connell & Slatyer (1997) and Platt & Connell (2003), the differences to the models appear to lie in facilitative replacement mechanisms. Facilitation is therefore considered in *S. aviculare* to be the incidental provision of more favourable environmental factors that provide for enhanced germination and growth of other species, as observed, such as sub-strate stabilisation, including reduction of rainfall effects and subsequent scouring of unstable ash, moisture retention, humidity and shading due to the survival growth tactics employed by *S. aviculare*. The differences to the classic succession models are

not unrealistic as Huston & Smith (1987) and Platt & Connell (2003) remark that no general predictive theory on population dynamics effects, and classic successional principles, has yet been proposed based on underlying common successional principles.

# PART B: SEED BANK, DISPERSAL TACTICS AND AGENTS

## 3.5 INTRODUCTION

Seed dispersal is a critical plant process in colonisation, establishment and species survival; and zoochory (seed dispersal via animals) is a very effective method of dispersing propagules over distance and area (Thorsen et al. in press). Crawley (1986) noted that in habitats where frequent disturbance is common, many short-lived species maintain a seed bank of dormant seeds. These seeds are generally small, and where spread by frugivorous birds the fruits, such as *S. aviculare*, can exhibit orange and red colouration. In New Zealand 9.1% of 'gradually declining' flora species and 6.5% of flora species that are recognised as 'data deficient', (i.e. unevaluated taxa in the Threatened Plants listing de Lange et al. 2008) are dispersed by frugivorous vertebrates, and knowledge of a species dispersal tactics can be important for ecological and conservation planning (Thorsen et al. in press).

Huston & Smith (1987) documented that early successional plants generally have large quantities of small seeds with a large dispersal range, often spread by birds, and long viability with induced dormancy (stasis). Denslow (1987) noted that many light demanding species generally have smaller seeds than shade tolerant species, and maintain dormancy under intact canopies for long temporal periods, as current seed rain accounts for only a small portion of germination from forest top soils. Denslow (1995) indicated that high diversity of secondary successive species in a disturbance gap can be produced from, and through soil seed stock germination from heterogeneous surrounding forest. Germination of these species is in response to the increased incident radiation and temperatures triggered by the canopy opening (Denslow 1987). Honda (2008) documented that life history and small seed size are closely related to the formation of a

persistent seed bank; defined as survival in soil for more than five years. Small seeded species were dependent on light for germination, burial preventing germination by light reduction. With soil depth there is a decline in temperature fluctuation, and the seeds ability to 'detect' soil depth is a factor in prevention of germination until depth decreases. Rowarth et al. (2007) noted that a seed bank, formed by bird dispersed species of between 10-100 seeds $m^{-2}$ was considered sufficient to maintain the species, and Ogden (1985) documented 'gap colonising' species with bird dispersed seed also maintain seed banks of 10-100 $m^2$.

In New Zealand endozoochory by frugivorous vertebrates is a dispersal mechanism in three *Solanum* species (Thorsen et al. in press). McEwen (1978), Dijkgraaf (2002) and Campbell (2006) documented kererū (*Hemiphaga novaeseelandiae*) as consuming *S. aviculare* fruits (seed 2 mm), and blackbirds (*Turdus merula*) are known to predate and disperse New Zealand small seeded (2-5mm) frugivorous species (McEwen 1978). Stanley & Lill (2002) documented the mean gut passage times of seed through silvereyes as 31.5 minutes, with 86.7% viability of *S. aviculare* seed recovered from silvereye faeces. Small birds have less gut retention times than larger birds, and viability can be reduced in dispersal over distance with larger birds, as longer retention times and gut treatment (maceration) can influence seed viability; seed retention time being a trade-off for a plant between seed viability and distance for dispersal (Wotton et al. 2008; Thorsen et al. in press).

Clout & Tilley (1992) concluded that any positive effect on germination of Miro (*Prumnopitys ferruginea*) seed through kererū gut passage was weak, and the most likely positive effect of kererū ingestion is long distance dispersal. Rowarth et al. (2007) noted in New Zealand that most bird-dispersed seeds do not need to have avian gut passage to aid germination, and Traveset (1994) noted internationally, that germination effects in most species also were not associated with gut passage times; other factors including the seed itself, such as the seed coating and surface may influence germination either positively or negatively after frugivore ingestion. The effects of glycoalkaloids in *Solanum* fruits on gut passage are not completely known, but may influence dispersal by either aiding or inhibiting gut retention time (Traveset 1994).

Williams et al. (2000), in feeding trials of wild ship rats documented that while fruit handling and eating behaviour differed between animals, they consumed the flesh and seeds of *S. aviculare*, passing the seed in large numbers while not destroying the small seeds. Viable germination of 64% (n=100) was documented in *S. aviculare* recovered seed. Non gut treated petri dish controls (n=150) returned up to 76% viable germination.

Burrows (1996, 1999) documented that *Solanum aviculare* and *S. laciniatum* are weak stemmed colonisers of disturbed sites, likely spread by bird endozoochory (dispersal via animal gut transmission) and with both species likely to form a medium term seed bank of seeds with induced secondary dormancy (dormancy not engendered by parent), or stasis due to environmental constraints.

This research aimed to answer the questions: 1) does a seed bank consisting of viable *S. aviculare* seeds exist, 2) what dispersal agents are involved in its formation and the spread of viable *S. aviculare* seed, and what transport mechanisms do they employ, and 3) what dispersal tactics, if any, are displayed by *S. aviculare*, occurring within a secondary disturbance regime in lowland podocarp-broadleaved forest of the eastern Bay of Plenty and Waikato, New Zealand? The aims of the research were undertaken by conducting *ex situ* soil seed bank cores, animal and bird seed viability and germination after gut transmission trials.

## 3.6 Methodology

### 3.6.1 Seed Bank

#### 3.6.1.1 Trial design and substrate collection

Soil cores were collected at the three Tarawera slip sites with a 100mm corer. Core sampling was adapted from Partridge (1989), and consisted of 40 cores per slip site for a total of 120 cores (± .03 $m^3$ total volume of soil). 20 cores (10 at 2.5cm depth and 10 at 2.5cm – 5cm depth) were taken randomly from beneath mature *S. aviculare* canopy within the slip site (Plate 3.8), and 20 cores (10 at 2.5cm and 10 at 2.5cm -5cm depth) from the ‘shade canopy’ expanded-gap area within surrounding heterogeneous forest either side of the slip site from within a

$50m^2$ area. The $50m^2$ consisted of one 5m x 5m ($25m^2$) randomly placed quadrat either side of the slip, with 10 cores (5 at 2.5cm and 5 at 2.5cm-5cm) taken from within each quadrat. Cores were selected randomly within each quadrat. Each quadrat, while being chosen randomly, was located a minimum of four metres from the edge of each slip for safety purposes.

The total volume of soil removed was calculated large enough to provide a reasonable representation of the substrate area prior to disturbance, and indicate possible seed numbers with a degree of precision at 0.3 - 0.4 (Forcella et al. 2003). The cores from the expanded-gap area were aimed to get a fair representation of the intact seed bank, and the cores from beneath the mature plants in the slips to represent both intact seed bank, and fresh seed from fruit drop.

*Ex situ* germination of seed bank material was conducted to standard conditions similarly to Burrows (1999) in an unheated glasshouse, with shade cloth around the sides to minimise direct sunlight, and open windows providing air movement to reduce overheating and ensure conditions were as close as possible to natural gap and seasonal conditions. Each tray was lined with shade-cloth material to ensure no loss of soil through drainage holes. Two core samples per site from the same depth were combined in a single tray as one germination sample, giving a total of 60 trays (Plate 3.9).

The trays were watered regularly to maintain moisture content, and air temperatures were recorded daily at 300mm and 1200mm height above the trays, with a standard Brannon brand maximum/minimum thermometer. Soil temperature readings were recorded daily with an RS Components electronic thermometer with a stainless steel probe. All temperatures were recorded at the same time and before watering commenced. The germination trial ran for 273 days (9 months) from 15$^{th}$ Sept.2008 – 14$^{th}$ June 2009, and temperatures were collated in three month increments to approximate spring, summer and autumn timings (Appendix 3).

Photo Moana Kapoor

**Plate 3.8: Soil core recovery under poroporo canopy TAR 3/2.**

Photo G. Weavers

**Plate 3.9: Soil cores combined in trays in glasshouse.**

3.6.1.2 Vegetation survey

Vegetation species composition from the expanded-gap were adapted from Partridge (1989), and recorded in modified Recce plots as per Leathwick (1987), and Hutcheson (1996. Recordings were taken from within the 25 $m^2$ randomly placed soil sample quadrat, one plot either side of the slip sites. All woody shrub and tree species were counted as well as tree ferns, *Cordateria* and *Cordyline* spp. (Appendix 4).

3.6.2 Seed viability and dispersal agents

Glasshouse germination and chemical seed viability trials were undertaken to determine quantities of, and the dispersal agents associated with spread, of viable *S. aviculare* seed. Bird recovered seed identification was determined under Olympus SD 30 binocular microscope using Webb & Simpson (2001) as a guide.

3.6.2.1 Bird recovery-trial design

Tarawera

Three fresh purple/red coloured faeces were recovered after observing an individual blackbird defecating on the Lake Tarawera to Tarawera Falls track (approximately 1 km from the Lake Tarawera campground) on 9th Jan. 2009. Two dried faeces, apparently from kererū, were collected from the base of a tawhero (*Weinmannia racemosa*) tree on the left (west) side of the same track approximately 10m from the deposited blackbird faeces, after observing a kererū roosting there and subsequently flying off. The base of the tree had numerous pigeonwood/poroporokaiwhiria (*Hedycarya aborea*) seedlings growing, with no adult tree in the vicinity, lending credence to the deposition by kererū.

Tauranga

A faeces collection point (used in conjunction with *S. aviculare* ripening) was located beneath a pigeonwood tree growing on the side of the main forest access track. This appeared to be a kererū roosting site as it had numerous *S. aviculare* seedlings growing directly beneath the overhanging branches (Plate 3.10). Inspection on 5th May 2009 showed *S. aviculare* plants spread over the whole property had scattered numbers of fruit ripening, and showed signs of predation. While the pigeonwood fruits were still green and no obvious predation or roosting had taken place, further inspection revealed that supplejack/kareao (*Ripogonum scandens*) fruits in the vicinity of the pigeonwood tree were fully ripe and being consumed. A large quantity of fresh, bright red faeces was collected along the track beneath tawa and *S. aviculare* plants. The faeces were considered to be kererū due to the large quantity and seed sizes contained within the faeces, as well as flight sounds heard in close proximity. In addition three blackbird faeces were collected containing small seeds.

All faeces were stored in a refrigerator until 9th May 2009 when they were washed with running water and sieved. Seeds and remaining pericarp skin were dried on paper towels in plastic petri dishes for 48 hours. Seeds were examined under Olympus SD 30 binocular microscope for identification. *Solanum aviculare* seeds were removed and dried on paper towels for 12 hours, and dry stored in home refrigerator in sealed containers.

Photo G. Weavers

**Plate 3.10: *S. aviculare* seedlings beneath mature pigeonwood tree, Tauranga.**

3.6.2.2 Rat feeding and recovery – experimental design

Approval for Experiments on Animals was obtained from the Animal Ethics Committee of The University of Waikato (protocol No. 754), and training in animal and unit protocols also undertaken (SOP #5). Rat feeding protocols consisted of two wild ship rats (*Rattus rattus*) housed in the Animal Behavioural Unit at The University of Waikato fed dried ripe *S. aviculare* fruit, and fresh ripe *S. aviculare* and *S. laciniatum* fruit over a five day period. The dried ripe fruit was fed for the first two consecutive days. Fresh ripe *S. aviculare* fruit was collected and fed on day three, and fresh ripe *S. laciniatum* fruit collected and fed on day four. Due to unavailability of fresh fruit for day five, 100 *S. aviculare* seeds, microscope sorted for estimated viability, were inserted in four fresh grapes, and fed for one day (Table 3.3). Faeces were collected every second day, then soaked in water until soft, sieved through a wire tea strainer, and dried on paper towels to recover seeds. Seeds were then removed and refrigerator stored.

**Table 3.3: Type and timing of feed presented to rats.**

| FEED | Day 1 | Day 2 | Day 3 | Day 4 | Day 5 |
|---|---|---|---|---|---|
| Dried ripe *S. aviculare* fruit | X | X | | | |
| Fresh ripe *S. aviculare* fruit | | | X | | |
| Fresh ripe *S. laciniatum* fruit | | | | X | |
| *S. aviculare* seeds in grape | | | | | X |

3.6.2.3 Seed viability-tetrazolium-experimental design

Tarawera seed viability was determined only by germination, and no identification by microscope or counts made. Tauranga bird and rat faeces seed viability was determined by using the tetrazolium chemical technique, and germination. The tetrazolium technique is a chemical staining process using 1g 2, 3, 5-triphenyl-2H-tetrazolium chloride in a solution of 5ml ethanol, diluted with water to 200ml to produce a 0.5 % solution. The technique acts through a reduction reaction of the tetrazolium chloride that detects dehydrogenase activity in active mitochondria of living tissues, indicating embryo tissue viability, living embryonic tissue stains red and dead tissue does not. The intensity of colour is related to the number of mitochondria present (Freeland 1976; Kearns & Inouye 1993; Stanley & Lill 2002; Grove 2005; Marrero et al. 2007).

Seeds were soaked overnight (17 hours) in tap water, cut in half longitudinally then soaked in the 0.5 % tetrazolium solution for four hours at $30^{o}$ C. A solution of 0.5 % solution rather than 1 % solution was used, as previous studies indicated that deeper staining was produced with a 0.5 % solution (Stanley & Lill 2002; Grove 2005). A control to determine seed viability without gut maceration consisting of 10 unstratified *S. aviculare* seeds was used. The seeds were randomly chosen from the Okataina plant 5 used in seed germination trial # 3 (refer Part C), and sorted under Olympus SD 30 binocular microscope to determine estimated viable seed.

3.6.2.4 Seed viability-germination-trial design

The Tarawera bird faeces were placed on standard commercial seed raising mix, lightly covered with mix, placed in an unheated glasshouse, and kept moist by

watering. The Tarawera trial was run for 235 days 33.5 weeks) starting on the 10th January 2009 and ending on 3rd September 2009. No control was used. The Tauranga bird and the rat recovered seed were spread on standard commercial seed raising mix and placed in a heated bed glasshouse at The University of Waikato glasshouse complex for 21 days, then removed and kept in the unheated glasshouse used for soil seed bank germination due to the author having to move off campus. The Tauranga trial was run for 65 days (10 weeks) starting on the 24th June 2009 and completed on the 3rd Sept 2009. A control of 30 *S. aviculare* seeds using the same seed from germination trial # 3, Okataina plant 5 was used. Soil and air temperatures were recorded daily through the duration of the trials.

## 3.7 Results

3.7.1 Seedbank

Eight *Solanum aviculare* plants germinated**.** Of the eight *S. aviculare* seeds that germinated, four germinated from under mature *S. aviculare* canopies, all at surface to 2.5cm depth, and four germinated from within established canopy gaps ('shade canopy'), two at surface to 2.5cm, and two at 2.5cm to 5.0cm depth (Table 3.4). The first *S. aviculare* to germinate was recorded on 10th Jan. 2009, 17 weeks (118 days) after the trial started, and germination continued over the following 9 week (61 day) period. No *S. aviculare* plants germinated after the 9 week (61 day) period ended, and the termination of the trial. The quantity of *S. aviculare* seed found in the soil seed bank was calculated from the 100mm core to approximate numbers per $m^2$, and is approximately 1-2 seeds 0.01 $m^2$, equal to100-200 seeds $m^2$.

**Table 3.4: Germination times, average soil temperature, quantity, site locations and depths from *S. aviculare* soil seed bank germination.**

| Date | $^{o}$ C | Quantity | Site | Core location & depth |
|---|---|---|---|---|
| 10.01.09 | 25.25 | 1 | TAR 3 | poroporo @ 2.5cm |
| 10.01.09 | 25.25 | 2 | TAR 3 | poroporo @ 2.5cm |
| 10.01.09 | 25.25 | 1 | TAR 2 | in canopy @ 2.5cn |
| 10.01.09 | 25.25 | 1 | TAR 1 | in canopy @ 2.5cm |
| 22.02.09 | 25.3 | 1 | TAR 1 | poroporo @ 2.5cm |
| 12.03.09 | 19.5 | 1 | TAR 2 | in canopy @ 5.0cm |
| 12.03.09 | 19.5 | 1 | TAR 3 | in canopy @ 5.0cm |

A total of 117 indigenous woody plants germinated, consisting of 11 genera. The most common being *Hebe stricta* (n=33), *Kunzea ericoides* (n=18), *Geniostoma rupestre* (n=15), *Weinmannia racemosa* (n=13), and *Carpodetus serratus* (n=12). Nine of the 11 genera had been recorded in the Recce vegetation plots (Appendix 3). Sixteen monocotyledon plants of rush (*Baumea* spp.), and eight plants of sedge (*Carex* spp.) also germinated, but were not recorded. The first plants to germinate on 23rd Sept. 2008, eight days after the trial started, were various weed species, and were not recorded. The first indigenous plant to germinate was recorded on 1st Dec. 2008; 11 weeks (78 days) after the trial started (Table 3.5).

**Table 3.5: Taxa, time and average soil temperature at germination of all indigenous species in seed bank germination trial.**

| | **Date & average soil temperature at germination** | | | |
|---|---|---|---|---|
| **TAXON** | **Dec 2008 23.9°C** | **Jan 2009 25.76°C** | **Feb 2009 25.39°C** | **Mar 2009 21.89°C** |
| *Aristotelia serrata* makomako | X | | | |
| *Carpodetus serratus* putaputawētā | X | | | |
| *Coriaria arborea* tutu | X | | | |
| *Geniostoma rupestre* hangehange | | | X | |
| *Hebe stricta* koromiko | | X | | |
| *Rubus cissoides* tātāramoa, bush lawyer | | X | | |
| *Kunzea ericoides* kānuka | X | | | |
| *Myrsine australis* māpau | X | | | |
| *Pseudopanax crassifolius* horoeka, lancewood | | X | | |
| *Solanum aviculare* poroporo | | X | X | X |
| *Weinmannia racemosa* tawhero | X | | | |

The highest air temperature recorded for the duration of the trials was $46.0^0$ C and $-3.5^0$C the lowest. The highest and lowest soil temperatures were $31.5^0$C and $3.5^0$C respectively. The average maximum soil temperature for the first increment was $18.66^o$C, and the average minimum was $14.59^o$C, although

temperatures were consistently above 20°C from 11th Nov. 2008. For the second increment the average maximum was 26.16°C, and minimum 22.94°C, and the third increment average maximum was 18.16°C, and minimum 15.76°C (full recordings in Appendix 3). From the 11th Nov. 2008–23rd Mar. 2009, a period of 19 weeks (133 days), the soil average maximum temperature was 25.97°C and average minimum 22.63°C. The germination of native seedlings starting on 1st Dec.2008 and *S. aviculare* on 10th Jan.2009 appears to coincide with the increase in soil temperatures averaging > 20° C within those times (Figure 3.6).

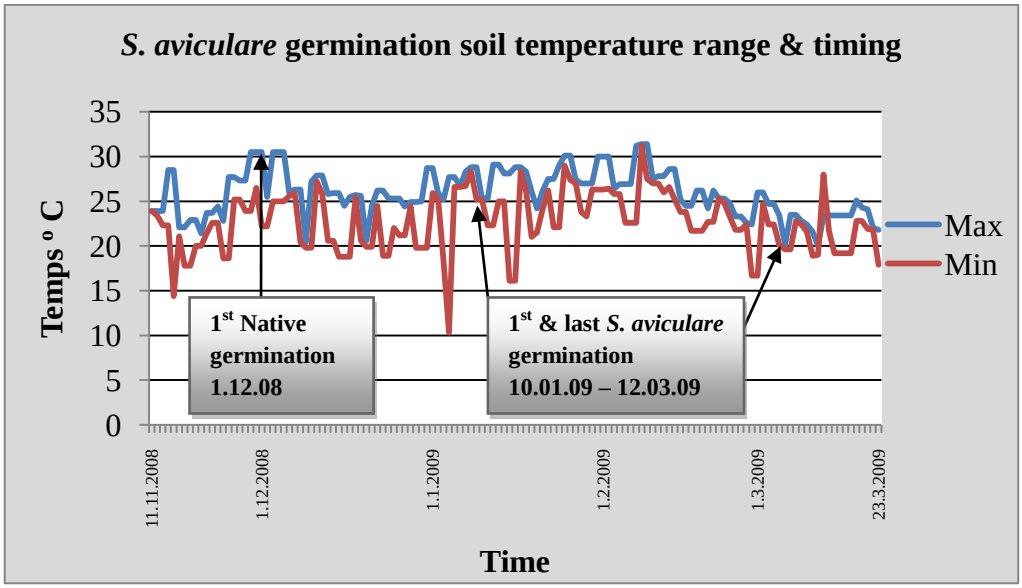


**Figure 3.6: Soil temperature range between dates averaging continuously above 20° C, with arrows highlighting start of germination of native plants and start and finish of *S. aviculare* plants.**

3.7.2 Seed viability and dispersal agents

A total of 702 seeds were recovered from kererū and blackbird faeces from the Tauranga sites. There were nine identified taxa, consisting of six genera and six unidentified taxa (Table 3.6). There were 126 *S. aviculare* seeds recovered, of which 83 were used for germination, and 43 used for the tetrazolium test. Due to my error the blackbird and kererū faeces from Tauranga were placed in the same container and could not be separated, and were counted together in the tetrazolium and germination trials, being referred to as kererū/blackbird. The

quantity and size of the seeds recovered indicated the majority of seeds were from kererū faeces (refer McEwen 1978).

**Table 3.6: Genera, species, seed numbers, size and shape of seed recovered from Tauranga bird faeces.**

| Genera | Number | Average seed length | shape |
|---|---|---|---|
| ***Coprosma grandifolia*** | 13 | 6mm | ovate |
| ***Coprosma lucida*** | 7 | 6mm | ovate- elongate |
| ***Coprosma repens*** | 8 | 5mm | ovate-elongate |
| ***Coprosma robusta*** | 8 | 4mm | ovate-elongate |
| ***Muehlenbeckia australis*** | 26 | 6mm | trullate |
| ***Pittosporum umbellatum*** | 131 | 2mm | ovoid |
| ***Prumnopitys ferruginea*** | 1 | 18mm | ovoid |
| ***Ripogonum scandens*** | 11 | 7mm | oval |
| ***Solanum aviculare*** | 126 | 2mm | obovate |
| **Others** | 371 | 1 – 4mm | various |

A total of 511 *S. aviculare* and *S. laciniatum* seeds were recovered from the combined rat faeces. There were 43 seeds used for the tetrazolium tests and remaining 468 used for germination. The seeds of *S. aviculare* and *S. laciniatum* were recorded together due to distinguishing difficulties associated with faeces recovery. Results of feeding behaviour showed individual differences between rats, similarly to Williams et al. (2000). Rat S14 ate the complete fruit at all feedings while S13 at first feeding initially ate only the pulp beneath the pericarp, leaving the seed cluster intact, eating most of the whole fruit at the second and third feedings, but still leaving some seed cluster and skin uneaten. The control in the tetrazolium test returned 100 % viable seed (n=10/10), and 76.75% (n=23/30) in the germination trial (Table 3.7).

3.7.2.1 Birds – Tarawera - germination.

A total of 13 tutu (*Coriaria arborea*) plants and one unidentified plant germinated between 10.1.09 and the termination of soil seed bank trials on 14$^{th}$ June 2009. A further 53 tutu plants germinated between 14$^{th}$ June 2009 and 3$^{rd}$ Sept. 2009 when the trials were completed in conjunction with the 10 week seed viability germination trials. No germination ensued from the kererū faeces. No *S.*

*aviculare* seed germinated from the kererū or blackbird recovered Tarawera faeces.

3.7.2.2 kererū/blackbird – Tauranga – tetrazolium

Using the tetrazolium test the kererū/blackbird recovered seed returned 18.6 % (n=8/43) viable seed. It was noted in the tetrazolium tests that a number of seeds recovered from birds were desiccated, and although many appeared viable, (i.e. not desiccated and displaying radicle and endosperm) these apparently viable seeds produced no staining. All stained seeds showed deep staining (Table 3.7).

3.7.2.3 kererū/blackbird –Tauranga – germination

Germination of the kererū/blackbird recovered *S. aviculare* seed after 10 weeks was 2.5% (n=2/83) (Table 3.7).

3.7.2.4 Rats – tetrazolium

Using the tetrazolium test the rat recovered seed returned 76.75 % (n=33/43) viable seed (Table 3.7). Two seeds showed light staining, the remainder were deeply stained.

3.7.2.5 Rats – germination

Germination of rat recovered seed after 10 weeks was 39% (n=182/468) (Table 3.7).

**Table 3.7: Number and percentage viability of recovered seeds using tetrazolium staining of embryonic tissue (T) and germination (G).**

| | **Number of seeds** | | **Number viable seeds** | | **% viable seeds** | |
|---|---|---|---|---|---|---|
| | **T** | **G** | **T** | **G** | **T** | **G** |
| **RAT** | 43 | 468 | 33 | 182 | 76.75 | 39 |
| **BIRD** | 43 | 83 | 8 | 2 | 18.6 | 2.5 |
| **CONTROL** | 10 | 30 | 10 | 23 | 100 | 76.75 |

## 3.8 Discussion

The seedbank trial (refer Tables 3.4 & 3.5) identifies that a soil seed bank of *S. aviculare* exists, consisting of viable seeds that appear to remain in stasis due to burial until disturbance induces germination, indicating secondary rather than primary dormancy (Burrows 1999). The seed bank consists of 1-2 seeds per 10cm x 10cm core equating to 100-200 seeds $m^2$, which reaches the threshold Rowarth et al. (2007) suggests is sufficient to maintain the species, although the results are predominantly at the lower scale at around 100 seeds. The seed bank germination cues indicate to be both a reduction in depth, shown by the core removal and placement in trays, and soil temperature increase due to increased incident sunlight warming the soil, as germination did not take place in *S. aviculare* or native species until soil temperatures rose on average above 20$^{o}$C (refer Figure 3.6); supporting Burrows (1999), Denslow (1987), and Honda (2008) results of germination in response to light, temperature and depth reduction.

Spread of viable seed by kererū/blackbird was confirmed in *S. aviculare* (Table 3.7) as well as *C. arborea*. The results indicate that kererū/blackbird distribute viable seed of *S. aviculare* to the forest floor, though not at high rates of viability; the germination and tetrazolium tests showing the viability of *S. aviculare* seed through kererū/blackbird predation and gut passage to be low at 18.6%, compared to the control. This is in contrast to results recorded by McEwen (1978) where kererū dropped seed was generally considered to have high percentage viability (specifically *Melicytus ramiflorus*, seed size ±2mm and *Rhopalostylis sapida*, seed size ±12mm produced 84% and 86% germination respectively).

Tetrazolium tests are an accepted research tool providing strong indications of seed viability, especially where confirmation is provided by subsequent germination (Freeland 1976; Kearns & Inouye 1993; Stanley & Lill 2002; Grove 2005; Marrero 2007). The results of the tetrazolium viability tests on each dispersal agent are consistent with, and confirmed by the recovered seed germination trials run for 10 weeks in potting media (refer Table 3.7). Differences can be expected between germination and tetrazolium due to potential predation and pathogens being present within potting media.

The results documenting high viability in control treatments shows large numbers of viable seeds exist within fruit before predation and fruit abscission. Although the viability trials provide no further information on whether differing gut passage times may relate to the bird recovered seed viability (Clout & Tilley 1992; Stanley & Lill 2002), the germination of viable *C. arborea* seed from Tarawera blackbird recovered seed may potentially indicate that blackbird predation and maceration may not be as destructive as kererū, similarly to Stanley & Lill (2002), where silvereye recovered seed viability was high in *S. aviculare* and other small seeded species; and Wotton et al. (2008) where small birds demonstrated less gut retention time than kererū. The unfortunate mixing of the kererū and blackbird faeces does not allow a comparison of viability between the two birds. Whether glycoalkaloids in the seeds may influence gut passage time remains unanswered.

The total germination of viable bird recovered seed does appear to indicate seed dispersal via bird endozoochory to be a colonisation tactic employed by *S. aviculare*, rather than a necessary requirement of gut passage for germination (refer Clout & Tilley 1992; Traveset 1994; Rowarth et al. 2007; Wotton et al. 2008; Thorsen et al. in press). The quantity of viable seed in the seed bank is enough to maintain the species, and as such may also highlight that plant tactics may relate to dispersal of large seed quantities rather than gut passage requirement. The production of large numbers of seed would ensure that despite ingestion and maceration, enough viable seed is dispersed and placed on the forest floor to maintain the species survival (refer Wotton et al. 2008; Thorsen et al. in press).

The results of the ship rat tetrazolium seed viability tests show a high rate of viability of 76.75%. The quantity of recovered *S. aviculare* and *S. laciniatum* seed from rats fed ripe fruit correspond to Williams et al. (2000) whose work documented that ship rats fed *S. aviculare* fruit passed abundant amounts of seed with little destruction, with a germination rate from the recovered seed of 64.0%. This research also therefore also indicates that ship rats can potentially be dispersers of viable *S. aviculare* seed, the quantity of seed dispersed, and the range being influenced by their behaviour including feeding habits as documented. The rat recovered seed similarly to the bird recovered seed also

displayed a portion of seeds (though less than bird recovered) that had radicles, endosperm, and appeared viable but produced no staining. While relationships between seed germination and gut passage time are unpredictable in small frugivorous mammals (Williams et al. 2000), the overall viability and germination being higher in rats than birds, suggests less potential maceration, and possible reduced gut passage time effects due to requirements for ejection of indigestible seed to increase food assimilation in small mammals.

# PART C: SEED GERMINATION

## 3.9 INTRODUCTION

Rowarth et al. (2007), in reviewing New Zealand indigenous species germination requirements identified that many species exhibited a flush of germination in spring followed by slow germination in autumn and winter, although many colonising species including *S. aviculare* and *S. laciniatum* exhibited germination patterns that relate to ecological conditions. Burrows (1996, 1999) tested variation in ecological conditions for seed germination in *S. aviculare* and *S. laciniatum*. Germination treatments in autumn-winter produced moderate results; *S. aviculare* partly exposed on soil mix produced germination of 46-58% (n=100), and *S. laciniatum* partly exposed on soil mix 56 % (n=50). Seeds of *S. aviculare* became dormant and remained in stasis when buried at 5cm for 22 months, with 56 % (n=25) of recovered seeds germinating when unearthed and placed in favourable conditions (Burrows 1999). *Solanum laciniatum* recovered seeds germinated readily after 18 months burial at 5cm at 100% (n=25) success (Burrows 1996). The delayed germination apparently being influenced by secondary dormancy pre-conditioning or stasis induced by reduction in light and factors associated with a burial environment, rather than primary dormancy from bio-chemical blocking.

The aims of this research were to determine and quantify seed germination characteristics of *S. aviculare* in relation with previous studies. The research trials were designed to assess and answer questions: 1) what preferential germination times does *S. aviculare* exhibit, 2) are any seasonal changes in day-length and temperature apparent, 3) what species differences may exist, 4) what

colonisation timing and germination tactics may relate to induced stasis and fresh seed, and 5) is there a germination response to depth cues in relation to ecological conditions such as exposure and reduced depth due to perturbation?

## 3.10 Methodology

Three seed germination trials were undertaken, Trial 1 commenced on 7th September 2008, Trial 2 commenced on 15th February 2009, and Trial 3 commenced on 20th April 2009. All germination assessments were for 10 week periods. Trials were undertaken in a commercial nursery to standard practice protocols similarly to Burrows (1996, 1999), using commercial mix with no added fertiliser. A control germination of fresh *S. aviculare* seed in petri dishes was conducted to compare with the first and third trials. Petri dish trials occurred at the author's home. Temperatures were not recorded.

### 3.10.1 Trial design and data analysis

Germination treatments were adapted from Burrows (1999). Each trial consisted of four treatments utilising dry stored stratified and fresh seed. There were 100 seeds per treatment (four replicates of 25 seeds), for a total of 400 seeds per trial (Table 3.8). Data was entered into Microsoft Excel 2007 and histograms produced.

**Table 2.8: Germination treatments undertaken for each seed germination trial.**

| # Seeds | # Replicates | Total seeds | Location in tray | Seed type |
|---|---|---|---|---|
| 25 | 4 | 100 | Surface scatter, partial burial | fresh |
| 25 | 4 | 100 | 2cm burial | fresh |
| 25 | 4 | 100 | Surface scatter, partial burial | dry |
| 25 | 4 | 100 | 2cm burial | dry |
| 25 | 2 | 50 | Petri dish control | fresh |

*Solanum laciniatum* seed, dry stored in a standard refrigerator to provide stratification and replicate stasis induction, and fresh collected *S. aviculare* seed were used in each trial and petri dish controls. The stratified *S. laciniatum* seed

used in trial one and two was 18 months old at commencement of the first trial, collected on 3rd March 2007. In trial three the stratified *S. laciniatum* seed was 14 months old collected on 5th Feb. 2008. Fresh *S. aviculare* seed for trial one, two, and for the first petri dish control were collected from ripe fruits on 25th June 2008 at Tarawera sites TAR 2 & TAR 3. The fresh *S. aviculare* seed for trial three and second control was collected from ripe fruits on 14th April 2009 from Okataina plant 5. Trial two used the same *S. aviculare* seed collected for the first trial due to lack of ripe fruit being available. To ensure stratification was not provided the fresh collected seed was not refrigerator stored.

Fresh seeds were removed from the pericarp by crushing the fruit carefully, and sieving with a standard kitchen sieve and cheese cloth, followed by drying on paper towels for four days at room temperature. Fresh seeds were examined under an Olympus SD 30 binocular microscope to exclude obvious damaged and considered non-viable seeds.

## 3.11 Results

In the first trial 101/400 seeds germinated, consisting of 44/200 fresh seeds, and 57/200 dry stored seeds for a total percentage germination of 25.25 %. In the second trial 18/400 seeds germinated, consisting of 13/200 fresh and 5/200 dry stored for a total germination percentage of 4.5%. In the third trial 107/400 seeds germinated, consisting of 62/200 fresh seeds and 45/200 dry stored seeds for a total germination of 26.75 %. A total of 17/50 seeds germinated in the initial petri dish trial for a germination of 34 %. The initial petri dish germination started 8th Oct. 2008, and no further germination took place after 16th Oct. 2008. There was no germination in the second petri dish trial at the cessation of germination trials on 22nd July 2009 (Table 3.9).

## 3.12 Discussion

The results of 25.25% and 26.75% in the seed germination trials replicating ecological conditions were lower than Burrows (1996) (n= 56%) and (1999) (n=46-58%) results (refer Table 3.9).

**Table 3.9: Seed type, numbers and total percentage of seed germination at differing depths in seed germination trials.**

| | Species & type | 2cm depth number | Surface number | Total number | % |
|---|---|---|---|---|---|
| **#1 Spring** | Dry-*S. laciniatum*<br>Fresh-*S. aviculare* | 43<br>38 | 14<br>6 | 101 | 25.25 |
| **#2 Summer** | Dry-*S. laciniatum*<br>Fresh-*S. aviculare* | 1<br>1 | 4<br>12 | 18 | 4.5 |
| **#3 Autumn** | Dry-*S. laciniatum*<br>Fresh-*S. aviculare* | 25<br>27 | 20<br>35 | 107 | 26.75 |
| **control #1** | *S. aviculare* | petri dish | | 17 | 34 |
| **control #2** | *S. aviculare* | petri dish | | 0 | 0 |

The trials timings and treatments corresponded to Burrows (1996, 1999), commercial potting mix was used, although not pasteurised, and replicates were higher in these trials than Burrows; therefore comparisons can be made. No explanation can be offered as to why the results were lower, except for lack of pasteurisation and the second trial where error in the trial protocol is considered to be the reason for the extremely low results. The second trial protocols were not supervised by the commercial operator who laid out the first trial. The third trial was supervised by the operator, and the similar results in the third to the first trial confirm operator error in the second trial as the likely cause. However sufficient similarities in protocols exist between the research trials to allow valid comparisons to be made between the trials, although the germination of *S. laciniatum* with stasis replicated and *S. aviculare* fresh collected were generally inconclusive as to any differences between the two treatments.

The results as shown in Table 3.9 documenting that both dry stored stratified and fresh seed produced similar germination rates when buried at 2 cm depth provide no indication of preferential germination from stasis over fresh seed in spring or autumn when seeds have been buried. Analyses of both fresh and dry stored stratified seed at surface scattering indicate some differences exist in germination, suggesting that stasis induced seed had an advantage over fresh seed in spring, with fresh seed having a germination advantage in autumn, when on the soil surface. These results can be considered somewhat parallel to Burrows (1999) results where fresh dispersed seed readily germinated in autumn-winter, and buried seed (stasis induced) in summer. Results of the *S. aviculare* control petri

dish trials were only 34% compared to 92% Burrows (1999) results (refer Table 3.9). The complete lack of germination in the second control petri dish trial has no explanation, although it is noted that results may relate to seeds being collected at different times in the fruiting period, different years or provenances, that Burrows (1999) noted may lead to exhibition of different germination behaviour.

# PART D: LEAF MORPHOLOGY, INSECT DAMAGE AND FLOWERING

## 3.13 INTRODUCTION

Baylis (1963) compared leaf morphology in six *Archaesolanum* species, and documented that a wide range of leaf forms can exist on a single plant, but between individual plants differences can be hard to define; *Solanum aviculare* and *S. laciniatum* were vegetatively indistinguishable, always bearing pinnately lobed leaves on juvenile plants, and entire leaves on adult plants, but with irregularly lobed semi-adult leaves re-occurring. Korneva et al. (1972) noted and named five variants of *S. aviculare* and *S. laciniatum* and Gerasimenko (1971) also documented and named five forms of *S. laciniatum*. Gerasimenko's observations noted the specific individual morphological features were practically indistinguishable in the forms, as noted by Baylis (1968), but were apparently heritable and considered to be genotypically influenced by geographical and ecological factors, rather than from directly their complex (sub-genus *Archaesolanum*). None of these variants were formally recognised botanically or common garden experiments undertaken to verify heritable characteristics.

James & Mantell (1994) confirming developmental phases of *S. aviculare* documented leaf heteroblastic development from seedling to sexual maturity. Progressive changes occurred in morphology, anatomy, phyllotaxis and other characteristics, that were correlated during development with their position on the plant and with each other, with axillary buds inheriting development potentials from their placement in relation to early and late shoot development. They also documented higher degrees of leaf lobing and lower length/breadth

ratio in the juvenile phase. Kerstetter & Poethig (1998) reviewed leaf modification during plant leaf cycles and noted short lived plants often continuously produce varied leaf shapes along shoots, and heteroblastic leaf development appears regulated by both genes and differential responses to physiological environmental factors affecting the whole plant, not just a leaf response to environmental factors such as shading or nutrient variability, which is known as heterophylly. They also noted that production of adult or juvenile leaves has been identified as being affected by mutations involving the regulation of adult and juvenile phases. Leaf production is independently regulated from leaf identity; leaf shape, particularly lobing, and leaf position on a shoot may be dependent on the time the leaf was initiated in relation to the time of the shoot developing, and adult shoots can be induced to produce juvenile leaf forms through a variety of conditions more readily than juvenile shoots are able to produce adult leaves (refer Kerstetter & Poethig 1998). Martin (1999) documented arthropods and molluscs associated with, and plant damage on, *S. aviculare* and *S. laciniatum* when in commercial production for solasodine extraction.

This research aimed, by documenting and comparing leaf development and flowering from radicle emergence in seedlings grown from the germination trials, to answer the questions: 1) what leaf morphological differences exist in and between *S. aviculare* and *S. laciniatum*, 2) how is heteroblastic leaf development determined and do any differences exist in and between species, 3) how can they be defined, 4) what ecological or other factors may appear to influence leaf morphology and development, 5) are there any leaf differences relating to adult and juvenile phase leaf production and how may they be influenced, 6) do any temporal flowering differences exist between species 7) what insect associations are apparent, and is any insect damage apparent similarly to that documented by Martin (1999)?

## 3.14 Methodology

3.14.1 Leaf morphology and heteroblastic development

Leaf morphology and growth was recorded from 50 dry stored *S. laciniatum* seedlings, and 44 fresh *S. aviculare* seedlings grown on from the 1st seed

germination trial, plus six fresh *S. aviculare* seeds grown from the 1st petri dish control, after being re-potted into PB 2 containers and kept at the author's home. Seeds used for documenting radicle emergence were *S. aviculare* seed germinated in the 1st petri dish control (Plate 3.11). Seedlings were first monitored for overall similarity and individual differences in initial leaf shape to the 4th leaf. Leaves were counted only to the 4th leaf as all subsequent leaves were deeply lobed. Leaves described as lobed included any protuberance (Figure 3.7, refer Figures 3.8 & 3.9).

Photos Moana Kapoor

**Plate 3.11: Radicle germination - note hairs on hypocotyl.**

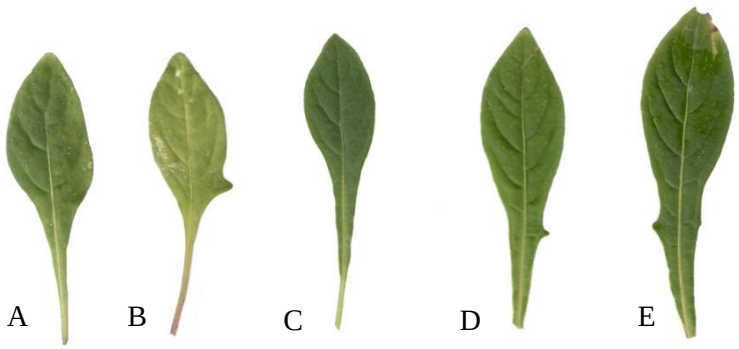


**Figure 3.7: Linear and lobed leaf shapes as determined for documentation of initial leaf to 4th leaf study. Left–right: A) *S. laciniatum* linear, B) lobed, C) *S. aviculare* linear, D) lobed, E) lobed. Not to scale.** (Scans G. Weavers)

Full observations of heteroblastic leaf growth from cotyledon to mature leaves continued with the 100 seedlings (refer Figures 3.10 & 3.11). Leaves were also monitored for variations in juvenile and adult growth from the same 100 plants as well as an *in situ S. aviculare* seedling at TAR 2. Observations were continually undertaken at the three Tarawera sites to monitor morphology in

relation to environmental conditions. Epidermal and leaf length (laminar to petiole base) differences on adult leaves between species were documented from 4th leaf to 8th leaf, as the leaf formation had stabilised by the 4th leaf, and was uniform to the 8th leaf to enable comparison. Data was entered into Microsoft Excel 2007 spreadsheets and histograms produced.

### 3.14.2 Insect damage

Six *S. aviculare* and six *S. laciniatum* plants of the 100 were grown on side by side for six months, and then two were planted out 12m apart, at the author's home. They were continuously monitored for insect attack and damage to the termination of the seedbank germination trials on 14th June 2009.

### 3.14.3 Flowering

The 50 *S. laciniatum* and 50 *S. aviculare* seedlings used for the leaf morphology trials were monitored for timing of flower set and flower opening until the termination of the seedbank germination trials on 14th June 2009.

## 3.15 Results

### 3.15.1 Leaf morphology

Senescence of plants at the Tarawera sites prevented complete monitoring of leaf morphology in conjunction with environmental conditions. Epidermal differences were noted between species; with *S. laciniatum* generally having a smoother epidermal texture with more regular reticulate venation than *S. aviculare* from both dorsal (lower) and ventral (upper) surfaces (Plate 3.12). *Solanum aviculare* generally had a longer laminar than *S. laciniatum* (Table 3.10), and leaf colouration was generally darker green in *S. aviculare* than *S. laciniatum* (Plate 3.12). Results also documented that individual monitored *ex situ* container grown seedlings and *in situ* monitored seedlings grew axillary adult leaves before flowering commenced and juvenile leaves post inflorescence (refer Plates 3.17, 3.18, 3.19, 3.20 & 3.21).

**Table 3.10: Laminar measurements of juvenile leaves at 5th- 8th node positions in *ex situ* seedlings of *S. aviculare* and *S. laciniatum*.**

| | *S. aviculare* | | *S. laciniatum* | |
|---|---|---|---|---|
| **Leaf** | **Length** | **Width** | **Length** | **Width** |
| **5th** | 280 mm | 24 mm | 285 mm | 44 mm |
| **6th** | 363 mm | 30 mm | 203 mm | 34 mm |
| **7th** | 361 mm | 30 mm | 194 mm | 23 mm |
| **8th** | 285 mm | 31 mm | 280 mm | 36 mm |

In regard pre-flowering axillary adult leaves forming, a count to the 4th leaf of the 50 *S. aviculare* and 50 *S. laciniatum* seedlings was undertaken, with 75/100 (75%) plants forming linear leaves of the adult shape in leaf axils at the 1st – 3rd nodes >65% of the time at 3 months of age (before sexual maturation; i.e. before flower formation and reproduction), reducing to 44% of the time at the 4th node (refer Baylis 1963; James & Mantell 1994) (Table 3.11, Plates 3.13 & 3.14). Differing numbers of lobed and linear leaves on adult plants were observed in all Tarawera sites, particularly TAR 3. All other sites observed had more predominantly linear leaves, with only small amounts of lobed leaves present on adult plants.

**Table 3.11: Numbers, percentage and location of axillary leaves that formed prior to flower formation, out of the 100 monitored *S. aviculare* and *S. laciniatum ex situ* seedlings.**

| Leaf | Quantity | Percentage |
|---|---|---|
| 1st | 67 | 67% |
| 2nd | 66 | 66% |
| 3rd | 66 | 66% |
| 4th | 44 | 44% |

The hypocotyl, and ovoid/linear initial true leaves produced epidermal hairs (trichomes) (refer Figures 2.3 & 2.5), subsequent lobed leaves did not. The results show 100% of initial first true leaves of *S. aviculare* and 92% of initial first true leaves of *S. laciniatum* were ovoid/linear. The numbers of ovoid/linear leaves dropping dramatically at second initial true leaf with the majority being more lobed/pinnatifid. All subsequent true leaves were lobed (Table 3.12, Figures 3.8, 3.9, 3.10 & 3.11).

Photos G. Weavers

**Plate 3.12: Epidermal venation and colour differences between *S. aviculare* (left top and bottom) and *S. laciniatum* (right top and bottom).**

Photos G. Weavers

**Plates 3.13 & 3.14: Adult linear leaves formed in leaf axils of *S. aviculare* (left) ca. 11 month old *in situ* seedling & (right) ca. 3 month old *ex situ* seedling, prior to flowering nodes and inflorescence development.**

**Table 3.12: Primary initial leaf shape from 1st to 4th true leaf, showing numbers and percentage of leaves linear and lobed (n=50 *S. aviculare*, n= 50 *S. laciniatum*).**

| | LINEAR | | | | LOBED | | | |
|---|---|---|---|---|---|---|---|---|
| | *S. laciniatum* | | *S. aviculare* | | *S. laciniatum* | | *S. aviculare* | |
| | number | % | number | % | number | % | number | % |
| 1st | 46 | 92 | 50 | 100 | 4 | 8 | 0 | 0 |
| 2nd | 2 | 4 | 10 | 20 | 48 | 96 | 40 | 80 |
| 3rd | 0 | 0 | 0 | 0 | 50 | 100 | 50 | 100 |
| 4th | 0 | 0 | 0 | 0 | 50 | 100 | 50 | 100 |

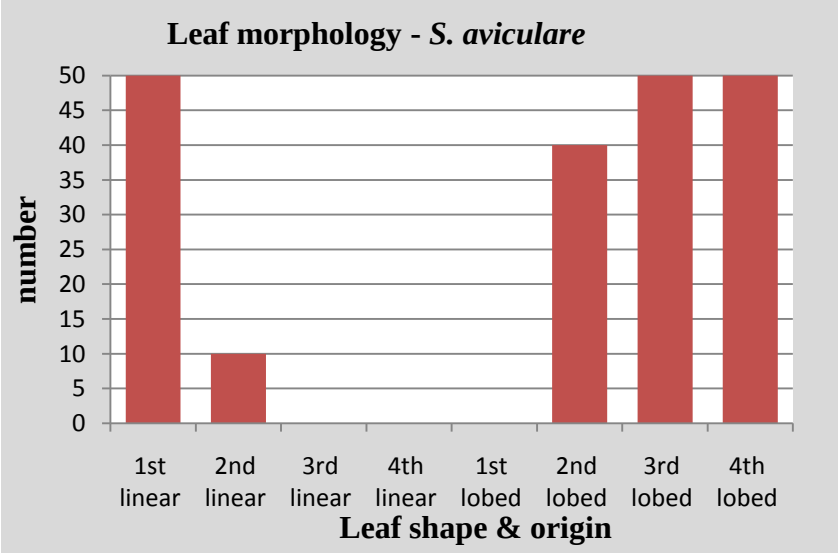


**Figure 3.8: *S. aviculare* leaf shape and origin morphology results from initial to 4th leaf; total number of leaves.**

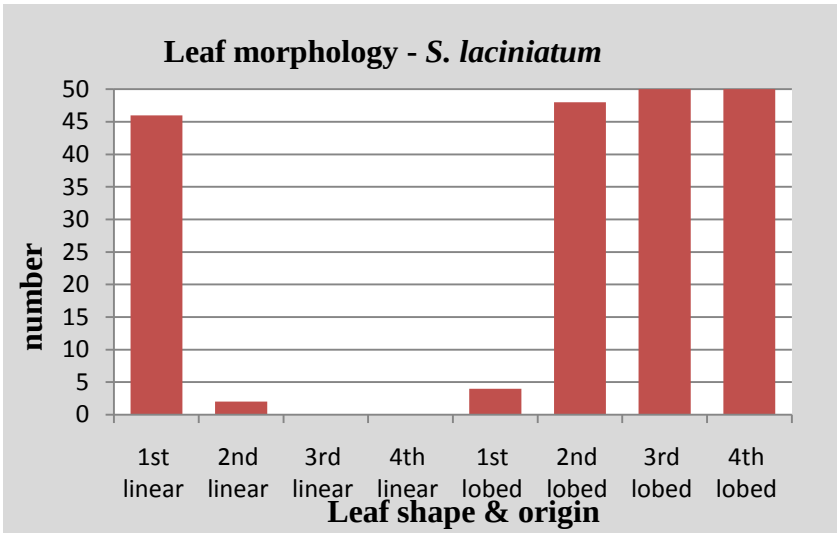


**Figure 3.9: *S. laciniatum* leaf shape and origin morphology results from initial to the 4th leaf; total number of leaves.**

3.15.2 Insect damage

*Solanum aviculare* plants, especially when flowering, were observed with numerous insect species on the leaves and flowers, although no exact monitoring was able to be undertaken within the scope and timing of the research. In the monitoring of the six *S. aviculare* and *S. laciniatum* plants five out of the six *S. aviculare* seedlings were attacked continuously by a stem boring caterpillar, whereas the six *S. laciniatum* seedlings were not attacked. The caterpillars were removed and the *S. aviculare* continued to be re-infested four times over a two month period, with *S. laciniatum* still unaffected. The caterpillars were tentatively identified as Codling Moth larvae (Lepidoptera *Cydia pomonella*) and were also found in adjacent apple fruits (Plates 3.15 & 3.16).

Photos G. Weavers

**Plate 3.15 & 3.16: Caterpillar and damage location on *S. aviculare ex situ* seedling.**

3.15.3 Flowering timing

Of the two species 60% of *S. laciniatum* seedlings flowered (n=30/50), after 16 weeks (112 days). Only one *S. aviculare* seedling had produced a single flower by the end of the third germination trial (22nd July 2009).

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**Figure 3.10**: ***S. aviculare*** **heteroblastic leaf development, representative leaves**: **Cotyledon, Juvenile leaves: 1st leaf unlobed, 1st leaf lobed, 2nd leaf, 3rd leaf, 4th leaf, 5th leaf, 6th leaf, 7th & subsequent leaves, adult pinnatifid leaf, adult pinnatifid leaf, adult linear leaf. Scale bar = 2.5cm** (Scans G. Weavers)


**Figure 3.11: *S. laciniatum* heteroblastic leaf development, representative 1eaves: Juvenile leaves; 1st unlobed leaf, 1st lobed leaf, 2nd leaf, 3rd leaf, 3rd leaf, 4th leaf, 5th leaf, 6th & subsequent leaves, adult linear leaf, adult linear leaf, adult pinnatifid leaf.**
**Scale bar = 2.5cm** (Scans G. Weavers)

## 3.16 Discussion

3.16.1 Leaf morphology

The results highlight some leaf shape and laminar epidermal differences, particularly in venation, occurring between the species, but as noted by Baylis (1963) are not easy to define. Heteroblastic leaf development is confirmed, the results documenting, in addition to Baylis (1963) that initial leaves of *S. aviculare* at 1st node, immediately following cotyledons, form as ovoid/linear with peripheral hairs always, and *S. laciniatum* predominantly. Lobing begins to form at the first node in *S. laciniatum* and in the majority of 2nd node leaves in both species, followed by fully lobed pinnatafid leaves forming from the 3rd & 4th nodes onward. Only ovoid/linear juvenile leaves produced trichomes, lobed leaves juvenile were glabrous. The hypocotyl also produced trichomes and the cotyledons were glabrous. Mature leaves follow Baylis (1963) in being glabrous lanceolate/linear, with glabrous pinnatafid leaves irregularly appearing. All the initial leaves exhibiting the ovoid/linear shape, and all initial leaves exhibiting the lobed shape were growing non-axillary prior to any terminal inflorescence development, therefore were considered juvenile (Baylis 1963; James & Mantell 1994).

Consideration is given to qualifying what is a juvenile and adult leaf in this study due to the documentation of both *S. aviculare* and *S. laciniatum* forming linear shaped adult axillary leaves at 3 months of age, before sexual maturation (refer Plate 3.12). It appears appropriate to consider all leaf growth not from leaf axils (non axillary) to be primary (currently called juvenile), and all leaf growth from leaf axils (axillary) to be secondary (currently called adult).This then removes doubt as to leaf growth in regard to morphology and the growth stage being juvenile or adult; with all leaves not grown from axils being primary and all leaves grown from axils being secondary. This can be further clarified by leaf growth to the second node to be initial primary leaves, and any axillary linear leaves appearing before terminal inflorescence nodes to be secondary juvenile. Secondary juvenile leaves were documented as growing continuously on a monitored *ex situ S. aviculare* non-flowering seedling (Plate 3.17) and an *in situ* flowering seedling prior to the inflorescence (Plates 3.18 & 3.19).

Non axillary leaves, with decreased lobing were also noted as growing from the axillary shoot growing above terminal flowering nodes in the monitored *in situ* seedling (Plates 3.20 & 3.21), therefore where this occurs these leaves growing non axillary from post flowering nodes would be known as primary adult. These primary adult leaves appear to correspond to the leaf shapes exhibiting deceasing degrees of leaf lobing growing on anthocladial shoots subtending the flowering terminal node, as documented by James & Mantell (1994). James & Mantell (1994) also documented that these leaf shapes on anthocladial shoots are relatively stable at the adult (secondary) and in the juvenile (primary) phase, though photoperiod had a greater influence on leaf shape in the juvenile (primary) phase. They also documented photoperiod influence on abscission of flowers, though the results of the flowering monitoring do not elaborate on this phenomenon.

Photo G. Weavers

**Plate 3.17: Non-flowering *S. aviculare* seedling showing continuous secondary juvenile leaf growth.**

Photos G. Weavers

**Plate 3.18 & Plate 3.19: Close-up of Plate 3.18: Secondary juvenile leaves forming axillary prior to inflorescence development on *in situ S. aviculare* seedling.**

Photos G. Weavers

**Plates 3.20 & 3.21: Photograph of the same plant in Plates 3.18 & 3.19, *in situ S. aviculare* seedling showing primary adult leaves (non-axillary juvenile straight arrow) with axillary secondary adult leaves forming post inflorescence (dotted arrow)**

Differing large numbers of pinnate adult leaves in relation to linear leaves, in comparison to plants at all other sites, were observed on immediately adjacent plants of similar height and crown spread growing in the same substrate conditions at all Tarawera sites, especially TAR 3. The appearance of these pinnate adult leaves may be associated with a combination of individual genotypic, and environmental phenotypic produced plant differences, displaying differing forms as documented by Gerasimenko (1971), rather than totally phenotypic causes. The differences observed may also be related to the fast initial vegetative growth where the shoots were initiated in the juvenile phase, similarly as documented by Kerstetter & Poethig (1998), thus producing more juvenile shaped leaves at the adult post inflorescence phase. No statistical analysis was attempted due to caution being applied due to the low data sets, and senescence of the plants making continued monitoring impossible after the initial observance. The other sites observed contained differing numbers of plants exhibiting varying amounts of leaf variation, but generally the majority of plants exhibited predominately linear leaves. The overlying interpretation is one of unpredictability to adult leaf shape in general observance

### 3.16.2 Flowering timing and insect damage

The results of the flower timing monitoring indicate that induction of stasis may play a role in the documented early flowering of *S. laciniatum* within 12 weeks. Both species are very closely related (Baylis 1954; Bohs & Olmstead 1997; Pozcai pers. comm. 2009), and no previous research has indicated that there are any species differences in flowering behaviour. Therefore it can be feasibly considered that the flowering of *S. laciniatum* at 12 weeks from stasis replicated seed (n = 30/50), and the non-flowering of *S. aviculare* from fresh collected seed (n = 1/50) may be related to stasis induction over fresh seed. Previous germination experiments document that both species readily germinate from fresh seed (Burrows 1996, 1999), but that research did not follow the seedlings to flowering. This research suggests that stasis induction as a species tactic may relate more to early flowering and reproduction rather than colonisation via germination; and in documenting the differing flowering behaviour between stratified and unstratified seed this research indicates that a specialised tactic of both species may be a predilection toward stasis induction following dispersal to

soil as a strategy for early, rapid fruit set after a disturbance provides germination cues to seeds.

The stem damage caused by Codling Moth larvae (refer Plates 3.15 & 3.16) was similar to that caused by the Solanaceous pest eggfruit caterpillar (Lepidoptera *Sceliodes cordalis*) in the commercial plantations noted by Martin (1999). The results of the monitoring provide new evidence suggesting *S. aviculare* is highly susceptible to stem boring insects, while *S. laciniatum* is not; the insect actively re-infecting *S. aviculare*, while not infecting *S. laciniatum* when side by side in pots and when planted out within 12 m. The quantities of alkaloids in the stems and leaves of *S. aviculare* have been documented as being higher than in *S. laciniatum* (Symon 1994), and no explanation is offered as to why *S. aviculare* was infected while *S. laciniatum* was not.

## 3.17 Conclusions and recommendations

Some new data on the seral dynamics of *S. aviculare*, seed viability, including a viable seed bank, and dispersal agents has been provided. Previous seed germination experiments have been confirmed. Some new data has been provided on heteroblastic leaf development and leaf morphology, flowering timing, initial leaf growth stages and epidermal differences between *S. aviculare* and *S. laciniatum*. A summary of the aims achieved and consideration of further research areas that can be proposed from these results follows.

The seral role of *S. aviculare* has been documented fully for the first time, describing a complete cycle of regeneration tactics and strategies of an early opportunist pioneer coloniser of New Zealand lowland forest which is dependent upon disturbance for survival. The research has provided new data on growth dynamics, documenting height and crown growth metadata correlations, showing cohort growth development which may relate to site resource availability and environmental exposure, indicating the flexibility of reproduction tactics to changes in ephemeral gap openings, documenting and discussing the successional position of *S. aviculare*. Further research is suggested in following growth and senescence between plants in tree fall gaps (less precarious) and earth slip sites (more precarious), to provide more complete information on the

flexibility of reproductive tactics in relation to precarious positions and reduced resources as discussed in Platt & Connell (2003). The non growth of *S. aviculare* seedlings beneath *S. aviculare* canopies despite fruit maturation and direct under canopy fruit fall suggests the possibility of either suppression of its own seedlings or a possible predilection toward primary dormancy. Further specific research is suggested as to whether this is truly suppression, a tactic indicative of a predilection toward primary dormancy, or truly secondary dormancy due to stasis induction cues.

The spread of viable *S. aviculare* and *C. arborea* seed by kererū/blackbird, and formation of a seed bank from surrounding heterogeneous forest was confirmed. The predominant dispersal agents are confirmed as birds and despite the mixing of specimen's, indicates kererū as a predominant disperser, and predominant colonisation strategies to be tactics of dispersal via bird endozoochory rather than a necessary gut passage requirement for germination. Differences in seed viability documented from the controls, and of recovered seed between bird species, in particular between indigenous and adventives, is suggested for further conservation research. Endozoochory effects including gut maceration are not fully explored and are considered to be unpredictable (refer Traveset 1998), and further research of this nature could provide valuable conservation information, not only for the enhancement of *S. aviculare*, but of many other bird dispersed species. Research into whether glycoalkaloids influence seed retention times is also suggested for consideration. The conservation consideration of high viability of rat recovered seed may imply changes need to be made to poisoning programs in regard timing and bird numbers within the sites to be controlled, and will be further addressed in chapter 6.

While the germination results were lower than previous research results (Burrows 1996, 1999), and did not provide any new data on germination behaviour in relation to stasis and fresh seed, they did confirm earlier research on temporal differences. The documentation of temporal flowering differences relating to apparent stasis induction between *S. aviculare* and *S. laciniatum* has not been documented before and further research is suggested to confirm this relationship and to what extent species differences exist. The repeated stem damage to *S. aviculare* and not to *S. laciniatum* has not been reported before, and

is considered relevant for further research in particular in regard to any exploitation for glycoalkaloid extraction.

The documentation of plant growth from radicle emergence to full leaf development has confirmed data on initial leaf shape with previous studies (Baylis 1963; James & Mantell 1994; Symon 1994), and provided new data highlighting some differences between *S. aviculare* and *S. laciniatum*. The data showed, in addition to previous studies, that the initial leaves formed on the first node directly following the cotyledons predominantly form in an ovoid/linear shape with trichomes. The lobed shaped leaves appear from the second node onward in both species, and are glabrous. Previous studies defined the initial ovoid/linear leaves as first foliage leaves, and subsequently more lobed leaves as juvenile phase (Symon 1994). This research used the nomenclature of initial leaves rather than first foliage to the 2$^{nd}$ node, and juvenile for all subsequent non-axillary leaves. A new nomenclature of primary and secondary has been suggested to clarify growth stages due to the documentation of differing adult and juvenile leaf growth occurring at flowering and non-flowering stages. The documentation of epidermal differences between the two closely related species has not been undertaken before, and warrants a more complete botanical study for confirmation of stability in differences. The documentation of differing numbers of pinnate adult (secondary) leaves on adjacent plants, and any relationship to environmental conditions or a genetic basis could not be substantiated further within the scope of this study due to senescence of the plants, and inability to monitor spatially distant groups of plants. Further research on the stability of this phenomenon is suggested.

In chapter four issues of genetics are studied and chapter five addresses issues surrounding the conservation status and cultural aspects in regard restoration and enhancement of *S. aviculare*.

Photo Moana Kapoor

***Solanum aviculare* in shadow with Lake Tarawera and Mount Tarawera background: TAR 3**

# CHAPTER 4

# CONSERVATION GENETICS OF *Solanum aviculare*

## 4.1 INTRODUCTION

*Solanum aviculare* is a pioneer secondary successional species appearing in New Zealand coastal habitats and lowland forest after disturbance events, and is considered in 'decline', 'sparse' and 'data poor' in the Threatened and Uncommon Plants of New Zealand Reappraisal list (de Lange et al. 2008). A total of 53% of plant species are listed as under threat, 30% are in lowland and 23% in coastal habitats; with 29.25% (n=792/2705) of all threatened and uncommon plants being found in the North Island (de Lange et al. 2009). The loss of species can be related to the loss of natural ecosystems and habitats through rural and urban development, particularly in lowland areas. Genetic diversity can play an important role in relationships between species, and diversity within a population can affect the productivity or fitness and long term survival and evolution of a population (Gemmill et al. 1998), having the potential to affect a wide range of population, community interactions, and ecosystem processes (Hughes et al. 2008). Little information is currently available about the range of potential ecological effects of genetic diversity, which can only be measured where genetic diversity data is available (McKay et al. 2005; Hughes et al. 2008). Therefore further additional data providing an understanding of how genetic variation is partitioned within and between populations, and including describing genetic distinctiveness between species, can support species conservation and restoration strategies and be important in helping maintain biodiversity and ecological linkages within wider ecosystems.

Restoration aims to re-establish as rapidly as possible a long-term viable population consisting of suitable species that can enhance ecosystem biodiversity, restore functions and processes, and protect vulnerable landscapes from damaging environmental effects (Lesica & Allendorf 1999). Conservation of a declining population involves dealing with living organisms that exhibit both genotypic and phenotypic variation, and it is considered that in successful restoration projects the fundamental forces of genetic variation (i.e., the regulatory and functional traits exhibited by gene expression) should be included

in the project design, as a diverse number of genotypes can be less vulnerable to allogenic events, and the long-term success of a project may depend on whether the genetic source is appropriate or not (Gemmill et al. 1998; Lesica & Allendorf 1999; Falk et al. 2001). Phenotypes (ecotypes) with wide ecological amplitudes (when identified) should be considered in project planning, as environmental factors may influence plant survival, and consideration of both environmental and genetic factors may contribute to overall success, particularly in short lived plants (Lesica & Allendorf 1999; Sackville-Hamilton 2001; Wilkinson 2001; McKay et al. 2005). A small or endangered population's ability to persist can be altered by changes in its genetic diversity and fitness due to genetic bottlenecks or inbreeding (Hughes et al. 2008), and the use of molecular markers to estimate variation within and between populations can measure historical levels of connectivity, allowing conservation and restoration planning to consider maintenance and restoration of habitat connectivity, reducing potential inbreeding depression (McKay et al. 2005).

### 4.1.1 Phylogenetic studies-*Solanum*

Worldwide, *Solanum* L. (Solanaceae) is a large and economically important genus containing many diverse species including important food crops and medicinal sources. DNA based studies have investigated the phylogenetic relationships between *Archaesolanum* and other *Solanum* sub-genera (Bohs & Olmstead 1997; Olmstead & Palmer 1997; Bohs & Olmstead 2001; Bohs 2005; Weese & Bohs 2007; Pozcai et al. 2008), but little work has concentrated on variation within singular species (intraspecies) and between species (interspecies).

The first cytogenetical studies undertaken comparing species within the *Archaesolanum S. aviculare* complex were by Prof. G. T. S. Baylis in Dunedin, New Zealand. Baylis (1963) produced results highlighting species differences and indicating, genetically, that from New Zealand through Australia to New Guinea *S. aviculare* in its "typical form" was "substantially uniform" and *S. laciniatum* was "identical" between New Zealand and Australia. Baylis (1968) documented that *S. aviculare* consisted of ecotypes where photoperiod was adapted to temperature and latitude, although *S. laciniatum* showed no

photoperiod latitudinal change. Recent phylogenetic work by Pozcai et al. (pers. comm. 2009) using random amplified polymorphic DNA (RAPD) and restricted fragment length polymorphism (RFLP) analyses indicates that *S. aviculare* is a highly monomorphic species and *S. laciniatum* is very closely related to *S. aviculare*, and may be a hybrid between *S. aviculare* and *S. vescum* as female parent, but is more closely related to *S. aviculare*. Monomorphic invariant genes have been demonstrated to be involved in species and genera differentiation rather than individual or geographic variation (Altukhov & Abramova 2001) and polymorphic genes associated more with individual genetic inheritance from DNA recombination associated with inheritance theory, than extranuclearly (Altukhov & Abramova 200; Allendorf & Luikart 2007).

### 4.1.2 Genetic diversity- techniques, previous studies and thesis aims

Genetic diversity in natural populations can be identified by the use of multi-locus markers, utilising a number of different techniques such as RFLP, RAPD, microsatellites, amplified fragment length polymorphism (AFLP), simple sequence repeats (SSR), and inter simple sequence repeat (ISSR). (McGregor et al. 2000; Camacho & Liston 2001; Wu et al. 2004; Xie et al. 2005; Archibald et al. 2006; Allendorf & Luikart 2007; Hughes et al. 2008; Venkatachalam et al. 2008). Microsatellites are short tandem repeats of 1-4 base pairs of DNA, dispersed throughout the genome, and usually anchored at the 3' or 5' end. ISSR's use microsatellite sequences as primers, in a Polymerase Chain Reaction (PCR) to generate the multilocus DNA markers used to estimate genetic diversity. ISSR's involve amplifying a DNA segment found between two identical repeat microsatellite/primer regions (loci) and are generally considered to be highly polymorphic due to high rates of mutation or evolutionary change taking place within microsatellites (Reddy et al. 2002; Allendorf & Luikart 2007). Previous studies in *Solanum* species have considered ISSR's to provide higher reproducibility, be less time consuming and have a lower cost than the other techniques (Prevost & Wilkinson 1999; Camacho & Liston 2001; Tikunov et al. 2003; Isshiki et al. 2008; Terzopoulos & Bebeli 2008). Specifically Tikunov et al. (2003) and Terzopoulos & Bebel (2008) in *Lycopersicon* species found ISSR a useful and powerful tool for documenting genetic diversity, additionally Prevost & Wilkinson (1999) found ISSR's generated more

polymorphism in potato cultivars than RAPD or RFLP, and Isshiki et al. (2008) in *Solanum melongena* L and related *Solanum* species found ISSR to be a powerful tool providing high discrimination.

This study utilises ISSR techniques to attempt to answer the questions: 1) what degrees of diversity are there within and between populations and groups of *S. aviculare*, 2) what DNA extraction protocols are the most effective for the species, 3) is any invariance identifiable in *S. aviculare* and *S. laciniatum* that relates to previous studies "uniformity", 4) what protocols are appropriate in conservation and restoration programs for *S. aviculare* and *S. laciniatum*. Therefore the first aim of this study was to estimate and determine genetic variation between and within eight populations of *S. aviculare* by extracting DNA from *in situ* (naturally occurring within the Bay of Plenty and Waikato, New Zealand) plants and using the DNA with *in vitro* ISSR techniques. The second aim was to determine and consider whether invariance exists within *S. aviculare* populations and between *S. aviculare* and *S. laciniatum.* These data will inform the third aim of considering conservation and restoration planning for *S. aviculare* and *S. laciniatum*.

## 4.2 Materials and Methods

### 4.2.1 Field Collection

Leaves were collected under the same permits and permission from the same sites used for growth data collection (refer chapter 3) (Table 4.1). Fresh leaf samples were collected from a minimum of 20 randomly selected healthy vigorous plants at each site. Two leaves were collected from each plant, for a total of 40 leaves per site. The second leaf was used as backup and stored separately. Where < 20 plants existed, samples were collected from all plants. Leaves of lesser quality (i.e., senescing or wind torn) were only collected where plant numbers were limited i.e. at Tarawera sites TAR 1, TAR 2 and TAR 3. There was one adult *S. laciniatum* plant and six seedlings on one property from which 14 leaves were collected. Leaves were immediately placed in individual sealable plastic bags labelled with a code identifying the site and plant number, and placed in an INNERCOOL™ carry bag (pre-frozen in a refrigerator containing freezer packs and bagged ice to keep the samples cool). All samples

were stored at -80$^{o}$C until the DNA extractions were performed. Voucher specimens, one per population, except Pirongia, due to access difficulties, were lodged at the University of Waikato Herbarium (WAIK).

**Table 4.1: Location, site code and quantity of leaves collected for DNA analysis and voucher specimens (WAIK).**

| Location | Site code | Total leaf number | WAIK |
|---|---|---|---|
| Tarawera | TAR 1-3 | 120 | √ |
| Rotoma | ROT 1-4 | 36 | √ |
| Okataina | OKA 1-4 | 12 | √ |
| Tauranga | TAU 1-15 | 30 | √ |
| Te Urewera | URE 1-3 | 52 | √ |
| Maungatautari North | MAU N 1 -3 | 30 | √ |
| Maungatautari South | MAU S 1-4 | 30 | √ |
| Pirongia Corcoran Rd | PIR 1-2 | 4 | X |
| Pirongia Kaniwhaniwha | PIR 3-5 | 6 | X |
| Stanley Road | LAC 1-7 | 14 | √ |

4.2.2 Laboratory-DNA extractions

DNA was extracted using a modified CTAB method (Doyle & Doyle 1987) (Appendix 5), and Invitrogen Purelink DNA extraction kits. The DNA extracts were visualised for quantity and quality of DNA on 100% agarose gels

4.2.3 Laboratory PCR-ISSR Production

Twenty ISSR primers were used (Table 4.2), with each being repeated a minimum of two times to test for reproducibility and to select primers that were polymorphic. The primers were selected from stock universal primers known to amplify across a wide range of taxa (Reddy et al. 2002; Allendorf & Luikart 2007). Optimisations of PCR utilised nine representative DNA samples, seven *S. aviculare*, including one sample with high quantities of RNA to test for the effect of RNA on the PCR process, one *S. laciniatum* sample to test variation between species, and a positive and negative control (Table 4.3). Upon any indication of polymorphism a further PCR was run using ten representative *S. aviculare* samples from each site, also including a sample with high RNA, and an *S. laciniatum* sample.

**Table 4.2: Sequences of primers used to document diversity between and within *S. aviculare* populations and *S. laciniatum*.**

**ISSR Primer Sequences**

| ISSR Primer | Sequence |
|---|---|
| 1 | 3’ CACACACACACAGG 5’ |
| 2 | 3’ CTCTCTCTCTCTCTCTAC 5’ |
| 3 | 3’ CTCTCTCTCTCTCTTG 5’ |
| 4 | 3’ CACACACACACAAC 5’ |
| 5 | 3’ CTCTCTCTCTCTCTCTGC 5’ |
| 6 | 3’ CACACACACACAAG 5’ |
| 7 | 3’ CACACACACACACT 5’ |
| 8 | 3’ GAGAGAGAGAGAGG 5’ |
| 9 | 3’ GTGTGTGTGTGTGG 5’ |
| 10 | 3’ GAGAGAGAGAGACC 5’ |
| 11 | 3’ GTGTGTGTGTGTCC 5’ |
| 12 | 3’ CACCACCACGCGCGC 5’ |
| 13 | 3’ GAGGAGGAGGC 5’ |
| 14 | 3’ CTCCTCCTCGCGCGC 5’ |
| 15 | 3’ GTGGTGGTGGCGCGC 5’ |
| A | 3’ CACACACACACACATC 5’ |
| B | 3’ CACCACCACCACGC 5’ |
| C | 3’ CTCTCTCTCTCTCTAC 5’ |
| E | 3’ CACACACACACAGC 5’ |
| F | 3’ GAGGAGGAGGAGGC 5' |

**Table: 4.3 Representative DNA samples used in PCR reactions.**

| # | DNA samples location & code |
|---|---|
| 1 | URE 3 & 6 |
| 2 | TAR 2/11 & 3/2 |
| 3 | ROT 2/7 |
| 4 | TAU 4, 5 & 6 |
| 5 | MAU N 4, N5 & S4 |
| 6 | PIR K1 & K2 |
| 7 | TAR 1/18 (RNA) |
| 8 | LAC 3, 4, 6 & 7 |
| 9 | *Pitt. corn.*, TAR 1/12,1/18, ROT 2/9 (Positive) |

PCR’s were set up in a fume hood to minimise contamination. Reactions contained 24 µl of reaction mix; final concentrations consisting of 16.92 µl Mq $H_2O$, 2.5 µl 10 x PCR Buffer, 4.0 µl Mg $Cl_2$, 0.08 µl dNTP’s, 0.2 µl BSA 1%, 0.2 µl Primer, 0.1 µl Taq, with 1 µl DNA added to make a total volume of 25 µl. Initial concentration of Mg $Cl_2$ was at 50 mM, the dNTP’s were at10 mM and the Primers were at 10 mM. The DNA was used @ 1 µl, 1:10 and 1:100 dilutions to provide comparative results during optimization. DNA @ 1:10 µl was used for

the majority of final PCR amplifications. A positive DNA sample at 1 µl and a control were included in all PCR's. A sample of *Pittosporum cornifolium* was used initially as a positive, but results were inconsistent and *S. aviculare* samples TAR 1/12, TAR 1/18 and ROT 2/9 were then used. PCR reactions were carried out in an Eppendorf 96-well Mastercycler thermocycler.

Gradient trials were undertaken to find the most suitable temperature range for annealing, a number of different reaction temperatures were trialled until a final temperature range was found that provided the most consistent results. Final cycling temperatures for all primers consisted of 94$^{o}$ C for 4 minutes, followed by 35 cycles of 94$^{o}$ C for 50 seconds, 50$^{o}$ C for 55 seconds, and 72$^{o}$ C for 1:30 minutes. A final amplification of 72$^{o}$ C for 5 minutes ended the cycles. The PCR products were separated on a 1.8 % agarose gel containing ethidium bromide; 10 µl of PCR product was mixed with 3 µl Loading Buffer (.25% Bromophenol Blue, .25% Xylenecynol FF, 15% Ficoll 400) and loaded into wells, with 3 µl Trackit™ 100 bp DNA Ladder included on all gels. Each ISSR fragment was to be scored according to the presence or absence of bright, reproducible and reliable bands, manually from a gel photograph using the size standards of the ladder to ensure even scoring of bands (Figure 4.1).

### 4.2.4 Sequencing

To ensure the species used were true to type, sequencing targeting the internal transcribed spacer (ITS) regions was undertaken using three representative *S. aviculare* (TAU 5, MAU 4 and TAR 3/2), and three representative *S. laciniatum* samples (LAC 4, LAC 6, LAC 7). The *S. laciniatum* sample LAC 7 consisted of leaves from the adult parent plant that seeds used in the germination trials (refer chapter 3) were collected from. The samples LAC 4 and LAC 6 were leaves taken from seedlings grown from the parent plant. The *S. aviculare* samples were selected from spatially distinct groups.

The ITS PCR products were produced containing a 24 µl reaction mix: final concentrations consisting of 12.2 µl Mq $H_2O$, 2.5 µl 10 x PCR, 3.0 µl mg $Cl_2$, 4.0µl dNTP's, 1.0 µl ITS 4, 1.0µl ITS 5HP, .2µl BSA and .1µl Taq with 1 µl DNA added to make a total of 25µl. Initial concentration of Mg $Cl_2$ was 50mM,

the dNTP's were 1mM, ITS 4 and 5 were 10mM. Thermocycling protocol was 94°C for 5 minutes initial, followed by 33 cycles of 94°C for 30 seconds, 55°C for 30 seconds, 72°C for 60 seconds, and final amplification of 72°C for 10 minutes. EXO-SAP cleanup protocols followed, consisting of a 20 µl solution of 1.2 µl SAP, 2.4 µl EXO, and 16.4 µl Mq $H_2O$. The products were separated on a 1.5% agarose gel containing 2 µl ethidium bromide after mixing 3 µl DNA PCR product with 2 µl Loading Buffer and loading 5 µl Trackit™ 100 bp Ladder. Results were compared on the GenBank National Centre for Biotechnology Information (NCBI) Basic Local Alignment Search Tool (Blast) program (http://www.ncbi.nim.nih.gov/Blast.cgi).

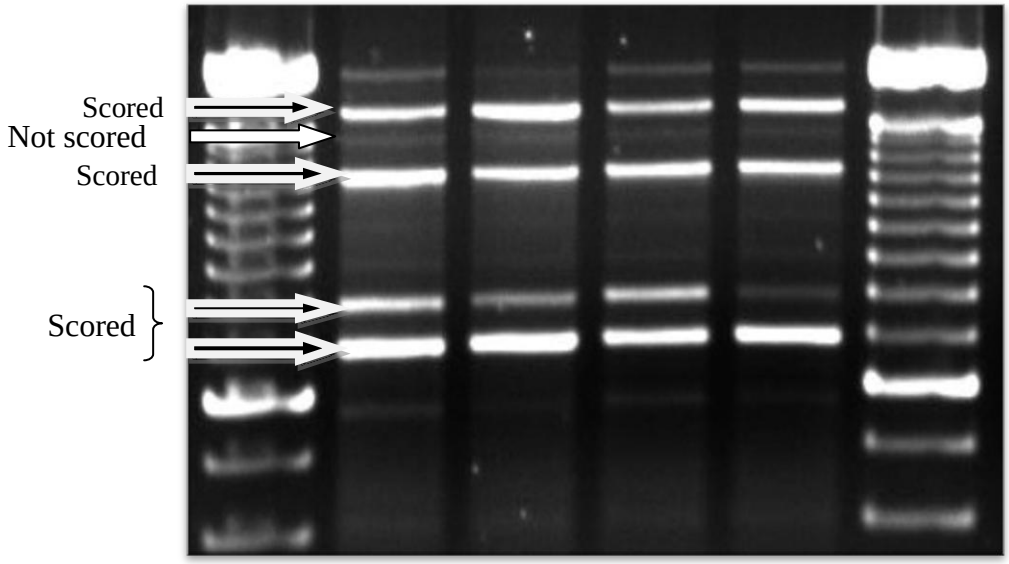


**Figure 4.1: Representative agarose gel showing ISSR bands of Primer A, Left to right: TAR 3/8, TAR 3/5, OKA 5 and ROT 2/5 indicating band strength used to determine presence or absence.**

## 4.3 Results

4.3.1 DNA extractions

The CTAB extractions produced continuously inconsistent results, with the majority of samples (n=62) containing large quantities of RNA despite RNAase treatment to preclude it. The CTAB RNAase protocol was modified at each extraction with no reduction in RNA. Many samples remained discoloured rather than clear, and in a number of samples the pellet produced at step 11 (Appendix

5) did not dissolve, requiring manual breaking up of the pellet. Seventy five samples were extracted, eleven producing good quantities of DNA alone, 35 producing high quantities of DNA with high quantities of RNA, two produced no DNA or inconclusive amounts, and 27 produced no DNA but high quantities of RNA (Table 4.4; Figure 4.2).

Invitrogen kits were trialled to 1) attempt more consistent extractions of high quantities of DNA, 2) remove RNA as no data was available on whether RNA would inhibit the PCR process with *S. aviculare*. The kits were consistent in extracting 120 samples, 94 producing high quantities of DNA with no RNA (including seven *S. laciniatum*), seven with DNA and RNA, and 19 producing no or inconclusive DNA results (refer Table 4.4; Figure 4.3), although a number of samples still retained discolouration the majority produced clear product. A total of 147 extractions with DNA were available for PCR.

**Table 4.4: Table showing DNA extraction results from CTAB and Kit procedures.**

| TYPE | DNA no RNA | DNA & RNA | RNA no DNA | NONE | Total |
|---|---|---|---|---|---|
| CTAB | 11 | 35 | 27 | 2 | 75 |
| KIT | 94 | 7 | | 19 | 120 |

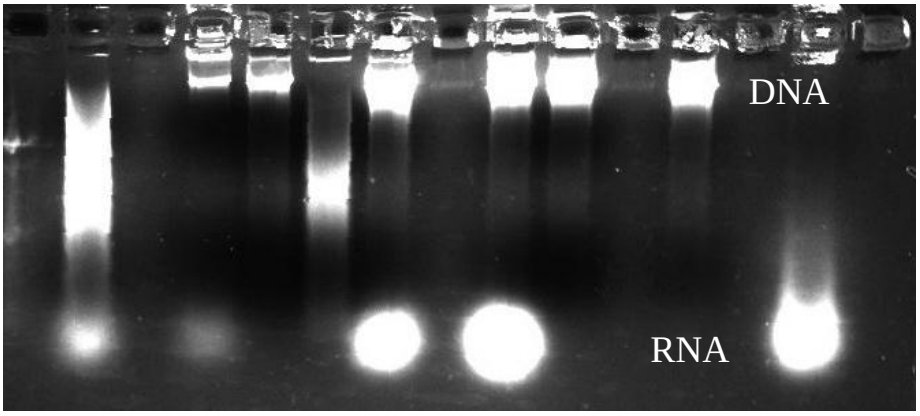


**Figure 4.2: Representative agarose gel of TAR 2: Left to right: ladder, 2/1, no sample, 2/3, 2/4, 2/5, 2/6, no sample, 2/7, 2/8, no sample, 2/9, no sample, 2/10 showing CTAB extractions with variable quantities of DNA and RNA.**

**Figure 4.3 Representative agarose gel showing Invitrogen kit extractions of previous CTAB extracted samples that failed or produced high quantities of RNA, showing consistent high quantities of DNA with no RNA. Left to right: ladder, ROT 2/10, ROT 2/11, OKA 1/1, TAR 2/10, TAR 3/18, TAR 2/2, TAR 2/17, TAR 1/19, TAR 2/17, TAR 3/20, ladder.**

### 4.3.2 PCR / ISSR Production

Results were continuously inconsistent from all thermocycling temperatures and timing protocols; reliable results would be produced on some primers, then not, and other primers would not amplify at all. Despite the final thermocycling protocols set to a standard *Pittosporum* protocol that provided very consistent polymorphic and informative band profiles on a wide range of indigenous species, the results remained inconsistent. However the *Pittosporum* protocol provided the most reliable and consistent results and informative band profiles overall.

Of the 20 tested primers, seven produced no or unreliable amplification (Table 4.5). Four of the seven provided some amplification and clear bands which showed no variation indicating monomorphism (XB). Two did not amplify enough to provide any bands or clear results (XA), and one (primer 15, XO) indicated some potential variation and polymorphic bands, but in seven attempts would not amplify enough to present a clear indication of polymorphism. The remaining primers produced clear distinguishable bands, however all were monomorphic (XC); one (primer 7, XP) producing some clear bands that had indicated variation (3 attempts) and potential polymorphism, but when amplified further (4 attempts) with increased samples, all the bands showed no variation

and were monomorphic (Figure 4.4). Where successful, amplification resulted in 2–13 clear, reproducible bands over all sites, with the percentage of polymorphic bands being zero (Table 4.6). The bands ranged in size between 300bp and 2100bp, with the majority of bands found between 600bp and 2072bp. Due to inconsistent and monomorphic results no final scoring of bands was undertaken.

**Table 4.5: Table showing primers and PCR amplification results for *S. aviculare*.**

| Primer | # attempts | Amplification | Amplification | Amplification |
|---|---|---|---|---|
| A | 7 | | | XC |
| B | 6 | | | XC |
| C | 4 | | | XC |
| E | 7 | | | XC |
| F | 5 | | | XC |
| 1 | 6 | | | XC |
| 2 | 4 | | | XC |
| 3 | 3 | | XB | |
| 4 | 2 | | | XC |
| 5 | 8 | | XB | |
| 6 | 3 | XA | | |
| 7 | 7 | | | XP |
| 8 | 4 | | | XC |
| 9 | 3 | | | XC |
| 10 | 9 | | | XC |
| 11 | 5 | | | XC |
| 12 | 2 | XA | | |
| 13 | 3 | | XB | |
| 14 | 2 | | XB | |
| 15 | 7 | XO | | |

Samples with RNA amplified as readily as samples with only DNA, where amplification succeeded (Figure 4.5). No amplification produced complete results where all samples amplified successfully (Figure 4.6).

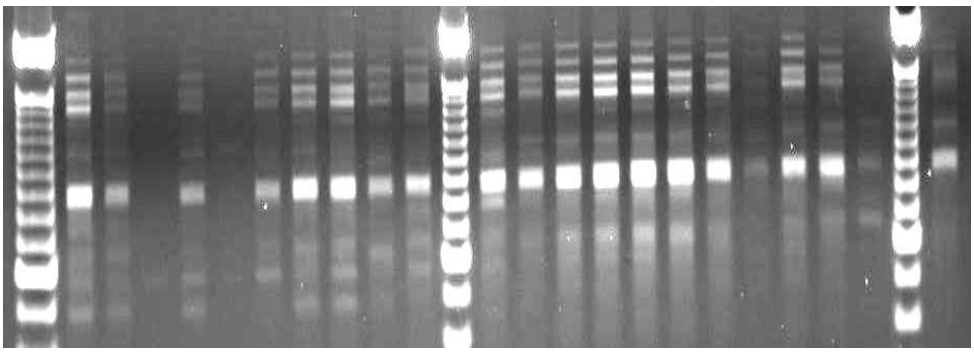

**Figure 4.4: Primer 7 gel showing monomorphic invariance with increased sample numbers. Left to right: ladder, URE 2, 3, 4, 6, 7, 10, 11, 12, 13, 14, ladder, TAR 1/3, 1/4, 1/11, 1/13, 1/15, 1/17, 1/18, 1/19, 1/8, 1/12, LAC 7, ladder, ROT 2/9 as positive control.**

**Table 4.6: ISSR primers used in analysis showing number of bands observed and levels of polymorphism.**

| Primer | Sequence | Number readable bands | % Polymorphic |
|---|---|---|---|
| A | $CA^{7}TC$ | 6 | 0 |
| B | $CAC^{4}GC$ | 5 | 0 |
| C | $CT^{7}AC$ | 4 | 0 |
| E | $CA^{6}GC$ | 9 | 0 |
| F | $GAG^{4}GC$ | 3 | 0 |
| 1 | $CA^{6}GG$ | 4 | 0 |
| 2 | $CT^{8}AC$ | 8 | 0 |
| 3 | $CT^{7}TG$ | 2 | 0 |
| 4 | $CA^{6}AC$ | 8 | 0 |
| 5 | $CT^{8}GC$ | 4 | 0 |
| 6 | $CA^{6}AG$ | 0 | 0 |
| 7 | $CA^{6}CT$ | 13 | 0 |
| 8 | $GA^{6}GG$ | 3 | 0 |
| 9 | $GT^{6}GG$ | 11 | 0 |
| 10 | $GA^{6}CC$ | 7 | 0 |
| 11 | $GT^{6}CC$ | 9 | 0 |
| 12 | $CAC^{3}GC^{3}$ | 0 | 0 |
| 13 | $GAG^{2}GC$ | 4 | 0 |
| 14 | $CTC^{3}GC^{3}$ | 2 | 0 |
| 15 | $GTG^{3}GC^{3}$ | 5 | 0 |

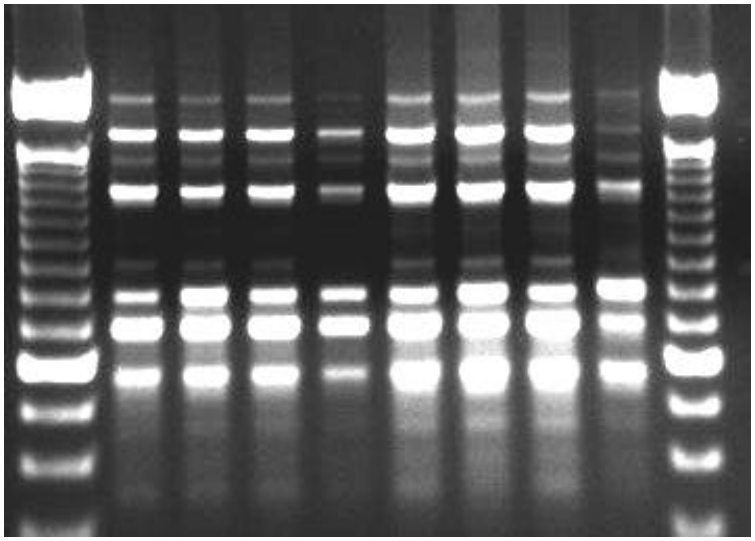

**Figure 4.5: Representative gel (primer 4) showing monomorphic bands with sample TAR 1/18 with RNA and *S. laciniatum* sample LAC 7, variable amplification with no variance indicated. Left to right: ladder, URE 6, TAR 3/2, ROT 2/7, TAU 6, MAU N5, PIR K2, TAR 1/18, LAC 7, ladder.**

The *S. laciniatum* samples produced more variable results than the *S. aviculare* samples in the same amplification. Where amplification was successful all monomorphic results indicated no variation between *S. aviculare* and *S. laciniatum*; although the more variable results from *S. laciniatum* made it difficult to be completely sure of the accuracy of reproducibility and that there being no band variation at all on every primer (refer Figures 4.4, 4.5 & 4.6).

DNA at 1 µl was used on 12 primers initially, with inconsistent results. Dilutions of DNA @ 1:100 were found to be unreliable, dilutions of DNA @ 1:10 were found to be generally as accurate as DNA @ 1µl, but overall consistency was not improved. As DNA at 1:10 µl dilution provided results with no less consistency than 1 µl, the 1:10 µl dilution was used for the full 20 twenty primers; the majority of results recorded therefore used DNA at 1:10 µl. DNA at 1 µl continued to be used for the positive control (refer Figure 4.6).

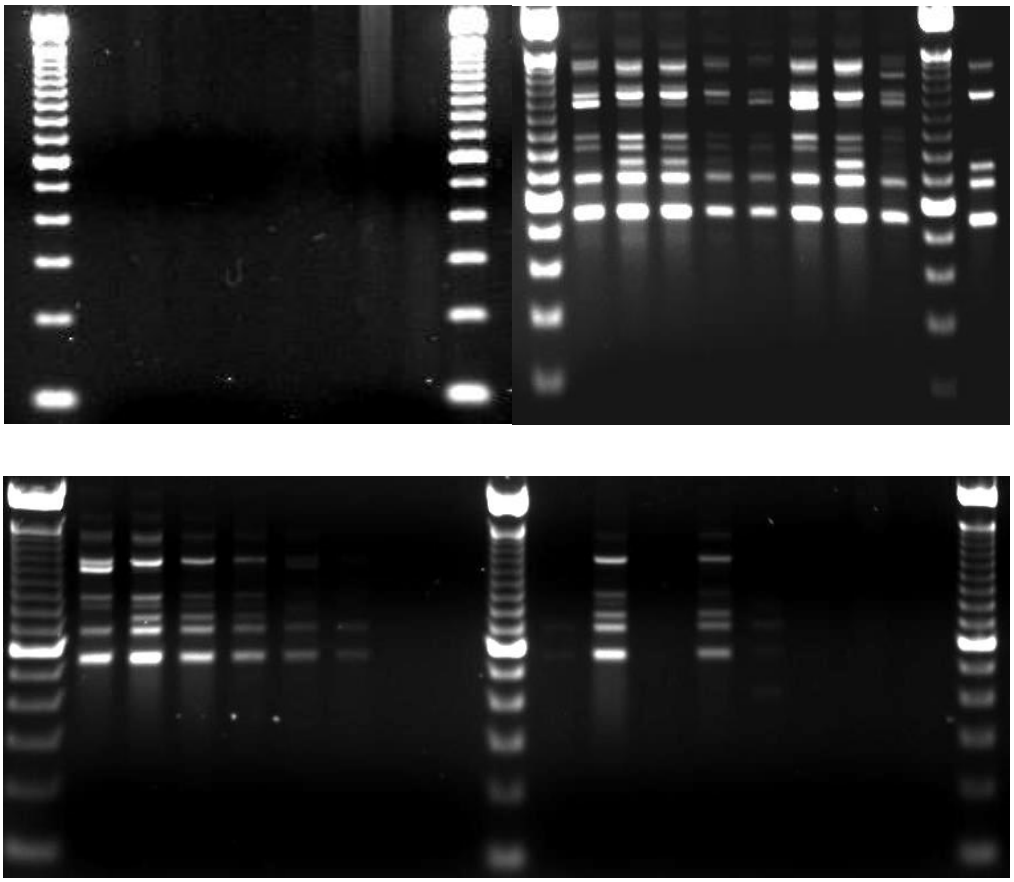

**Figure 4.6: Representative agarose gels on Primer 10 showing inconsistent results, even with different personnel and dilutions Key: <u>Top left</u> 6.05.09 made up by 2nd personnel, DNA @ 1µl; Left to right: ladder, URE 6, TAR 3/2, ROT 2/7, TAU 5, MAU N4, PIR K2, TAR 1/18 (RNA), LAC 7, ladder, *Pitt. corn*. positive control @ 1µl: <u>Top right</u> 28.06.09 made up by author, DNA @ 1:10µl: Left to right: ladder, URE 6, TAR 3/2, ROT 2/7, TAU 6, MAU N5, PIR K2, TAR 1/18 (RNA), LAC 7, ladder, TAR 1/12 positive control @ 1µl. <u>Bottom</u> 24.06.09 made up by the author, left gel DNA @ 1:10µl, right gel DNA @ 1µl: Left to right: ladder, URE 6, TAR 3/2, ROT 2/7, TAU 6, MAU N5, PIR K2, TAR 1/18 (RNA), LAC 7, ladder, same samples lanes 1-8, ladder, *Pitt. corn*. positive control @ 1µl.**

4.3.3 Sequencing

Results of the sequencing were:

LAC 4 returned a result of 713 sequence letters. The NCBI Blast results identified the sequence at 98% *S. laciniatum* GenBank accession AF244744.1, Taxonomy ID 45835, with 97% relationship values to *S. aviculare.*

LAC 6 returned a result of 442 sequence letters. The NCBI Blast results identified the sequence at 96% *S. laciniatum* GenBank accession AF244744.1, Taxonomy ID 45835, with 95% relationship values to *S. aviculare*.

LAC 7 returned a result of 173 sequence letters. The NCBI blast results identified no significant similarities. Changing the parameters produced no further identification.

TAR 3/2 returned a result of 723 sequence letters. The NCBI Blast results identified the sequence at 94% *S. aviculare*, GenBank accession AF244743.1, Taxonomy ID 4114, with 81% relationship values to *S. laciniatum*.

TAU 5 returned a result of 207 sequence letters. The NCBI Blast results identified no significant similarities. Changing the parameters produced no further results.

MAU 4 returned a result of 600 sequence letters. The NCBI Blast results identified the sequence at 99% *S. aviculare*, GenBank accession AF244743.1, Taxonomy ID 4114, with 98% relationship values to *S. laciniatum* (Appendix 6).

## 4.4 Discussion

4.4.1 DNA extractions

Results of the *S. aviculare* DNA extractions indicate that the CTAB extraction formula protocols (n=46) produces less consistent extraction results than DNA kits (n=101 including seven *S. laciniatum*) in extracting high quantities of DNA, and did not efficiently remove RNA. The DNA kits removed RNA more efficiently from the majority of extractions. The leaf material of *S. aviculare* is soft and has a high alkaloid content that may be the cause of discolouration, both protocols producing samples that retained discolouration after extraction. The discolouration did not appear to restrict the PCR process from either CTAB or Kit extractions. In regard the use of CTAB or Kit extractions in PCR, where amplification was successful the sample containing CTAB extracted DNA with RNA amplified just as readily as Kit samples with only DNA, therefore the PCR results (4.4.2) indicate that RNA is not a limiting factor in PCR and ISSR

production with *S. aviculare* plant material and CTAB extractions are adequate for PCR/ISSR production in evaluating *S. aviculare*, particularly where cost is a factor.

### 4.4.2 PCR / ISSR production

ISSR's have been successful in estimating inter and intra specific levels of genetic diversity and providing insights into levels of polymorphism in a wide range of taxa, including various *Solanum* species (Prevost & Wilkinson 1999; Camacho & Liston 2001; Reddy et al. 2002; Tikunov et al. 2003; Wu et al. 2004; Xie et al. 2005; Allendorf & Luikart 2007; Isshiki et al. 2008; Terzopoulos & Bebeli 2008; Venkatachalam et al. 2008). The results of this research using ISSR's found no polymorphism, and the various PCR amplification procedures attempted have highlighted that *S. aviculare* and *S. laciniatum* are difficult plants to produce consistent PCR/ISSR results from. No single amplification produced complete sample results that did not require further amplification to produce adequate bands that could attempt to provide consistent indications of any variation or not. The use of an *S. laciniatum* sample in each reaction produced variable results. Where amplification was successful the *S. laciniatum* sample generally produced less reliable bands than the *S. aviculare* samples, although overall results were generally readable enough to provide an indication of variability (refer Figures 4.4, 4.5 & 4.6).

The results included undertaking a number of checks to attempt to understand why the PCR amplification results were inconsistent. The protocols involving quantities and concentrations of PCR materials were checked to ensure accuracy, and any adjustments made. A number of dilutions of DNA were run to check if differing dilutions of DNA would provide more consistency, with overall consistency not improving. Differing thermocycler temperatures and times were trialled, over and above gradient trials undertaken. Results were still inconsistent, procedures working for a time, then not producing further reliable results. Final thermocycling changes were undertaken to a standard *Pittosporum* protocol that was providing very consistent ISSR polymorphic results and informative band profiles on a wide range of indigenous species. Results remained inconsistent, although the modified procedure provided the most reliable results and

informative band profiles overall, and was the final reaction protocol used to produce the results for analysis. Different personnel were trialled to ensure operator error from the author was eliminated. Three different laboratory personnel undertook trials, all with more PCR experience than the author. No different personnel produced more consistent results than the author.

Monomorphism in a species population is described as 1) common genes or alleles being invariant (n =1), identical across the population where virtually all individuals have the same identical genotype or allele at a single locus, or 2) the presence of a common allele at high frequency (>99%) (Falk et al. 2001; USGS 2008). Polymorphism in a population is described as 1) the presence of two or more genotypes or alleles (n = >1) at a locus, or 2) the frequency or proportion of loci that are genetically variable, the most common allele being less than <99% (Allendorf & Luikart 2007).

Primers with AG, GA, CT, TC, AC and CA repeat sequences are generally considered to show high polymorphism (Reddy et al. 2002). This research predominantly used primer repeats CA, GA, CT, and GT, therefore based on previous studies the sequences should have revealed polymorphic bands. The primer sequences used were anchored at the 5' end, and as such are usually considered to reveal more bands, and a high degree of polymorphism, although anchoring at the 3' end can give clearer banding (Reddy et al. 2002). These selected stock universal primers also produced high quantities of polymorphic bands consistently on *Pittosporum* spp. and other indigenous taxa previously in the PBRL; therefore considering those universal primers are known to amplify across a wide range of taxa (Reddy et al. 2002; Allendorf & Luikart 2007) the lack of polymorphic bands is suggestive of *S. aviculare* having predominant monomorphic loci rather than the research investigating with the wrong primers.

Sample size is also considered to influence the detection of genetic variation at a locus; the more individuals from a population sampled increases the likelihood of detecting variation and any rare alleles. The minimum sample size to achieve this is considered to be >50 (Allendorf & Luikart 2007). This research used nine spatially distinct samples per preliminary PCR/ISSR amplification and investigation, and increased this to >60 where any potential polymorphism was

indicated. The increase in samples amplified with the primer (primer 7) that indicted polymorphic bands, did not reveal any polymorphism, only confirming monomorphism. This continued lack of variation on the primer that indicated potential polymorphism, with the increased samples, indicates that the use of the nine samples per primer for initial investigation was valid, and did not mask the detection of polymorphic loci. It was not feasible within the timeframe and financial constraints, coupled with the high incidence of inconsistent results requiring continuous repeats, of this research to run >60 samples per preliminary investigation.

The ISSR production used chloroplast DNA, and all indications from previous research on *Solanum* spp. (Prevost & Wilkinson 1999; Tikunov et al. 2003; Isshiki et al. 2008; Terzopoulos & Bebeli 2008) and other genera (Camacho & Liston 2001; Wu et al. 2004; Xie 2005; Venkatachalam et al. 2008) indicated leaf material was appropriate to use. Whether in *S. aviculare* the haploid or diploid status of the DNA is important in detecting polymorphic variable loci was beyond the scope of this research. The overarching result remains that *S. aviculare* was a difficult species to undertake PCR amplification from, with no consistent technical answers as to why. The detection of polymorphic loci and subsequent genetic variation in *S. aviculare* is indicated to require more intensive searching with the use of increased numbers of primers, possibly a minimum of 100. This further research may include the investigation, design, and generation of specific primers to aid in the location of polymorphic loci (Pozcai pers. comm. 2009).

The results of the PCR/ISSR production indicate that *S. aviculare* is a highly monomorphic species, consisting of predominant monomorphic loci with only small amounts of polymorphic loci; 17 of 20 primers trialled producing indicated or identified monomorphic results (refer Table 4.4), with no confirmed polymorphic loci. This result is in alignment with Pozcai et al. (Pozcai pers. comm. 2009) who also found monomorphic patterns in *S. aviculare* DNA sequences in *trn*S-G, GBSSI, *trn*T-F, *ndh*F, SCoT, Intron Targetting (IT) and ISSR analyses; polymorphic loci existing “but not much” (Pozcai pers. comm. 2009).

Altukhov & Abramova (2001) utilising RAPD analysis on the haploid endosperm cells of nine conifer species, considered whether gene polymorphism may be hidden behind dominant homo or heterozygous alleles; specific analytical work being required to uncover them. Genetic variation in *S. aviculare* appears from this research's results to be confined to polymorphic loci that have not been located. This variation may show itself as moderately expressed ecotypes or forms, expressing differing leaf shapes within a population that would take specific observance and growth trials to determine. Sackville-Hamilton (2001) noted that geographic and genetic distance can have little relationship; plants that appear superficially to be a continuous population can be genetically distant, and differently adapted within a few meters of each other. Findings associated with this research (in particular differences in *S. aviculare* leaf shape and quantity between adjacent plants at specific locations, refer chapter 3) may be suggestive that forms or ecotypes exist that are closely adapted to their local environment (Gerasimenko 1970; Baylis 1968; Lesica & Allendorf 1999) and are moderately expressed through polymorphic loci that are difficult to locate, or hidden beneath dominant monomorphic alleles.

Altukhov & Abramova (2001) also documented structural duality in finding both polymorphic and monomorphic loci, which this research did not. No polymorphic loci were found in any of the primers trialled; however whether polymorphic loci are hidden behind dominant invariant alleles is not answered by this research. The use of the same universal primers located polymorphic loci, and produced successful results across a wide range of other indigenous taxa in the PBRL, further suggesting that any polymorphic loci in *S. aviculare* are either hidden, or exist in low numbers.

The research undertaken by Altukhov & Abramova (2001) confirmed that monomorphic loci are intraspecies invariant, displaying neither individual intrapopulation nor geographic (interpopulation) variation; being determinants of differentiation in species traits within a genus and higher taxa, and suggesting an important role in evolution. This current ISSR research appears to confirm the monomorphic loci findings of Altukhov & Abramova (2001), whereby the results documenting the monomorphic loci found in this research showed no intraspecies, interpopulation or geographic variance in spatially distinct

populations of *S. aviculare*, or interspecies variance with *S. laciniatum*. The research results are suggestive that monomorphic loci are, as described in Altukhov & Abramova (2001), determinants of traits at generic level rather than intraspecies differentiation, and as such may be the evolutionary determinants of change within *Archaesolanum*. Thus, monomorphic genes may be the determinant markers of the differentiation of the *Archaesolanum* sub-genus within the *Solanum* genus, marking and maintaining the unique status of *Archaesolanum* within the evolution of *Solanum*.

*Solanum aviculare* and *S. laciniatum* do not appear to be changing at species level, beyond potentially localised ecotypes or forms, possibly resulting from a high incidence of self fertilisation. Whether any inbreeding is the cause of the apparent predominant genomic monomorphic homozygosity, with any possible resultant inbreeding depression producing fitness reduction, warrants further research. Polymorphic loci appear to be the determinants of variation at species and population level, and the lack of variation documented between and within populations, due apparently to the low incidence of polymorphic loci, is indicative the species' may not be readily adaptable to change. Therefore any onset of changes to environmental and climatic conditions, pathogen incidence, or sudden random occurrences associated with photoperiod changes, could further their decline; small populations being especially vulnerable (Allendorf & Luikart 2007).

The invariance and lack of polymorphism between *S. aviculare* and *S. laciniatum* was not expected, and of further interest (Pozcai pers. comm. 2009). Results suggest that the known similarities between, and the high bootstrap values connecting *S. aviculare* and *S. laciniatum*, exist on monomorphic loci, therefore the differences that highlight the species differentiation may only be located on polymorphic loci. Evidence suggests these are small in number and difficult to locate, and further specific work on this phenomenon and the location of the specific loci is suggested.

### 4.4.3 Sequencing

The results of the sample sequencing and GenBank analysis indicate the species used in the PCR/ISSR production were true to type, and the PCR results produced indicating lack of variation between *S. aviculare* groups and *S. laciniatum* were resultant from the use of true to type samples. The failure of analysis and lack of identification on sample LAC 7 and TAU 5 are most likely caused by error in the ITS PCR process, due to lack of amplification of the starting template not producing enough template sequence length for the sequencing reaction to read. The Tauranga samples (TAU) proved to be generally the most problematic and difficult samples to amplify, but where amplification succeeded the results were invariant with all other samples, and the leaf and flower morphology identified them as *S. aviculare*, therefore the results are considered valid. The *S. laciniatum* sample LAC 7 was the sample used the most, due it being the parent plant the other samples came from, and the lack of identification is unfortunate. However the sample is considered valid, as the flower and leaf morphology identify it as *S. laciniatum*. Herbarium voucher specimens lodged at the Waikato University herbarium (WAIK) are available to refer against other existing specimen data bases.

### 4.4.4 Conservation and restoration

*Solanum aviculare* is documented as being ‘in decline’, ‘sparse’ and ‘data poor’. Surveys of population numbers (refer chapter 5) indicate that when found, *S. aviculare* generally exists in isolated small groups or individuals within open areas such as stream and forest edges, or tree fall gaps rather than populations. Populations of *S. aviculare* appear only in larger ephemeral disturbance areas such as earth slips or regenerating areas following post pine tree production (refer chapter 5). Therefore with *S. aviculare* documented as existing in ‘declining data poor’ populations, the overarching aim of this research was to provide requisite data on the genetic diversity of *S. aviculare*, including any incidence of inbreeding generated by isolation, that may enhance restoration programs for the species by identifying any local provenance, gene flow connectivity between spatially distinct populations, and identifying genotypes to increase the gene pool of isolated groups. The production of this data depended

on the location of polymorphic loci that would identify any variation, allowing recommendations to be made.

By not finding any polymorphic loci this research has identified what appears to be a predominant monomorphic state of *S. aviculare*, with only the suggestion of local provenance such as ecotypes. This monomorphism suggests that *S. aviculare* is uniform in its genotype over the extent of its range as described by Baylis (1963). This incidence of monomorphism suggests that *S. aviculare* may be transplanted between regions with no deleterious effects, seeds being utilised from any region for use in restoration of the species. This is tempered by caution that, as applied to other genera, some local adaption may exist as ecotypes that may not be suited to particular translocations. An example being plants located at altitude 566m in Whirinaki Forest may not readily translocate and perform well at sea-level for dune restoration. Further research on ecotypes that may help ensure successful restorations is considered warranted.

This research appears to highlight the close relationship between *S. aviculare* and *S. laciniatum*, suggesting the possibility of their combined use in restoration; however the results of Baylis (1963) show that crosses between the two taxa can produce non-viable seed. These two species are not easy to distinguish vegetatively, and therefore caution is suggested to be applied in restoration plantings where seedlings are not fully identified as *S. aviculare* or *S. laciniatum.* Full identification of the origin of the taxa being planted is essential to ensure that the objective of providing a viable long-term population to achieve the aims of increased biodiversity and ecosystem functioning.

## 4.5 Conclusions and recommendations

Aims of this research have been fulfilled, with results confirming earlier studies regarding the 'uniformity' status of *S. aviculare* and *S. laciniatum*, and subsequent applications for restoration planning. New information regarding PCR/ISSR production, DNA extraction, monomorphic and polymorphic loci status, interspecies and intraspecies invariance of *S. aviculare* and *S. laciniatum* has also been produced. Conclusions, summary of the achieved aims, and

consideration of further research areas that can be proposed from these results follows.

The extraction of DNA from *S. aviculare* leaf material was successful from both CTAB and DNA Kits. The CTAB extraction protocols were unable to remove RNA, although the use of a sample containing high quantities of RNA in conjunction with high quantities of DNA (TAR 1/18) in each reaction provided results that indicated RNA was not an inhibiting factor in PCR/ISSR production in *S. aviculare*. While DNA kits produced more consistent extraction results than CTAB, and removed RNA successfully, they also cost a great deal more (ca. $300 per 50 extractions). Where cost is a factor the use of CTAB protocols is recommended, as the presence of RNA did not appear to inhibit PCR production in any of the results.

*Solanum aviculare* and *S. laciniatum* are difficult species to produce consistent reliable PCR/ISSR results from. The use of three different laboratory personnel experienced in PCR/ISSR production, coupled with the results continuously varying widely using the same protocol on the same primer, but on different days, suggests the difficulties associated with PCR production appear to be associated with the species' rather than operator or protocol error. No polymorphic loci were identified, nor any intraspecies or interspecies genetic variation documented, despite the use of universal primers that located polymorphic loci on a number of indigenous species. This research's ISSR results indicate, through the primers trialled, that the populations of *S. aviculare* between Te Urewera in the Eastern Bay of Plenty, and Pirongia on the West Coast of Waikato, in the North Island of New Zealand are monomorphic, consisting of invariant genotypes on multiple loci; where virtually all individuals have the same genotype (gene /allele) at the same locus; and is invariant with *S. laciniatum* on these monomorphic loci. *Solanum laciniatum* also potentially consists of invariant genotypes.

This is consistent with Baylis (1963) where it was stated that *S. aviculare* genetically is "substantially uniform" with no geographic variation, not just through New Zealand but over the whole of its range including Australia and New Guinea; *S. laciniatum* also being "identical" in Australia and New Zealand.

Baylis's (1963) description of uniformity was confirmed by Gerasimenko (1971), who also suggested that intra specific variations existed that were moderately expressed as forms in *S. laciniatum*, and were stable and heritable as individual features "not of their complex" (subgenus *Archaesolanum*). Gerasimenko suggested that these forms are caused by geographic and ecological factors, similarly to the suggested ecotypes relating to day length existing in *S. aviculare*, but not proven in *S. laciniatum* by Baylis (1968). This research indicates that ecotypes and forms may be present (refer chapter3), but remain unproven until polymorphic loci are located. Specific research to investigate forms and ecotyping is recommended. Further research may provide information on whether the lack of variation observed may be also be related to connectivity of populations when forest was contiguous.

Monomorphic loci appear from the results to be the determinants of generic level traits, and are possibly locations of the evolutionary markers of *Archaesolanum* differentiation within *Solanum*. The results also suggest that polymorphic loci appear to be the areas where variation exists at population and species level in both taxa; although the presence of polymorphic loci in both species appears to be low, difficult to locate, and may be hidden behind dominant monomorphic loci. These polymorphic loci are most likely determinants of variation that, due to their low presence, is moderately expressed, and therefore difficult to discern. Further research to uncover the extent of polymorphic loci, whether they are hidden behind monomorphic loci, or require diploid rather than haploid DNA to investigate variance within populations, that may also help discern and confirm forms, ecotypes and potential inbreeding, is recommended. This research may also include primer sequence investigation, and the possibility of creating specific primers. Research to determine whether monomorphic loci are the determinants of generic level differentiation is also highly recommended, adding further to the phylogenetic studies that are determining the relationships within the *Solanum* genus. The invariance of *S. laciniatum* with *S. aviculare* is suggested for further research, including whether this phenomenon is due to, and confined only to monomorphic loci.

The monomorphic results suggest that *S. aviculare* does not deviate from the expected Hardy-Weinberg values, as the allelic frequencies appear to be constant,

and is therefore not evolving. However the potential appearance of forms that may be heritable and phenotypically derived (refer Gerasimenko 1971) within *S. aviculare* populations (refer chapter 3) suggests that microevolution is may be taking place via deviation from the Hardy-Weinberg equilibrium, and *S. aviculare* is evolving. Further research is suggested that will entail the location of polymorphic loci to fully document variation, and confirm the evolutionary status. This information may help determine further conservation ranking in regard the species protection and enhancement.

The results highlighting the predominant monomorphic status, and low presence of polymorphic loci in both taxa, suggests that both species are unable to modify or change quickly in event of environmental or climatic conditions changing, making them vulnerable to extirpation or extinction. Therefore conservation and restoration programs for both species are important to be undertaken throughout the country, and in particular the North Island to ensure that viable seed is produced and spread, to help reverse the decline, and continue the survival of the species. Requisite data gained from the results, in consideration of conservation and restoration planning, suggests that both *S. aviculare* and *S. laciniatum* are able to be transplanted within regions, and potentially nationally. However due to the unresolved status of ecotypes, and the known low viability of seed from cross breeding, it is recommended that *S. aviculare* and *S. laciniatum* are positively identified, not planted in close proximity to each other, and that climatic and geographic factors such as altitude, and the potential for local adaption are considered in all planting programs. The use of local seed sources for planting programs, considering local adaption, is recommended to ensure long term success of restoration programs.

# CHAPTER 5

# CONSERVATION AND CULTURAL STATUS

# PART A: CONSERVATION STATUS

## 5.1 INTRODUCTION

### 5.1.1 Background

Conservation on Public Conservation Land in New Zealand is undertaken by the Department of Conservation (DOC) which administers 24 Acts of Parliament, and functions under various other Acts and General Policy; the primary legislation being the Conservation Act (1987). Part I Section 4 of the Act states that; "This Act shall so be interpreted and administered as to give effect to the principles of the Treaty of Waitangi". The General Policy is designed to help integrate conservation management by providing criteria for decision making, determine the direction of the legislation, provide national consistency in management, determine whether an issue is best addressed at national, local or another level, and decide on how the Department will engage with tangata whenua and the public in conservation management; ensuring "that decisions are not predetermined by restricting the possibilities provided for in the legislation". All policies are to be considered in conjunction with other relevant legislation including the Wildlife Act 1953, the Marine Reserves Act 1971, the Reserves Act 1977, the Wild Animal Control Act 1977, and the Marine Mammals Protection Act 1978. District and Regional Councils undertake conservation and restoration work on other Public Lands, operating within the Conservation Act (1987) and the Resource Management Act (1991).

Conservation functions as defined in the Act are: "to manage for conservation purposes, all land, and all other natural and historic resources held under the Act". The Department also determinations the threatened plant status list. Conservation General Policy considers that we need to conserve and care for species because of their recognised values. As many of our plants are endemic they are often perceived by many to be central to our sense of self and way of life. Conservation has been necessary to prevent the irreparable loss of these unique plants, and integrated conservation management can only occur when all

areas of conservation activity contribute towards objectives that are consistent with the relevant legislation and General Policy, are not inconsistent with each other, and resolve conflicts between potentially conflicting objectives and interests.

While implementation of conservation policy has been successful in preventing extinction of many endangered species and ensuring protection and restoration of many important ecosystems (New Zealand Biodiversity Statement (NZBS) 2000; Atkinson 2001), the basis of the General Policy is rooted in complex political documentation, which does not always provide for effective implementation (Parliamentary Commissioner for the Environment (PCE) 2004). Lack of effectiveness can often in part be due to political changes in Government, subsequent political policy being at cross-purposes with environmental need. Selectively used modern science-based policies can depict the environment and ecosystems as quantitative, often mechanical and distinct from human interaction rather than qualitative and holistic (Lyver et al. 2008). The NZBS (2000) acknowledges a problem has existed where there has been a tendency to separate species management from their habitats and is attempting to address this through a stronger ecosystem focus in management programs.

### 5.1.2 Threat classification

Indigenous New Zealand taxa are listed within two broad taxonomic groupings for conservation threat classification assessment; taxonomically determinate and taxonomically indeterminate. These broad groupings are further classified into two evaluated categories: 1) threatened: a) nationally critical, b) nationally endangered, and c) nationally vulnerable; and 2) at risk: a) declining, b) recovering, c) relic and d) naturally uncommon. Sub-categories further define the exact status of the species. Unevaluated taxa are listed as ‘data deficient’ (Townsend et al. 2008; Figure 5.1). The New Zealand Threat Classification system differs from the world Conservation Union (IUCN) (2000) Red List categories system, more accurately depicting the uniqueness of the New Zealand taxa (de Lange et al. 2009). Compilation of the threatened plant list is undertaken by the New Zealand vascular plant panel, appointed by DOC after extensive botanical consultation. This is made up of six experts: three from DOC, one from

a University, one from a Museum herbarium, and one from a Crown Research Institute, who gathers and collates the information including submissions from the New Zealand population, then make a final status determination and publish the list.

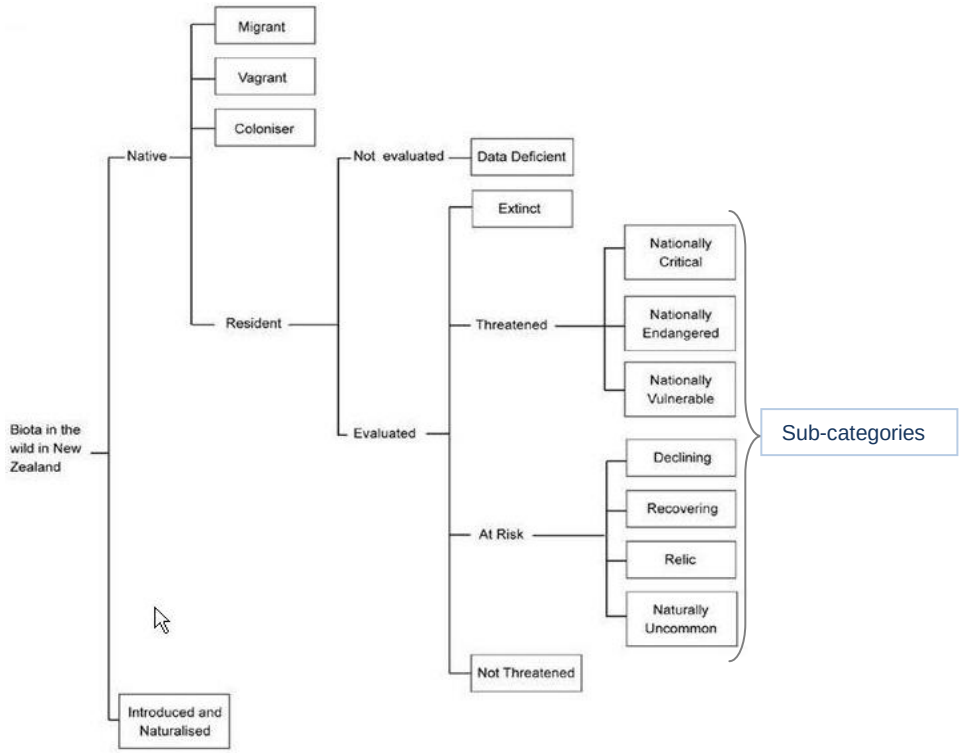


**Figure 5.1: Diagrammatic representation of the structure of the New Zealand Threat Classification System. (**Adapted from Townsend et al. 2008**)**

The largest proportion of plants listed as under threat (53%), are in lowland (30%) and coastal (23%) habitats; with 29.25% (n=792/2705) of all threatened and uncommon plants being found in the North Island (de Lange et al. 2009). The overriding conclusion of the de Lange et al. (2009) listing is that the status of New Zealand's indigenous vascular plants is worsening, and unless further action is taken in the near future New Zealand stands to lose a number of species from the wild.

The New Zealand Biodiversity Statement (2000) warns that the decline in indigenous biodiversity is New Zealand's most pervasive environmental issue; the loss of species is related to loss of natural ecosystems and habitats, particularly in lowland areas in which a once contiguous landscape has been fragmented, isolating communities. The implementation of new breakthrough

techniques in species management, while having an impressive effect, has not halted the decline of species, and a downward trend continues. The challenge is to halt the decline and turn it upward. To achieve a halt in species decline, and to maintain and restore viable populations of all indigenous species and subspecies across their natural range, including maintaining their genetic diversity, the implementation of an integrated number of measures is considered to be vital. This implementation includes improvements in technical knowledge, increasing good information on critical species to ensure efforts are targeted successfully, reducing a paucity of knowledge, increasing use of the Māori world-view and the important role traditional knowledge can play in sustainably managing our biodiversity. These aims cannot be achieved by Government alone: they require involvement from the whole New Zealand Community.

The New Zealand Plant Conservation Network (NZPCN 2005) lists *S. aviculare* as 'non-threatened', although it recognises *S. aviculare* as having become less common in the northern North Island. As the reasons for the decline are unclear the NZPCN suggests a nationwide assessment of its exact status is warranted. However the updated Threatened and Uncommon Plants of New Zealand Reappraisal (2008 revision) lists *S. aviculare* as, 'At Risk', 1) 'Declining', sub-categories 'sparse and data poor', as well as 'threatened overseas' (de Lange et al 2009). This is confirmed by Tangata Whenua, many of whom were raised in forests, and when asked today do not know this species (refer Part B).

The criteria for *S. aviculare* to qualify as an 'At Risk' plant rather than 'Threatened' is for it to be 'declining' (although buffered by population size or slow decline rate); biologically scarce; or surviving in relic populations of either a) 5000-20,000 individuals occupying ≤ 1000ha ($10km^2$) with an ongoing or predicted decline of 10-30% over the following 10 years or three generations, b) 20,000-100,000 individuals occupying ≤ 10,000ha ($100km^2$) with an ongoing or predicted decline of 10-50% over the following 10 years or three generations, or c) >100,000 individuals >10,000ha and with an ongoing or predicted decline of 10-70% over the following 10 years or three generations. The criteria relating to the status of *S. aviculare* being sub-category 'data poor' means that confidence in the listing is low due to poor-quality data, so collection of sufficient data may confirm or change the listing. The criteria for a sub-category 'sparse' listing is

that *S. aviculare* typically occurs in small and widely scattered populations, and sub-category 'threatened overseas' implies that *S. aviculare* is threatened in its natural range outside of New Zealand (Townsend et al. 2008).

The decline of *S. aviculare* is occurring despite its known ingestion by common bird species such as blackbird (*Turdus merula*) (Burrows 1999), dissemination by starlings (*Sturnus vulgaris*) (Fergusson & Drake 1999) and defecation of viable seed by silvereyes (*Zosterops lateralis*) (Stanley and Lill 2003). The observed apparent lack of colonisation in areas where pest species are not dominant is also unexplained, and may relate to reductions of dispersal agents such as kererū (*Hemiphaga novaeseelandiae*) (Lyver et al. 2009) and kaka (*Nestor septentrionalis*).

The overarching goal of this research is to contribute to the Threatened Plants listing and requirements of the Conservation Act (1987) and the NZBS (2000) by providing information to add to the knowledge base of the species including its current distribution and habitats, thereby assisting in management practices, especially ecosystem based, aimed at halting the decline of *S. aviculare*. The goals will be achieved by documenting and providing information on the current conservation status of *S. aviculare* (and *S. laciniatum*) currently recognised as 'data poor', 'sparse' and an 'at risk declining' species (de Lange et al. 2008).

## 5.2 Methodology

The methods used were field searches, e mail surveys and discussions with key informants (private individuals and ecologists) following Human Ethics approval by the Human Ethics Committee of Waikato University (10.02.2009).

1. Field searches to locate *S. aviculare* were undertaken by the author. These included the research sites at Te Urewera, Tarawera, Rotoma/Okataina, Tauranga, Maungatautari and Pirongia (refer Ch 3), areas of Manawatū, Horowhenua, and following discussions with a local Wellington nurseryman, Fred Allen of Kiwi Plants Ltd, the Pauatahanui Estuary (Wellington).

2. Surveys were undertaken via e mail (Appendix 7) to DOC Offices throughout New Zealand, and to private ecologists in areas where *S. aviculare* and *S. laciniatum* were known or considered to be found, based on information provided by the Manaaki Whenua-Landcare Research New Zealand Flora (http://nzflora.landcareresearch.co.nz), and Allen Herbarium databases. Some of these initial contacts were followed up by phone conversations. In addition after publicity surrounding the research a letter was received from Mr. Ron Vanstone of Ohiwa Harbour in the eastern Bay of Plenty regarding places he had observed 'poroporo' growing.

The survey questions were designed to provide an estimation of the commonality of the species; the nature of their appearance (i.e. individually or in groupings); site (location within or outside forest) and habitat, to provide a considered estimation of numbers of individuals rather than a complete survey of numbers, and habitats which was beyond the scope of this research.

All survey participants were asked the same questions: In regard their conservation/observed areas:

**Q.1:** Is *S. aviculare* seen a) commonly (regularly observed >50%), b) uncommonly (regularly observed <50%) or c) rarely (regularly observed <10%).
**Q.2:** Does *S. aviculare* appear commonly, uncommonly or rarely in a) disturbed areas within intact forest e.g. slip sites, b) canopy gaps, c) forest edges, d) open areas outside of intact forest, or e) other.
**Q.3:** When observed as per Q.2 does *S. aviculare* occur as a) single plants, b) small groupings or c) populations > 20 + plants, commonly, uncommonly or rarely.
**Q.4:** Does *S. laciniatum* also occur?

The classification of the regularity criteria used for question one (Q.1) relates to whether the informant observed the species almost always (>50%) on every journey into an area, and so considered it to be a common plant; <50% relates to whether the species was observed ½ or less of the time on every journey into the area, but considered it still to be somewhat less common than a); and <10%

relates to whether the species is seldom observed, and considered scarce, being seen rarely (<10% of the time) on a journey into the area.

Questions two (Q.2) and three (Q.3) were related, and the regularity criteria for both questions was whether the informant observed *S. aviculare*, when located, occurring in habitats and groupings in a criteria manner similar to Q.1, but specifically: common being *S. aviculare* when located, regularly appeared and occurred in that habitat commonly, and was almost always seen in that habitat >50%; uncommon being *S. aviculare* when located, less regularly appeared and occurred in that habitat uncommonly, (seen <50%), and rare being *S. aviculare* when located, rarely appeared and occurred in that habitat (seen <10%).

## 5.3 Results

Field searches in areas of Manawatū, Horowhenua were confined to main road locations, including reserve areas. No *S. aviculare* was found in Manawatū or Horowhenua, only one population of >25 plants of *S. laciniatum* located on a farm on the true right of the highway ±1km south of Hunterville, Manawatū. There was one plant growing beside a shearing shed, the remainder were in an adjacent grove of mature pine trees (*Pinus radiata*). No *S. aviculare* was located at the Pauatahanui Estuary only *S. laciniatum*. Discussions with Fred Allen of Kiwi Plants Ltd established that *S. laciniatum* appeared also to be the species he had found in valleys alongside the Hutt Highway, but it was not established whether the *S. laciniatum* plants at Pauatahanui had been planted, or were naturally occurring from populations in the Hutt hills.

Mr. Ron Vanstone in his letter confirmed he had observed the plants on the Tarawera Falls track, labelled by the writer TAR 1 (refer chapter 3), and had seen plants growing on “the scrubby hillsides of the Hutt valley”, and possibly in the Hikutaia Domain inland from Opotiki. Plants were also seen growing in the old Opotiki Hospital grounds and seedlings from this site had been transplanted by him to the Ohiwa Harbour. Neighbours of his had planted a ‘poroporo’ (*S. laciniatum*?) plant, and it had spread via birds under avocado trees. The writer

has been unable to confirm if the species growing in the avocados, the old Hospital grounds or the Hikutaia Domain was *S. laciniatum* or *S. aviculare*. Although results from the Opotiki DOC office suggest they may be *S. aviculare* in the Hikutaia Domain, and coupled with the descriptions provided by Mr. Vanstone may be *S. laciniatum* in the old Hospital grounds, and the avocados. The plants on the Tarawera falls track are confirmed as *S. aviculare*, and the plants in the Hutt Valley are most likely *S. laciniatum* as observed by Fred Allen.

Replies were received from Oratia Nurseries in Waitakere Ranges, DOC offices at Kaitaia, Waipoua, Opotiki (including Te Urewera and Aniwaniwa), Murupara (Whirinaki), Wairoa (Lake Waikaremoana/Southern Te Urewera National Park), South Marlborough, Kaikoura (Hundalee Ecological Area), and North Westland. The results from the reported observances indicate that *S. aviculare* is generally found more rarely than commonly, and *S. laciniatum* is very rarely found (Table 5.1). The results also show a key theme in observation that *S. aviculare*, when located was found in all habitats, but generally observed more rarely than commonly, and in low numbers in those habitats (Table 5.2 & 5.3).

**Table 5.1: Observed general occurrence of *S. aviculare* and *S. laciniatum* from 10 survey replies.**

| Common | Uncommon | Rare | *S. laciniatum* | |
|---|---|---|---|---|
| | | | Yes | No |
| 1 | 1 | 8 | 1 | 9 |

**Table 5.2: Appearance in localised habitats of *S. aviculare* within observed area, in Q.2.**

| Habitat | Common | Uncommon | Rare |
|---|---|---|---|
| **a) Disturbed areas in intact forest** | 1 | 2 | 3 |
| **b) Canopy gaps** | | 1 | 6 |
| **c) Forest edges** | | 2 | 6 |
| **d) Open areas outside intact forest** | 2 | | 5 |
| **e) Other** | 2 | 1 | 2 |
| **Total observances** | **5** | **6** | **22** |

**Table 5.3: Population size occurrence of *S. aviculare* at Q. 3, in relation to habitat areas in Q.2.**

| Habitat & Occurrence | Common | Uncommon | Rare |
|---|---|---|---|
| **Single plants a) in disturbed areas** | 1 | 2 | 1 |
| **Single plants b) canopy gaps** | 1 | 2 | 3 |
| **Single plants c) forest edges** | 1 | 2 | 4 |
| **Single plants d) open areas o/s forest** | 1 | 2 | 3 |
| **Single plants e) other** | 1 | 2 | 2 |
| **Small groupings** | 3 | 3 | 2 |
| **Populations >20+ plants** | 2 | | 2 |
| **Total observances** | **10** | **11** | **17** |

The full results from each of the DOC regions are as follows:

**Q.1:** *Solanum aviculare* was generally found rarely (<10%) in Waitakere, Kaitaia, Waipoua, Te Urewera/Aniwaniwa, Opotiki, Whirinaki, Kaikoura and North Westland, and where found was mainly as patchy individuals. It was uncommon (<50%) in Waikaremoana and common (>50%) in South Marlborough. Comments made were: a) Waitakere "only seen occasionally, three times in nine years, surrounded by weeds, usually where trees have fallen over", b) Kaikoura was not clear on how much of *S. aviculare* found was naturalised from tangata whenua introduction, or how much resulted from spread of natural populations, c) Opotiki stated it was "not common in any of conservation areas here, seems to be observed in more northern localities"(coastal/lowland?) d) South Marlborough stated it was observed only "In lowland/coastal areas", e) Waipoua only observed *S. aviculare* in one of the Waipoua River catchments, "I have only observed it in one of the river catchments (the Waipoua River) where it is common in places in disturbed alluvial flats", f) North Westland noted that "Individuals are patchy. The only areas where the plants have been observed by me are in coastal situations in north Westland", g) Whirinaki only found one small grouping within a large area of tawa (*Beilschmiedia tawa*) forest.

**Q.2:** Where *S. aviculare* was found results and comments were: a) Kaitaia found it rarely in any location: b) Waipoua noted it was uncommon in disturbed areas, and rarely seen in canopy gaps, forest edges and open areas, c) Waitakere found

it rarely in any location: “not enough plants over broad areas to consider any site to be common”, d) Te Urewera/Aniwaniwa found it rarely, and only in disturbed areas, canopy gaps and forest edges, e) Opotiki found it commonly in disturbed areas and open areas outside of intact forest, uncommonly in forest edges and any other areas, and rarely in canopy gaps: “very scattered, short-lived, and a good bird species”, f) Waikaremoana noted it as uncommonly found only in disturbed areas, canopy gaps and forest edges, g) Whirinaki found it rarely in all sites except ‘other’(e.g. stream edges), h) Kaikoura found it rarely, and only in forest edges and open areas outside of intact forest, i) South Marlborough said it was commonly observed only in open areas outside of intact forest and ‘other’ ‘Riparian areas’, “Common in regenerating areas/disturbed sites in lowland, coastal, and riparian areas”, and j) North Westland found it rarely, in forest edges and open areas outside of intact forest: but only near the coast.

**Q.3:** When *S. aviculare* was observed results and comments were: a) Kaitaia it was rarely observed, b) Waipoua observed it rarely as single plants, and commonly as small groups and populations >20+ plants, c) Waitakere noted it appeared only as single plants and small groupings: “no more than 6 plants found at one time, and they are mostly seedlings”, d) Te Urewera/Aniwaniwa observed it rarely and only as single plants, e) Opotiki observed it uncommonly as single plants, in small groupings commonly, and as populations >20 + rarely: only small groups being observed, f) Waikaremoana observed it as common as single plants, uncommon as small groupings, g) Whirinaki observed only as single plants and small groups, h) South Marlborough observed it as common as small groupings and populations >20+ plants in the described areas. “In Marlborough there is very little forest remaining; many of the ecosystems are fragmented and this tends to favour species such as poroporo. After the large fires in the Wither Hills in 2000, poroporo was noted as one of the main regenerating species in the riparian zones where fruit had washed into and accumulated. From our recently published PNA report *S. aviculare* was recorded in all the lowland ED’s within the Wairau Ecological Region (Blenheim, Grassmere, Flaxbourne, Wither Hills)”, i) Kaikoura it was observed rarely, and only as single plants, and j) North Westland observed it uncommonly only as single plants.

**Q.4:** Results and comments were: Only Opotiki noted *S. laciniatum* as being present: but "very rarely encountered". All other areas noted *S. laciniatum* as not being present. Comments from Waitakere were that "Despite the naturalisation of *Solanum laciniatum* in developed areas, it has not established successfully in the wild", and South Marlborough noted that results were from the Flaxbourne ED only.

## 5.4 Discussion

As an in-depth survey was beyond the scope of this research the DOC survey's were designed only to provide some insights into the decline and uncommon appearance, as well as further details of the status of *S. aviculare* and potential suggestions for further research. Because results were not received from all conservation areas throughout the country and the Human Ethics approval from the University did not allow further contact if the initial approach was unsuccessful, the North Island the results are representative only of the eastern and small areas of the northern North Island. However replies from the South Island are broad enough to be considered representative of all areas in which *S. aviculare* occurs. The central and lower North Island status comes from searches by the author, and information supplied from a single nurseryman. The key point to emerge from the results is that *S. aviculare* is an uncommon and rarely seen species, which has reduced in number since the NZPCN status listing of 2005. It currently appears to exist mainly as single plants or small groups in a varying number of habitats within conservation areas. The exception is in South Marlborough where *S. aviculare* is common in large populations, but confined to regenerating areas in riparian reserves.

The results confirm the New Zealand Threatened Plants listing, i.e. that *S. aviculare* is an 'At-risk declining', 'sparse' and uncommon species, 'data poor' and with further decline likely. The main addition to the existing status from this research is that the 'at-risk declining' and uncommon status is highlighted as being widespread throughout New Zealand, not just in the northern North Island as noted by the NZPCN (2005). It is therefore suggested that the NZPCN (2005) listing of 'non-threatened' be updated to the 'threatened at-risk declining' category. The results also reflect that *S. aviculare* is still present, although much

less commonly, in locations documented by earlier writers such as Cockayne (1958), and Allan (1961) who highlighted the common presence of *S. aviculare* in North and South Island coastal and lowland habitats and communities.

The results from searches in Manawatū, Horowhenua and Pauatahanui coupled with the observances of Fred Allen, suggest that *S. laciniatum* is the dominant species in those areas, not *S. aviculare*, and the question of whether any earlier reports of *S. aviculare* in the area were actually *S. laciniatum* is suggested as part of further research. This further suggests that *S. aviculare* is rare in those areas, as also indicated in the northern and eastern North Island. Taken together these results may provide preliminary evidence that the decline of *S. aviculare* is occurring over the whole of New Zealand, not just the northern North Island.

The survey results have provided useful and valid data from the observances of experienced field staff and ecologists, but some interpretative flaws are highlighted when presented. Question one was straight-forward and provided useful data on the general occurrence of *S. aviculare* within the DOC informant's conservation area. The results indicate that *S. aviculare* does not commonly occur in the conservation areas surveyed, being observed in the majority of surveys irregularly and rarely (<10%) on journeys into the conservation areas. Question two was interpreted by the informants in the manner as planned, and provided some useful insights into the habitats where *S. aviculare* is found within each conservation area. The results suggest that *S. aviculare* is found less commonly in all habitats within each conservation area, and appears to relate to and reinforce the overall general view indicted by Q.1, of *S. aviculare* being an uncommon species within the conservation areas surveyed, even in disturbed and open habitats. While the survey results have provided useful data from the observances of experienced field staff and ecologists, some interpretative flaws present when tabulating the results. These appear to relate to the use of the descriptions. For example where *S. aviculare* is observed only rarely in canopy gaps (Q.2b), it is not clear whether that means it is more commonly observed in other habitats. The results on the survey sheets are clear in their reporting of common, uncommon, and rare appearance in each habitat, but have proven to be less clear when presented in table form.

This is further highlighted in Q.3, which while being answered well by the informants, the question presented difficulties in tabulating and interpretation, as it was structured in a way that did not make it easy to compare with Q.2, which was the intention. This arises from the survey wording where common, uncommon, rare, single plants, small groupings and populations are not directly related to a habitat in which the occurrence was observed. This again was not apparent on the form, but became apparent when being tabularised. However the results are still indicative of the proportions of plants occurring within different habitats. Q.4 four was straight-forward and simple, and the results indicate that *S. laciniatum* is not found in the majority of conservation areas surveyed.

The replies from Q.2 b) and c) and the uncommon and rare occurrence of single plants at Q.3 differ from the author's observances and records from the study areas in the Bay of Plenty and Waikato (refer chapter 3), where single plants and small groups predominantly occur, and tree falls/canopy gaps and forest edges are the predominant habitats where *S. aviculare* is found. This may relate to other areas simply being different, or interpretive flaws in the survey questions. However the dominant theme is still rarity of the species in all habitats, and the overarching aim of the limited scope of the research in this area, which was to provide data to further the knowledge base of the species, and thereby provide an indication of the confidence level of the Threatened Plant listing, has been achieved. It has not been possible within the scope of this survey to gain an accurate population level of mature individual plants; however based on the population numbers of the plants studied in the Bay of Plenty and Waikato (chapter 3), coupled with the survey results, the 'At-risk' category is considered accurate, although *S. aviculare* may be at the high end of risk and it is suggested that *S. aviculare* falls into the category of 'Declining a)' rather than b), c) or d), as populations appear to be at the lower scale of category 'Declining a)'.

The results from other chapters of this research suggest that bird dispersal may not be as widespread or as dominant a dispersal mechanism as it once was, and taken together may explain the non-appearance of *S. aviculare* in areas outside of conservation reserves. For example although genetic invariance results (chapter 4) suggest that bird dispersal was wide spread when forest was contiguous, the seed bank data (chapter 3), while being considered sufficient for the species to

remain viable, are at the low end of viability. In addition the documented large scale reduction in populations of kererū throughout the North Island allied with a DOC classification of this bird in Te Urewera National park as 'in gradual decline' (Lyver et al. 2009) may be an important factor in a reduced spread of *S. aviculare*. Kererū are rarely seen in areas outside of intact forest, and small bird species that predate and spread *S. aviculare* seed are generally localised within forest habitats, contributing to a lack of wide spread dispersal. Other factors include grazing, weed infestations such as kikuyu (*Pennisetum clandestinum*) and weed clearance measures such as herbicide spraying, all of which are possibly preventing the natural spread and establishment of *S. aviculare* outside of intact forest reserve areas.

# PART B: CULTURAL STATUS

## 5.5 INTRODUCTION TO THE MĀORI WORLD VIEW

Culture as defined by the Collins English Dictionary is "the total of the inherited ideas, beliefs, values, knowledge, range of activities and ideas of a group of people with shared traditions, transmitted and reinforced by members of the group; and which may constitute social action". The DOC's General Policy and NZBS (2000) do provide a basis for recognition of, and active complementary practice of traditional mātauranga Māori (Māori traditional knowledge) and koiora Māori (Māori biological knowledge) alongside modern science in addressing conservation management, particularly where taxa of cultural importance to tangata whenua may be endangered. The intention is that effective partnerships with tangata whenua can achieve enhanced conservation of natural resources, historical and cultural heritage.

An important interface in this relationship is the differing interpretation of objectives, values, and interpretation of 'endangered' as defined by the Act, as well as conservation practices required in rectifying a species decline. In part due to Eurocentric colonial interpretation of tangata whenua concepts (Lyver et al. 2008), which the General Policy attempts to address, as the epistemologies of politics, science and mātauranga are very different (Moller 2009). European

society, including the scientific community, has often discounted the importance of traditional mātauranga knowledge, or attempted to integrate only those components of traditional knowledge perceived by them as relevant (Hepi et al. 2007; Lyver 2009), even though modern society and its science has roots in traditional cultures (Given & Harris 1994).

Mātauranga Māori coupled with wānanga Māori (metaphysical knowledge) is a complete understanding of the world in its entirety (Roberts & Wills 1998). For this reason many authors have argued that it should not be an adjunct or assimilated into modern scientific knowledge, but included in its totality alongside science to provide effective collaborative and integrated co-management policies and practices to advance sustainability, biodiversity and conservation goals (Harmsworth 2003; Moller et al. 2004; PCE 2004; Ngati Awa/DOC Joint Management Committee 2005; Beyschlag & Ryel 2007; Lyver et al. 2008; Lyver et al. 2009; Moller 2009; Moller et al. 2009). Successful collaborative research has often resulted from the fact that it was primarily based on the terms set out by tangata whenua in which both tangata whenua and scientists are aware of and recognise the strengths in each other's world views, and enter into meaningful relationships (PCE 2004).

As with all documented indigenous knowledge systems, many of the underlying attributes of Mātauranga Māori can be considered similar to those of modern science e.g. empirical observations, experimentation/practice, research, and formulation of explanatory theories, as a basis for understanding (Roberts 1998; Harmsworth 2003). Through the passage of time observations and experiences were assembled, tested and adapted, and it can be argued that the use of traditional knowledge can be likened to adaptive management, capable of actively responding to environmental signals (Moller et al. 2004). However traditional mātauranga Māori is also a qualitative ontological world view where everything is interrelated, not separated, and springs from a common source. Considered from an indigenous perspective the universe is holistic and not a closed system, mātauranga includes knowledge and understanding of all phenomena both visible and invisible, which interpenetrate each other within the universe (Figure 5.2) (Best 1903; Irwin 1984; NZBS 2000; Harmsworth 2008). Mātauranga koiora/ngahere (knowledge/lore and biology of forests) includes the everyday

knowledge based on empirical observations of climate, seasonality changes, ecosystem interrelationships, and responses of species and resources (Haami & Roberts 2002), encompassing a number of qualitative concepts and practices pertaining to kaupapa Māori (philosophy and practice of being Māori), including and involving kaitiakitanga, mauri, wairua, whakapapa, tikanga, mana, tapu and kawa (Appendix 1).


**Figure 5.2: Representative outline of Māori ontological holistic cosmological world view, highlighting that the realms are not closed systems but are interpenetrative of each other continuously.** (Irwin 1984)

Tangata whenua conservation responsibilities are embodied in the ethic of kaitiakitanga, which as exercised by tangata whenua, embraces all ancestral lands, water, sites, resources and other taonga within the tribal area or rohe. Maintaining the mauri of their tribal resources was the most important responsibility of kaitiaki, and if and where it was depleted to restore it to its usual state of well being. Mauri is an immaterial life principle or force that is possessed by and permeates all things of the natural environment whether animate or inanimate, without which an entity has no life, and is able to be 'sensed' by the elements of the natural world (atua). Mauri can be subsequently

enhanced or diminished (Best 1922; Best 1954; NZBS 2000; Clarke 2007; Lyver et al. 2008; Lyver et al. 2009). Wairua is also possessed by all things of the natural world, considered a vital sentient spirit without which no entity can possess form. Although wairua is most generally concerned with human beings, equated with the soul, leaving the body at death, wairua is considered to be within all phenomena emanating from creation, including trees, animals, water, and inanimate objects such as rocks. Wairua, although an immaterial phenomenon can also be partially material, is active and transferable, capable of maintaining interactions within and between the human world, natural and spiritual realms. Wairua can be equated with 'spirit' or energy capable of being interpreted, and is often considered in association with mauri, as decline in mauri can lead to death, and the wairua leaving a physical entity (Best 1922; Best 1954; Moon 2003).

Whakapapa relates as a genealogical record connecting and tracing the origins and relationships of all phenomena from the primeval cosmological 'parent' (IO) and state of potential (Te Korekore), through becoming and expansion (Te Po), to creation of the primal parents (Ranginui & Papatuanuku) and release of potential through their children (nga atua), followed by the development from nga atua of all life forms, to the present day (Te Ao Marama) (Figure 5.3). Whakapapa is not considered to be a linear progression but more of a circular and cyclical whole, where both the present, past and future exist within the participating moment (Irwin 1984).

Whakapapa reveals no distinction or break between the spiritual and material worlds, and that because all things descend from the gods (nga atua) and are related, they all contain a shared life force or mauri. The close relationship between humans and the rest of the world encourages a Māori world view in which the environment is personified and understood in terms of human values and behaviours such as respect and reciprocity. These values form the basis of kaitiakitanga and equate with similar values between people (manaaki/care & awhi/nurturing), all of which preclude any unrestrained exploitation of resources (Haami & Roberts 2002).

**Stages in the creation of the Universe and all things on Earth**

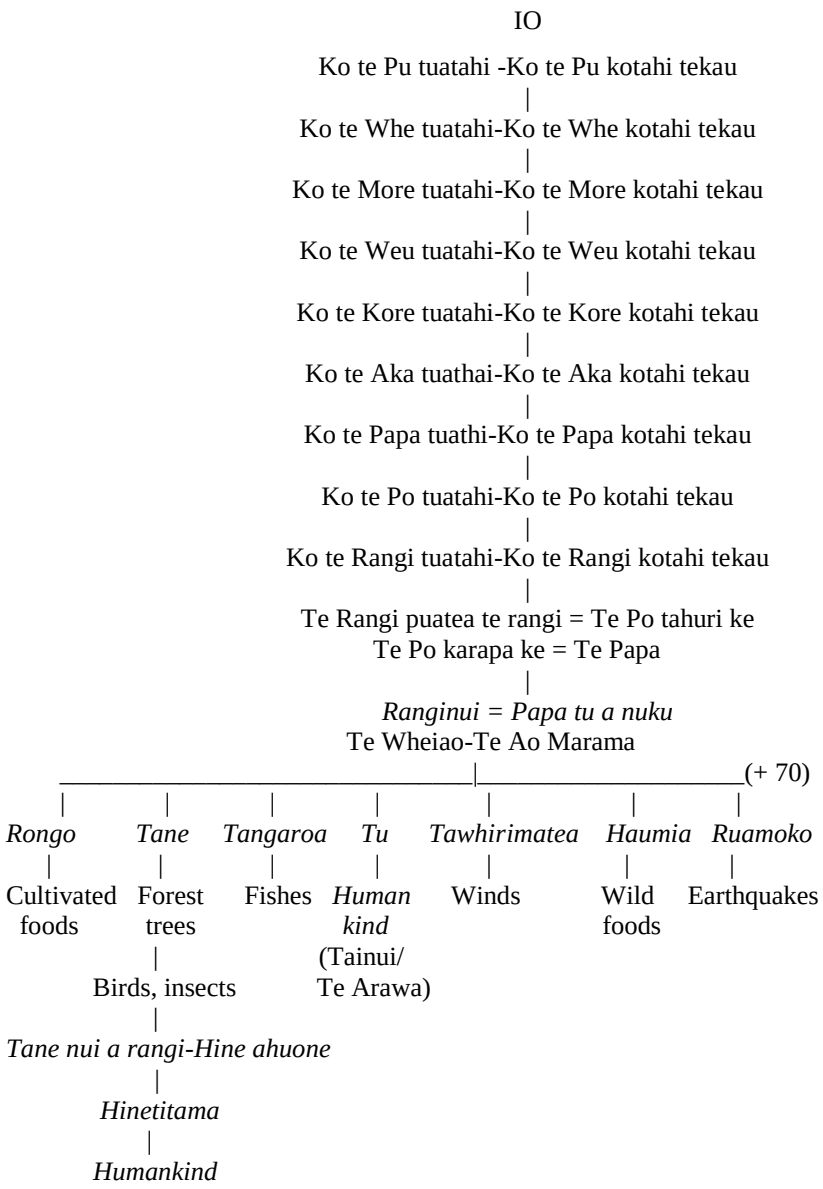


**Figure 5.3: Representation of cosmological origin and connectivity of all phenomena as whakapapa lineage (Mataatua). Emphasising the source of all things above and below the world of Man, the number of periods (generations) until the emergence of the world of Light, birth of Ranginui and Papatuanuku and subsequent development of Man and all things from the children (atua) of Ranginui and Papatuanuku; and that there is no end to mātauranga Māori in as much as the ancestors (atua) who were parted from the primeval parents, and their children, still exist above and below the world of Man. Note the complexity of descent, personification of elements and differing numerous developmental stages. Italics denote atua personifications and human beings.** (Adapted from Best 1896 & Roberts et al. 2004)

Thus kaitiakitanga is a concept grounded in principles, values and beliefs. Those who practice kaitiakitanga (the kaitiaki) inherit their role from nga atua, and thence through whakapapa to tīpuna and, if chosen, to themselves. The root word

is tiaki which describes the understanding of a resource and its indicators, and the close association with, and care and protection of a resource. Kai denotes the agent by which tiaki is performed; therefore kaitiaki is a person (or agent not necessarily human) that performs guardianship tasks, with the mana (spiritual authority/control) required. Kaitiakitanga is the active practice of the guardianship, and is a duty of Māori who act in accordance with their tikanga and kawa. Tikanga can be described as cultural principles in practice, including tapu (restriction), that are observed to maintain the mauri of the world around. Kawa are the protocols relating to successful application of tikanga (Mead 2006). The observance of tikanga and kawa form the basis of kaitiakitanga, again emphasising the circular nature of tangata whenua conceptual thought.

This aims of this section of the thesis are to utilise tangata whenua information from surveys, informal personal communications, formal interviews, and published sources to explore and determine the current conservation, cultural and rongoā status of poroporo, discuss management and enhancement procedures for poroporo from a tangata whenua perspective, determine if conservation decline is related to cultural decline of *S. aviculare* and forward collaborative practices which if implemented may help reverse the ecological decline of this species.

## 5.6 Methodology for investigating mātauranga concerning poroporo

### 5.6.1 Interviews and surveys

The formal interviews for this research were undertaken over a five month period and the informal over a 12 month period. Informal interviews consisted of kōrero with the participant as and when the opportunity arose at various hui, tangi and other social occasions. All the participants were tangata whenua of the eastern Bay of Plenty, and had grown up utilising and working in and with the forests, and so had an intimate knowledge of their rohe. None of these participants were interested in taking part in a formal interview process, and did not want their names used, although they were happy for their information to be used in the author's research. The importance of respecting the participant's wishes was fundamental, and having the privilege of their kōrero, and being able

to use it in my research is the reason behind the twelve anonymous personal communications.

The formal interviews took place with four people who are acknowledged as having expertise in their particular fields, two with rongoā knowledge, one with ngahere rākau (forest and tree) and rongoā knowledge, and one of tribal history (Table 5.4). The interviews primarily focussed on their personal experiences of poroporo and included referral to experiences and knowledge passed to them by their tīpuna (ancestors) and elders (kaumātua), and was not given on behalf of their hapū or iwi. Tribal affiliations are noted in Table 5.4 to indicate the rohe from which the knowledge is derived. The participants lived predominantly in the eastern and central Bay of Plenty regions, therefore it is accepted that the knowledge may not reflect the use of, and mātauranga pertaining to poroporo in other areas in New Zealand.

**Table 5.4: Research participants: name, age range, regional affiliations and relationship with use and mātauranga of poroporo.**

| Name | Age range | Tribal affiliation | Relationship to use of poroporo |
|---|---|---|---|
| Korotau PehoTamiana | 60-80 | Tūhoe | Tohunga rākau |
| Deborah Huia Heke-Kaiawha | 40-60 | Tauranga Moana | Tohunga rongoā |
| Diana Wikitoria Tupe | 40-60 | Tūhoe ki Waiohau | Rongoā practitioner |
| Mandy Home | 20-40 | Ngai Tahu ki Waitaki/Temuka | Iwi environmental liaison |
| John Waaka | 40-60 | Tūhourangi | Iwi leader |
| Anonymous (12) | 40-80 | Tūhoe/Te Whānau a Apanui/Ngati Awa/ | Forest dwellers |

Survey forms were forwarded via e mail to two individuals, following a request by the author for help locating poroporo and associated mātauranga at a hui of Māori environmental managers at Onuku marae, Akaroa, South Island. One of these persons forwarded the form onto a number of whanau and hapū within their iwi rohe. The form was the same as that sent to the DOC field staff, with the exclusion of question four and the inclusion of three further questions. One

asked whether purple or orange berried species occur, and two asked about the rongoā pertaining to poroporo (Appendix 7). Only one reply was received (M. Home).

The same questions were used with all participants, and were approved by the University of Waikato Ethics Committee (10/02/2009). The interviews were undertaken between February 2009 and June 2009. All participants, except one were unknown at the time to the interviewer, being introduced by third party referral. Two interviews were set up by third parties without telephone introduction, and two with an introductory phone call. These were followed by a letter of introduction including a participant information form, consent form and list of proposed questions for the participant to read before the interview (Appendix 8). The locations of the interviews were chosen by the participants and took place as a form of discussion/kōrero in which the interviewer would introduce the topic and discuss the questions he wished to be answered. The participant was then given time to kōrero as they wished about poroporo, while at appropriate times the interviewer introduced the questions. This allowed for a relaxed conversation in which many varied but related topics were discussed, including mutually shared knowledge and whakapapa so as to increase the understanding of each other. Family members if present were included to tautoko (support) the participant, and add kōrero to the conversation if they so wished.

The interviews were not taped so as to keep formalities of the occasion to a minimum, although participants stated that they were not averse to recording being done; instead notes were taken and annotated to each written question. Direct quotes were avoided so as to attempt to ensure that no misinterpretations took place. Following each interview the conversation was written up and each participant provided with a copy of the script along with an invitation to comment, and make any changes to the dialogue that they considered was a more accurate interpretation of their kōrero. These conversations were then scrutinised for common themes as well as any other information concerning poroporo.

5.6.2 Published material

A search of published material pertaining to tikanga and mātauranga Māori of poroporo was conducted through the library and data bases at the University of Waikato and wider on line searches and referenced against the oral knowledge and nomenclature provided by the participants.

## 5.7 Results

The five formal participants and twelve informal participants provided some interesting information with regard to the current conservation status of poroporo, aspects relating to its decline and preservation, and traditional and extant knowledge concerning rongoā practices. Certain key themes that emerged are summarised in Table 5.5.

**Table 5.5: Emergent themes from interviews and surveys.**

| | **Key theme** |
|---|---|
| 1 | *S. aviculare* was unknown by three participants as poroporo |
| 2 | Other *Solanum* species e.g. *Solanum nigrum*, were mainly known as poroporo rather than *S. aviculare* |
| 3 | Poroporo was no longer in regular use as a rongoā or kai by the majority of informants |
| 4 | Knowledge of poroporo preparation and use was known by few people |
| 5 | Many only knew of the historic use of poroporo |
| 6 | Loss of poroporo was related to raupatu and land clearances |

5.7.1 Mātauranga associated with presence and abundance of poroporo

Two of the formal participants and one of the informal participants did not know *S. aviculare* (orange berried species) as poroporo, having not seen it and having no knowledge of it; they only knew a purple berried species (*S. americanum* or *S. nigrum*) as poroporo. The remaining participants knew of the orange berried species (*S. aviculare*) and the purple berried species, both of which they called poroporo. Many of those informants had never seen *S. aviculare* and only knew

of it from kōrero, the remainder had seen it in their youth but considered that it was no longer present or common. The purple berried species were identified as being the one with shiny, glossy berries (*S. americanum* & *S. nigrum*?), or with dull purple berries (*S. nodiflorum*?). Participants from Te Whānau a Apanui noted that both purple species had occurred in their rohe in conjunction with orange berried poroporo, but noted the glossy purple one was not seen much anymore. Diana Tupe noted that the orange *S. aviculare* was unknown in her rohe, and poroporo was known only as a purple berried species, which she identified as most likely *S. americanum*. It was considered to have been in the rohe for many generations, and also thought not to be as common as it once was.

Mandy Home in reply to the survey stated that within te rohe o Waitaki/Oamaru/Herbert Forest that *S. aviculare* Q.1 was uncommon (<50%), Q.2 was observed in open areas outside of intact forest and other areas, with comments: " Stripped forest land and coastal lands plus road side verges", Q. 3 when observed occurred as single plants or small groupings, along with Q. 4 purple berried species. Mandy also noted that *S. aviculare* had disappeared from South Canterbury, and around her home in Temuka, since the 1940's. Korotau Tamiana identified that *S. aviculare* was not primarily known as poroporo within Ruātoki rohe, but at least five other various *Solanum* species were known as poroporo, including two purple berried ones, but he was unsure about the species the author was referring to. When a single *S. aviculare* plant was located at the head of the Ruātoki Valley by the author, he stated that he knew now which plant the author was referring to, and where they were located. He knew of plants at Ruatahuna, Waikaremoana and Maungapōhatu, where he considered they were uncommon (<50%), not rare but scarce, existing mainly as single plants, and small groups. Deborah Heke-Kaiawha stated that *S. aviculare* was common on her whanau home block which was a rongoā reserve, but observed that generally it was rarely found now days in the wider Tauranga Moana rohe. All participants considered that raupatu (confiscation) of traditional lands, subsequent clearances and development, coupled with grazing pressures and weed spraying were probable reasons behind the decline of orange berried poroporo.

### 5.7.2 Rongoā use

Discussions involving the historical uses and future potential for commercial exploitation of poroporo arose in the majority of informal and formal interviews. None of the contributors were comfortable with commercial exploitation of rongoā. The general theme was that traditionally rongoā was a personal practice, which essentially included collection, preparation, and contact with the forest environment as part of a personal holistic healing process. Following further discussion around the supply of rongoā to people that were not now able to collect and prepare a rongoā themselves, yet to whom its use would be advantageous, the general feeling was that while making rongoā available to others was worthy, to do so required that it was not commercial in manner, and control was retained within whanau and hapū, not with iwi or any outside persons or organisations.

Of the formal participants, Diana Tupe and John Waaka stated that *S. aviculare* as poroporo was not known by them or used as a rongoā of any kind within their rohe or rōpū (group). John Waaka acknowledged it may have been known to his people in the past. He further stated that the eruption of Tarawera in 1886 had removed not only any physical evidence of māra (gardens) around Tarawera Pā sites, but the loss of lives may have contributed to loss of mātauranga concerning plants and their uses. Mandy Home noted in survey Q. 6 and Q.7 that poroporo had been known traditionally within women that *S. aviculare* was an alternative similar to the pill/contraceptive, but since its disappearance from the rohe it was no longer used.

Korotau Tamiana's father had been a Tohunga rongoā in Tūhoe Tuawhenua (hinterland/interior) areas of Maungapōhatu and Ruatahuna, and he had acted as a collector under instruction from his father when he was a young man. He related that he had never once been instructed to collect for rongoā use the plant he now knew as poroporo (*S. aviculare)*, and therefore did not know of its rongoā uses. In discussion surrounding this information he considered that as a number of plants he had collected were used for similar treatments as poroporo, quite possibly it had not been used due to the other plants doing the same job, and which may have been more prevalent than poroporo in the rohe. He also

suggested that as other areas such as lowland Tūhoe had been known to use poroporo for rongoā purposes, perhaps its use had been confined within Tūhoe to those that lived closer to the coast and lowland forests, rather than inland.

Deborah Heke-Kaiawha was the only participant who knew of the range of poroporo rongoā uses, and still utilised it. She had been trained as a rongoā practitioner by her father, now deceased, who had spent much of his life protecting and ensuring that their whanau land remained in forest cover for use as a rongoā resource. She knew that poroporo had been used orally for contraception purposes, but that use had not been practised within her whanau. She used it for joint pain and some skin disorders, but only externally never internally. The main use for joint pain was where ripe berries were crushed and placed on the afflicted area, held in place with a bandage, and changed at various intervals. The leaves could be used in skin treatments as a bath, but had never been utilised much by her whanau.

All of the informal participants bar one knew of the rongoā uses of poroporo, and a number knew also that the purple berried species could be and were used for similar rongoā uses as *S. aviculare*. These uses were for contraception, skin disorders and joint pain, but no one knew of anyone still using it, or of the exact process in preparing and use of the rongoā.

### 5.7.3 Other uses

The participants who knew of *S. aviculare* as poroporo recognised the berries were edible, although the majority had not consumed them. All of the participants recognised that the purple berried species were used for kai, with the berries being consumed, and a number used the leaves as a vegetable, although the name raupeti was not known. Mandy Home said that in her rohe the purple berried species were more readily eaten than *S. aviculare* due to the *S. aviculare* berries being bitter. She also noted that a Pākehā lady she knew made jam out of the purple berries. Diana Tupe stated that the purple berried species, when abundant was eaten readily as a kai tamariki, usually on the way to school. John Waaka documented that the purple berried species were used regularly as a

vegetable, although without the name raupeti. None of the participants knew of any current or historical cultural uses of *S. aviculare*.

### 5.7.4 Reasons for decline

Discussions with the participants concerning their views on the decline of poroporo related without exception that raupatu (confiscation) was the most likely primary cause. The subsequent development for farmland and roads necessitates clearance of vegetation, maintenance with chemical spraying and animal grazing. Hence the āhua (appearance/condition/essence) of the land was changed, and therefore became unsuitable for growth of the species. Mandy Home noted that the agriculture in South Canterbury was considered to be a major cause of poroporo decline. Korotau Tamiana in further more in-depth discussions regarding the decline, suggested that an aspect of mātauranga Māori understanding (mōhio/māramatanga) was that the intimate relationship Māori had with nature (through a spiritual link with atua) meant that nature had provided all things that Māori needed for their use, but if those things were no longer required or used by Māori then nature noticed and took them away; nature sensing that Māori no longer needed them. So in addition to raupatu and land clearances earlier in the century he considered the decline in use and knowledge of poroporo was a contributing factor in the continued decline of the species. As a result the mauri of the plant is being taken back by nature, along with a decline in its wairua, hence the plant is suffering ecological decline.

An example of this belief in practice was given in regard to a ceremony undertaken by his father and elders that he had observed in youth. Following the passing of laws in the early part of the last century prohibiting he harvesting of kererū the relationship with this species was interrupted leading to a decline in tikanga (traditional practices) promoting the well being of this species. So the mauri of the kererū was given back to nature, until humans renewed their use of kererū again. This has been recorded by Lyver et al. (2008) and Lyver et al. (2009), who suggested that a decline in tūturu (real/authentic/fixed) Māori practices was associated with a subsequent decline in a species such as kererū, and this was possibly due to the colonisation process taking away the mana (authority) over kererū from Tūhoe. Hence recovery of the species was seen as

being directly related to a return of mana over the land, whereby traditional harvesting practices would signal to nature that the species was still valued and needed, and therefore the species would again become abundant.

The issue of wairua in regard to species maintenance was discussed with Deborah Heke-Kaiawha. The author having observed when taking measurements and leaf samples of poroporo plants on her whanau land block, that one particular plant, which her nephew described as being the one that was harvested, and originally close to where the old homestead once stood, was much older than any other poroporo plant observed within the research. It was hidden inside other vegetation, and on closer inspection consisted of multiple large, in width and length, branches that were estimated by the writer, based on observances and ring analysis of other mature plants at Tarawera (refer chapter 3), to be potentially >20 years of age; an unheard of age for the species.

In discussion with Deborah I mentioned this particular plant and suggested that there appeared to be a wairua associated with its state of longevity, and thought that this may have been associated with aroha (love), care and associated karakia (ritual incantation/prayer) associated with the use of this plant when undertaking harvest. She believed that the plant in question was to her knowledge a minimum of 25 years of age, and quite possibly up to 35 years of age, and that it was the original plant used for rongoā harvest over the years. She further explained that the wairua was connected with her father (a tiaki or guardian) who had dedicated his life to saving the land from being sold or developed, instead retaining it as a natural resource especially for rongoā (kaitiakitanga). The wairua was the key reason for the health and abundant growth observed within the block, rather than simply science based management practices of pest control and species enhancement. The mana of the land still remained with the whanau, with no diminishment of her father's wairua, thereby maintaining and enhancing all species wairua and mauri. In contrast other lands had lost their mana by being sold or confiscated, with subsequent reduction in species mauri and wairua; hence the diversity of native species was no longer present on those lands.

## 5.8 Discussion

5.8.1 Mātauranga associated with presence and abundance of poroporo

The results demonstrate that any mātauranga associated with *S. aviculare* as poroporo is scarce among Māori alive today. Reasons for this scarcity are considered to be associated with the decline in availability of poroporo which has taken place during their lifetimes, equating to the time period associated with land clearances. Many of the participants had observed poroporo in their youth, albeit not commonly, but have no longer observed it in their adult lives. In areas where *S. aviculare* was documented as being found and common in the 19$^{th}$ and early 20$^{th}$ centuries, (refer chapter 1) participants now have either little or no knowledge of the plant growing there, or of its uses. The causes of the loss of poroporo is considered to be directly related to removal of mana over the land due to confiscation and land clearance; correspondingly the return of the mana via return of land is also considered to be a primary factor in restoration of the species. The results have also confirmed that the name poroporo is now mainly associated with purple berried species that were either introduced by early Polynesian voyagers or post-European colonisation (refer chapter1).

The possibility of loss of traditional knowledge associated with poroporo in the Tarawera district thought to be caused by the eruption of 1886, was not known to the author before, and highlighted the tragedy that befell the people of the area. In a modern context the damage caused by the eruption highlights the potential of the increased vulnerability of *S. aviculare*, in the face of small populations and the predicted continued decline, where any changes to environmental conditions, including sudden random events such as large volcanic eruptions, could dramatically alter species survival.

5.8.2 Rongoā and other cultural uses

The use of poroporo for rongoā has virtually ceased, being maintained only in specific whanau situations particularly where land has been dedicated for rongoā purposes and poroporo still grows readily with easy access. Poroporo may have grown and been used as a rongoā in the wider areas in the past, but knowledge of it and its use has disappeared, only existing in memory. No found oral or written

record remains of its preparation, and there is no wider current use apart from occasional use of berries as kai. It appears that purple berried species have substituted for orange poroporo rongoā uses in some instances, although the former appears to have ceased as well. The use of purple berried species appears now to be only the leaves as a vegetable, use of the berries as a food has reduced with younger generations. The non-use of poroporo and knowledge of any uses in inland Tūhoe despite published information that shows a terminology for differing fruiting phases (refer chapter 2), and the common occurrence of the species in that area in the 19th Century (Colenso 1868) is interesting. Korotau Tamiana's father would have grown up and trained in rongoā at a time when poroporo was more common, and perhaps his non-use of the species highlights that the published information regarding the rongoā uses was indeed gathered from lowland and coastal areas. No cultural use in the modern era has been located, or memories of its cultural use remaining.

### 5.8.3 Reasons for decline

An overarching theme in all participant's korero, in regard to the decline, loss of knowledge, and use of poroporo, is the loss of mana associated with land being confiscated, and subsequently sold and cleared for non-traditional uses. A direct result of the loss of mana over the land is the loss of the intimate connection with nature, and hence the mātauranga and tikanga practice, due to restrictions being placed upon the use of traditional practices; such as harvesting of native species. The cycle of decline in use and knowledge of poroporo appears to have begun with the removal of people, and their connection to the land. Subsequent clearing of the land removed poroporo physically and prevented its regeneration. As traditional practices declined, poroporo was seen by nature as if it was no longer required by the people, and so nature has taken it back; hence the observed decline, similarly as documented for kererū by Lyver et al. (2009). Further decline of poroporo is the physical manifestation of the decrease in the mauri and wairua, as they are returning to nature. The wairua of the species being the interactive 'energy' that nature responds to, will also therefore be the vehicle to 'inform' nature that the species is valued once again, and a reversal of the decline begun. The decline is therefore also cyclic, not linear, and can be reversed.

The issue of personal wairua associated with a human being directly relating to the maintenance and enhancement of mauri and wairua of the land is not an unusual concept to Māori (Best 1925; Best 1954); the interrelatedness of all things being integral to an understanding of mātauranga Māori. The relationship of wairua and mauri, and the ability for the wairua of humans and nature to interact, especially through traditional vehicles such as karakia, is also an acknowledged and accepted part of mātauranga Māori (Best 1954; Irwin 1984; King et al. 2008). Lyver et al. (2008) explained it was common for people who base their actions on traditional knowledge to attribute events involving species and environmental issues to spiritual mechanisms, and the use of mātauranga techniques and indicators has been recognised as being valuable in species studies, potentially providing information regarded as reliable.

Empirical evidence is very hard to obtain in attempting to understand the age of the plant on the Heke-Kaiawha block without the addition of core sampling, and this is suggested for further research, if acceptable to the whanau. However based on the knowledge and observances of Deborah who lived, and practiced on the Heke-Kaiawha land block, wairua has an effect on the physical plant by producing a specimen that is 3-4 times the normally accepted age of seven to ten years for *S. aviculare* in a natural environment (Gerasimenko 1971; Wardle 1991; Symon 1994).

The results show the importance and the central nature of mātauranga concepts, especially wairua and mauri, to the Māori world view relating to conservation and biodiversity. The acknowledgement by Government and collaborative researchers of the value of these concepts being incorporated in conservation planning and practice is reinforced by the research results. The plant documented on the Heke-Kaiawha rongoā reserve highlights the inter relationship that is possible between plants, nature and humans within traditional practice. The philosophy and practice of kaitiakitanga on the land, is the central vehicle for the functioning of the relationship that has produced a plant surviving to such an advanced age, while still being viable. The mauri and wairua of the plant has been maintained and enhanced through use, manaaki and karakia, by kaitiaki; the personal tiaki wairua acts as the connecting vehicle binding the whenua and all that dwells upon the whenua. The practices of kaitiaki which in addition to

spiritual practices now includes scientific methods of integrated pest management, bird counts and species enhancement programs, has resulted in a holistic and collaborative approach providing for sustainable management.

This example of the interpenetration and activity of wairua with the physical world demonstrates that wairua is a vehicle by which nature is able to 'take notice' of issues. Therefore enhancement of a species can begin to take place in as simple a manner as having the will to enhance, and the use of karakia to provide potential to the purpose. The physical act of restorative planting combined with karakia, will (purpose), and mātauranga insights relevant to the area, when allied with scientific practices such as ecotyping, acts as a vehicle or signal to 'get nature to take notice'. It then provides an integrative and collaborative solution to species loss and decline, involving the planting of large numbers of a species known to be suitable to a site, along with the assurance that they will supported by nature and returned to people. Both processes combined, providing for the strengthening and enhancement of the species mauri and wairua.

The Māori concepts as expressed by the participants have metaphysical connotations, but are also equated with practical values, in that an everyday meaning is also incorporated. This is exemplified by mana, mauri, wairua and karakia, which while acknowledged and understood as metaphysical concepts emanating from the divine, also include everyday practical understandings of that knowledge. More simply they can be interpreted as the need to pay respect to all things of the natural world, including how one operates within it (Patterson 1992). This interpretation of metaphysical concepts in a practical manner, can allow science and traditional thought and practices to interface, where a mutual basis of awareness and recognition of the strengths in each other's world views is undertaken by entering into meaningful relationships, without necessarily completely sharing in each other's beliefs (PCE 2004); the ecological problem attempting to be solved being the overarching concern.

A practicality associated with these metaphysically based concepts can be further exemplified by the fact that return of land, therefore mana, may not guarantee or equate to return of species that were lost, as modern requirements in the use of

land often provided by legislation such as Te Ture Whenua Act 1993, will not always be conducive to traditional practices predominating. Therefore in many situations Māori will have to make practical choices that may not include provision for their land to be returned to forest and traditional use. The inclusion of mana whenua holders in collaborative decision making processes for all lands deemed for conservation and restoration may be a trade off for the incorporation of traditional concepts.

Whakapapa is a construct utilised by Māori to encapsulate their world view. Among its many functions it acts as a taxonomy of all things of value to Māori. When acting as a taxonomy whakapapa group's plants, animals and other phenomena perceived as being related according to the ecosystem or habitat in which they occur (Roberts et al. 2004). In addition whakapapa seeks to identify the ancestral origins of each species from Ranginui and Papatuanuku, via their many offspring. Whakapapa thus acts as genealogies and taxonomies (systems of classification) similar but analogous to scientific phylogenies which also seek to assign a common ancestor to each lineage, as well as group "like" species with "like". However, whakapapa relationships and groupings are based on habitat and other aspects e.g. utility and cultural reasons (Roberts et al. 2004), whereas scientific phylogenies seek to identify and group things according to their genetic relationships, emphasising the connections of that plant via genera and species to the rest of the world (Papatuanuku). While the underpinning philosophies and theories upon which each approach (whakapapa and phylogeny) is based may differ, both share the same belief that all living organisms are related and each major lineage descends ultimately from a common ancestor.

Attempts to locate published material or elicit an unpublished whakapapa of poroporo, were however, unsuccessful. This may suggest that it was not regarded as a particularly important resource or more simply that this knowledge has been lost. It is recommended however that more research be conducted into the mātauranga of rongoā plants in general on the basis that there may be a whakapapa which is inclusive of a number of different rongoā species including poroporo**.** For plants such as *S. aviculare* that have threatened status in overseas areas where they also have originated (Australia), the use of common dialogue and indigenous knowledge potentially may advance the species survival in both

Australia/New Guinea, and New Zealand. Therefore the use of whakapapa, systematics and scientific nomenclature can be complimentary, and may be a valuable starting off point for collaborative relationships.

## 5.9 Conclusions and recommendations

5.9.1 Conservation

The surveys undertaken among DOC field staff, scientists/ecologists and interviews with Māori confirm that the current status of *S. aviculare* in the Threatened and Uncommon Plants list (de Lange et al. 2008) as 'At Risk, Declining', 'sparse' and 'data poor' is warranted. They suggest that *S. aviculare* is a plant that is generally found uncommonly in conservation areas and in low numbers, particularly, as noted by the NZPCN (2005), in the northern half of the North Island, and rarely outside of those conservation reserves. The results reinforce that *S. aviculare* most likely falls into the category of Declining a). The national population if extrapolated from results in this research appears at a minimum to fit the population size of 'Declining a'), with *S. aviculare* appearing in moderate rather than large populations, and mainly as either single plants or small groups, with an on-going predicted population decline. This research's results further suggest the possibility that populations of *S. aviculare* may even be lower, less wide spread, and not regenerating as much as than the current category suggests.

The observance of *S. laciniatum* in Manawatū and Wellington rather than *S. aviculare* indicates that plants reported in those districts for the Threatened Plant list may actually be *S. laciniatum* not *S. aviculare*, further indicating a decline in *S. aviculare* particularly in the lower North Island. Identification surveys are recommended to be undertaken in the lower North Island to confirm whether *S. aviculare* or *S. laciniatum* is prevalent. The inclusion of *S. laciniatum* in future Threatened Plant lists appears warranted from the results, therefore it is recommended to include *S. laciniatum*, with *S. aviculare* and var. *latifolium* in future listings. While observances made by the author located *S. laciniatum* in >20+ population sizes in the Manawatū, and Wellington Districts, no surveys were returned from those areas. Replies from other districts suggest that the

species is not prevalent, and may therefore warrant the inclusion in the list. The inclusion of *S. aviculare* in the category 'threatened overseas' suggests that collaboration and information exchange with Australian ecologists may provide insight into control measures that will help reverse the predicted decline in both countries, particularly involving utilisation of local Traditional knowledge.

A further restoration Recommendation Category is suggested to be included as an appendix to the Threatened Plants list. Based on the information gathered by those with expertise it is hoped that this will provide accurate guidance and confidence to authorities, organisations and individuals, not just as to the appropriate plants to use in restoration, but also that the plantings undertaken are of threatened indigenous species so that their use in restoration plantings may help to enhance the biodiversity of wild populations.

It is noted however, that the interpretation of 'threatened' may be completely different when cultural aspects are included. While the quantitative scientific data points to *S. aviculare* being 'at-risk' rather than 'nationally endangered' or 'vulnerable'; the status of this plant as viewed by tangata whenua can be considered qualitatively to be either 'nationally critical', or even in some areas 'extinct'. This conclusion is based on the interviews which suggest that while tangata whenua who are regularly in the forest environment do not consider *S. aviculare* to be necessarily critically endangered (5.7.1), a majority of Māori do not live in such areas, and so with *S. aviculare* being confined generally to reserve areas which require permits to harvest, coupled with the reduction in knowledge of the species, these people consider *S. aviculare* to be critically endangered, or even extinct. While those issues may not represent how the Threatened Plant list is complied, it may be relevant for tangata whenua aspects to be included as a section in the new Recommendation Category so as to assist authorities to choose species for restoration projects based on bicultural not just monocultural perspectives. This new section therefore would recommend plants that are threatened from a cultural perspective, be compiled by tangata whenua along with scientists and ecologists, to provide a guide for restoration programs.

Evidence suggests that *S. aviculare* and *S. laciniatum* are especially vulnerable to grazing from ungulate species such as deer, goats and cattle, and control

measures are recommended to be maintained and increased where animal populations are high. Population surveys are recommended to be undertaken prior to the next Threatened and Uncommon List publication, to identify the accuracy of population numbers, and whether population numbers of *S. aviculare* and *S. laciniatum* are lower than 'Declining a'). Restoration programs for *S. aviculare* and *S. laciniatum* are recommended to be undertaken, and advanced throughout the North and South Islands. Both species are easy to propagate from seed in large numbers, and are easy to maintain with pruning in park situations if desired. They tolerate a wide range of environmental conditions, being able to be planted from coastal dune areas, as is being currently undertaken by Environment Bay of Plenty Coast Care, to inland lowland forest areas throughout the majority of the North and South Islands. It is recommended that the species are positively identified and not planted close together due to the potential production of non-viable seed where they cross pollinate (Baylis 1963). An example of collaborative restoration is in Rotorua, where John Waaka and Tūhourangi Tribal Authority are working with the Rotorua City Parks department in distributing and planting out locally numbers of *S. aviculare* seedlings grown from the Tarawera study areas by the author.

### 5.9.2 Cultural

The decline in use of poroporo for rongoā and cultural purposes has been confirmed, being either unknown or confined to memory of historic use. Only one current practitioner was able to be located, and no cultural uses documented. The scope and time frame of this research was such that areas outside of the Bay of Plenty were only able to be investigated in a restricted manner through electronic surveys, hence it is recognised that the lack of documented current use within the Bay of Plenty may not relate to whanau in other areas. However the replies from DOC in the South Island are relatively widespread, and coupled with the reply from Mandy Home as an environmental liaison coordinator within her iwi Ngai Tahu suggests that her reporting the lack of use of poroporo in South Canterbury due its disappearance, may also generally relate to other areas in the South Island, while still acknowledging that individual whanau may utilise poroporo where it is still found. The loss of knowledge of poroporo in the Tarawera district attributed in part to being influenced by the eruption of 1886,

has highlighted the vulnerability of a species that is already in decline to random occurrences that may cause localised extinction, not only of a species itself but also of knowledge pertaining to that species. The failure to find any published records of whakapapa involving poroporo, or of any knowledge of such from the interviewees could suggest several things: one is that the plant was not considered important enough to warrant its own whakapapa (unlike the kumara for example; Roberts et al. 2004), or it could also be that such knowledge has been lost, which is true of many aspects of traditional mātauranga (M. Roberts pers. comm. 2009).

Mātauranga Māori and tangata whenua kaitiaki practices have been shown to provide enhancement to the growth and survival of *S. aviculare*, poroporo. These practices in conjunction with scientific techniques have highlighted the success that is possible with collaborative programs, especially where they are primarily based around mātauranga practices. Collaborative restoration and enhancement programs are recommended to be undertaken as a matter of course, fulfilling not only the requirements of legislation but also of the land and plants. This research has highlighted not only previous collaborative programs that have successfully operated, but documented a highly successful sustainable whanau program incorporating traditional and scientific elements, that provides evidence of how integrated kaitiakitanga practice can successfully operate. Supported by scientific assessment techniques, mātauranga can help in understanding population changes, recovery and sustainability; the evidence provided in this research is highly suggestive that mātauranga Māori kaitiaki practices are valuable to the success of environmental programs, and recommended to be incorporated in all enhancement and restoration programs. Local mātauranga and practices associated with kaitiakitanga such as karakia are recommended to be included with mana whenua in all planting programs, as the research evidence suggests that these practices are important success factors for sustainable restoration, and enhancement programs. Plantings are further recommended to be undertaken for cultural purposes in areas where access is easy, and do not require permitting for cultural harvest. These processes combined may help in reversing the ecological decline, revive cultural knowledge, and rongoā practices, including consideration of how Māori concepts can be interpreted (Patterson 1992).

Whilst tangata whenua participants were not in favour of commercial exploitation of rongoā, and the author does not recommend such, discussions around the issue of supplying rongoā to others made it clear that traditionally rongoā was also about self healing, and part of the healing was the collection and preparation of the rongoā, including the need to connect with the forest and its wairua. However, this was acknowledged as often being not possible with the decline, and location of poroporo within areas that needed permits to harvest. Further discussions suggested that controlled use by hapū and whanau collaborations may be an acceptable way of supplying rongoā to those who are not able to collect and produce it themselves. This possibility is therefore suggested for further discussion among tangata whenua, that would help in reviving traditional mātauranga, as well as contraceptive and rongoā uses for ailments such as arthritis and skin disorders that poroporo is grown for and used internationally.

Photo G. Weavers

**Poroporo (*S. laciniatum*) plant growing beside Onuku Marae, Akaroa, Banks Peninsula.**

# CHAPTER 6

# SUMMARY:

# CONCLUSIONS AND RECOMENDATIONS

## 6.1 Conclusions

The overall aims and goals of this research have been achieved: having documented the ecological dynamics and morphological characteristics, genetic diversity status, and highlighted the declined conservation and cultural status of *S. aviculare*, providing additional conservation data, forwarding some reasoning as to why the decline has taken place, and potential solutions. The results from this research highlight optimal information and solutions that may bring us somewhat closer to reversing the declining trend, and reviving not only a valuable ecological species, but also increasing and maintaining the mātauranga associated with the traditional role of poroporo. Successful restoration programs integrating scientific and tikanga based knowledge and practices underpin the likelihood of reversing the decline of the species, from both a conservation and cultural perspective. The findings of the research are summarised as follows.

6.1.1 Biology, ecology, plant dynamics, successional status, seed bank, seed viability and dispersal agents.

*Solanum aviculare* is an ancient flowering shrub, whose phylogenetic connections suggest an early radiation from a common land mass. The successional status has been determined and shown to conform to the definitions of an opportunist pioneer coloniser in earlier studies. Cohort development data showed the development and maturity of populations and indicated the possibility of population growth and development being influenced by resource availability, site precariousness and environmental exposure. Metapopulation growth structure data showed significant correlation between height and crown spread providing evidence of the position of *S. aviculare* as a seral prisere species with a rapid expansive growth strategy providing a dominant advantage for light availability and propagule dispersal, as well as ecological stability and facilitation for regenerating species. The documentation of the full cycle of

regenerative tactics provides further insight into how the species has spread and survived. Survival tactics appear to be flexible specialisations that can allow *S. aviculare* to complete a life cycle quickly, especially where disturbance conditions may be precarious. Senescence of mature plants at ± three years in low resource slip sites, with observed survival of similar aged plants for longer periods in higher resource sites such a canopy gaps, is highly suggestive of this possibility, and worthy of further research. The initial colonisation tactic of *S. aviculare* regenerating via stasis induced seed from a seed bank through germination due to soil disturbance and associated environmental conditions has been definitively identified. Soil seed bank results indicate that sufficient seeds are being supplied to the seed bank to maintain the species at current population levels; however the results were on the low side of viability, therefore may be a contributing factor in *S. aviculare* population's current moderate appearance (de Lange et al. 2009), and if further reductions in *S. aviculare* population size, and/or reductions in dispersal agents take place, it is suggested the ability for the species to maintain itself will be further compromised.

Spread of viable seed via kererū and blackbird was definitively confirmed, indicating bird endozoochory to be the most likely predominant method for seed dispersal and new population establishment. Results highlight that the viability of the bird deposited seed was not high, in comparison to controls, suggesting gut maceration in larger birds may influence seed viability, providing further indication that large seed numbers, coupled with fruit colouration, and placement for bird predation are important survival tactics for *S. aviculare* regeneration. The tetrazolium trial results were confirmed by germination trials, and revealed higher viability than the germination trials, possibly relating to stochastic variance when using soil media, further indicating the usefulness of the tetrazolium treatments for seed viability estimation.

The results of the ship rat gut passage seed viability trials confirmed earlier studies that ship rats predate and ingest *S. aviculare* and *S. laciniatum* fruits and seed, documenting a higher rate of viable seed being passed than kererū/blackbird, confirming that rat feeding and gut passage do not destroy small seeds such as *S. aviculare* readily. Extrapolated implications of these results suggest that in situations where bird distance dispersers such as kererū are

in low numbers, and adventive species such as blackbird are less predominant in deeper inland forest areas, then rats may be the predominant local dispersers of not only *S. aviculare* and *S. laciniatum*, but other frugivorous shrub species as well. Subsequent control measures being undertaken for rats may then have to be timed to ensure that seed of *S. aviculare* and other species is still being dispersed until it is known that bird numbers have risen. On the contrary where bird numbers are high, and spread of *S. aviculare* is observed, then control of rat populations is suggested prior to fruit maturation.

### 6.1.2 Seed germination.

The results confirm, in alignment with earlier studies, temporal differences in germination of stasis induced seed in spring, and fresh seed in autumn/winter, and that the species readily germinates throughout summer, autumn and winter. Although, the seed germination trials were generally inconclusive, only revealing new information suggesting preferential germination of surface scattered rather than buried stasis induced seed in spring over fresh seed; fresh seed has an apparent advantage in autumn. The overall results were lower than previous published studies, despite taking place in a commercial nursery using similar protocols. No explanation is able to be offered as to why the results were lower than expected, apart from any influence of provenance, and/or temporal collection protocols.

### 6.1.3 Flowering timing, insect damage and leaf morphology

The documentation of *S. laciniatum* flowering, from stasis replicated seed, within a 12 week period, while fresh collected *S. aviculare* seed did not flower after 36 weeks, indicates that stasis induction following seed dispersal to soil is a species strategy for early flowering, fruit set, and subsequent dispersal, possibly existing to compensate for the precarious survival situations involved with early colonisation of disturbance sites. The very close relationship of the two species, coupled with no previous studies indicating any differential in flowering, indicates the validity of the results, although further research on this phenomenon is suggested.

*Solanum aviculare* is generally considered to have higher alkaloid content that *S. laciniatum*, and the repeated stem damage to *S. aviculare* and not *S. laciniatum* is an unexplained phenomenon, unless there is a predilection to high alkaloids in the predator caterpillar species, or there is some unknown difference in the stems of the species. Research on this would be recommended if both species were considered for any further commercial exploitation.

Differences in leaf morphology between species were documented, confirming earlier studies, highlighting that the differences are not easy to define. Further additional evidence of heteroblastic leaf development was documented, documenting the glabrous nature of cotyledons, the appearance of trichomes on the hypocotyl and initial leaves of the ovoid shape to the 2$^{nd}$ node, and confirming lobed glabrous leaves appear predominantly from the 2$^{nd}$ node onward. The observed growth of axillary adult linear shaped leaves prior to inflorescence development forwarded consideration to the use of further nomenclature, primary and secondary leaf growth, to clarify documented axillary and non-axillary leaf growth differences. Coupled with studies to uncover potential forms exhibiting differing leaf shapes, the new nomenclature is suggested for further research observation and confirmation.

### 6.1.4 DNA extractions, PCR/ISSR production and restoration

The genetic status of *S. aviculare* and *S. laciniatum* and the role that may play in restoration has been documented. The extraction of high quantities of DNA from the leaves of *S. aviculare* was more consistent from kit extractions than CTAB. CTAB protocols were unable to remove RNA from the extractions, despite modifications to RNAase protocols; however the results from the PCR production indicated that RNA did not inhibit the PCR process, providing information that CTAB extractions are adequate for *S. aviculare* genetic studies where cost is a factor.

The production of ISSR's through PCR generation proved to be fraught with difficulty. Consistent, reliable and reproducible results were unable to be produced. Many attempts had to be made with the same primer and samples to produce enough banding to indicate the polymorphic or monomorphic status of

the locus. Protocol modifications, and checks on the modifications were constantly undertaken, differing experienced personnel trialled in attempts to improve consistency, but all attempts proved to be unable to provide the improved consistency desired. The overarching conclusion is that *S. aviculare* is a difficult species to produce ISSR bands from, with no current technical reasoning as to why, warranting further research.

The results identified no polymorphic loci, intra or interspecies variation on the 20 primers trialled, despite the universal primers used producing consistent polymorphic results on a wide range of indigenous taxa in the PBRL laboratory and similar sequences producing consistent polymorphic results on other *Solanum* species in previous studies. Where polymorphic loci were considered to potentially be present, continued inconsistency in band production precluded any reliable detection. However results either indicated, or definitively confirmed monomorphic loci predominate in *S. aviculare*, suggesting invariance and uniformity exists across all populations of the species, in alignment with previous studies. Invariance with *S. laciniatum* on the monomorphic loci studied suggests that *S. laciniatum* is also highly monomorphic.

The differences documented in phylogenetic studies between *S. aviculare* and *S. laciniatum* are indicated to exist on polymorphic loci, as the similarities associated with the high bootstrap values connecting them appear to be located on monomorphic loci, as revealed by this research. Polymorphic loci appear to be low in frequency, may be hidden behind dominant homozygous alleles; and any genetic variation is indicated to be determined on polymorphic loci that are low in presence, therefore difficult to detect, and may only be moderately expressed. These moderately expressed differences may be the forms or ecotypes as reported in earlier research that will require the detection of polymorphic loci to locate and document fully. This research has highlighted that monomorphic loci may be the determinants of generic level traits, and as such may be the evolutionary markers differentiating *Archaesolanum* species from other *Solanum* taxa, as described in phylogenetic studies; and further research is suggested that may add to the determination of relationships within *Solanum*. Whether the haploid or diploid status of plant material may help in the detection of

polymorphic loci, along with the possible production of primer sequences is suggested for further study.

The monomorphic results indicate that both species may not be able to readily adapt to change, and coupled with the confirmed decline in *S. aviculare* population numbers, strongly highlights that restoration programs should commence. The invariance reported, suggests that both species are able to transplant between regions for restoration purposes. However due to the known low production of viable seed from interspecies crosses, and the still unknown incidence of forms or ecotypes, it is suggested to positively identify, and plant each species separately, utilising local provenance in all restoration programs until further research is undertaken.

### 6.1.5 Conservation status and cultural status

The conservation status results confirmed the 'At Risk', 'Declining' category, and sub-categories 'sparse' and 'data poor' of *S. aviculare* in the Threatened Plants Listing, further suggesting that 'Declining a') may reflect the actual current status. The inclusion of *S. laciniatum* is suggested for future plant listings. Identification surveys, including confirmation of species identity in the lower North Island, are important to be undertaken prior to the next plant listing to confirm the actual population levels, as this research suggests that species' range is shrinking, and dispersal and regeneration levels may also be reduced.

A further Recommendation category, including species relevant to tangata whenua is suggested to be included in the next plant listing as a guide to relevant species for inclusion in restoration programs to enhance wild populations, and cultural uses, as tangata whenua may view endangered differently to science due to restricted access for plant collection, lack of easy access to plants growing close to current living areas, and loss of mātauranga regarding collection and use. Restoration programs are suggested to be important for *S. aviculare* and *S. laciniatum* in the North and South Islands. Correct identification of species is suggested prior to planting to ensure the production of viable seed is returned to the seed bank.

The decline in knowledge of and use of *S. aviculare* and *S. laciniatum* as poroporo has been confirmed, appearing in relation to the ecological decline. The ecological decline in *S. aviculare* appears to be related to removal from tangata whenua of control over their land, corresponding is the decline in its use as a rongoā, other *Solanum* species having substituted for poroporo, with even the use of those species having now ceased. Only one current rongoā practitioner was able to be located, and no cultural uses or memory of those uses found. The return of mana over the land, even as primary collaborative decision making, is considered essential to the reduction in decline and future enhancement of the species. Mātauranga Māori, spiritual concepts and associated kaitiaki traditional practices have definitively been shown to be valuable for the enhancement of species and sustainable success of conservation and restoration programs. The use of collaborative programs, mana whenua kaitiaki and their tikanga with scientific techniques, has been shown to be highly effective and is suggested to be undertaken as a matter of course. Collaborative tikanga based restoration programs, including planting poroporo in urban areas, are therefore suggested to be essential to the revival of the use of poroporo for rongoā, and the mātauranga pertaining to the species and its uses.

## 6.2 Recommendations

Research for this thesis has forwarded specific recommendations for further related study as follows:

### 6.2.1 Ecology and plant dynamics

To further the ecological knowledge base of the species research could include:

- Providing information on the flexibility of reproductive tactics in relation to precarious positions such as earth slips and reduced resources by documenting growth and senescence between plants in areas of higher resource availability such as tree fall gaps and the lower resource earth slip sites
- Whether seedling suppression, primary dormancy, or secondary dormancy is initiated due to stasis induction cues beneath *S. aviculare* canopies

- Whether *S. aviculare* has any nitrogen fixing ability

### 6.2.2 Seed viability, seed bank and dispersal agents

Further research into the viability of seed dispersal agents and seed bank could include:

- Documenting the status of both indigenous and adventive birds within forest areas where *S. aviculare* is declining and further analysing seed bank levels
- Documenting differences in seed viability from endozoochory and gut retention times between bird species, in particular between indigenous and adventive species known to ingest and spread *S. aviculare*
- Whether glykoalkaloids influence seed retention times and maceration in bird guts

### 6.2.3 Flowering timing, stem damage and leaf morphology

To increase the knowledge base in regard reproduction and plant morphology further research could include:

- Documentation of temporal flowering differences in relation to stasis induction and fresh seed, in and between *S. aviculare* and *S. laciniatum*
- Whether the repeated stem damage to *S. aviculare* and not to *S. laciniatum* is related to species stem differences or glykoalkaloid tolerance of caterpillar species
- Documentation of the stability of epidermal differences between *S. aviculare* and *S. laciniatum*
- Investigation of the basis regarding leaf shape differences on adjacent plants within a population
- Investigation of how adult axillary leaves form prior to inflorescence development and juvenile non-axillary leaves form on sub-tended post inflorescence shoots and whether they may be temporally related to fast initial vegetative growth where the shoots were initiated in the juvenile phase, producing more juvenile shaped leaves at the adult post inflorescence phase

### 6.2.4 Population genetics, monomorphism and polymorphism

Further research on the genetic status of populations and documentation of genetic diversity should include:

- Documentation of specific forms and/or ecotypes
- The genetic basis to differing leaf morphology, and forms or ecotypes, and whether the observed differences in leaf morphology are from inbreeding, or are particularly forms and/or ecotypes that are expressing variance within populations
- The extent that self fertilisation may influence inbreeding, leaf forms and monomorphism
- Locate and document the extent of polymorphic loci, whether they are hidden behind monomorphic loci or require diploid rather than haploid DNA, and production of specific primers and sequences to fully document variation and confirm the evolutionary status of *S. aviculare* and *S. laciniatum*
- Documentation of whether monomorphic loci are the determinants of generic level differentiation of the relationships within the *Solanum* genus, and *Archaesolanum* sub-genus.
- Whether the invariance between *S. laciniatum* with *S. aviculare* is confined only to monomorphic loci, or is apparent also on polymorphic loci

### 6.2.5 Cultural, conservation and restoration

Conservation and restoration programs for species protection and enhancement of *S. aviculare* and *S. laciniatum* should include the following:

- Restoration and enhancement programs for *S. aviculare* and *S. laciniatum* to be undertaken throughout the North and South Islands
- In all planting programs *S. aviculare* and *S. laciniatum* are positively identified, not planted in close proximity to each other to avoid non-viable seed production from cross pollination
- The use of local seed sources, and consideration of climatic and geographic factors such as altitude, and the potential for local adaption

are considered in all planting programs due to the unresolved status of ecotypes

- Identification surveys are recommended to be undertaken in the lower North Island to confirm whether *S. aviculare* or *S. laciniatum* are present
- A Recommendation Category included as an appendix to the Threatened Plants list, and tangata whenua aspects to be included as a section to assist in choosing species for restoration projects, that will help enhance wild populations and cultural uses
- Maintenance of, and increase in ungulate control measures
- Control measures for rats localised and timed in relation to bird numbers and fruit maturation of *S. aviculare*
- Population surveys to be undertaken prior to the next Threatened and Uncommon List publication, to identify the accuracy of population numbers, and whether population numbers of *S. aviculare* and *S. laciniatum* are lower than 'Declining a')
- Collaborative restoration and enhancement programs by mana whenua incorporating local mātauranga Maori and kaitiaki practice in primary position to be undertaken as a matter of course

Photo Meredith Te Atawhai

**Mount Tarawera, with snow: winter 2009.**

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# APPENDICES:

## Appendix 1: Māori glossary

NB: only brief meanings have been supplied, for more in depth descriptions please refer to dictionaries and alternative sources.

| | |
|---|---|
| Aotearoa | New Zealand |
| āhua | condition, to form, to make, appearance |
| aroha | love, feel concern for, empathise, pity |
| atua | gods, supernatural beings, ancestors |
| e noho kainga | place where residing |
| hapū | sub-tribe, clan |
| Hawaiki | ancient Māori homeland from whence migrated |
| iwi | extended kinship, tribe, nation |
| kai | food, meal, to consume |
| kai tamariki | food of children |
| kainga | residence, home, habitat |
| kaitiaki | guardian, custodian |
| kaitiakitanga | act of guardianship |
| kaumātua | elder, man or woman |
| kaupapa | ideology |
| kawa | protocol, customs |
| kererū | native pigeon |
| kiore | native rat |
| kōkōwai | red ochre |
| kōrero | speak, address, discussion, conversation |
| kowhaiwhai | painted panels |
| mana | authority, control, power, spiritual power, prestige |
| manāki | support, take care, hospitality |
| māra | garden, cultivation |
| māramatanga | understanding, enlightenment, significance |
| mātauranga | knowledge, wisdom, understanding |
| mātauranga koiora | biology |
| mauri | life principle, material symbol of life principle |
| moa | extinct ratite bird |
| mōhio | to know, understand, comprehend, realise |
| ngahere rākau | forest tree |
| Ngati Awa | tribal group of Whakatane and Te Teko |
| Ngai Tahu | tribal group of the South island |
| Ngai Tūhoe | tribal group of Te Urewera, Ruātoki, Waimana, Waikaremoana, Kutarere |
| Pā | village, fortified village |
| Pākehā | New Zealander of European descent |
| Papatuanuku | Earth mother, wife of Ranginui |
| poroporo | *Solanum aviculare* or *Solanum laciniatum* |
| Rangatira | revered, chief, chieftainess, aristocracy |
| Ranginui | sky father, husband of Papatuanuku |
| raupatu | confiscation |
| raupeti | leaf of *Solanum* species used as a vegetable |
| Rekohu/Wharekauri | Chatham Islands |

| | |
|---|---|
| rohe | boundary, district |
| rongoā | remedy, medicine |
| rōpū | group, association |
| tangata whenua | indigenous people to the land, local people |
| taonga | something prized, |
| tapu | restricted, prohibited, set apart, under atua protection |
| Tauranga Moana | tribal group of Tauranga area |
| tautoko | to support, agree, verify |
| Te Ika a Maui | North Island of New Zealand |
| Te Wai Pounamu | South Island of New Zealand |
| Te Whanau a Apanui | tribal group from east coast of North Island |
| tiaki | conserve, look after |
| tikanga | correct procedure, lore, method, reason, plan |
| tīpuna | ancestors, grand parents |
| tuawhenua | interior, hinterland |
| Tūhoe ki Waiohau | Tūhoe group from Waiohau area |
| Tūhourangi | tribal group from Tarawera area |
| tūturu | true, actual |
| umu | earth oven |
| waka | canoe, vessel, kinship groups |
| wairua | spirit, soul, quintessence |
| wānanga | tribal knowledge, seminar, to meet, discuss institution |
| whakapapa | genealogy, lineage, descent |

**Appendix 2: Study site descriptions.**

**Tarawera** ($38^o$ 08'12.24"S $176^o$ 27'18.84"E)

Tarawera contained three sites:

**TAR 1** contained >30 plants located below the Tarawera Falls, 50m from the car park on the left side of the track, with an east-west aspect: elevation 190m, GPS E 2818607 N 6332396. Consisting of a shallow slump slip ($<10^o$), substrate of scoria and ash, with intact bush either side and above. The Tarawera Falls walking track lies immediately at the base, with the Tarawera River 15m further to the west from the track.

**TAR 2** contained >30 plants located above the Tarawera Falls 15 minutes walk up the Tarawera Falls to Lake Tarawera track with an west-east aspect: elevation 288m (+/- 14m), GPS E 2818488 N 6331244. Consisting of a steep slip ($>30^o$) crossing through the track, with the *S. aviculare* plants located above and below the track. The substrate consisted of ash and some scoria. Intact forest surrounds the site above, below and on either side.

**TAR 3** contained >30 plants located 1.5km west from the Lake Tarawera campground on the Humphries Bay track. Consisting of a very steep slip ($>45^o$) with a south facing aspect: elevation 309m at the base to 350m at the top (+/- 7m), GPS E 2815942 N 6329391. The substrate consists of ash, scoria and fused obsidian rock. The surrounding intact forest is located above and to the sides, with Lake Tarawera directly at the bottom.

**Okataina** ($38^o$ 02'46.69" S $176^o$ 25'57.43" E)

At Okataina there were four sites.

**OKA 1** located approximately 1km along the Lake Rotongata and Lake Rotoatua track beginning opposite the Okataina Outdoor Education Centre (OOEC) entrance. The site contained four plants, one larger (1.95m) and two smaller in a small tree fall canopy gap directly on the left (west) side of the track, with a further small plant located on the right (east) side of the track directly opposite the others. The substrate was ash, the GPS reading was considered inaccurate due the enclosed canopy cover.

**OKA 2** located on the Patotara crater track, 500m down from the small car parking area on the right side of Lake Okataina road 1.3 km before OOEC entrance, consisting of a single plant on the left side of the track, below a slip which crosses through the track. The substrate was ash and intact scrub and forest surrounded the track above and below: elevation 375 m, GPS E 2809878 N 6341962.

**OKA 3 and 4** single plants located in the forest edge, on the left hand side of the Lake Okataina road 1km and 1.5 km from the OOEC entrance: site 3 elevation 395 m, GPS E2810731 N6340873, site 4 elevation 392 m GPS E2810876 N6340873.

**Rotoma** (38°00'42.26"S 176°37'50.96"E)
There were four sites located at Rotoma.

**ROT 1** located on the Rotorua – Whakatane highway approximately 44km from Whakatane, on the hill leading up to Lake Rotoma from Whakatane, consisted of a single plant on the right hand (north) side of the road. Surrounded above and to the sides with intact re-growth bush: elevation 329m, GPS E 2827223 N 6341967.

**ROT 2** located .3km further up the road also on the right hand side of the road facing south in a cool, damp tawa (*Beilschmiedia tawa*) / rewa-rewa (*Knightia excelsia*) / kohekohe (*Dysoxylum spectabile*) grove within a tawa tree fall canopy gap, consisted of 18 plants: elevation 349m, GPS E 2826970 N 6342153.

**ROT 3** located 1km further up the road on a bend to the left (south) side of the road above a siding, 100m before the Matahi Rd turn-off to Manawahe, consisted of a single plant: elevation 348m, GPS E 2826344 N 6342247.

**ROT 4** located on the edge of the Matahi Reserve on the north side of Lake Rotoma in lake edge tutu (*Coriaria arborea*) / tātarāmoa (*Rubus cissoides*) / karamu (*Coprosma robusta*) / *Buddleia davidii* scrub, 500m from the northern

end of the beach car park adjacent to Matahi Lagoon, consisted of a single plant: elevation 335m, GPS E 2826170 N 6343245.

**Tauranga (**37$^{o}$ 41'03.04"S 176$^{o}$ 14'00.44"E)

This site is a private whanau (family) rongoā reserve located on Mountain Road in the O Tane Wainuku Block inland from Tauranga, consisted of thirteen plants variously located individually on forest edges throughout the block: elevation 402m, GPS E 2789536 N 6366242.

In addition there were a number of *S. aviculare* seedlings growing beneath a pigeon wood (*Hedycarya arborea*) tree which was identified as a kereru roosting site, to be used for faeces collection, but were not included in the count.

**Te Urewera / Ika Whenua** (38$^{o}$ 17'46.91"S 176$^{o}$ 52'41.91"E)

Sites are located on the Ohutu Stream off Troutbeck Road, Galatea at the head of the farm used to access the Park, consisting of 3 locations.

**URE 1** contained four plants, one large (2.08m) and three smaller stunted and broken plants, scattered over a steep scree slip on the right (south) side of the stream around ½ hour walk from the farm car park area: elevation 293m, GPS E 2845741 N 6304566.

**URE 2** contained two plants directly on the left (north) side of the stream further upstream from site 1, growing in stream gravel and humus accumulation with *Buddleia davidii*: elevation 297m, GPS E 2846102 N 6304456.

**URE 3** contained >24 plants further up stream, found on a large steep scree slip within a tawa grove over the top of a ridge after rising up a steep sidle: elevation 380m, GPS E 2846387 N 6304182.

**Maungatautari (**38$^{o}$ 01'18.55"S 175$^{o}$ 34'38.51"E)

**MAU N**: Maungatautari northern enclosure consists of three locations.

**MAU N1** is a single plant 30m on the right from the beginning of the Te Ara Tirohia loop track, after leading left.

**MAU N2** consisted of >3 plants in a tawa tree fall canopy gap either side of the track 10 minutes easy walk from the first location.

**MAU N3** consisted of >25 plants in a large tawa tree fall gap, 10m off to the right side of the track, 7–8 minutes easy walk leading right from the beginning of the Te Ara Tirohia track.

Elevation at the northern enclosure main gate 339m (+/- 7m), GPS E 2736497 N 6352580.

**MAU S**: Southern enclosure consisted of four locations.

**MAU S1** consisted of five individual plants located on the right side of the Nikau track 5 minutes from the beginning of the track.

**MAU S2** contained two plants off the left of the Nikau track in a tawa tree fall canopy gap, a further 5 minutes up the path.

**MAU S3** contained three plants 15 minutes up Nikau track and leading right onto the Rata track, in a tawa tree fall canopy gap 5 m off the track.

**MAU S4** located at the aviary on the Rimu track under a tawa tree contained >20 plants.

Elevation at main gate 323 m, GPS E 2734885 N 6346492.

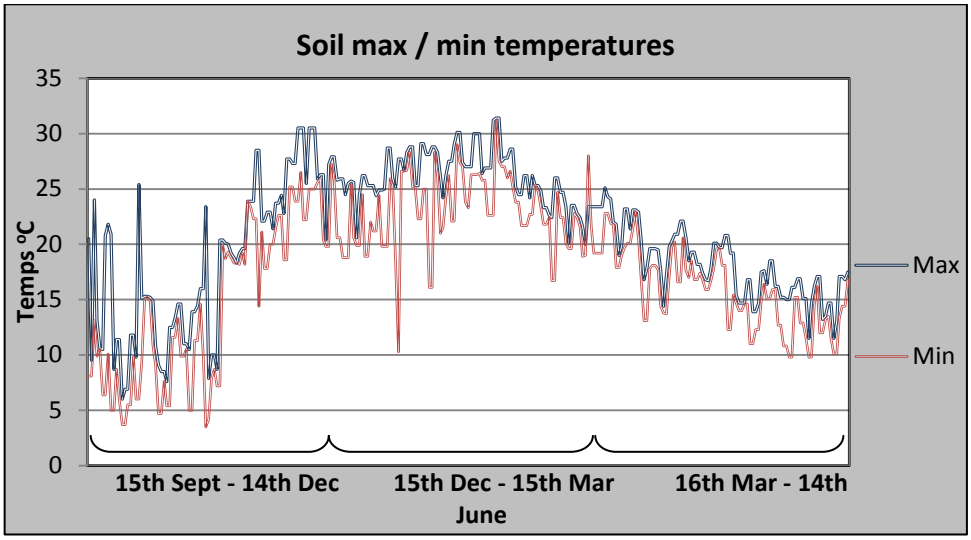
**Pirongia** ($37^{o}$ 58’27.17”S $175^{o}$ 06’23.14”E)

The Pirongia sites consisted of two locations.

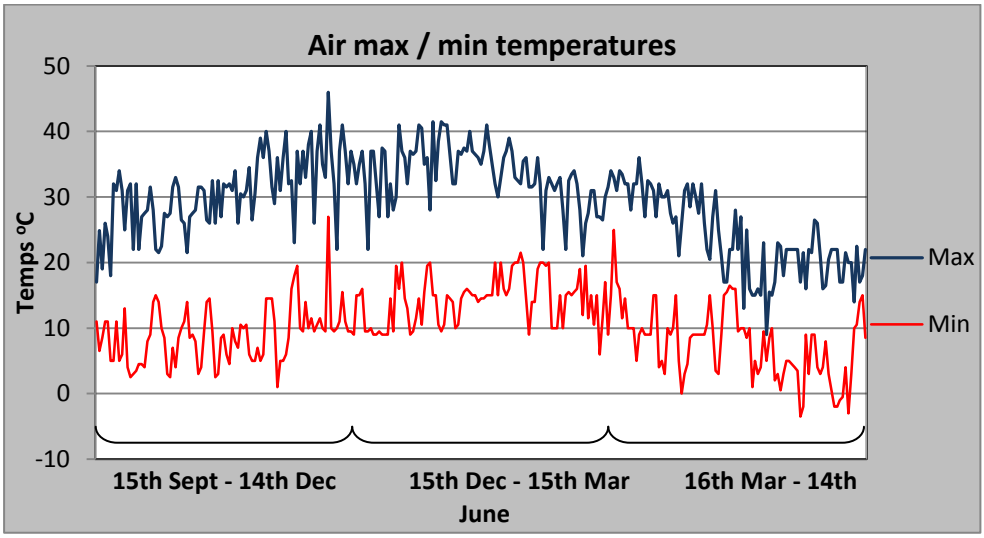
**PIR 1 & 2** at the Corcoran Road car park entrance containing two small plants directly at the beginning of the track: elevation 335m (+/- 7m), GPS E 2696511 N 6357644.

**PIR 3-5** located in the Kaniwhaniwha reserve containing three plants individually off the right side of the track after entering from the track leading from the car park and turning right after the gate: elevation is 116m (+/- 6m), GPS E 2692633 N 6361062.

## Appendix 3: Total temperature ranges for seed bank trial




**Soil temperature range in 3 month increments for the duration of the soil seed bank germination trial.**




**Air temperature range in 3 month increments for the duration of the soil seed bank germination trial.**

**Appendix 4: Site locations and species identified in Tarawera vegetation plots**

| SPECIES | TAR 1 | TAR 2 | TAR 3 |
|---|---|---|---|
| *Beilschmiedia tawa* | X | X | |
| *Brachyglottis repanda* | X | | |
| *Carpodetus serratus* | | | X |
| *Coprosma lucida* | X | | |
| *Coriaria arborea* | | X | |
| *Cortaderia fulvida* | | X | |
| *Cyathia dealbata* | X | X | |
| *Cyathodes fasciculata* | X | X | X |
| *Dicksonia squarrosa* | X | X | X |
| *Dracophyllum strictum* | | | X |
| *Gaultheria antipoda* | | X | X |
| *Geniostoma rupestre* | X | X | X |
| *Hebe stricta* | | X | |
| *Hedyacarya arborea* | | X | X |
| *Knightia excelsia* | X | X | X |
| *Kunzea ericoides* | | X | X |
| *Leptospermum scoparium* | | | X |
| *Litsea calicaris* | X | X | X |
| *Macropiper excelsum* | X | X | |
| *Melicytus ramiflorus* | X | X | X |
| *Metrosideros excelsa* | X | | |
| *Myrsine australis* | X | X | |
| *Oleria albida* | | | X |
| *Oleria rani* | X | | |
| *Pittosporum tenuifolium* | X | | |
| *Pseudopanax crassifolius* | X | X | |
| *Ripogonum scandens* | X | | |
| *Rubus cissioides* | | | X |
| *St Streblus banksii* | | | X |
| *Weinmannia racemosa* | X | X | X |

**Appendix 5: CTAB extraction standard protocols**

# ISOLATION OF DNA

Adjusted from Ray and Dicks original procedure!

**HOMOGENISATION BUFFER:**

- EDTA Ethylenediaminetetraacetic acid 20mM
- CTAB Hexadecyltrimethylammonium bromide 2 %
- PVP-40 Polyinvylpyrrolidone MW 40,000 w/v
- DIECA Diethyldithiocarbamic acid 4mM
- Tris- HCl (pH 8) 100mM
- NaCl 1.42M

**100ml solution of Homogenisation Buffer:**

- 10ml 1M Tris-HCl
- 4ml 0.5M EDTA
- 2g CTAB
- 28.4ml 5M NaCl
- 2 g PVP-40
- 0.069 DIECA

**Other Chemicals needed for procedure:**

- 2-mercaptoethanol (3.49ul per reaction)
- RNase A (10mg/ml)
- Chloroform: Isoamyl alcohol (24:1)
- Isopropanol
- 100% ethanol
- 70% ethanol
- TE buffer

**PREPARATION:**

- Label and U.V 2x 1.5ml Microcentrifuge tubes per sample
- Collect ice and Liquid Nitrogen
- Collect Mortar and Pestles (one per sample)
- Add 500ul (X no. of samples) of Homogenisation Buffer to a15ml Conical tube, Then add 3.49ul of 2-mercaptoethanol (X no. of samples) to the conical tube and Invert to mix
- Aliquot 500ul of H. Buffer + 2-mercaptoethanol mix into the first set of U.Ved microcentrifuge tubes and sit on ice.

**1:** Weigh out about 0.7g of frozen leaf material per sample then grind to a fine powder using Liquid Nitrogen. Scrape powder into microcentrifuge tube on ice, then return to ice until all samples have been ground. Then take off ice and add 5ul of RNase A to each sample.

**2:** Vortex samples @ high speed then incubate in the Thermomixer @ 60°C for 10-15 minutes (more for fibrous leaves)

**3:** Add 500ul of Chloroform: Isoamyl alcohol (24:1), cap tube and mix vigorously using Vortex @ high speed

**4:** Centrifuge @ Max speed for 10 minutes – you should now have two distinct layers in the tube with an interface between them that may look like skin. The DNA is in the top (Aqueous) layer, Debris type plant material in the interface and proteins etc in the lower (Chloroform) layer

**5:** Recover DNA by gently sucking off the top layer and transferring it to a fresh 1.5ml microcentrifuge tube – Try to recover as much of the top layer without sucking up any of the interface or lower layer (NOTE: if supernatant appears cloudy or had debris left in it REPEAT steps **3 – 5**)

**6:** Add equal volume of Isopropanol (400ul?) and invert to mix, then sit samples for 10 minutes

**7:** Centrifuge @ max for 10 minutes

**8:** Locate the fine whitish pellet near the bottom of the tube, then use a fine tip pipette to remove all supernatant without disturbing pellet (which contains the DNA!)

**9:** Add 500ul of 100% ethanol, centrifuge @ max for 1 min and again locate the pellet and remove the supernatant as above in **8**

**10:** Add 500ul of 70% ethanol to samples, centrifuge @ max for 1 min – again locate pellet and remove supernatant as in **8** – Re-spin @ max briefly to bring liquid from sides down – remove excess liquid without disturbing pellet

**11:** Sit on bench for 5 mins to dry off any ethanol. Then add 50ul of TE buffer – Shake tube in thermomixer @ 37 °C for 10-20 minutes to resuspend DNA

## Appendix 6 Sequences

>0314_**Lac4(**ITS5HP).ab1
CCTAGCTCAGCGGCAGGTCATTGTCGAACCTGCAAAGCAGAACGACCCGCGAACTTGTTTGAA
CACCGGGGGGGCCGCGC
GGCGGGGGTGCTTCGTCCCGAGTTGAGTGTCCGCCACCACTTGCCGCGACGTCCGTCGACGTG
GACTCGCATTTAGGCCA
GCCGCGAGCTCGAGGCCCACGGGAGGCCAGCTTCCGCCCCCACGACGCCTGGCGGCGTGCGGG
GGGCGACGCGATGCGTG
ACGCCCAGGCAGACGTGCCCTCGGCCAAATGGCTTCGGGCGCAACTTGCGTTCAAAGACTCGA
TGGTTCACGGGATTCTG
CAATTCACACCAAGTATCGCATTTCGCTACGTTCTTCATCGATGCGAGAGCCGAGATATCCGT
TGCCGAGAGTCGTTTTT
GTTTCAAAAGAAGCACACGTCCCCCCACGCACTCCGCGGACGGGGCGCGGGGGGCGGGCCGTC
AATTCAGTATTCCTTGG
CGCTTTCCGCGCCGGGGTTCGTTAGCCGCCCGAACGAGCGCGCAAGCGCGCGTCGGGGGCGGG
AGGGAGACGCGCGGACG
GGAGGGGCCGAAGCACCCCCGCCGCGCGGCCCCCCCGGTGTTCAAACAAGTTCGCGGGTCGTT
CTGCTTTGCAGGTTTCG
ACAATGATCCCTTCCGCAGGTTCACCTACGGAAAACCTTGTTACGACTTCTCCTTCCAA

>0314_**Lac6**(ITS5HP).ab1
TATAAGCGGGCGACTCGTGGAGAACATTGTCGAACCTGCAAAGCAGAACGACCCGCGAACTTG
TTTGAACACCGGGGGGG
CCGCGCGGCGGCTGCGAAACATCGAGAGTTGAGTTTGAACCACCACTTGGTGTCAGATACGAC
GACGTGGACTCGCATTT
AGGCCAGCCGCGAGCTCGAGGCCCACGGCCCCCCAAGAAACCGCCCCCACGACGCCTGGCGGC
GTGCGGGGGGCGACGCG
ATGCGTGACGCCCAGACAGACGTGCCCTCGGCCAAATGGCTTCGGGCGCAAATTGCGTTCAAA
GACTCGATGGTCACGGG
ATTCTGCAATTCACACCAAGTATCGCATTTCGCTACGTTCTTCATCGATGCGAGAGCCGAGAT
ATCCGTTGCCGAGAGTC
GTTTTTGTTTCAAAGAAGCACACGTCCT

>0314_**Lac7**(ITS5HP).ab1
TACGGCTATAGTATACTCCGCGAACTGCAAGCAGACGACCGCGACTGTGACACGCGCTCAGTC
GTGACATGACAGCATAG
TGACTCACACACGCTGACAGCGGTACGATAGCCGACACCCTCGCCACGAGGCTGGATCAATCA
GGTAGAGTGCCAGTCG

>0314_**MAU N4**(ITS5HP).ab1
TTAGCGGGGTTGGTATATCTGAGAAGGAGCGGGGGAATGAATGAAGAAGAGCGCAAACTTGTT
TGAACACCGGAGGGGCC
GCGCGGCGGCCGTGAAACATCGAGAGTTGAGTTTCAACCACCACTTGCCGCGACGTCCGTCGA
CGTGGACTCGCATTTAG
GCCAGCCGCGAGCTCGAGGCGCACGGGAGGCCAGCTTCCGCCCCCGCGACGCCTGGCGGCGTG
CGGGGGGCGACGCGATG
CGTGACGCCCAGGCAGACGTGCCCTCGGCCAAATGGCTTCGGGCGCAACTTGCGTTCAAAGAC
TCGATGGTTCACGGGAT
TCTGCAATTCACACCAAGTATCGCATTTCGCTACGTTCTTCATCGATGCGAGAGCCGAGATAT
CCGTTGCCGAGAGTCGT
TTTTGTTTCAAAAGAAGCACACGTCCCCCCGCGCACTCCGCAGACGGGGCGCGGGGGGCGGGC
CGTCAATTCAGTATTCC
TTGGCGCTTTCCGCGCCGGGGTTCGTTAGCCGCCCAAACGAGCGCGCAAGCGCGCGTCGGGGG
CGGGAGGGAGACGCGCG
GACTGGGGGGGCCGAACCACCCCCGC

>0314_**TAR 3/2**(ITS5HP).ab1
AAAACGGTTGTAAGGGTTTTCTTTGGAAGATTGCTCGGACTTGCGAGTAGAACGACCCGCGAA
ACTTATTAGAACACCGG
AGGGACTGCATGTGCGCTTGTTTTTGCATTCGGTCGGCTAACAACCCTCAGTGTGTAAAGCAC
CATTGAATACCCAATGG
ATGGCCCGCTCTCTGTGCCCCATCTGTGAAGGGGGTATGGGGACGTGTGCTTCTTTTGAAATA
AAAATGACTCTCGGCAA
TGGATATATAGGCTCTTACATCGATAACGAACGTAAAGAAATGCGATACTTAATGTGAATTGT
AGAATCCTGTGAACCGT
AGAGTCTTTGAATGCAAGTTATGCCTAAAGTCATTTGACCGACGACACGTCTGCCTGGGCCTC
ATGCATCACATCCCCCC
CGCATTCTTCCAGGCATCGTGGGGTGGAAGCTGGCCTCCCATGCACCTTGAGCTCGCAGCTGG
CCTAAATGCGATTCCAC
TTCTACGGACATCACGCTAAGAAGTGTGTGAAACTTAACTCTTGATGTGCCGCGACCGATCCT
CATCGCGCGGCTAGCCT
CCCAGACCCACCGTGCACTCCTGCACTCTAACTGCGACCCCATCTCACGCAGGATTACCCACT
GAGTTTAATGATATCAA
TACGCGAACCAAATCCATGAGCGGTTAACCACTGATTATAATCATATCATTAATCGGAGGAAG
AACTTC

>0314_**TAU5**(ITS5HP).ab1
TGTTGCGGGGTTAAGTATAAAACACTCATGGCGGTCGCTGAACGGTACTCTCTCAATCAGCGG
TAGGCATGGTGGCGGTG
GCACGGGGGGACGCGTCAGCAGTTTTTGCGCAGGCCGTGGCGCGGGCAAGCACCGCTTGTAGA
ATGAAAGGCGGCGTGCT
ATCGCATTTGGGCCAGCCGGGTCTCGAGGCACTCGGGCCTGTCCGAT

**Appendix 7: Sample e survey forms A) DOC, B) Tangata whenua**

**A)** Population survey of *Solanum aviculare* for MSc Thesis, Waikato University.

Please answer as noted all questions considered relevant, and return via e mail: mishin.env@clear.net.nz by December 2008.

**In your conservation / observed area: Opotiki**

| | |
|---|---|
| **1** | Is *S. aviculare* seen as: (1 answer - highlight in bold)<br>a) common (regularly observed >50% ),<br>b) uncommon (regularly observed <50%)<br>c) rare (regularly observed <10%) |
| | Comments: not common in any of conservation areas here seems to be observed in more northern localities |

| | | | | |
|---|---|---|---|---|
| **2** | Does *S. aviculare* appear in: (all relevant answers - yes) | common | uncommon | rare |
| | a) disturbed areas within intact forest eg slip sites | yes | | |
| | b) canopy gaps | | | yes |
| | c) forest edges | | yes | |
| | d) open areas outside of intact forest | yes | | |
| | e) other: | | yes | |
| | Comments:<br>Very scattered ,short lived ,good bird species | | | |

| | | | | |
|---|---|---|---|---|
| **3** | When observed as per Qu. 2 does it occur: (all relevant – yes) | common | uncommon | rare |
| | a) as single plants | | yes | |
| | b) small groupings | yes | | |
| | c) populations > 20 + plants | | | yes |
| | Comments: only small group obs | | | |

| | | |
|---|---|---|
| **4** | Does *S. laciniatum* occur also: (highlight in bold) | Yes / No |
| | Comments: very rarely encountered | |

**Please state area that plants were observed:** in disturbed sites in seral forest on conservation or scenic reserves peripheries-Opotiki

Thank you for your time and effort.

Graeme Weavers: MSc Waikato University

**B)** Population survey of *Solanum aviculare* (orange berried poroporo) for MSc Thesis, Waikato University.

Please answer as noted all questions considered relevant, and return via e mail: mishin.env@clear.net.nz

**In your rohe:**

| | |
|---|---|
| **1** | Is *S. aviculare* seen as: (1 answer - highlight in bold)<br>a) common (regularly observed >50% ),<br>b) **uncommon** (regularly observed <50%)<br>c) rare (regularly observed <10%) |
| | Comments: |

| | | | | |
|---|---|---|---|---|
| **2** | Does *S. aviculare* appear in: (all relevant answers - yes) | common | uncommon | rare |
| | a) disturbed areas within intact forest eg slip sites | | | |
| | b)canopy gaps | | | |
| | c) forest edges | | | |
| | d) **open areas outside of intact forest** | | | |
| | e) **other:** | | | |
| | Comments: Stripped forest land and coastal lands plus road side verges. | | | |
| | | | | |
| **3** | When observed as per Qu. 2 does it occur: (all relevant – yes) | common | uncommon | rare |
| | **c) as single plants** | | | |
| | **d) small groupings** | | | |
| | c) populations > 20 + plants | | | |
| | Comments: | | | |

| | |
|---|---|
| **4** | Do other orange berried or purple berried species occur also?: (highlight in bold) Yes / No |
| | Comments: Yes I watch them change colour. See both colours |

| | |
|---|---|
| **5** | Do you, or have you or your whanau used *S. aviculare*, orange berried poroporo for kai or rongoa?<br><br>(highlight in bold) **Yes** / No |
| | If yes, is the use modern / current or historic? (ie Present generation or previous generations)<br><br>Comments: Was known within the woman that it was an alternative similar to the pill/contraceptive, but disappeared around the South Canterbury district in the 40's.<br><br>I chew the purple berries if I find them becos the other colours are bitter.<br><br>A Pakeha lady I know made jelly out of the purple berries.<br><br>If no, is there knowledge of its previous use?<br><br>Comments: |

| | |
|---|---|
| **6** | To your knowledge is the orange berried poroporo used for kai or rongoa by hapu or iwi within your rohe, and if so is the use current or historical? |
| | Comments: Not current becos they have disappeared from our district. |

**Please state rohe and area that plants were observed:**

Waitaki/Oamaru/Herbert Forest.

Thank you for your time and effort. G. M. Weavers – Ahu Whenua o Ngai

Tama Tuhirae ki Waimana; MSc Waikato University21 Tuwharetoa Rd,

Kawerau 3127Ph: 07/ 3238190 - e mail: mishin.env@clear.net.nz

**Appendix 8: Sample letters and questions**

G. M. Weavers
21 Tuwharetoa Rd
Kawerau 3127
Ph: 07 323 8190
E mail: mishin.env@clear.net.nz

13/02/2009

Tena koe he Rangatira,

Dear ……

I am writing to ask for your permission to interview you in regard the cultural uses of poroporo (*Solanum aviculare*) and to include any information from that interview and / or personal conversations in my MSc thesis research study, for publication. This study involves investigating and documenting the population dynamics and community structure of poroporo in slip sites within Tarawera Scenic Reserve. In conjunction with that work will be an investigation outside of Tarawera of the genetic diversity and connectivity of populations and groups to the plants at Tarawera. This will entail the taking of leaf samples for genetic analysis.

A major part of my study, which will be a whole chapter, is regarding the cultural uses, both historic and modern, of poroporo including its current environmental and rongoā status. That is the main focus of my korero to you. I am wishing to discuss 1) the traditional cultivation practices (if any) that you may know 2) any additional rongoā uses in addition to those published 3) if the plant is made more widely available again in amenity plantings would that be an advantage and would you consider the re-use of the plant ? 4) discuss with you the published information, to consider its accuracy.

Your participation in this study will remain confidential to me and any information that you provide that I use in my thesis or other publications will be reported without use of your name. I would like to audio-tape our conversation so that I can recall it later and I am able to provide you with a written record of our interview for you to verify if you wish. I expect our interview to take no longer than one hour.

I hope that my research can 1) add to the understanding of the ecological

knowledge of the plant 2) consider appropriate restoration and conservation measures 3) encourage more accessible plantings and 4) encourage a revival of the rongoā uses.

To that end I am attempting to tie together the known science of the plant and additional knowledge from this study to traditional practices, to consider how any similarities between the two can be aligned to modern restoration and conservation programs, not just for poroporo but for other species as well. I would like to use any information that you may have in the publishing of my work, but fully accept that if you are not comfortable with that then I will not proceed.

I would greatly value discussing these issues with you, even if you are only comfortable with korero, without publication. If you do not feel comfortable with either proposal, I fully understand and please feel free to say no. If you are happy to participate in an interview please read the attached consent form, sign and return to me.

If you have any concerns arising from this proposal please feel free to contact my Supervisors at the University of Waikato, Dr. Chrissen Gemmill Ph. 07 838 4053 or Professor Bruce Clarkson Ph. 07 838 4148

Pai marie

Graeme Weavers B.E.S.

**Research Consent Form**

I have read the attached letter of information.

I understand that:

1. My participation in the project is voluntary.
2. I have the right to withdraw my consent at anytime.
3. Data can be collected from my korero in the ways specified in the accompanying letter. This data will be kept confidential, securely stored and reported anonymously unless as stated below.

I give my consent conditional to the following (tick box):

☐ Data obtained from my korero during the research project may be used in the writing of reports or published papers and making presentations about the project.

☐ My name may be used in publication.

☐ The issue of Intellectual Property rights has been fully discussed and I am content with the arrangements as attached.

I can direct any questions to Graeme Weavers

For any unresolved issues I can contact Dr. Chrissen Gemmill.

E mail: gemmill@waikato.ac.nz Tel: 07 838 4053.

I give consent to be involved in the project under the conditions set out above.

Name:_____

Signed:_____

Date:_____

## SUGGESTED INTERVIEW QUESTIONS FOR POROPORO

**1.** Poroporo was previously noted as being common in te Urewera / Whirinaki. Indications from DOC surveys and conversations are that it has become much less common over the past 70 years. Have you observed this decline?

Have you knowledge from your tīpuna or elders that the previous commonality was correct? Have tīpuna or elders conveyed knowledge of the decline?

**2.** Some reasons for the decline have been attributed to deer and goat grazing, and possible reductions in bird species. Do you or your tīpuna / elders have any consideration for the reasons that may have contributed to the decline?

**3.** Do you have, or can you report on others, any ideas how the decline may be halted?

**4.** Do you have knowledge of whether poroporo was cultivated at kainga of Pa sites in the area in previous generations?

**5.** I believe that in previous generation's tangata whenua conducted a 'type' of forest management practice of a manner similar (but possibly less urgent) reasons that DOC attempts at the present. Do you have any knowledge of any previous generation's cultivation and forest management practices in regard poroporo?

**6.** Do you know of any traditional ecological management practices that could be aligned to present modern environmental practices to benefit the conservation and restoration of poroporo?

**7.** Poroporo was used for kai tamariki and rongoā including skin and joint conditions as well as contraception. Do you have any knowledge that suggest poroporo was used in te Urewera for the same purposes?

**8.** The use of poroporo for rongoā appears (from discussions) to have declined not only in the BOP but widely around the country. That decline in use appears to correspond to the ecological decline, and subsequent availability for harvest outside of reserves for which permits are required. Does this appear to be a similar situation in Te Urewera?

**9.** If poroporo was available for re-planting in areas outside of permitted reserves, do you consider this to be an advantage to renewed use of it as a rongoā?

**10.** Do you know of poroporo as orange or purple berried?