

Identification of the Area of Origin in Southern Africa of the Australian Form of Fireweed, Senecio

On

Project number CS.274

Meat and Livestock Australia Ltd

Locked Bag 991

North Sydney NSW 2059

ISBN 174036239X

MLA makes no representation as to the accuracy of any information or advice contained in this document and excludes all liability, whether in contract, tort (including negligence or breach of statutory duty) or otherwise as a result of reliance by any person on such information or advice.



**Identification of the area of origin in
southern Africa of the Australian form of
fireweed, *Senecio madagascariensis* (Poir)**

Final report for MRC project CS.274

August 1997



1. Genetic relationships between Australian fireweed and South African and Madagascan populations of *Senecio madagascariensis* Poir.

Ian Radford, CSIRO Tropical Agriculture, Townsville, Australia

Abstract

An isozyme study of the Senecio madagascariensis Poir. species complex was used to investigate the most likely region of origin for Australian fireweed within southern Africa and Madagascar as part of a biological control study. Collections of seed and voucher specimens were made in Australia, Natal, Eastern and Western Cape Provinces and South Eastern and South Western Madagascar to sample the full range of variation (excluding exotic populations found in Argentina). Specimens were classified using taxa recognised by Hilliard (1977) and using putative groups based on observations of voucher specimens. Fresh leaf and apical cell tissue from plants grown from seed collections were used for isozyme electrophoresis. Australian fireweed populations were found to be most closely related to S. madagascariensis from Natal and the Eastern Cape. Future biological control collections should target this region to maximise the likelihood of finding host specific agents for fireweed.

Introduction

Fireweed (believed to be *S. madagascariensis* Poir.) is an important weed in south eastern Australia (Watson et al. 1984, 1994, Sindel 1986, 1996). This weed is considered important because it is toxic to cattle (Walker and Kirkland 1981, Kirkland et al. 1982), is competitive with pasture species (Sindel and Michael 1992b) and can cause losses in pasture production

(Radford et al. Unpublished Report 1997), because it is invasive and continues to spread in many areas (Watson et al. 1984, 1994, Sindel and Michael 1992a, Radford et al. 1995a), is expensive or difficult to control (Watson et al. 1994, Sindel 1996) and is conspicuous and highly visible over large areas of the NSW and South Eastern Queensland coastal plains and ranges (Sindel and Michael 1988). For these reasons fireweed is currently the subject of a biological control programme in Australia.

Biocontrol agents so far tested for this weed have unacceptably wide host ranges. Several native species, including *S. laetus* Forst. f. ex Willd. (*sensu lato*, Belcher 1993), have been found to act as hosts for proposed biocontrol agents (McFadyen and Sparks 1996). Biological control impact assessment studies have shown that fireweed is temporally and spatially coincident (as per Cullen 1990) with *S. laetus* populations in the field, increasing probability of spread and damage to native communities (Radford 1997). This means that only highly host specific biological control agents are likely to be acceptable for release in Australia.

Although fireweed is identified as *S. madagascariensis* from Africa (Michael 1981) the exact provenance of our weedy variant within this region is still controversial (Marohasy 1993, Radford et al. 1995). Original collections for potential biocontrol agents occurred in Madagascar (Marohasy 1989), presumably because the species name suggested its origin was in that country. *S. madagascariensis* plants, however, occur in Madagascar, South Africa, Swaziland, Mozambique and other areas in the region (Hilliard 1977). Which of these regions Australian fireweed comes from is not known. There are also questions concerning the taxonomic identity of fireweed as there are several morphologically similar species within the greater *S. madagascariensis* species complex (e.g. *S. inaequidens*, *S. burchellii* and *S. skirrhodon*) (Marohasy 1993). Preliminary observations of herbarium specimens (Radford et al. 1995) suggest that Australian plants were more similar to plants from Natal (South Africa) than to plants collected in Madagascar.

This paper reports on genetic studies initiated for *S. madagascariensis* and allies in Madagascar, South Africa and Australia. Seed and plant collections were made from the major areas of distribution in southern Africa (Muller, MRC Report 1996) and morphological and isozyme studies undertaken to investigate relationships with Australian plants. It is hoped

that future predator and pathogen collections from these populations will increase the chance of finding a host specific biological control agent for fireweed.

Materials and Methods

Sampling strategy.

Plant and seed collections were made from within the known geographic range of *S. madagascariensis* in Natal, Eastern and Western Cape Provinces in South Africa, from South East and South West Madagascar and from south eastern Australia where this species is a weed. Localities, region and latitude and longitude of collection sites are found in Table 1. Seed collections were made from several well spaced individual plants at collection sites in order to sample a representative range of the variation in that population. A voucher specimen was dried and pressed to provide information on the morphological form of plants at a given population site. Seeds (achenes) were separated from pappus and capitulum material by winnowing and stored in paper envelopes in sealed glass jars with silica gel at constant 20°C. Maps of collection sites and further details on the collection procedure can be found in Muller (MRC Report 1996).

Plants were grouped according to distribution and morphology for hierarchical analysis. The first level of the hierarchy was country of origin; South Africa, Madagascar and Australia. Accessions were then divided into regional groups: Natal and Eastern/Western Cape populations within South Africa; south east and south west populations in Madagascar; and Australia. Within regions plants were then grouped morphologically using the key developed by Hilliard (1977) based on floral and leaf attributes (Table 1). Where known taxa did not correspond clearly to observed variation, putative groups (variants) were assigned on the basis of their morphology. Eastern Australian populations were not divided regionally or morphologically due to their uniform morphology throughout the region (Radford 1997). The following key defines groups used to classify plants in this study.

Table 1. Variant, localities, state and latitude and Longitude of sites of populations used in morphometric analyses. Locality data is from Muller (MRC Report 1996). Taxonomic and varietal interpretations are based on the treatment by Hilliard (1977).

Locality	Region	Lat-Long	Variant
1. Nottingham Rd	Natal, SA	29°24'S 29°57'E	UNKNOWN sp.
2. Laingsnek Pass	Natal, SA	27°27'S 29°52'E	<i>S. madagascariensis</i>
3. Fort Mistake	Natal, SA	28°11'S 28°58'E	<i>S. madagascariensis</i>
4. Cedara Agric. Instit. *	Natal, SA	29°33'S 30°16'E	<i>S. skirrhodon</i> ?
5. Injasuti, Drakensberg	Natal, SA		<i>S. inaequidens</i>
6. Mnyama, Sepong *	Swaziland, SA	27°21'S 31°47'E	<i>S. madagascariensis</i>
7. Volkrust	Natal, SA	27°24'S 29°53'E	<i>S. madagascariensis</i>
8. Monks Cowl	Natal, SA	29°03'S 29°24'E	<i>S. madagascariensis</i>
9. Mbabane	Swaziland, SA	26°18'S 31°10'E	<i>S. madagascariensis</i>
10. Fort Dauphin *	SE Madagascar	24°47'S 47°10'E	OBLANCEOLATE V.
11. Twin Streams	Natal, SA	28°58'S 31°45'E	<i>S. skirrhodon</i>
12. Umlalazi	Natal, SA	28°13'S 31°04'E	<i>S. madagascariensis</i>
13. Rooi Els **	Western Cape, SA	34°20'S 19°02'E	<i>S. burchellii</i>
13. Fairbreeze	Natal, SA	29°02'S 31°36'E	<i>S. madagascariensis</i>
14. Wynberg Park **	Western Cape, SA	33°57'S 18°27'E	<i>S. burchellii</i>
15. Sea Acres **	Eastern Cape, SA	34°10'S 24°50'E	14 BRACT V..
16. Port Elizabeth **	Eastern Cape, SA	34°22'S 24°25'E	14 BRACT V..
17. Cape Point **	Western Cape, SA	34°12'S 18°25'E	<i>S. burchellii</i>
32. Durban Airport	Natal, SA	29°58'S 30°58'E	<i>S. madagascariensis</i>
33. Bluff Nature Reserve *	Natal, SA	29°55'S 31°00'E	<i>S. madagascariensis</i>
34. Shaka's Rocks *	Natal, SA	29°31'S 31°13'E	<i>S. madagascariensis</i>
36. Mtunzini Chalets	Natal, SA	28°58'S 31°45'E	<i>S. skirrhodon</i>
37. Mtunzini Field *	Natal, SA	28°57'S 31°45'E	<i>S. madagascariensis</i>
38. St Lucia *	Natal, SA	28°22'S 32°26'E	<i>S. skirrhodon</i>
39. Fannies Island *	Natal, SA	28°08'S 32°25'E	<i>S. madagascariensis</i>
40. Empangeni Road *	Natal, SA	28°43'S 31°38'E	<i>S. madagascariensis</i>
41. Mvoti Toll Gate	Natal, SA	29°24'S 31°17'E	<i>S. madagascariensis</i>
42. Oribi Gorge	Natal, SA	30°44'S 30°16'E	<i>S. madagascariensis</i>
43. Hibberdene	Natal, SA	30°40'S 30°31'E	UNKNOWN sp.
44. Midmar Dam	Natal, SA	29°30'S 30°12'E	<i>S. madagascariensis</i>
49. Fort Dauphin Airport	SE Madagascar	25°02'S 46°57'E	OBLANCEOLATE V.
50. Fort Dauphin Mission	SE Madagascar	25°01'S 46°57'E	OBLANCEOLATE V.
51. East Fort Dauphin	SE Madagascar	25°02'S 46°39'E	OBLANCEOLATE V.
52. Taolagnaro Hotel	SE Madagascar	25°02'S 46°59'E	OBLANCEOLATE V.
53. Toliara Centre	SW Madagascar	23°21'S 43°40'E	OBLANCEOLATE V.
54. Toliara Hotel	SW Madagascar	23°29'S 43°45'E	OBLANCEOLATE V.
55. Taolagnaro Dunes	SE Madagascar	25°08'S 46°23'E	OBLANCEOLATE V.
59. Riversdale	Western Cape, SA	34°12'S 21°36'E	14 BRACT V.
61. St Frances Bay *	Eastern Cape, SA	34°08'S 24°49'E	UNKNOWN sp.
62. Sea View *	Eastern Cape, SA	34°00'S 25°21'E	UNKNOWN sp.
63B. Sea Acres, Erdoff *	Eastern Cape, SA	33°58'S 25°39'E	DISSECTED V.
63A. Sea Acres Tennis *	Eastern Cape, SA	33°58'S 25°39'E	<i>S. madagascariensis</i>
64B. Cannon Rks Mead. *	Eastern Cape, SA	33°44'S 26°33'E	UNKNOWN sp.
64A. Cannon Rocks	Eastern Cape, SA	33°44'S 26°33'E	DISSECTED V.
65. Port Alfred	Eastern Cape, SA	33°36'S 26°40'E	<i>S. madagascariensis</i>
66. Maccassar *	Eastern Cape, SA	33°19'S 27°20'E	UNKNOWN sp.
69. Kenton on Sea	Eastern Cape, SA	33°40'S 26°40'E	14 BRACT V.
70. Alexandria *	Eastern Cape, SA	33°38'S 26°27'E	DISSECTED V.
72. Grahamstown	Eastern Cape, SA	33°19'S 26°31'E	<i>S. madagascariensis</i>
73. Sedgfield *	Western Cape, SA	34°01'S 22°49'E	UNKNOWN sp.
74. Groot Brak *	Western Cape, SA	34°01'S 22°13'E	UNKNOWN sp.

* Accessions for which electrophoresis results were not included (5 or fewer plants analysed). ** Accessions for which no seed was available.

Key to collections from the African *S. madagascariensis* complex

- 1. Natal plants, 19-21 involucral bracts
 - 2. broad lanceolate to succulent leaves,
irregularly toothed *S. skirrhodon*
 - 2* linear-lanceolate leaves, regularly and
finely toothed
 - 3. small linear to linear lanceolate,
woody stems *S. inaequidens*
 - 3* large leaves, linear-lanceolate
regularly toothed..... *S. madagascariensis*

- 1* Eastern and Western Cape plants
 - 4. 19-21 involucral bracts
 - 5. linear-lanceolate leaves, regularly toothed... *S. madagascariensis*
 - 5* leaves lobed or dissected..... DISSECTED V.
 - 4* 12-16 involucral bracts,
 - 6. linear to narrow linear-lanceolate leaves..... 14 BRACT V.
 - 6* fine linear to filiform leaves (measure)..... *S. burchellii*

- 1# Madagascan plants, 12-16 involucral bracts,
oblanceolate to broad lanceolate..... OBLANCEOLATE V

Electrophoretic Procedures.

Randomly chosen seeds from collections were placed on moist filter paper (Whatman® No. 1) and imbibed with 300 ppm gibberellic acid (GA₃) to stimulate seedling germination (overcome possible innate dormancy). Seedlings were then transplanted into pots in a standard 1:1 gravel/peat mixture and fertilised with slow release Osmacote® pellets. Seedlings were grown to at least the 4 leaf stage (excluding cotyledons) to provide sufficient leaf material for protein extraction and to increase levels of enzyme activity to allow good staining. Plants were harvested and/or destroyed upon maturity of first flowers to prevent any seed production. Plants were kept in Quarantine facilities at the University of Sydney (AQIS specifications) to prevent escape of potentially weedy plants into surrounding areas.

The electrophoretic procedures used were mostly as described in Soltis et al. (1983). Actively growing young leaf material (usually 1 to 3 leaves) was ground on ice in two drops of Tris-HCl buffer (0.01M EDTA, KCl, MgCl₂·4H₂O, pH=7.5) containing 0.1% v/v 2-mercaptoethanol and 10% w/v PVP 40000 (polyvinylpyrrolidone) to inhibit the breakdown of proteins by secondary metabolites (Kephart 1990). Protein extracts were then absorbed onto 10 mm by 3 mm filter paper wicks (Whatman No. 1®) and loaded onto 13% starch gels. Gels were made according to procedures described by Kephart (1990).

Electrophoresis was conducted by applying an electric current across gels to separate out charged proteins or isozymes. BIO-RAD Power pacs were used to apply a constant current of 50-60mA through gels via platinum electrodes in filled buffer trays and electrolyte bridges made of Wettex® cloth. Isozymes migrated towards the positive electrode.

Electrophoresis was run for six hours at a time to allow for sufficient protein separation, at 4°C (refrigerated with ice packs) to prevent build up of heat through gel resistance, which could cause denaturation of proteins. When electrophoresis was complete gels were cut into 4 horizontal slices to be stained for four different enzyme systems. Two gels were run simultaneously to stain for a total of seven enzyme systems. Details of the enzyme systems used for this analysis are found in Table 2.

Genetic interpretation of stained isozyme bands followed Pasteur et al. (1988). Alleles were interpreted as bands with different migration rates (different distance from the origin) after a six hour run. Single bands were interpreted as homozygotes for a particular allele (e.g. AA), while triple bands (or double bands in the case of shikimate dehydrogenase, Pasteur et al. 1988) were interpreted as heterozygotes (AB) following simple Mendelian patterns of inheritance (Warwick 1991). Groups of independent bands were interpreted as genetic loci. Full documentation of the genetic basis of variation through controlled crosses and segregation studies (Warwick 1991) were not attempted in this study.

The absence of bands for loci where they had previously been expressed were interpreted as "null" alleles so that otherwise complete data could be included in analyses (BIOSYS-1 cannot analyse populations with missing data, Swofford and Selander 1989). Null alleles were interpreted as a non-staining enzyme polymorphism. We assumed that failure of

expression of a loci was due to one allelic polymorphism. However, as some null alleles were probably not genetically based, but caused by environmental fluctuations, differentiation between populations in some cases may be exaggerated by conditions in which plants were kept. Controlled environment facilities, which may have reduced variability in enzymatic activity, were not available for this study.

Table 2. Enzyme systems used for isozyme analysis and number of loci for each for *S. madagascariensis* and *S. lautus*.

Enzyme System	Buffer System	No. Loci
Aspartate Amino Transferase (AAT)	Boric acid/Tris-Citrate*	1
Phosphoglucoisomerase (PGI)	Boric acid/Tris-Citrate*	2
Triosephosphate isomerase (TPI)	Boric acid/Tris-Citrate*	1
Isocitrate Dehydrogenase (IDH)	Tris/ Citric Acid #	1
Malate Dehydrogenase (MDH)	Tris/ Citric Acid #	2
6-Phosphogluconate Dehydrogenase (6-PGD)	Tris/ Citric Acid #	2
Shikimate Dehydrogenase (SkDH)	Tris/ Citric Acid #	1
	Total	10

Inconsistent or unsuccessful systems: Acid Phosphatase (AP), Aconitase (ACT), Esterase (EST), Fructose-1,6-diphosphatase (F-1,6-DP), Glucose-6-phosphate dehydrogenase (G-6-PDH), Glutamate dehydrogenase (GDH), Glyceraldehyde-3-phosphate dehydrogenase (G-3-PDH), Leucine aminopeptidase (LAP), Malic enzyme (ME) and Phosphoglucomutase (PGM).

* Electrode buffer 0.100 M NaOH, 0.300 M Boric Acid; gel buffer 0.015 M Tris, 0.004 M Citric Acid

Electrode buffer 0.223 M Tris, 0.086 M Citric Acid; gel buffer 0.008 M Tris, 0.003 M Citric Acid

Data analysis.

Data analyses of isozyme genotype arrays were performed using BIOSYS-1, a FORTRAN-77 package (Swofford and Selander 1981, 1989). Individual genotype data was entered and used to calculate allele frequencies. Allele frequencies were then used to calculate gene diversity

measurements, allele distribution classes, genetic diversity within and among populations, hierarchical genetic distance statistics and genetic relatedness of populations to one another.

BIOSYS-1 was used to calculate gene diversity statistics. Genetic diversity was quantified as the number of alleles per locus (A), percentage of loci which were polymorphic (P), observed heterozygosity (H_o), and expected heterozygosity in a panmictic or random mating system (H_e). These were calculated independently for each population, and mean values calculated for Australian, South African and Madagascan populations.

Genetic diversity within and between populations were calculated using the hierarchical gene diversity statistics of Nei and Wright (Swofford and Selander 1989). Nei's (1973) mean within population diversity (H_s) was taken as the mean expected panmictic (random mating in an infinite population) heterozygosity (H_e) (Swofford and Selander 1989). The proportion of genetic diversity found between populations (G_{ST}) (Nei 1973) was calculated as Wright's F_{ST} , defined by the following formula:-

$$1-F_{IT}=(1-F_{IS})(1-F_{ST})$$

where F_{IT} and F_{ST} are the fixation indices of individuals relative to the species and its populations, and F_{ST} measures the amount of differentiation among populations (Swofford and Selander 1989). This was calculated separately for each locus and averaged for both species. Total species genetic diversity (H_T) (Nei 1973) was calculated as Wright's total limiting variance averaged over all loci (Swofford and Selander 1989).

Relationships between southern African, Madagascan and Australian populations were determined using hierarchical analysis of genetic distances between groups identified on the basis of distribution and morphology. Nei (1978) unbiased genetic distance was used to calculate a matrix of similarity/distance between groups at all three levels of the previously constructed hierarchy (Country, Region, Variant).

Relationships among individual populations were determined using cluster analyses. Unweighted pair group method with arithmetic averaging (UPGMA), using Nei's (1978) unbiased genetic distances between population pairs was used to construct clusters of related

populations. An UPGMA cluster was also constructed to directly assess relationships between regions, pooling all accessions into one population for each region for the analysis. The robustness of groups formed was tested by comparing results with cluster analyses using the complete linkage method, the single linkage method, the weighted pair group method (Swofford and Selander 1989). Rogers (1972) genetic similarity, Nei's (1972) genetic identity and modified Rogers distance (Wright 1978) (cited by Swofford and Selander 1989) were also used for comparison with the original method to test for group robustness.

Due to limits on population number which can be analysed by BIOSYS-1 (40 populations per analysis) not all were included in the final analysis. Populations were excluded for which viable seed was not collected, for which five or less plants were analysed (where sampling effects may have influenced results out of proportion to numbers) and for populations for which no voucher specimens were available. Accessions for which vouchers were not sent from Africa were also excluded from analysis as they could not be assigned taxonomic or varietal status.

Results

Genetic Diversity Statistics

Mean gene variability measurements are consistently higher for South African and Madagascan populations than for Australian fireweed (Table 3). Greater between population genetic variation is also found in South Africa and Madagascar (30% and 18%) compared to that found in Australia (12%) reflecting observations of morphological variability in these regions in previous studies (Hilliard 1977, Marohasy 1993, Radford et al. 1995, Radford 1997). Reduced overall genetic variation and low differentiation between populations in Australia as compared to native regions is consistent with recent introduction of relatively small numbers of individuals and common ancestry of exotic populations, leading to reduced genetic diversity.

Table 3. Gene diversity statistics of Australian fireweed and variants from South Africa and Madagascar. Calculations were performed using BIOSYS-1 (Swofford and Selander 1989).

Gene Statistics	South Africa	Madagascar	Australia
(Population Number)	(27)	(7)	(6)
(Mean plants/population)	(7.6)	(8.4)	(7.7)
Alleles per Locus (A)	1.6	1.6	1.5
Percentage loci polymorphic (P)	47%	49%	45%
Observed heterozygosity (H_o)	0.174	0.179	0.149
Within population diversity /Exp. heterozygosity ($H_s=H_e$)	0.215	0.213	0.205
Total genetic diversity (H_T)	0.292	0.350	0.261
Between population diversity (G_{ST})	31%	18%	12%

Hierarchical Genetic Relationships

Relationships between groups of the *S. madagascariensis* complex are found in Tables 4, 5 and 6. Average genetic distance between countries of origin (the highest level in the hierarchy) are found in Table 4. Australian fireweed populations are much more closely related to plants from South Africa (genetic distance of 0.086) than to plants from Madagascar (0.132). Average population distance (0.106) and the range of population differentiation within South Africa (0-0.475) was greater than within Madagascar (0.051 and 0-0.131) or Australia (0.063 and 0.013-0.143), indicating greater differentiation among populations here.

Table 4. Matrix of genetic distance coefficients averaged by country of origin. Genetic distance was calculated using Nei (1978) unbiased genetic distance using BIOSYS-1 (Swofford and Selander 1989). Bracketed figures represent the range of genetic distance between accession pairs.

Country	1. South Africa	2. Madagascar	3. Australia
1. South Africa	0.106 (0.000-0.475)		
2. Madagascar	0.119 (0.009-0.328)	0.051 (0.000-0.131)	
3. Australia	0.086 (0.000-0.377)	0.132 (0.036-0.254)	0.063 (0.013-0.143)

Genetic distances between regions in southern Africa and Australia are recorded in Table 5. Populations from Natal are the more closely related to Australian populations (genetic distance 0.072) than to populations from the Eastern and Western Cape (0.118), south east Madagascar (0.124) and south west Madagascar (0.152). While regions in Madagascar are fairly similar genetically (genetic distance 0.061), regions within South Africa (0.122) and between South Africa and Madagascar (0.112, 0.126, 0.121 and 0.143) are more genetically distant.

Table 5. Matrix of genetic distance coefficients averaged by region of origin. Genetic distance was calculated using Nei (1978) unbiased genetic distance using BIOSYS-1 (Swofford and Selander 1989).

Region (popn. no.)	Natal	E & W Cape	SE Madagascar	SW Madagascar
Natal (19)	0.083			
E & W Cape (8)	0.122	0.156		
SE Madagascar (5)	0.112	0.121	0.046	
SW Madagascar (2)	0.126	0.143	0.061	0.000
Australia (6)	0.072	0.118	0.124	0.152

Within South Africa plants most closely related to Australian fireweed populations are those identified as *S. madagascariensis* (unbiased genetic distance 0.071 in Natal and 0.070 in Eastern and Western Cape Provinces), *S. inaequidens* (0.030) and unidentified plants from Natal (0.066) (Table 6). *S. skirrhodon* plants were found to be genetically more distant from Australian fireweed (0.096) while the DISSECTED and 14 BRACKT variants from the Cape were still further removed.

Table 6. Average genetic distance between Australian and Southern African variants of the *S. madagascariensis* complex. Genetic distance was calculated using Nei (1978) unbiased genetic distance using BIOSYS-1 (Swofford and Selander 1989).

Taxa/Variant	Population Number	Genetic Distance from Australian Accessions
Natal		
<i>S. madagascariensis</i>	13	0.071
<i>S. inaequidens</i>	1	0.030
<i>S. skirrhodon</i>	3	0.096
UNKNOWN sp.	2	0.066
Eastern and Western Cape		
<i>S. madagascariensis</i>	3	0.070
DISSECTED VARIANT	3	0.142
14 BRACKT VARIANT	2	0.155

Cluster Analysis

UPGMA cluster analyses show strong geographic segregation of plants into groups made up of Australian, Natal and Eastern Cape accessions (top two major clusters), and groups made up predominantly of Madagascan populations (fourth and fifth clusters) (Figure 1). These groups were separated by approximately 12% variation. Only one Australian population was found to cluster within a group made up predominantly of Madagascan populations (Breakwell). The majority of accessions identified as *S. madagascariensis* were also found to cluster together in the top two clusters of the dendrogram. Two of the three *S. skirrhodon* accessions were found to fall into a different cluster (third down). An out-group was formed by Alexandria from Eastern Cape (divergence of >25%). High genetic distance may be the result of sampling effect caused by the small number of plants analysed ($n=5$) in this population. Apart from Alexandria, all accessions were found to diverge by a maximum of 15% with no major genetic disjunctions, indicating that the *S. madagascariensis* complex is quite closely related overall.

Figure 2 shows a UPGMA cluster analysis of genetic relationships between regions. This clearly shows close affinities of Australian fireweed with plants from Natal. Though Eastern Cape plants are also closely related to Australian populations, Madagascan plants are clearly separated on the basis of genetic distance measurements.

Analyses using alternative clustering methods (e.g. complete linkage, single linkage and weighted pair group method) and using alternative genetic similarity/distance calculations resulted in similar groupings to those found using UPGMA. This indicates that groups formed are robust and non-sensitive to the cluster analysis used.

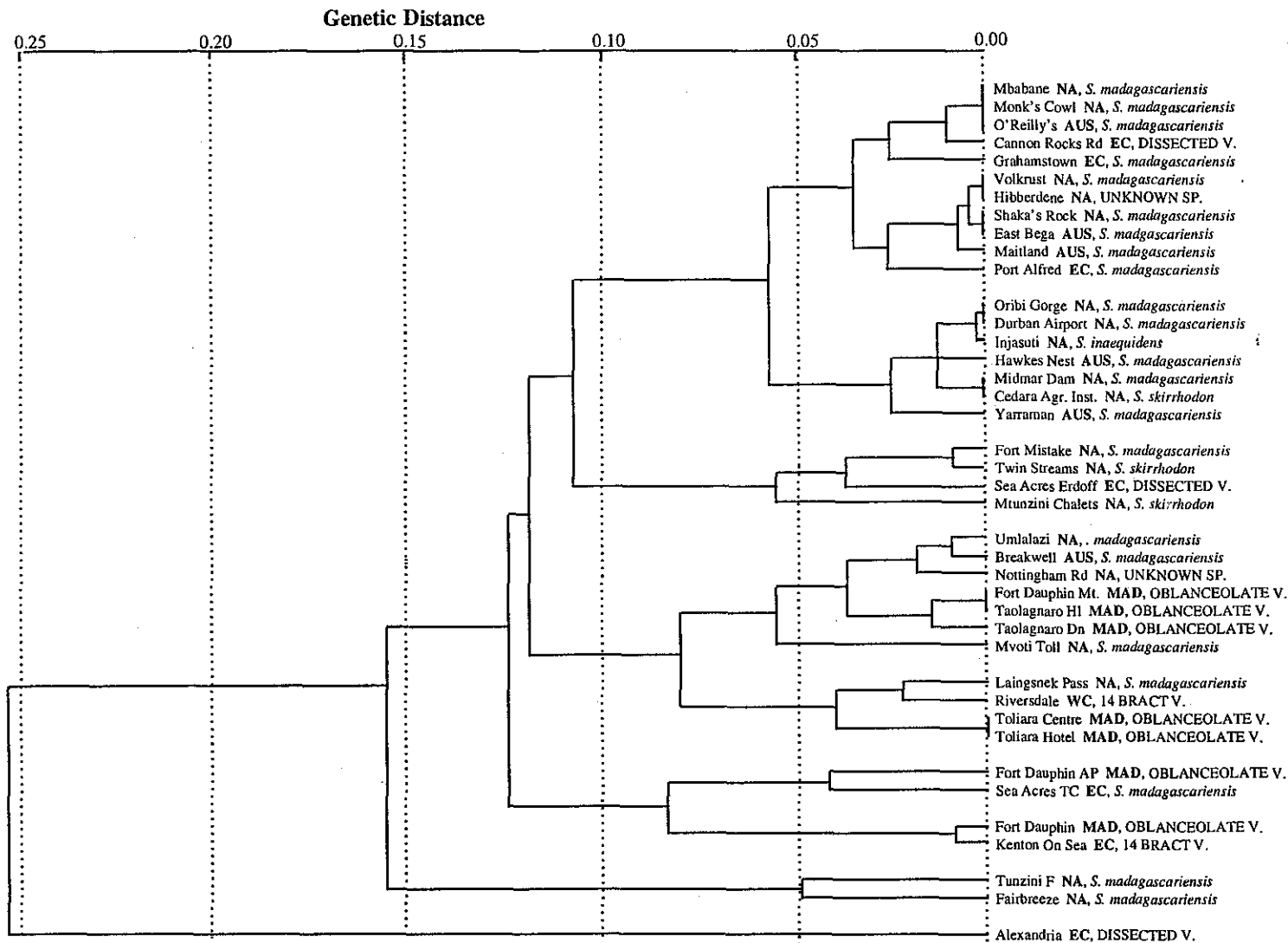


Figure 1. Cluster analysis using UPGMA of Nei (1978) unbiased genetic distance (Swofford and Selander 1989)
 NA=Natal, EC=Eastern Cape, MAD=Madagascar.

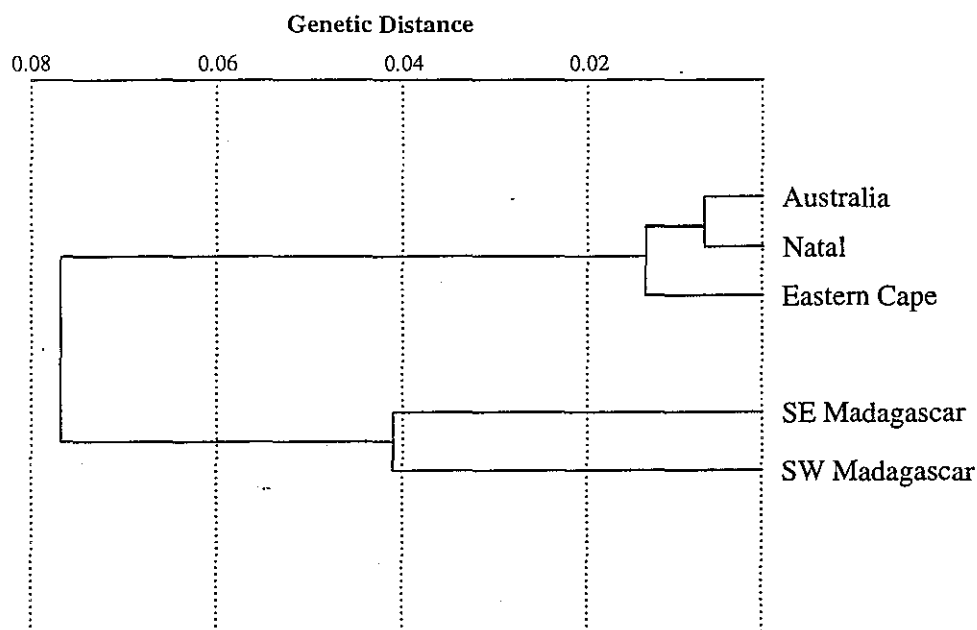


Figure 2. Cluster analysis of regions using unweighted pair group method using Nei (1978) unbiased genetic distance (Swofford and Selander 1989).

Discussion

Australian populations of fireweed were found to be genetically most closely related to *S. madagascariensis*/*S. inaequidens* from Natal in South Africa. Plants from Natal had greater genetic similarity to Australian plants than either Madagascan or Eastern/Western Cape populations. Although a small number of Eastern Cape populations of *S. madagascariensis* have equally close affinities with Australian populations (Table 6), Natal appears to be the major centre for the distribution of closely related *S. madagascariensis* populations in the region. Because few populations in the Eastern Cape have close affinities with fireweed, care should be taken during future biocontrol collections, to differentiate *S. madagascariensis* from other variants in this region (e.g. DISSECTED V.).

Isozyme results are similar to results and observations from previous studies. ITS1 DNA sequence data showed that *S. madagascariensis* and *S. inaequidens* specimens from a single location in South Africa (Pietermaritzburg) were more closely related to Australian plants than plants from Fort Dauphin in Madagascar (Scott et al. in press). Morphological observations of plants collected during this and previous studies (Radford et al. 1995, Muller, Report 1996) also indicate that Australian plants are morphologically

more similar to those from Natal and Swaziland than to plants from Madagascar (Marohasy 1989).

S. skirrhodon and *S. burchellii* are clearly more distantly related to Australian fireweed than with *S. madagascariensis* in Natal/Swaziland. *S. skirrhodon*, differentiated using leaf characters described above (see Key), was found to be genetically more distant from fireweed than *S. madagascariensis* (Table 6). Although populations of *S. burchellii* were not able to be analysed for isozyme variation because viable seed was not available, plants are clearly differentiated on the basis of involucral bract number (14 compared to 21) and leaf characters (fine linear or filiform compared to broad linear-lanceolate). *S. burchellii* is also found away from the central part of *S. madagascariensis* distribution in the Eastern Cape region.

Future biocontrol collections should target *S. madagascariensis* populations within Natal. Australian and Madagascan predators/pathogens of *S. madagascariensis* so far studied have proved to have low host specificity (Holtkamp and Hosking 1993, McFadyen and Sparks 1996). The Natal region, where *S. madagascariensis* is endemic and common is much more likely to have highly adapted and specialised predators/pathogens than areas such as the Eastern Cape where most populations are not so closely related to Australian fireweed.

Future taxonomic studies are needed to elucidate relationships between *S. madagascariensis* and *S. inaequidens* within South Africa. This study, and DNA analyses described by Scott et al. (in press) show that *S. inaequidens* is genetically as close, or more closely related to Australian fireweed than to plants identified as *S. madagascariensis*. This has important implications for resolution of the nomenclature of Australian fireweed, but also for future fine tuning of search efforts for biological control agents for fireweed and weedy plants known as *S. inaequidens* in Europe. Although chromosome studies of European specimens of *S. inaequidens* clearly separate it from Australian specimens of *S. madagascariensis* ($2n=40$, Harland 1955 and Chichiricco et al. 1979, instead of $2n=20$, Radford et al. 1995) morphological differentiation is problematic (Hilliard 1977). Genetic analysis and examination of only one accession identified as *S. inaequidens* in this study would not allow us to reliably separate these groups.

Acknowledgments.

Financial support for this project was provided by the Meat Research Corporation. Thanks are due to Sue Fiffer for electrophoretic work performed. Thanks also to Dr Peter Michael for taxonomic determinations of some of the South African specimens.

References

- Belcher, R.O. (1993). The '*Senecio* aff. *lautus*' complex (Asteraceae) in Australia I. Criteria for exclusion of lautusoid *Senecio* of Australia from *S. lautus* sensu stricto of New Zealand. *Australian Systematic Botany* 6, 359-363.
- Chichiricco, G., Frizzi, G. and Tammaro, F. (1979). Numeri cromosomici per la Flore Italiana. *Inform. Bot. Ital.* 11, 3-35. In 'Index to Plant Chromosome Numbers 1979-1981', ed. P. Goldblatt, pp. 598-601. (Missouri Botanic Gardens: St Louis.)
- Harland, S.C. (1955). The experimental approach to the species problem. In 'Species Studies in the British Flora', ed. J.E. Lousley, pp. 16-20. (Botanical Society of the British Isles: London.)
- Hilliard, O.M. (1977). 'Compositae in Natal,' (University of Natal Press: Pietermaritzburg.)
- Holtkamp, R.H. and Hosking, J.R. (1993a). Insects and diseases of fireweed, *Senecio madagascariensis*, and the closely related *Senecio lautus* complex. In Proceedings I. 10th Australian Weeds Conference, and 14th Asian-Pacific Weed Sciences Society Conference, pp. 104-106.
- Kephart, S.R. (1990). Starch gel electrophoresis of plant isozymes: a comparative analysis of techniques. *American Journal of Botany* 77, 693-712.
- Kirkland, P.D., Moore, R.E., Walker, K.H., Seaman, J.T. and Dunn, S.E. (1982). Deaths in cattle associated with *Senecio lautus* consumption. *Australian Veterinary Journal* 59, 64.
- McFadyen, R. and Sparks, D. (1996). Biological control of fireweed. Proceedings of the Eleventh Australian Weeds Conference, pp. 305-308.
- Marohasy, J.J. (1989). A survey of Fireweed (*Senecio madagascariensis* Poir.) and its natural enemies in Madagascar with a view to biological control in Australia. *Plant Protection Quarterly* 4, 139-140.
- Marohasy, J.J. (1993). Are we justified in considering fireweed (*Senecio madagascariensis*) an exotic? In Proceedings II. 10th Australian and 14th Asian-Pacific Weed Conference, Brisbane, Australia, pp. 122-127.
- Michael, P.W. (1981). Alien plants. In 'Australian Vegetation', ed. R.H. Groves, pp. 44-46. (Cambridge University Press: Cambridge.)
- Nei, M. (1973). Analysis of gene diversity in subdivided populations. *Proceedings of the National Academy of Science U.S.A.* 70(12), 3321-3323.

- Nei, M. (1978). Estimation of average heterozygosity and genetic distance from a small number of individuals. *Genetics* **89**, 583-590.
- Pasteur, N., Pasteur, G., Bonhomme, F., Catalan, J. And Britton-Davidian, J. (1988). 'Practical Isozyme Genetics.' (translator: M. Cobb). (Ellis Horwood Limited: Chichester.)
- Radford, I.J. (1997). Impact Assessment For The Biological Control of *Senecio madagascariensis* Poir. (Fireweed). PhD Thesis, University of Sydney.
- Radford, I.J., King, D. and Cousens, R.D. (1995a). A survey of *Senecio madagascariensis* Poir. (fireweed) density in pastures of coastal New South Wales. *Plant Protection Quarterly* **10**, 107-111.
- Radford, I.J., Liu, Q. and Michael, P.W. (1995b). Chromosome counts for the Australian weed known as *Senecio madagascariensis*. (Asteraceae). *Australian Systematic Botany*, **8**, 1029-1033.
- Radford, I.J., King, D. and Cousens, R.D. (1997 Unpublished Report). Pasture production impact of *Senecio madagascariensis* (fireweed). Dairy Research and Development Final Report.
- Sindel, B.M. (1986). The ecology and control of fireweed (*Senecio madagascariensis* Poir.). *Plant Protection Quarterly* **1**, 163-172.
- Sindel, B.M. (1996). Impact, ecology and control of the weed *Senecio madagacariensis* in Australia. Proceedings of the International Compositae Conference, Kew **2**, 339-349.
- Sindel, B.M. and Michael, P.W. (1988). Survey of the impact and control of fireweed (*Senecio madagascariensis* Poir.) in New South Wales. *Plant Protection Quarterly* **3**, 22-28.
- Sindel, B.M. and Michael, P.W. (1992a). Spread and potential distribution of *Senecio madagascariensis* Poir. (fireweed) in Australia. *Australian Journal of Ecology* **17**, 21-26.
- Sindel, B.M. and Michael, P.W. (1992b). Growth and competitiveness of *Senecio madagascariensis* Poir. (fireweed) in relation to fertiliser use and increases in soil fertility. *Weed Research* **32**, 399-406.
- Swofford, D.L. and Selander, R. (1989). BIOSYS-1, a computer program for the analysis of allelic variation in population genetic and biochemical systematics. *Release 1.7 David Swofford Natural History Survey*.
- Swofford, D.L. and Selander, R.B. (1981). BIOSYS-1: a FORTRAN program for the comprehensive analysis of electrophoretic data in population genetics and systematics. *The Journal of Heredity* **72**, 281-283.
- Walker, K.H. and Kirkland, P.D. (1981). *Senecio lautus* toxicity in cattle. *Australian Veterinary Journal* **57**, 1-7.
- Watson, R., Launders, T. and Macadam, J. (1984). Fireweed. Agfact P7.6.26. First Edition. Department of Agriculture New South Wales.
- Watson, R., Allan, H., Munnich, D., Launders, T. and Macadam, J. (1994). Fireweed. Agfact P7.6.26. Second Edition. NSW Agriculture.

2: Report on the 1996 CSIRO survey conducted in South Africa, Madagascar and Australia of *Senecio madagascariensis* (Poir) and associated potential biological control agents

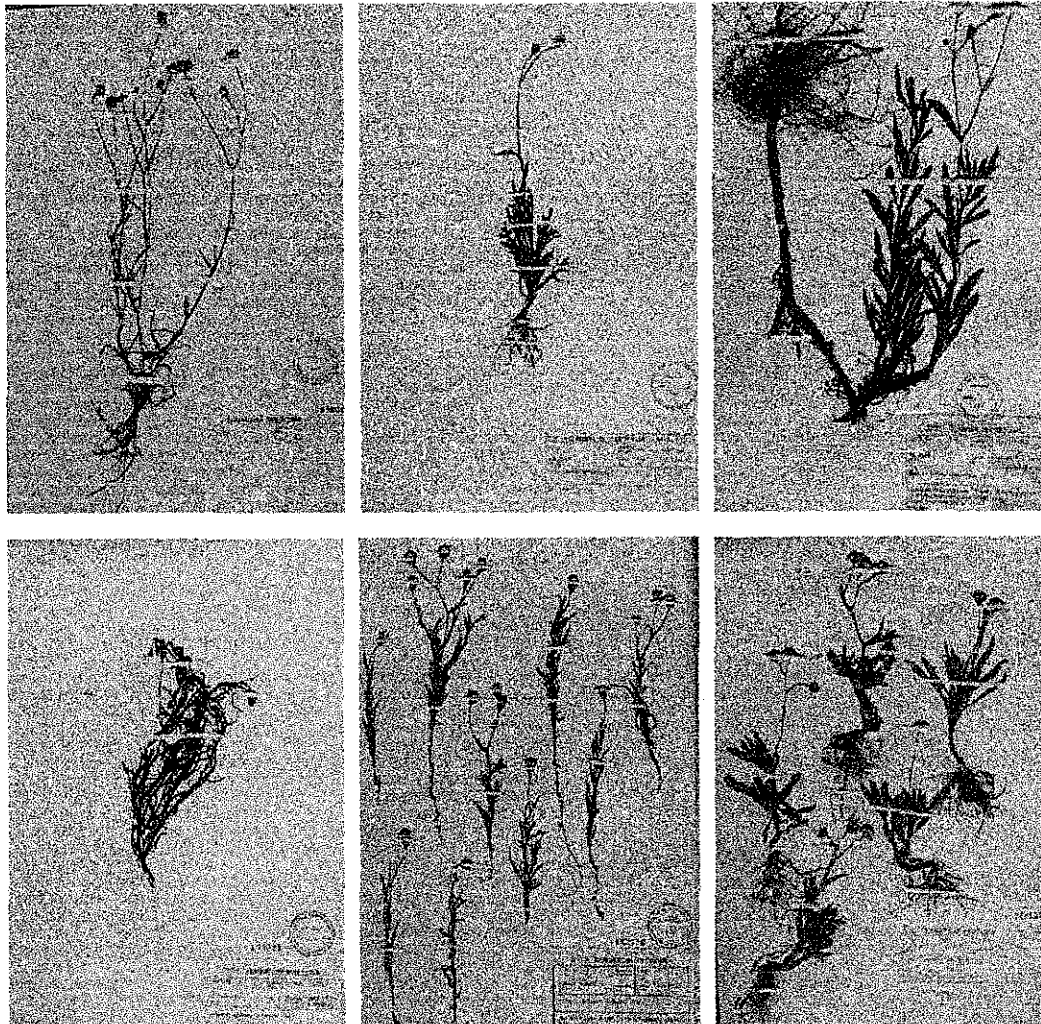
Petra Müller, CSIRO Entomology, Cape Town, South Africa

In 1989 the Australian Meat Research Council (MRC) commissioned a project with Queensland Lands Department entomologist J. Marohasy to find agents for the biological control of Fireweed (*Senecio madagascariensis* Poir) in Australia. Fireweed is a major weed of coastal pastures in New South Wales and southern Queensland where it reduces pasture growth and is toxic to livestock. Fireweed specimens from Australia were sent to O.M. Hilliard in South Africa who identified them as *Senecio madagascariensis* (Sindell 1989) and a biocontrol project was initiated on the basis of this taxonomic determination.

In 1996 MRC approached the CSIRO Division of Entomology to undertake a preliminary study of this target plant. The CSIRO Biological Control Unit in Cape Town, in conjunction with PPRI Pretoria, was contracted to do a survey of *S. madagascariensis* in southern Africa and Madagascar to establish the origin of the weed found in Australia. It is of prime importance in any biological control program to ensure that potential agents are associated with and are host specific only to the introduced target plant.

In early 1996 Petra Müller (CSIRO Entomology, Cape Town) visited most of the herbariums within southern Africa to study specimens labelled as *Senecio madagascariensis* Poir and other closely related *Senecio* species. Her opinion was that many of these specimens had been misidentified (Fig.1), hence she undertook to collect these closely related *Senecio* species from as many localities possible to clarify this taxon.

Fig 1. Examples of southern African herbarium specimens identified as *S. madagascariensis* (Poir) showing morphological variation.



In South Africa there is a distinct difference between leaf shape and florets of the closely related *S. skirrhodon* and that of *S. burchellii* and *S. madagascariensis*. There is a marked overlap in the distribution of *S. burchellii* and *S. madagascariensis*, making rapid differentiation of the two species difficult, however *S. burchellii* commonly has narrow thin pinnate leaves and when flowering, typically 12-13 involucre bracts on the florets as opposed to *S. madagascariensis* which has large variable, often auriculate leaves and 19-21 involucre bracts on the florets. In South African herbariums, *S. inaequidens* and *S. madagascariensis* appear to have been grouped under one species. (Previously *S. inaequidens* was grouped with *S. burchellii*). There is a distinct similarity between *S. madagascariensis*, *S. inaequidens* and the Australian *S. lautus* complex and

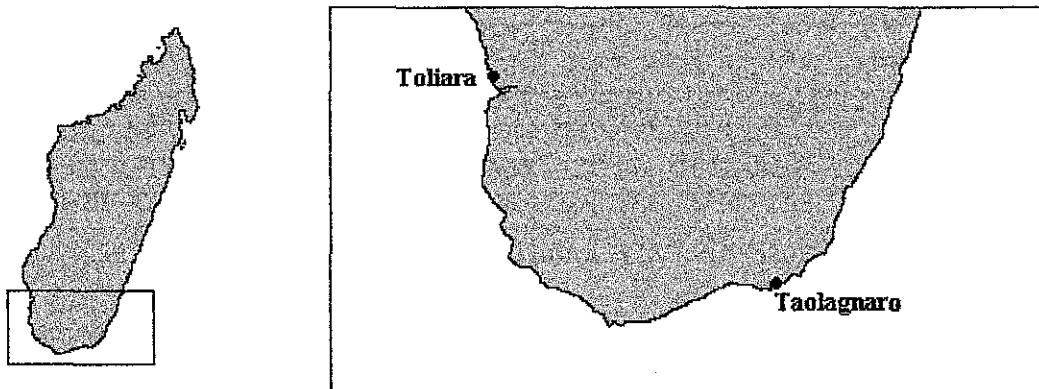
in most cases plants cannot be separated with certainty on gross morphological attributes. Examples of *Senecio* habitat in South Africa and Madagascar are shown in Fig. 2., and maps of all collection sites are shown in Fig.3.

Fig. 2a-b. (a) Typical *Senecio* sampling regions showing common roadside occurrence in Madagascar, (b) and in a field in South Africa.

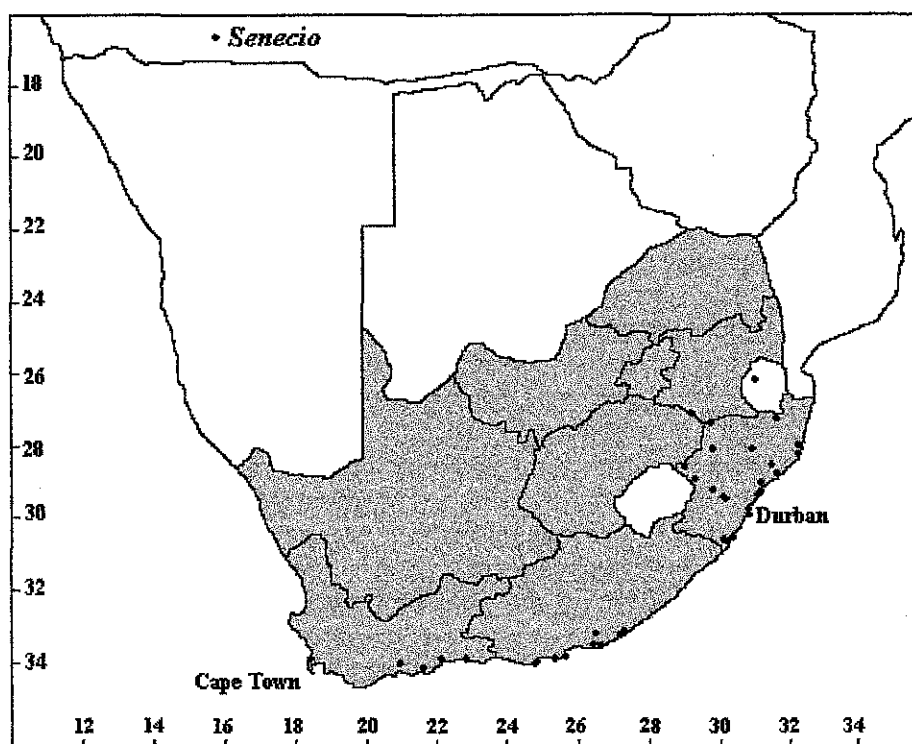
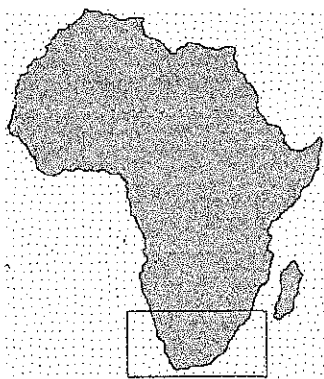


Fig. 3a-b. Distribution maps of 1996 *Senecio* survey sites.

(a). Madagascar: South west and south east



(b). Southern Africa: Western and Eastern Cape, Kwazulu-Natal and Swaziland.



In Madagascar and South Africa sampling was done at every available opportunity. At least five or more replicates of a range of the most physically variable plants per site were collected, pressed and labelled with reference numbers and locality details using GPS (Appendix I). This will enable easy location of sites for the assessment of biological control agents, once the taxonomy of *S. madagascariensis* and the origin of the Australian material have been resolved. At least 20 seed heads of each of these sampled plants were collected for the electrophoresis study at the University of Sydney and a larger mix of seed heads from many different plants per site was collected to determine population diversity. Any potential biological control agents (insects or pathogens) present were also recorded or collected.

Within species of the genus *Senecio* there is extreme variability in leaf shape and plant size according to locality, habitat, soil type and regional rainfall. As with most weeds, *S. madagascariensis* is well adapted to disturbance and stress which contributes to marked variability in physical appearance. Most of the specimens in herbariums within South Africa show a wide range of different morphological features, particularly in leaf shape. It was thus necessary to obtain fresh field material from Australia as a comparison for target plants in the survey. Australian collection sites of *S. madagascariensis* are shown in Fig. 4, and a typical infestation is shown in Fig. 5.

Fig. 4. Collection sites of *Senecio madagascariensis* in Australia (1996)

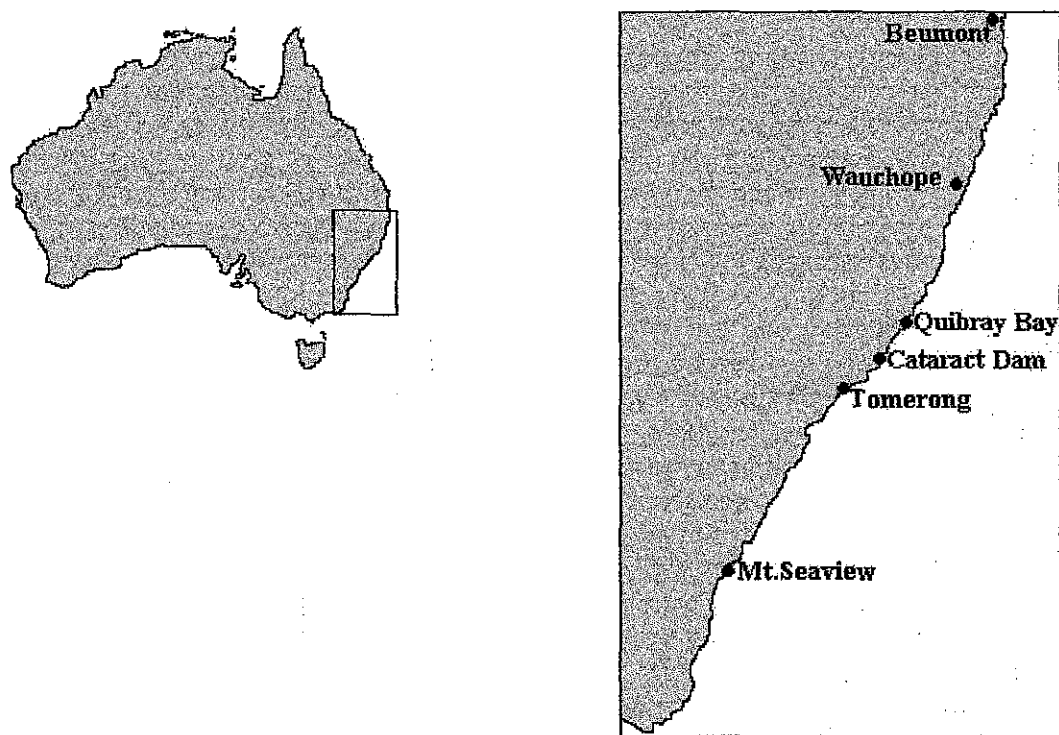


Fig. 5. *Senecio madagascariensis* in a pasture in New South Wales, Australia



The CSIRO collection resulting from this study is currently housed in the Bolus Herbarium at the University of Cape Town, with duplicate specimens of many of the collections housed at the University of Sydney where isozyme analyses are being undertaken. Specimens from Australia, Madagascar and South Africa have been incorporated into this study to make it the most comprehensive investigation into *Senecio madagascariensis* to date (Fig. 6). A morphometric study on herbarium material together with the isozyme results should clarify the status of all these species, as well as answer the key question concerning the origin of Australian *S. madagascariensis*.

Fig. 6a-b. (a) Australian herbarium material consulted for identification purposes, and (b) part of the southern African collected material housed at CSIRO in Cape Town



Ian Radford (University of Sydney) has clearly differentiated Australian *S. madagascariensis* from the native *S. lautus* using electrophoretic techniques. (Leon Scott of the Queensland University of Technology also undertook a DNA study of the two species using ITS1 sequencing, the results of which suggest hybridisation may be occurring between the Australian *S. lautus* and introduced *S. madagascariensis*). Preliminary results from the current isozyme study of all the collected material thus far seems to indicate that Kwazulu-Natal is the most likely area of origin of *S. madagascariensis* in Australia (I Radford, pers. comm.). The status of the Madagascan material will not be known until the isozyme analyses are complete, but on morphometric grounds (particularly number of bracts) it appears unlikely that it matches *S. madagascariensis* in Australia as closely as does the Kwazulu-Natal material. In conclusion, on completion this study will result in a better clarification of the *Senecio* taxa, providing more reliability in the identification of the target weed *S. madagascariensis* for future assessment of potential biological control agents. In particular, the most likely source of origin of Australian Firweed (*S. madagascariensis*) will be identified.

Notes on potential biological control agents

A rust fungus, *Puccinia* sp. has been observed on material collected from both South Africa and Madagascar. (*Puccinia lagenophorae* has been identified on Australian material and has not been recorded from South Africa). In most cases the infections have been prolific and plants were noted to be stressed and in bad condition (Fig. 7-8). This fungus seems promising as a potential biological control agent, however a thorough investigation will have to be made to differentiate and identify the *Puccinia* species for any potential testing. Dr. L. Morin (CSIRO Entomology) has prepared slides of material from all three countries for preliminary identification purposes.

Fig 7a-c. (a) *Senecio* sp. heavily infected with *Puccinia* sp. in Madagascar, (b) *Puccinia* sp. damage on the stem of *S. madagascariensis* in South Africa and (c) a microscopic view of aecia of *Puccinia* sp.

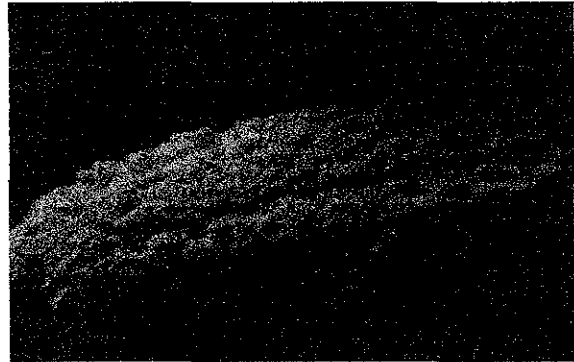


Fig.8. Rust fungus damage on *S. madagascariensis* plant in Kwazulu-Natal, South Africa.

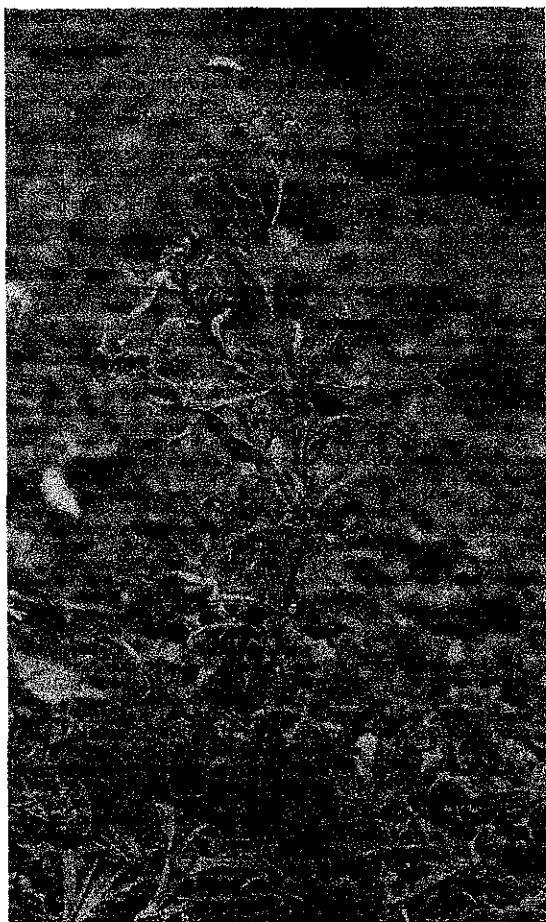


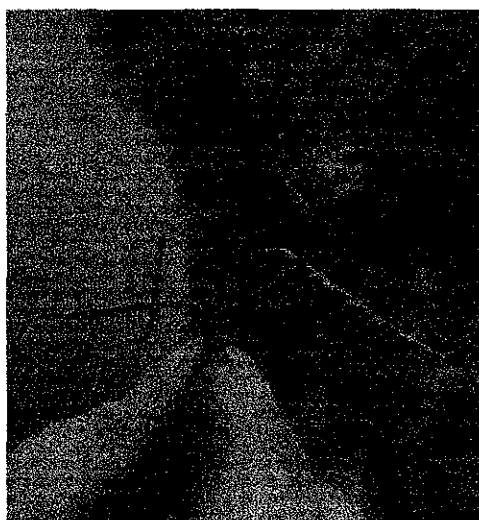
Fig 9. Leaf miner damage on *Senecio sp.* in Madagascar.



Surveying in both south west and south east Madagascar yielded quite a few sites where *Senecio* plants were attacked by an as yet unidentified leaf miner (Fig. 9). All plants in

the immediate surrounding region were checked for similar leaf miner damage with no success so it is likely that this insect is probably specific to the *Senecio* species. Unfortunately permits (which are extremely difficult to obtain), were only issued for restricted botanical sampling, so this insect could not be collected for rearing and identification. In South Africa, and particularly from Kwazulu-Natal, a seed-feeding tephritid (as yet unidentified) has been reared from numerous flower head collections. A stem tip boring cerambycid (Fig. 10) has been found in the field but it has not yet been reared through for identification. Numerous collections of flowers and seed from the Eastern Cape have also yielded a species of an as yet unidentified cecidomyiid (pinned for identification).

Fig. 10. Stem boring cerambycid damage on *S. madagascariensis* in Kwazulu-Natal, South Africa.

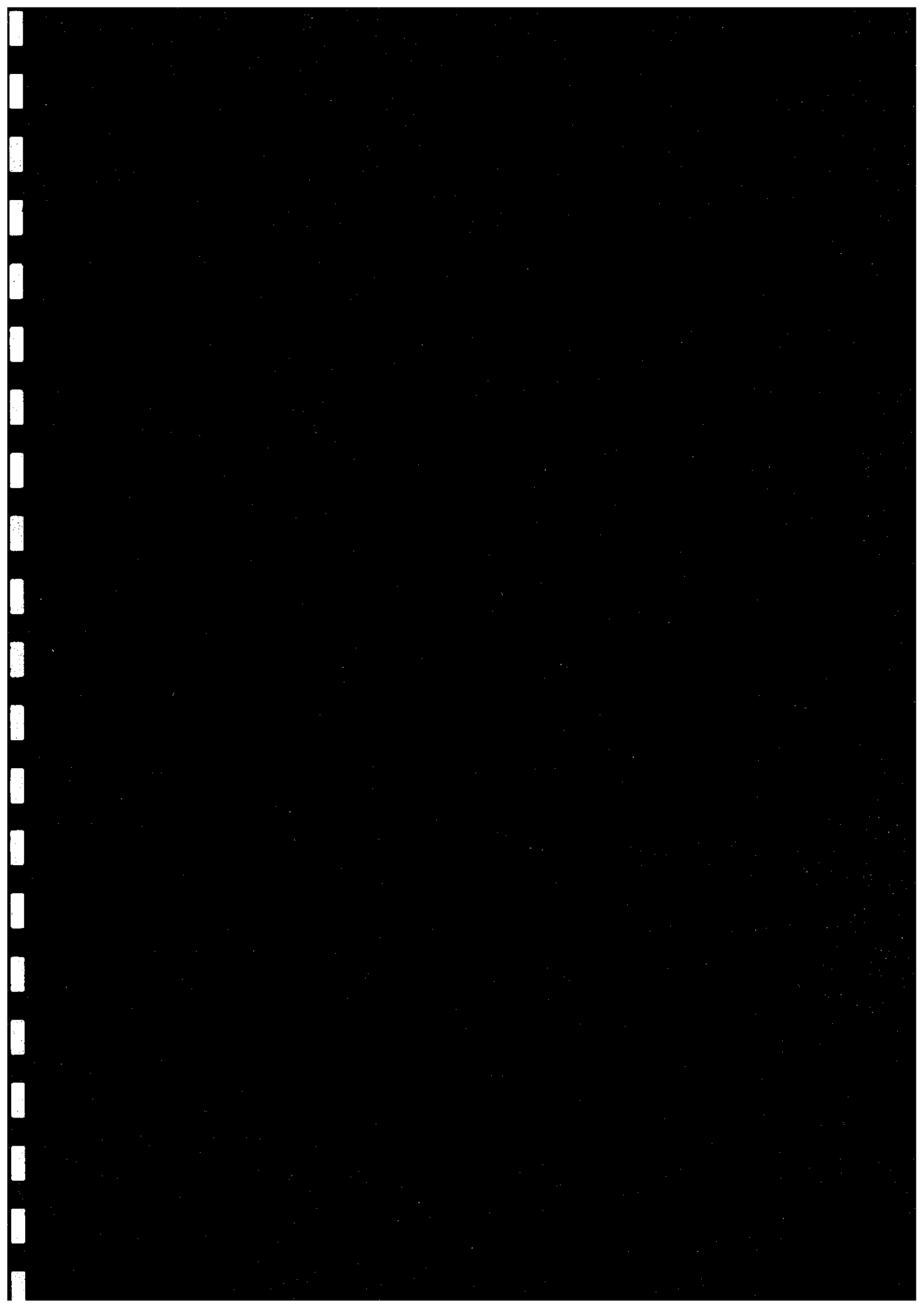


This work was funded by the MRC and aided with invaluable assistance from:

Australia: P.B.Edwards, S.Fiffer, R.H.Holtkamp, J.R.Hoskings, L.Morin, W.J. Wanjura and S.Young.

Madagascar: N.Raharijaona

South Africa: I.Davidson, J.H.Hoffmann, S.Neser, T.Olckers, A.B.R.Witt.



Appendix I : List of detailed herbarium records of all material collected.

Senecio madagascariensis Poir

CSIRO Biological Control Unit / Date: 15.07.1996
PPRI Natal (Acc.No. Sen 1) Coll: T.Olckers
Experimental Research Material

Loc: 29° 24' S 29° 57' E Nottingham Road, Natal, South Africa
Notes: On roadside. 2km along road to Fort Nottingham.
10 - 70 cm high branched herb. Small yellow flowers.
(1 x Seed Samples & 1 duplicate plant specimen collected.)

Senecio madagascariensis Poir

CSIRO Biological Control Unit / Date: 24.07.1996
PPRI Natal (Acc.No. Sen 2) Coll: T.Olckers
Experimental Research Material

Loc: 27° 27' S 29° 52' E Laingsnek Pass, Natal, South Africa
Notes: On road side. 23 km from Volksrust on road to Newcastle.
10 - 70 cm high branched herb. Small yellow flowers.
(1x Seed Samples & 1 duplicate plant specimen collected.)

Senecio madagascariensis Poir

CSIRO Biological Control Unit / Date: 24.07.1996
PPRI Natal (Acc.No. Sen 3) Coll: T.Olckers
Experimental Research Material

Loc: 28° 11' S 29° 58' E Fort Mistake, Natal, South Africa
Notes: Growing along stream, under bridge. 55km from Newcastle
on Road to Ladysmith. 10 - 70 cm high branched herb.
Small yellow flowers.
(1x Seed Samples & 1 duplicate plant specimen collected.)

Senecio madagascariensis Poir

CSIRO Biological Control Unit / Date: 06.08.1996
PPRI Natal (Acc.No. Sen 4) Coll: T.Olckers
Experimental Research Material

Loc: 29° 33' S 30° 16' E Cedara Agric. Institute, Natal, South Africa
Notes: Growing along fence of paddock on Farm. Small yellow flowers.
(1x Seed Samples & 1 duplicate plant specimen collected.)

Senecio madagascariensis Poir

CSIRO Biological Control Unit / Date: 22.08.1996
PPRI Natal (Acc.No. Sen 5) Coll: T.Olckers
Experimental Research Material

Loc: ?S?E Injasuti, Drakensberg Mountians, Natal, South Africa
Notes: On road side. High altitude. 4 km from Natal Parks Board Gate to
Reserve. 10 - 70 cm high branched herb. Small yellow flowers.
(1x Seed Samples & 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit / Date: 25.05.1996
PPRI Gauteng (Acc.No. Sen 6) Coll: S.Neser
Experimental Research Material

Loc: 27° 21' S 31° 47' E Mnyama, Sepong, Swaziland
Notes: Small yellow flowers.
(No Seed Samples collected but duplicate plant specimen with
flowers sent to I.Radford.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit / Date: 18.05.1996
PPRI Gauteng (Acc.No. Sen 7) Coll: S.Neser
Experimental Research Material

Loc: 27° 21' S 29° 25' E Volksrust, Natal, South Africa
Notes: On road side. 10 - 70 cm high branched herb. Small
yellow flowers.
(1x Seed Sample & 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit / Date: 17.06.1996
PPRI Gauteng (Acc.No. Sen 8) Coll: S.Neser
Experimental Research Material

Loc: 29° 03' S 29° 24' E Monks' Cowl, Natal, South Africa
Notes: Just above road to Sphinx in Drakensberg. High Altitude.
10 - 70 cm high branched herb. Small yellow flowers.
(1x Seed Sample & 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit / Date: 25.05.1996
PPRI Gauteng (Acc.No. Sen 9) Coll: S.Neser
Experimental Research Material

Loc: 26° 18' S 31° 10' E Mbabane, Swaziland
Notes: In Hills above the West edge of town. Branched herb.
Small yellow flowers.
(1x Seed Sample & 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit / Date: 19.04.1996
PPRI Gauteng (Acc.No. Sen 10) Coll: D.S.Hardy
Experimental Research Material

Loc: 24° 47' S 47° 10' E Fort Dauphin, S.E.Madagascar
Notes: Growing as wayside weed. Rare at this time of year.
(Note: P.A.Muller disagrees with "rarity" due to herbarium records
in Antananarivo indicating otherwise.)
Small yellow flowers. 15 - 19 involucre bracts.
(No seed samples. 1 poor duplicate plant specimen at PPRI Gauteng)

(No Acc.No. Sen 11 Allocated)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 12)
Experimental Research Material

Date: 15.07.1996
Coll: P.A.Muller
A.B.R.Witt

Loc: 28° 13' S 31° 04' E Umlalazi, Natal, South Africa
Notes: Common on dunes. 10 - 70 cm high branched herb.
Small yellow flowers. 19 - 22 involucre bracts.
(3 x Seed Samples & 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 13)
Experimental Research Material

Date: 28.06.1996
Coll: P.A.Muller
A.B.R.Witt

Loc: 34° 20' S 19° 02' E Rooi Els, Western Cape, South Africa
Notes: Common on roadsides. 10 - 70 cm high branched herb.
Very thin leaves. Small yellow flowers. 13 involucre bracts, perhaps
more like *S.burchelli* ?
(No seed samples. 1 duplicate specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 14)
Experimental Research Material

Date: 24.03.1996
Coll: P.A. Muller

Loc: 33° 57' S 18° 27' E Wynberg Park, Western Cape, South Africa
Notes: Abundant in pine forest. 10 - 70 cm high branched herb.
Very thin leaves. Small yellow flowers. 13 involucre bracts, perhaps
more like *S.Burchelli*?
(No Seed Samples, 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 15)
Experimental Research Material

Date: 20.03.1996
Coll: P.A. Muller

Loc: 34° 10' S 24° 50' E Sea Acres, Eastern Cape, South Africa
Notes: Few plants on edge of dunes/seashore. 10 - 70 cm high branched herb.
Leaves abundant and compact. Small yellow flowers. 13 - 16 involucre bracts.
(No Seed Samples, 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 16)
Experimental Research Material

Date: 21.03.1996
Coll: P.A. Muller

Loc: 34° 22' S 24° 25' E Port Elizabeth, Eastern Cape, South Africa
Notes: Common on roadsides in suburb. 10 - 70 cm high branched herb.
Small yellow flowers. 13 - 16 involucre bracts.
(No Seed Samples, 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 17)
Experimental Research Material

Date: 04.09.1996
Coll: P.B.Edwards
S.Young

Loc: 35° 03' S 150° 35' E Tomerong, New South Wales, Australia
Notes: Along roadside, near coast. Plants small. Small yellow flowers.
19 - 22 involucre bracts. (Aphids collected - Mary Carver to I.D.)
(No seed samples. Only specimen.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 18)
Experimental Research Material

Date: 12.09.1996
Coll: P.B.Edwards
S.Young

Loc: 34° 15' S 150° 48' E Cataract Dam, New South Wales, Australia
Notes: Collected in disturbed area within nature reserve. Only a few
plants. Flowering. Aphids and rust fungus present.
(No seed samples. Only specimen.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 19)
Experimental Research Material

Date: 26.06.1996
Coll: P.B.Edwards
W.J.Wanjura

Loc: 34° 01' S 150° 10' E Quibray Bay, New South Wales, Australia
Notes: On sand dunes by estuary. Small yellow flowers. 19 - 22
involucre bracts.
(No seed samples. Only specimen.)

(No Acc.No Sen 20 - Acc.No. Sen 31 have yet been allocated.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 32)
Experimental Research Material

Date: 20.08.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 29° 58' S 30° 58' E Durban Airport, Natal, South Africa
Notes: Abundant on road sides. 10 - 70 cm high branched herb. Leaves
pinnately lobed. Small yellow flowers. 19 - 22 involucre bracts.
(5x Seed Samples & 1 duplicate plant specimen.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 33)
Experimental Research Material

Date: 20.08.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 29° 55' S 31° 00' E Bluff Nature Reserve, Natal, South Africa
Notes: Common on road sides. 10 - 70 cm high branched herb. Leaves
pinnately lobed. Small yellow flowers. 19 - 22 involucre bracts.
(5x Seed Samples & 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 34)

Date: 20.08.1996
Coll: P.A. Muller

Experimental Research Material

A.B.R.Witt

Loc: 29° 31' S 31° 13' E Shaka's Rock, Natal, South Africa
Notes: Abundant in fields. Heavy Rust. 10 - 70 cm high branched herb.
Leaves pinnately lobed. Small yellow flowers. 19 - 22 involucre bracts.
(5x Seed Samples & 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 35)
Experimental Research Material

Date: 20.08.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 29° 02' S 31° 36' E Fairbreeze, Natal, South Africa
Notes: Abundant on road sides. 10 - 70 cm high branched herb.
Leaves pinnately lobed. Small yellow flowers. 19 - 22 involucre bracts.
(5x Seed Samples & 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 36)
Experimental Research Material

Date: 21.08.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 28° 58' S 31° 45' E Mtunzini Chalets, Natal, South Africa
Notes: Abundant on dunes. Heavy Rust. 10 - 70 cm high branched herb.
Leaves pinnately lobed. Small yellow flowers. 19 - 22 involucre bracts.
(5x Seed Samples & 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 37)
Experimental Research Material

Date: 21.08.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 28° 57' S 31° 45' E Mtunzini Field, Natal, South Africa
Notes: Abundant in sugar cane fields. 10 - 70 cm high branched herb.
Leaves pinnately lobed. Small yellow flowers. 19 - 22 involucre bracts.
(5x Seed Samples & 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 38)
Experimental Research Material

Date: 21.08.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 28° 22' S 32° 26' E St.Lucia, Natal, South Africa
Notes: Abundant on dunes. 10 - 70 cm high branched herb. Leaves
pinnately lobed. Small yellow flowers. 19 - 22 involucre bracts.
(5x Seed Samples & 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 39)
Experimental Research Material

Date: 21.08.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 28° 08' S 32° 25' E Fannies Island, Natal, South Africa
Notes: Abundant in Blue Gum Plantations. 10 - 70 cm high branched herb.

Leaves pinnately lobed. Small yellow flowers. 19 - 22 involucral bracts.
(5x Seed Samples & 1 duplicate plant specimen collected .)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 40)
Experimental Research Material

Date: 22.08.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 28° 43' S 31° 38' E On Empangeni Road(R34), Natal, South Africa
Notes: Abundant on road sides & in Sugar Cane Fields. 10 - 70 cm high
branched herb. Leaves pinnately lobed. Small yellow flowers.
(5x Seed Samples & 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 41)
Experimental Research Material

Date: 22.08.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 29° 24' S 31° 17' E Mvoti Toll Gate, Natal, South Africa
Notes: Abundant on road sides. 10 - 70 cm high branched herb. Leaves
pinnately lobed. Small yellow flowers. 19 - 22 involucral bracts.
(5x Seed Samples & 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 42)
Experimental Research Material

Date: 22.08.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 30° 44' S 30° 16' E Oribi Gorge Reserve, Natal, South Africa
Notes: Abundant on road sides. 10 - 70 cm high branched herb. Leaves
pinnately lobed. Small yellow flowers. 19 - 22 involucral bracts.
(5x Seed Samples & 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 43)
Experimental Research Material

Date: 23.08.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 30° 40' S 30° 31' E Hibberdene, Natal, South Africa
Notes: Common on road sides & dunes. 10 - 70 cm high branched herb.
Leaves pinnately lobed. Small yellow flowers. 19 - 22 involucral bracts.
(5x Seed Samples & 1 duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 44)
Experimental Research Material

Date: 23.08.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 29° 30' S 30° 12' E Midmar Dam Reserve, Natal, South Africa
Notes: Common on road sides. 10 - 70 cm high branched herb. Leaves
pinnately lobed. Small yellow flowers. 19 - 22 involucral bracts.
(5x Seed Samples & 1 duplicate plant specimen collected .)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 45)
Experimental Research Material

Date: 27.03.1996
Coll: R.H. Holtkamp

Loc: 31° 27' S 152° 44' E Wauchope, New South Wales, Australia
Notes: Occasional herb to 50 cm high. Flower heads yellow. Roadside
weed in alluvial soil 1km East of Wauchope. 19 - 22 involucre bracts.
(Duplicate to CANB.NSW Australia; TARCH No.5445)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 46)
Experimental Research Material

Date: 27.03.1996
Coll: R.H. Holtkamp

Loc: 31° 24' S 152° 16' E Mt.Seaview, New South Wales, Australia
Notes: Occasional herb to 50 cm high. Flower heads yellow. Roadside
weed in basalt on edge of steep slope. 19 - 22 involucre bracts.
(Duplicate to CANB.NSW Australia; TARCH No.5446)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 47)
Experimental Research Material

Date: 31.01.1996
Coll: R.C. Galayhen

Loc: 28° 10' S 153° 15' E Beuhmont, Queensland, Australia
Notes: Altitude 1850 ft. Growing in red earth in Kikuyu grass paddock.
Small yellow flowers. 19 - 22 involucre bracts. (Duplicate at PPRI, Pta)

(No Acc.No Sen 48 allocated.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 49)
Experimental Research Material

Date: 01.11.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 25° 02' S 46° 57' E Taolagnaro (Fort Dauphin), S.E.Madagascar
Notes: In grassland at airport. Scattered on road sides. Red dry earthy soil.
Small branched herb. Small yellow flowers. Lots of seed. 19 - 22 involucre
bracts. (5x Seed Samples & duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 50)
Experimental Research Material

Date: 01.11.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 25° 01' S 46° 57' E Taolagnaro (Fort Dauphin), S.E.Madagascar
Notes: At Mission Station. Common in shady forest next to rice fields.
Rich peat-like soil. 10 - 70 cm high branched herb. Small yellow flowers.
19 - 22 involucre bracts. Lots of Rust on stems and leaves. Photographed.
(5x Seed Samples & 1 duplicate plant specimen collected)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit

Date: 03.11.1996

(Acc.No. Sen 51)
Experimental Research Material

Coll: P.A. Muller
A.B.R.Witt

Loc: 25° 02' S 46° 39' E Taolagnaro (Fort Dauphin), S.E.Madagascar
Notes: Very abundant next to soccerfield on Cliffs above sea. In sunny cultivated potato field. Soil like dune sand. Plants low and very branched. Lots of Rust on stems and leaves. Small yellow flowers. 19 - 22 involucral bracts. (5x Seed Samples & 1 duplicate plant specimen collected)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 52)
Experimental Research Material

Date: 03.11.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 25° 02' S 46° 59' E Taolagnaro (Fort Dauphin), S.E.Madagascar
Notes: On lawn at hotel. Not many plants in dunes, very flat and branched. Flowers yellow, 19 - 21 involucral bracts. Quite healthy, no rust. (Only one large general seed sample taken.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 53)
Experimental Research Material

Date: 05.11.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 23° 21' S 43° 40' E Toliara (Tulear), S.W. Madagascar
Notes: Few plants in garden in centre of town. Dry hard red soil. 10 - 70 cm high branched herb. Small yellow flowers almost finished flowering. 19 - 21 involucral bracts. Plants healthy. No rust evident. (5x Seed Samples & duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 54)
Experimental Research Material

Date: 07.11.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 23° 29' S 43° 45' E Toliara (Tulear), S.W.Madagascar
Notes: Few plants on roadside in town near Hotel. Soil red and dry. 10 - 70 cm high branched herb. Leaves broad and plants tall. Lots of small yellow flowers. 19 - 21 involucral bracts. Plants heavily attacked by leafminers. (Photographed). (5x Seed Samples & duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 55)
Experimental Research Material

Date: 03.11.1996
Coll: P.A. Muller
A.B.R.Witt

Loc: 25° 08' S 46° 23' E Taolagnaro (Fort Dauphin), S.W. Madagascar
Notes: Abundant on dunes and lawn on town square overlooking the sea. Plants very low on ground and branched. Flowering profusely. Small yellow flowers. 19 - 22 involucral bracts. Lots of rust present on plants. (5x Seed Samples & duplicate plant specimen collected.)

(No Acc.No. Sen 56 allocated.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 57)
Experimental Research Material

Date: 19.11.1996
Coll: P.A. Muller
P.B. Edwards

Loc: 34° 06' S 20° 56' E Heidelberg, Western Cape, South Africa
Notes: Scarce on road side. 3km West of Heidelberg/Witsand Beaufort
turnoff. Brown stony clay next to wheat fields. Small yellow flowers.
13 involucre bracts, possibly *S.Burchellii*.
(No Seed Samples & only 3 plants collected.)

(No Acc.No. 58 allocated)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 59)
Experimental Research Material

Date: 19.11.1996
Coll: P.A. Muller
P.B. Edwards

Loc: 34° 12' S 21° 36' E Riversdale, Western Cape, South Africa
Notes: Abundant in field. On N2 8km West of R325 to Gouritsmond.
Loose sandy soil (lots of moles). Lots of flowering. Leaves variable, small
and thin, to broad. Lots of rust (photographed). Small yellow flowers.
13 - 15 involucre bracts, possibly *S.Burchellii*.
(5x Seed Samples & duplicate plant specimens collected.)

(No Acc.No. Sen 60 allocated)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 61)
Experimental Research Material

Date: 20.11.1996
Coll: P.A. Muller
P.B. Edwards

Loc: 34° 08' S 24° 49' E St. Francis Bay, Eastern Cape, South Africa
Notes: Abundant in grassfields next to canals at Riverhouse. Black clay
fertile soil. Seems like a mix of *Senecios*. (Not sure if this is our fireweed).
Leaves large at apex of stems, and very variable, long and digitate. Quite
abit of rust. Yellow flowers, 19 involucre bracts.
(5x Seed Samples & duplicate plant specimens collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 62)
Experimental Research Material

Date: 21.11.1996
Coll: P.A. Muller
P.B. Edwards

Loc: 34° 00' S 25° 21' E Sea View, Eastern Cape, South Africa
Notes: (East of P.E.) Common on road sides and in park on beach
front in front of Hotel. Sandy soil. Plants low on ground. Rust present.
(Photographed). Small yellow flowers. (Don't think it is our fireweed.)
(5x Seed Samples & duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 63a)
Experimental Research Material

Date: 21.11.1996
Coll: P.A. Muller
P.B. Edwards

Loc: 33° 58' S 25° 39' E Sea Acres, Eastern Cape, South Africa

Notes: On edge of tennis court, near Beach Front KFC. Small yellow flowers. Leaves variable from plant to plant. Tall herb 70cm high.

No rust seen..

(5 x Seed Samples & duplicate plant specimens collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 63b)
Experimental Research Material

Date: 21.11.1996
Coll: P.A. Muller
P.B. Edwards

Loc: 33° 58' S 25° 39' E Sea Acres, Eastern Cape, South Africa

Notes: Abundant fields of the plant along highway on N2 West of Port Elizabeth. Plants small due to mowing. White dune sand. Lots of flowering. Leaves variable, small and thin, to broad. Small yellow flowers.

(5x Seed Samples & duplicate plant specimens collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 64a)
Experimental Research Material

Date: 21.11.1996
Coll: P.A. Muller
P.B. Edwards

Loc: 33° 44' S 26° 33' E Cannon Rocks, Eastern Cape, South Africa

Notes: Common on road sides next to paddocks. Looks more like our fireweed. Small yellow flowers, 19 involucre bracts. Some rust. Photographed.

(5x Seed Samples & duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 64b)
Experimental Research Material

Date: 21.11.1996
Coll: P.A. Muller
P.B. Edwards

Loc: 33° 44' S 26° 33' E Cannon Rocks, Eastern Cape, South Africa

Notes: Abundant in grassfields in meadows. Rich clay soil. Green lush cattle pastures. Photographed. Some rust. (Two types of plants, one very hairy, possibly not fireweed.) Small yellow flowers.

(5x Seed Samples & duplicate plant specimens collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 65)
Experimental Research Material

Date: 22.11.1996
Coll: P.A. Muller
P.B. Edwards

Loc: 33° 36' S 26° 40' E Port Alfred, Eastern Cape, South Africa

Notes: A few plants on Beach Road in Sandy dunes. Quite abit of variation between plants. Rust present. Small yellow flowers.

19 involucre bracts.

(5 x seed samples & duplicate specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 66)
Experimental Research Material

Date: 22.11.1996
Coll: P.A. Muller
P.B. Edwards

Loc: 33° 19' S 27° 20' E Maccassar, Eastern cape, South Africa

Notes: On road side near Craft Store (maccassar Wholesales) East of Fish River. Rust. Plants small. Small yellow flowers. 19 - 22 involucre bracts. Photographed.
(5 x seed samples & duplicate specimen collected.)

(No Acc.No. Sen 67 and Acc.No. Sen 68 allocated.)

Senecio madagascariensis Poir

CSIRO Biological Control Unit Date: 23.11.1996
(Acc.No. Sen 69) Coll: P.A. Muller
Experimental Research Material P.B.Edwards

Loc: 33° 40' S 26° 40' E Kenton on Sea, Eastern Cape, South Africa
Notes: Scarce in grass on hill behind Hotel. Brown compact sand.
10 - 70 cm high branched herb. Small yellow flowers. 19 - 22 involucre bracts. No rust seen. Photographed.
(5x Seed Samples & duplicate plant specimen collected.)

Senecio madagascariensis Poir

CSIRO Biological Control Unit Date: 23.11.1996
(Acc.No. Sen 70) Coll: P.A. Muller
Experimental Research Material P.B.Edwards

Loc: 33° 38' S 26° 27' E Alexandria, Eastern Cape, South Africa
Notes: Abundant on road side 5km East of Alexandria. 10 - 70 cm high branched herb. Small yellow flowers. 19 - 21 involucre bracts. Looks alot like the Australian fireweed. Rust present. Photographed.
(5x Seed Samples & duplicate plant specimen collected.)

(No Acc.No. Sen 71 allocated.)

Senecio madagascariensis Poir

CSIRO Biological Control Unit Date: 23.11.1996
(Acc.No. Sen 72) Coll: P.A. Muller
Experimental Research Material P.B.Edwards

Loc: 33° 19' S 26° 31' E Grahamstown, Eastern Cape, South Africa
Notes: Abundant on road side in car park next to Grahamstown Monument. 10 - 70 cm high branched herb. Small yellow flowers. 19 - 22 involucre bracts. No rust seen. photographed.
(5x Seed Samples & duplicate plant specimen collected.)

Senecio madagascariensis Poir

CSIRO Biological Control Unit Date: 24.11.1996
(Acc.No. Sen 73) Coll: P.A. Muller
Experimental Research Material P.B.Edwards

Loc: 34° 01' S 22° 49' E Sedgefield, Western Cape, South Africa
Notes: Small plants in median strip, but collected large plants East of Sedgefield in open veld. Lots of moles and disturbance. Very Sandy. Very tall plants 70 - 100cm high branched herb. Small yellow flowers. 19 - 22 involucre bracts. Rust present and stem galls. Photographed.
(5x Seed Samples & duplicate plant specimen collected.)

***Senecio madagascariensis* Poir**

CSIRO Biological Control Unit
(Acc.No. Sen 74)
Experimental Research Material

Date: 24.11.1996
Coll: P.A. Muller
P.B.Edwards

Loc: 34° 01' S 22° 13' E Groot Brak, Western Cape, South Africa

Notes: Common on road side median strip. Red compact soil. 70 cm
high tall branched plants. Small yellow flowers. 19 - 22 involucral bracts.

No rust seen. Site photographed.

(5 x Seed Samples & duplicate specimen collected.)

For further information contact:

Petra Müller
CSIRO Biological Control Unit
c/o Zoology Department
University of Cape Town
Private Bag
Rondebosch 7701
SOUTH AFRICA

Tel: (021) 650 3650

Fax: (021) 650 4096

Email:PMULLER@BOTZOO.UCT.AC.ZA