

PT354

**Use of natural sprouting inhibitors for
potato storage**

Andrew Baker

Sunrise Agricultural Pty Limited



Know-how for Horticulture™

PT354

This report is published by the Horticultural Research and Development Corporation to pass on information concerning horticultural research and development undertaken for the potato industry.

The research contained in this report was funded by the Horticultural Research and Development Corporation with the financial support of the potato industry.

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Cover Price \$20.00

HRDC ISBN 1 86423 481 4

Published and Distributed by:



Horticultural Research and Development Corporation
Level 6
7 Merriwa Street
Gordon NSW 2072

Telephone: (02) 9418 2200
Fax: (02) 9418 1352

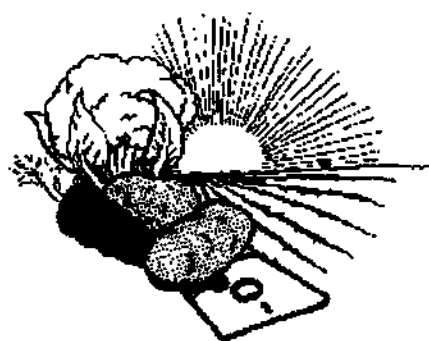
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HORTICULTURAL RESEARCH &
DEVELOPMENT CORPORATION
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Australian Horticultural Industries

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Industry Summary

Talent® (carvone)

A semi-commercial demonstration of Talent® was conducted at the Forthside Vegetable Research Station during 1994/95. The purpose of this trial was to demonstrate the efficacy of Talent® and to gather residue data in the event that registration in Australia would proceed. During 1995/96 this trial was repeated as part of the NRA requirement for residue studies. Additionally five tonnes of potatoes were bulk stored to further test the efficacy of this product.

An efficacy and residue data package has been completed for Talent®; this data is in line with previous studies conducted in the United Kingdom and The Netherlands. These studies show that after storage most of the residue is contained within the skins (<5 ppm) and that the cortex residue levels are <0.01 ppm. An organoleptic assessment conducted during the 1994/95 trial failed to detect the presence of Talent® after cooking.

Talent® displayed greater efficacy in the bulk storage situation, with completed removal of emerging sprouts within 7 days. This agrees with overseas work which showed that the efficacy of this product was dependent on the ratio between the volume of stored potatoes and the volume of air contained within the store.

Whilst Talent® has been shown to be as effective as CIPC as a sprout suppressant, the use in a "hot fogging" system is far too expensive to be considered whilst either CIPC or other cheaper alternatives are available. The cost has been estimated at \$AUS 15 per tonne of stored potatoes; this is some 3-4 times the current cost of CIPC.

During these trials Talent® has been shown to display other characteristics that are not directly applicable to the French Fry industry but may benefit other sectors of the potato industry. Talent®, unlike CIPC, doesn't permanently damage the sprouting mechanism of potato tubers. Potatoes that were stored during 1995 were planted back to assess the effect of this sprout control on subsequent crop performance. The sprout suppressant treatments appeared not to affect yield and improved the tuber size distribution.

A secondary benefit which has been reported overseas and observed in trials at the University of Tasmania is the fungistatic property displayed by Talent® against some species of *Fusarium* and against Silver Scurf. Both these diseases have caused considerable losses in the fresh, seed and processing potato sectors of the industry.

The current technology of "hot-fogging" has been found to reduce the amount of effective active ingredient that was applied to the store. This was found to be chiefly due to oxidation of the sprout suppressant during application.

A "cold fogging" system for the application of Talent® has been developed over the past 12 months; this system produces ultra-fine droplets through high pressure equipment. Residue studies after application have shown concentrations to be 2-3 times higher than when the same amount was delivered via the "hot-fogging" system. This development potentially reduces the amount of carvone that needs to be applied to the store and would make this product a commercially viable option.

It is recommended that this technology be trialed during the 1996 storage season in containerised trials at the Forthside Vegetable Research Station.

Currently Talent® is registered for use in the Netherlands; registration is imminent in both the United Kingdom and Germany. During March 1996 BV Luxan resolved to seek registration of Talent® in Australia. The New Zealand potato industry has been following the progress in Australia and will begin their own residue studies during 1996.

1-4 SIGHT® - (1-4 Dimethylnaphthalene)

Dimethylnaphthalenes were identified in potatoes and potato stores by British researchers during the 1980's. The concentration of this substance decreased with time during storage and it was postulated that this could be the potatoes' own sprout suppressant. This compound is also a by product of the petroleum industry and occurs naturally as number of isomers. When these compounds were applied to potato stores, excellent sprout suppression was achieved. Registration was not pursued due to the expense of creating toxicological data for all of these isomers and the risk of flavour taints in finished products. In recent times the most effective isomer was identified and production or purification commenced. This isomer was 1-4 dimethylnaphthalene and a toxicological package was developed in the USA.

This product was registered for use in the USA as 1-4 Sight®, EPA No.67727-1 during 1995 and was trialed at the Forthside Vegetable Research Station during the 1995/96 storage trials. It was found to be an effective sprout inhibitor and would be considered superior to Talent®. Preliminary residue data was collected at the end of the trial and will be used if further developments in Australia are to occur.

1-4 Sight® was jointly developed by D-I-1-4 ,Inc (USA) and Sumitomo Corporation (Japan). The commercial production of this material has now been finalised and D-I-1-4 ,Inc will support the development and registration of this product in Australia. An approximate cost of \$AUS 100 per litre has been quoted which if used at rates of 20-40 mls/tonne equates to costs of \$AUS 2-4 per tonne of potatoes.

This product was registered as a "nature identical" product and as such some of the toxicological data that is normally required may be waived. From a residue point of view the NRA are likely to consider DMN similar to Talent® but from an OH&S point of view a full toxicological package may be required. An extensive list of studies is available from the USA.

Like Talent®, 1-4 Sight® is a reversible sprout inhibitor; studies in the USA have also shown this product to enhance suberisation of cut or damaged potatoes at low temperatures. This factor could prove useful in the curing of stored potatoes and in the storage of pre-cut seed. Tubers stored with DMN during 1995 were planted out in a demonstration trial and performed similar to the Talent® treated tubers.

PSS-25 - Imazethapyr

This chemical was originally developed as a pre-emergent and post-emergent herbicide for soybeans. The active ingredient *imazethapyr* was developed by the American Cyanamid Company and its use as a potato sprout suppressant has been trialed by Cyanamid-UK and the Potato Marketing Board.

The UK efficacy data show this inhibitor to be effective after one application. *Imazethapyr* is applied at store loading as a mist (1 litre/tonne containing 1 gram ai / tonne). The mode of action is systemic; cell division is prevented in the meristem by inhibition of amino acid biosynthesis.

A preliminary trial conducted during 1995 only showed moderate sprout control and warrants further investigation. This product has now been registered in the UK and is particularly suited to their system where potatoes are treated as they are loaded into boxes prior to storage. Cyanamid-Australia have expressed no interest in the development of this chemical in Australia at the present time.

CIPC - Chloroprotham

Review update

The NRA has confirmed that it now has the necessary data to review CIPC. The original package required some clarification and additional data which has been completed. A decision regarding the use of CIPC as a potato sprout inhibitor was due at the end of June 1996.

Given a favourable decision Aceto will have proprietary rights for the distribution of their formulation of CIPC within Australia for a period of up to seven years. During this period no other company can register an "image product" using this data unless some compensation has been negotiated with the primary registrant. No proprietary rights exist for the active ingredient alone. Another company could register an alternative formulation providing they have their own complete data package.

The issue of price increases whilst Aceto has proprietary rights is beyond the control of the NRA. The Prices Surveillance Authority (PSA) may control this issue and it is recommended that the potato industry seek advice from the PSA prior to the granting of proprietary rights.

During the past twelve months BV Luxan has developed a CIPC formulation which is based on a light natural oil. This is sprayed onto potatoes as they are loaded into bulk stores and is said to prevent sprouting for six months. They have also developed the technology to vary the application of this product as the volume of potatoes change on the pile loader. A maximum of 200 mls of fluid (formulation + water) per tonne is applied. BV Luxan is also considering registering their fogging formulation of CIPC which is based on dichloromethane.

Technical Report

I. Introduction

The national annual potato production is approximately 1 million tonnes and of this 360,000 tonnes are held for storage after harvest. The majority of these stored potatoes are destined for processing as either potato crisps or potato fries. Storage of potatoes is necessary as the capacity of the Australian processing sector is exceeded during the main harvest period.

After the storage period the potato tubers destined for processing must be in a similar condition to freshly harvested tubers. The tubers must be unsprouted, have low reducing sugar content and have retained turgidity. During sprouting, water is lost at a high rate through the sprout and simple sugars built up as starch reserves are mobilised. Additionally, sprouted tubers also have elevated levels of solanine, a potato alkaloid which is poisonous if consumed in large quantities.

To maintain the quality of stored potatoes sophisticated technology is required to store potatoes long term. Water loss is restricted by storing the tubers in a high humidity environment, usually in excess of 95% relative humidity. Potatoes after harvest have a dormancy period, which for practical purposes is defined as the period between harvest and when the potatoes begin to sprout. To prevent sprouting during storage the temperature is maintained at 7-10 °C which is the minimum temperature at which potatoes can be held if they are destined for processing. Storage at lower temperatures than this will increase the build-up of reducing sugars, which is referred to in industry as "low temperature sweetening". Potatoes with high reducing sugar content are rejected as they produce unacceptable darkening of products during the cooking process.

Additionally to these storage parameters, various chemicals have been applied to potato tubers to prevent them from sprouting. During the 1950's the herbicide *isopropyl N-(3-chlorophenyl) isopropylcarbamate* (CIPC or Chloroprotham) was demonstrated as an effective sprout inhibitor. This chemical is a mitotic inhibitor and prevents cell division as potato sprouts begin to develop; it is used worldwide in long term potato storage and is usually applied as a fog after potato stores have been filled.

During the past 15 years some questions as to the safety of this chemical in the food chain have been raised. This was due to the fact that during the biodegradation of this herbicide, a potent carcinogen was identified. The chemical identified was chloroaniline and is a by product of the soil microbial digestion of CIPC. Extensive studies have been conducted in Europe and USA to determine whether chloroaniline can be found in stored potatoes, its products or in the processing residues. Feedlot studies on cattle and poultry have been completed to test for residues picked up from potato wastes ex factory.

These concerns have led to the support for the investigation of new sprout inhibitors that can be shown to be free of these risks. In particular, naturally occurring chemicals have been extensively investigated in the USA and Europe. For example (d)-carvone, a monoterpene extracted from caraway seed has been investigated by Dutch scientists during the past five years. This chemical has been shown to be a potent sprout inhibitor of potatoes and was registered by BV Luxan during 1995 in the Netherlands as Talent®. Workers in the USA have extended earlier British work on the potato's own sprout suppressants. A number of chemicals found in the head space of stored potatoes have been identified and, when applied in a concentrated form, have been reported to inhibit sprouting. This group of chemicals include a wide range of substituted naphthalene compounds. The mode of action of these chemicals is less understood. D-I-1-4, inc. in the USA has registered 1-4 Sight® (1-4 dimethylnaphthalene) as a potato sprout suppressant.

During 1993 the Chemical Safety Unit in Canberra gave notice to the suppliers of CIPC in Australia to provide supporting safety data that was not submitted in the 1960's when this active ingredient was registered in Australia. Initially the industry had until November 1994 to provide this information. At the time there was some doubt as to the quality of the data that existed overseas and the proprietary right issues that might have been attached to this data. The loss of this chemical provided the stimulus to investigate the alternative sprout suppressants for the potato industry.

The aim of this project was to test the efficacy of alternative sprout suppressants that had been identified by overseas researchers and to collect residue data in the event that these products may be registered in Australia. Potatoes were stored in modified refrigerated containers over a six month period. The efficacy of the sprout suppressants and quality of the stored tubers were monitored throughout the trial.

In smaller scale trials conducted at the University of Tasmania the effect of these sprout suppressants on the sprouting physiology was examined. This included the effect treatment had on morphological changes and the mobilisation for starch reserves during the sprouting process.

This project has confirmed the overseas development of Talent® and 1-4 Sight® as two potential alternative sprout suppressants. Data has been collected that will support the registration of these products if the manufacturing companies choose to pursue this in Australia.

II. Materials and methods

1.0 Semi-commercial Storage trials 1994/95 and 1995/96

1.1 Storage containers

1994/95

Refrigerated containers were hired from Wreckair Container Services P/L. These were modified by building a false floor in each container that was vented in such a way that the air only flowed through the trial bins. Bulkheads were constructed that allowed the fogging of containers whilst the main doors were open. The temperature was maintained at 7-8 °C and air flow was maintained at approximately 20m³/tonne/hour. The containers were also fitted with data loggers which monitored temperature and humidity throughout the trial.

1995/96

The containers were set up the same as the previous year with the exception that two containers were bulk filled to test the efficacy of these suppressants at a larger scale. The potatoes from McCain Foods (Aust) Pty Ltd were used for the bulk filled containers and the potatoes from Simplot Australia Pty Ltd and Smith's Snackfood Company were packed in the trial bins and used for the collection of residue data.

1.2 Potatoes

1994/95

Russet Burbank and Denali were used to represent the most common potato cultivars stored in the Australian industry. Denali was sourced from Smiths Snackfood Company from a crop in Pinnaroo, South Australia. Russet Burbank was sourced from McCain Foods (Aust) Pty Ltd and was from three production areas, Circular Head, Weslyvale and Cressy. Included in the trial were small quantities of new cultivars emerging in the Australian breeding program.

1995/96

Russet Burbank potatoes were sourced from Simplot Australia Pty Ltd and McCain Food (Aust) Pty Ltd . Denali was Sourced from Smiths Snackfood Company from the same region as in 1994.

1.3 Sprout suppressants and application methods

1994/95

Three sprout suppressants were used; Tato-Vapo® (N-(3-chlorophenyl)-isopropylcarbamate); Talent®, ((d)-carvone) or 1-methyl-4-isopropyl-6-cyclohexene-2-one) and (d)-fenchone (1,3,3-trimethyl-2-norcamphanone).

Tato-Vapo® (500 mg/l CIPC) was supplied by Serve-Ag P/L and applied at the current industry rate of 60 mls/tonne in a split application. Talent® was supplied by BV Luxan, The Netherlands and applied at the label rate of 300 mls/tonne and at 600 mls/tonne; this was split over three applications at six weekly intervals. (d)-fenchone is a by product of the local essential oil industry and was used at the same rate as Talent®.

1995/96

Three sprout suppressants were used; Talent® ((d)-carvone or (1-methyl-4-isopropyl-6-cyclohexene-2-one)), 1-4 Sight® (1-4-dimethylnaphthalene) and Spinnaker® (Imazethapyr).

As in 1994/95 trials the crates were treated with Talent® at 600 mls per tonne. For the bulk filled trials, a total of 300 mls/tonne was applied to the first container and 600 mls/tonne to a second container over three applications. In contrast to 1994, the applications were not made until sprout activity was observed. Similarly, 1-4 Sight® was applied at a rate of 20 mls/tonne when sprout activity was observed.

For both 1994/95 and 1995/96 trials Tato-Vapo®, Talent® and 1-4 Sight® were applied by Smiths Pest Control P/L using a swinfogger. The container fans were turned off and the vents closed during treatment. After application the containers were turned on and air was recirculated for 24 hours then ventilation was resumed at approximately 20 m³/tonne/hour. Spinnaker® (250gm/l imazethapyr) was made up as a solution in which 1 gram of imazethapyr was applied as a mist to potatoes at a rate of 1 litre per tonne.

2. Mode of action of Talent®

2.1 Storage conditions

During 1994 an experiment that examined the physiological effects of the potato sprout suppressants Tato-Vapo® and Talent® was conducted at the University of Tasmania. Potatoes were held in 20 litre containers with a source of humidified air which passed through the containers at a rate of 20 m³/tonne/hour. This was similar to the airflow expected under commercial storage conditions.

2.2 Potatoes

Russet Burbank potatoes were sourced from Mr Hugh Morris's property at Kindred, Northwest Tasmania. These were graded and tubers weighing between 100 and 250 grams were used for the trial.

2.3 Sprout suppressants

Two sprout suppressants used were Tato-Vapo® (N-(3-chlorophenyl)-isopropylcarbamate) and Talent® (1-methyl-4-isopropyl-6-cyclohexene-2-one)

The potatoes for this experiment were treated with the large trial in 1994 and then replaced in the storage containers at the University of Tasmania. An untreated control was also included.

2.4 Morphological Changes

The apical bud of a marked potato tuber was examined each week; an impression of this bud was taken using a dental polymer. From this impression, a copy of the bud was cast using West System 504 resin. These models were examined under the environmental scanning electron microscope to monitor any morphological changes which occurred during treatment and during the subsequent storage period. Further records of macroscopic changes were taken down a light microscope after treatment.

2.5 Sugar Analysis

Three tubers were selected at random each week for sugar analysis. An 8 mm core through the apical bud to the middle of the potato tuber was taken. This core was subdivided into three sections and labelled periderm, mid cortex and deep cortex. These were then freeze dried in preparation for sugar analysis.

Sugar extraction was a modification from the Lambrechts et al (1994) method. 100 mg of freeze dried potato powder was extracted at 60 °C in 2 mls of 80% ethanol (three times, 1hr per extraction). After each extraction the suspension was centrifuged at 1750G for 5 minutes. The supernatants were pipetted off and combined. The ethanol was removed using a vacuum rotary evaporator. The residue was made up to 5 ml with water and partitioned with 3 ml of chloroform. The water phase was collected for analysis of sucrose, (d)-glucose and (d)-fructose.

The sugar assay was an enzymatic analysis using the Boehringer Mannheim sucrose/(d)-glucose/(d)-fructose test kit. The sugars were progressively digested to glucose and a colorimetric measurement was made at 340nm.

The extraction efficiency was calculated by preparing potato cores not previously used for analysis. 100 mg of this material was spiked 377 mg glucose, 372 mg fructose and 750 mg sucrose and extracted and analysed as per the experimental samples. The percentage recovery of these sugars after extraction was calculated and the experimental data transformed.

2.6 Disease Assessment

The remaining tubers at the end of the storage trial were examined for the presence of the storage disease *Helminthosporium solani* (silver scurf). This was recorded on a presence or absence basis and the severity of this disease was based on the surface area infected. The grades were Nil, 1 (<5% infection) and 2 (>5% infection).

3. General methods

The following describes the methods that were common to the residue and efficacy studies and the mode of action study.

3.1 Residue analysis

Potato residue studies included the measurement of residues in the whole tuber (WT), the peeled tuber (PT) and the potato peelings (PP). Talent® and 1-4 Sight® residues were extracted from the tissues using HPLC grade hexane.

For each residue analysis a 10 kg sample was taken from each trial bin. The 10 kg sample was sub-divided into two 5 kg lots and the WT, PT and PP preparations were made. 10 grams from each preparation were taken for residue extraction.

3.2 Residue analytical method

Selected ion monitoring using an HP5890 GCMS was used to assess the level of Talent® and residue after storage. Each tissue sample was extracted with 20 mls of hexane containing dodecane as a standard at 70 mg/litre. Dodecane was chosen as it has a similar retention time to (d)-carvone and a clearly defined peak.

A non-polar phase HP1 coil was chosen which would resolve dodecane and carvone; helium was used as the carrier gas at 5 psi. The run program was:

oven 60°C 120°C @ 10°C/min
injection 250°C
detector 2890°C
selected ion monitoring
m/z* 99.1 dodecane
m/z 82 (d)-carvone
multiplier 2200V
solvent delay 2.4 minutes

* m/z = molecular wt./charge

(d)-carvone has an unique ion m/z = 82 and the closest dodecane ion (without causing interference) was m/z = 99. Between these two ions exists a linear relationship that allows the calculation of carvone residues after the response factor has been calculated by running a series of calibrations.

response factor = ($\mu\text{g. C12} * \text{Ratio } 82/99 \text{ peak area}$)/ $\mu\text{g carvone}$

For each residue study a calibration series was run which encompassed the expected range of carvone concentrations to be found. This range was 0.5 to 5 ppm carvone.

$$\begin{aligned} \text{eg. R.F} &= (70 * (11968741/14828529))/5 \\ &= 11.29 \end{aligned}$$

The method for the analysis of 1-4 Sight® residues was the same as for Talent® except that the unique ion for the active ingredient of this sprout suppressant, 1-4 dimethylnaphthalene, was $m/z = 109$

3.3 Recovery of residues

The method and efficiency of the above extraction method was proved by the following recovery experiment.

As Talent® is highly soluble in hexane and sparingly soluble in water, the extraction of residual Talent® was expected to be high. This was checked by running blanks with Talent® added and extracts of potato tissues spiked with Talent® at concentrations near the expected residue levels.

100 gm of potato peels were spiked with 100 µg of Talent® dissolved in 2 mls of methyl alcohol. After the peels were macerated 10 gm sub-samples were taken, these were extracted with 20 mls of hexane containing a 70 mg/litre dodecane standard. For the blanks, 10µg of Talent® in methyl alcohol was added directly to 20 ml of the extracting solution.

These samples were analysed using the above method and checked for consistency.

3.4 Head space concentrations (1994 only)

During 1994 the headspace concentration of Talent® was monitored after application using a Dragar Gas Detection test kit. One litre of air was drawn through sample tubes containing activated carbon. These samples were then extracted and residues measured using the same procedure developed for residue analysis.

3.5 Efficacy - Degree of Sprouting

The degree of sprouting was assessed at the end of the trial by selecting fifty tubers at random from each crate. The length of the longest sprout was measured and recorded in one of the following five categories.

- (1) not sprouted
- (2) less than 3 mm
- (3) 3- 10 mm
- (4) 10-20 mm
- (5) greater than 20 mm

The index was calculated using the mid-point values of these categories and the frequency of tubers in the five categories.

eg. Sprouting index = $(A*0)+(B*1.5) + (C*6.6) + (D*15) + (E*40)$ where A...E were the proportion of tubers in the categories 1 to 5

3.6 Tuber Quality

Fry colour

As a measure of the sugar accumulation within the tubers during and after storage, the fry colour was assessed. For the cultivar Russet Burbank, fifty tubers were selected from each crate and sent to the Simplot factory for testing. This was done by removing the central fry from ten potatoes selected at random, cooking them at 191 °C for 2 minutes 45 seconds and comparing the fry colour with a standard chart. There was five colour grades 0, 1, 2, 3, and 4. The proportion of fries in each of these categories were recorded and any load with chips in the 4th or 5th grade were rejected.

For the cultivar Denali fifty tubers were sent to the Smiths Snackfood Company factory in Tynong, Victoria. Slices taken with a crinkle cut slicer and fried at 185 °C for 1 minute in tub 1 and then fried for a further one and a half minutes in tub 2 at 185 °C. Each slice was compared with a company standards chart and graded as defective or processable. If the sample contained greater than 20 % defective slices the load was rejected.

Organoleptic assessment

Crisps were prepared from both the Tato-Vapo® and Talent® treated potatoes and a panel of thirty three DPIF staff and industry representatives were given samples to taste. The samples were presented in a double blind format. Each panelist was given three samples in random order, two of these samples were from the same treatment. The panelist was then asked to pick which two samples were the same and explain any differences they detected.

III. Results

1. Semi-commercial storage trials

1.1 Efficacy of the sprout inhibitors

1994/95

The efficacy of the sprout inhibitors Talent® and (d)-fenchone were compared to Tato-Vapo® by calculating the sprouting index after 172 days storage. Table 1. below shows the average for each storage container .

Container	Treatment	Average sprouting index
1	Tato-Vapo®	1.94
4	Tato-Vapo®	1.77
2	Talent® 300 mls/tonne	4.94
5	Talent® 300 mls/tonne	6.81
3	Talent® 600 mls/tonne	1.86
6	Talent® 600 mls/tonne	1.64
7	(d)-fenchone 600 mls/tonne	14.5

Table 1. Sprouting indices 1994/95 storage trial

In comparison between containers; Talent® and Tato-Vapo® were equivalent sprout suppressants as the observed differences were not significant. The lower rate of Talent® produced a greater sprouting index chiefly due to the failure to control the sprouting in the cultivar Denali ($P < 0.05$). Denali has a thinner skin than Russet Burbank and was stressed by the low humidity conditions at the end of the trial. Sprout control of the cultivar Denali was achieved when the higher rate of Talent® was used; the differences in the sprouting index within these containers was not significant.

In comparison (d)-fenchone failed to control sprouting after the 172 day storage period. Whilst sprouts that were present when (d)-fenchone was applied were damaged after treatment, there was little time delay in the development of auxiliary buds at the affected eyes or the sprouting of lateral buds. This contrasts with Talent® in which the development of sprouts was retarded after treatment.

The ability of Talent® to prevent sprouting was also demonstrated in the mixed cultivars. These cultivars displayed a wide range of characteristics that included different dormancy periods and degrees of apical dominance. In all cases sprouting was controlled. It was observed that the cultivars with strongest apical dominance produced a proliferation of lateral shoots after treatment which were also controlled by subsequent treatments.

In contrast to Tato-Vapo® treated buds where sprout development is inhibited by the interruption of mitotic cell division, Talent® treated potatoes were not permanently affected. After a delay sprouting developed normally. In comparison to untreated potatoes, more lateral sprouts were observed to develop.

1995/96

Talent® was again shown to be an acceptable potato sprout inhibitor. The sprout indices for the crate filled trial were higher in comparison to the 1994 trial. This was a result of the different application methodology used in the 1995 trial in which sprout suppressants were withheld until visible sprout activity occurred.

The bulk filled trial was treated the same as the 1994 trial with the first application being applied at the beginning of the storage period. This resulted in excellent sprout control as indicated by the very low sprout indices. Talent® used at half the label rate produced equivalent sprout control in comparison to the label rate under these storage conditions.

1-4 Sight® was shown to be a potent sprout inhibitor at 20 ppm as demonstrated by the low sprouting indices. In comparison to Talent®, the application of this suppressant when visible activity was observed did not effect the efficacy of this product. This indicates a different mode of action, which, apart from observation, was not studied in this project.

Imazethapyr controlled sprouting for a short time after application: the growth of the sprouts was retarded and they turned purple 30 days after application. During storage auxillary and lateral buds began to emerge which appeared unaffected by the treatment. After 150 days sprout control was unacceptable with sprouting indices in excess of 40 being recorded.

Table 3. summarises the sprouting indices for Talent®, 1-4 Sight® and Spinnaker® treated potatoes.

Container	Treatment	Sprouting index (mm)
1	Denali Talent® 600 mls/tonne	7.53
2	Russet Talent® 600 mls/tonne	3.4
3	Denali 1-4 Sight® 60 mls/tonne	2.54
4	Russet 1-4 Sight® 60 mls/tonne	1.87
5	Russet Talent® 600 mls/tonne	1.25
6	Russet Talent® 300 mls/tonne	1.35
Cool Room	Spinnaker®	40 +

Table 2. Sprouting Indices 1995/96 storage trial

Although the degree of sprouting was acceptable in all containers, the unsuitability of this storage system for Denali was demonstrated by the higher sprouting index. This was confirmed later in poor fry colour tests.

There were also visual differences between the Talent® and 1-4 Sight® treated potatoes. As in the 1994 trial, Talent treated potatoes were damaged and there was a proliferation of auxiliary and lateral buds. In comparison, the sprouting on 1-4 Sight® treated potatoes appeared stunted, without any physical damage having been done. Morphologically these sprouts appeared normal, unlike Tato-Vapo® treated potatoes in which the sprouts were deformed.

1.2 Residue Studies

1994/95

Talent®

The actual residue data cannot be reported here as it is protected by secrecy agreements. In general terms most of the residue was found in the periderm at concentrations less than 5 ppm. The concentration found in the cortex was in the parts per billion range.

These results confirmed the studies in the Netherlands and the United Kingdom and showed that the residues found were at one hundred times lower than commonly found in European food stuffs such as caraway seed cake and beverages.

This data has been compiled and will form part of the data package that will be required by the National Registration Authority for an Australian registration of this sprout suppressant.

(d)-fenchone

(d)-fenchone produced unacceptable sprout control under the conditions used and residue studies were not pursued.

1995/96

Talent®

The residue levels of Talent® were similar to the 1994 storage trial and evenly distributed throughout the container. This data completes the second study required by the National Registration Authority.

1-4 Sight®

Similarly this residue data is protected by secrecy agreements and is only available with the agreement of D-I-1-4, Inc USA. Similarly to Talent® the bulk of the residue was contained within the periderm layer after storage at low levels. The cortex layer had a concentration one hundred times lower than the periderm layer.

The level of 1-4 Sight® residue found in the cortex could not be separated from endogenous 1-4 dimethylnaphthalene. This was demonstrated by the assay for 1-4 dimethylnaphthalene in Talent® treated potatoes. The concentration found was similar to that found in the cortex of 1-4 Sight® treated potatoes.

1.3 Head space concentrations of Talent®

The head space concentrations in the storage containers were measured over the first 2 months of storage. The results are presented in Table 3. below.

Container	Head Space Concentration (µg/litre)			
	2	3	5	6
Date				
25/5	19.4	51.3	61.8	65.0
1/6	0.07	0.07	0.12	0.1
6/6	0.28	0.01	0.08	0.1
22/6	<0.01	<0.01	<0.01	<0.01
30/6	<0.01	<0.01	<0.01	<0.01
8/7	71.6	84.4	75.3	79.4
11/7	8.1	8.0	7.5	12.5
15/7	<0.01	<0.01	<0.01	0.08

Table 3. Head space concentration of Talent® 1994/95

The head space concentrations rapidly decreased soon after the containers were ventilated, with concentration falling below detectable levels within three weeks.

1.4 Fry Quality

1994/95

Samples from each of the trial bins were taken for fry quality assessment. There was no statistical difference between treatments. The mean fry colour from each treatment is plotted below. Overall 12 of the 54 samples had a small proportion of tubers which exceeded the colour specifications (ie. grades 3 &4). Seven crates of these were from Tato-Vapo® treated containers. Figure 1., plots the fry colour profile for these treatments.

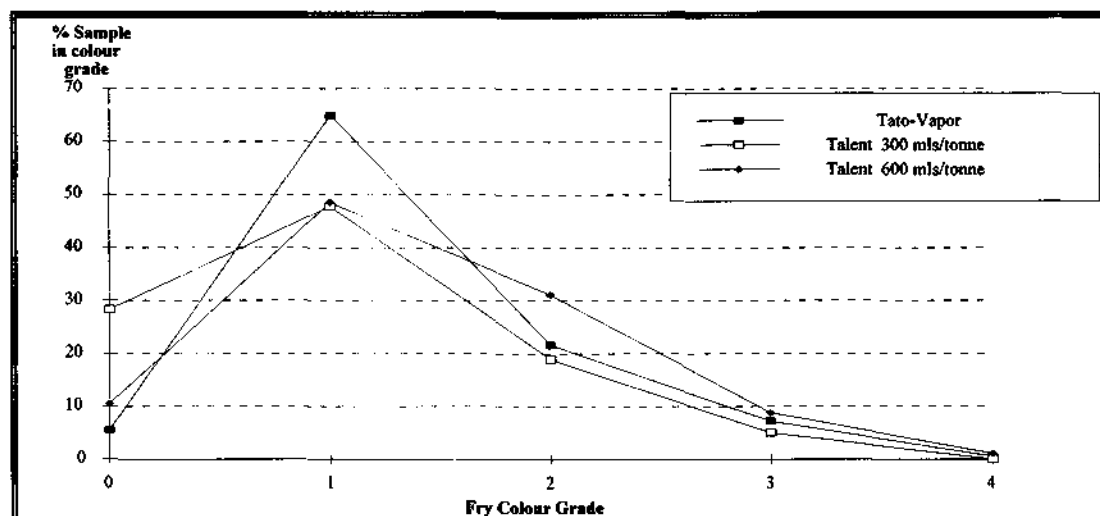


Figure 1. Fry colour assessment 1994/95 storage trail

In contrast only three from the eighteen crates of Denali sampled met the upper defect limit of 20%. The poor condition of these potatoes and the advanced sprouting in the low Talent treatment after storage are indicative of sugar mobilisation and the build up of reducing sugars.

1995/96

The fry colour assessment conducted by Simplot during 1995 after the storage period showed that both Talent® and 1-4-Sight® do not affect the fry colour quality of the potatoes. Figure 2. below shows that the potatoes after storage were of a higher quality than those stored during 1994 and were similar in quality to the Tato-Vapo® stored potatoes in that trial.

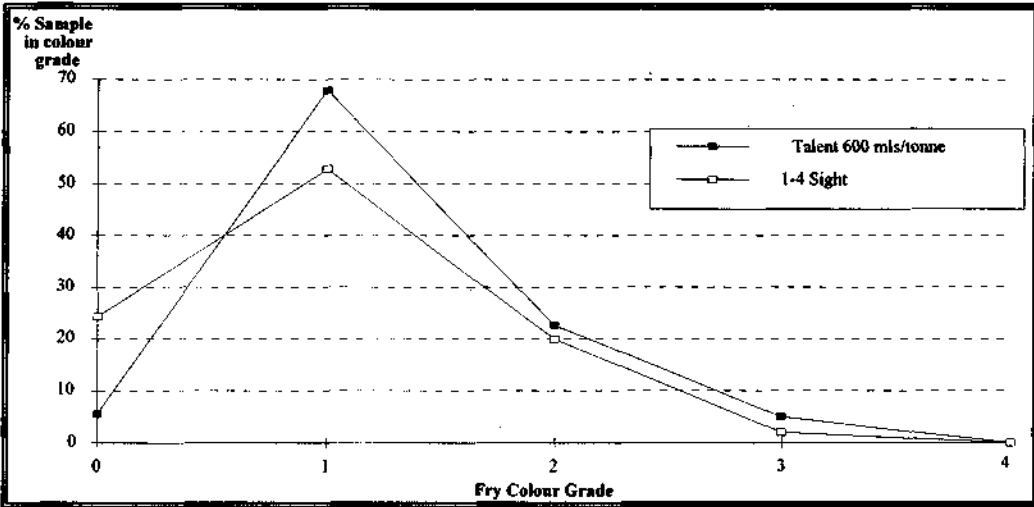


Figure 2. Fry colour assessment 1995/96 storage trial.

Organoleptic Assessment -1994/95

From the thirty-three panellists subjected to the double blind taste test only eight isolated the Talent treated potatoes. When asked to comment on their choice, seven noted visual differences between samples as the major factor, the remaining panellist commented on flavour. The visual difference was attributed to the shape or pattern on the crisp after cooking. None of the panellist identified the flavour or odour of Talent ® in the samples

This result shows that this monoterpene can be used without having adverse effects on the final product. This differs from many other monoterpenes which show good sprout control but leave strong flavour taints after processing. Examples of these are (l)-carvone common in the *Mentha* family and 1-8 cineole derived from eucalyptus oil.

2.0 Mode of action of Talent®

2.1 End of dormancy

The untreated tubers were the first to show signs that the dormancy period had ended when sprouting was observed on 26th July, 2 weeks after storage. The Talent® treatment showed some activity by 23rd August, 6 weeks after storage and prior to the second sprout control treatment. The Tato-Vapo® treated tubers began to show sprout activity by 6th September, 8 weeks after storage .

2.2 Efficacy of Talent®

Talent® exhibited acceptable sprout control with a sprout index of 9.66 in comparison to the untreated control which had an index of 28.67. Tato-Vapo® completely controlled sprouting with an index of 0.46. Talent® was not as effective in this trial in comparison to the larger storage trial as indicated by the higher sprouting index.

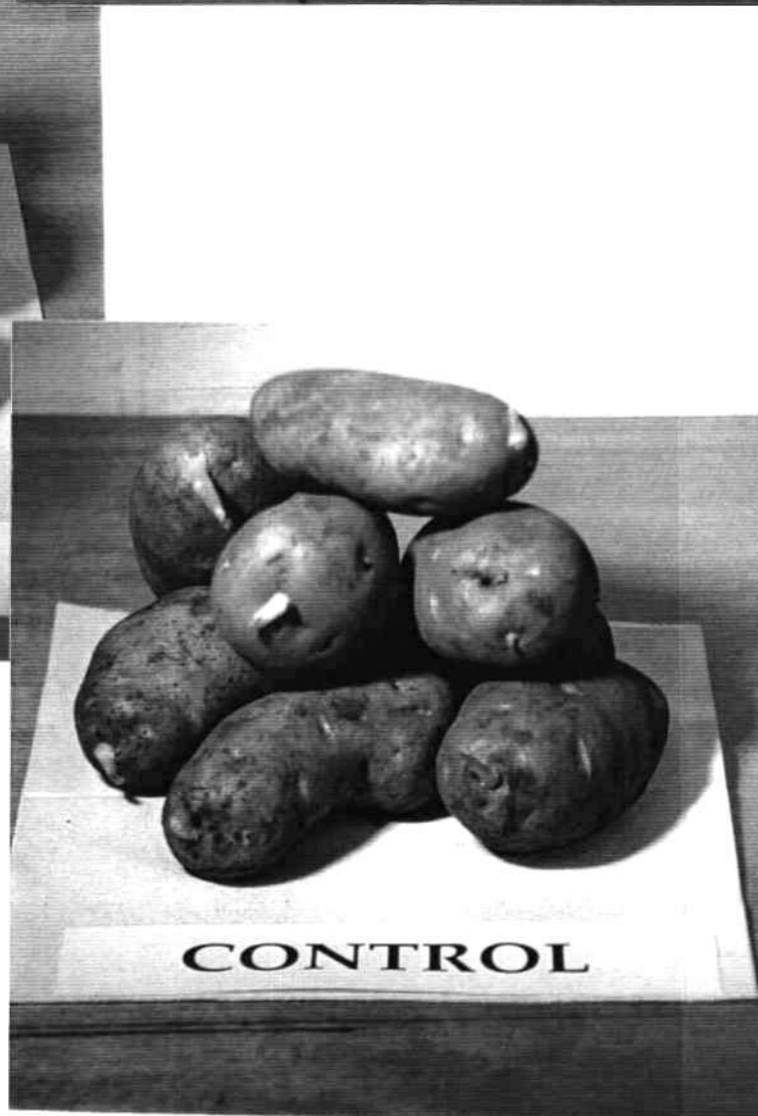
As in the larger scale trials the presence of Talent® on the surface of tubers was detected and generally the apical sprout was destroyed; only weak auxillary and lateral sprouts were beginning to emerge at the end of the trial. Most eyes on these tubers were beginning to sprout.

In contrast, the untreated tubers exhibited normal apical dominance with one large uninhibited apical sprout, and only a few small lateral sprouts generally less than 3 mm long. The Tato-Vapo® treated potatoes had deformed sprout growth at all eyes, less than 2mm in length. Under microscopic examination this growth appeared as callus tissue which is consistent with its mode of action as a mitotic inhibitor.

Figure 3. illustrates the difference between the treatments at the end of the trial.

see over for photographs

Figure 3. Sprouting tubers at the end of the trial



2.3 Morphological changes during storage

Macroscopic changes

The following series of photos demonstrates the efficacy of Talent® and the morphological changes that take place during sprouting. The photo of the apical sprout of an untreated potato (figure 4) shows normal development of the stem and root tissues. These tissues are undamaged and expanding rapidly.

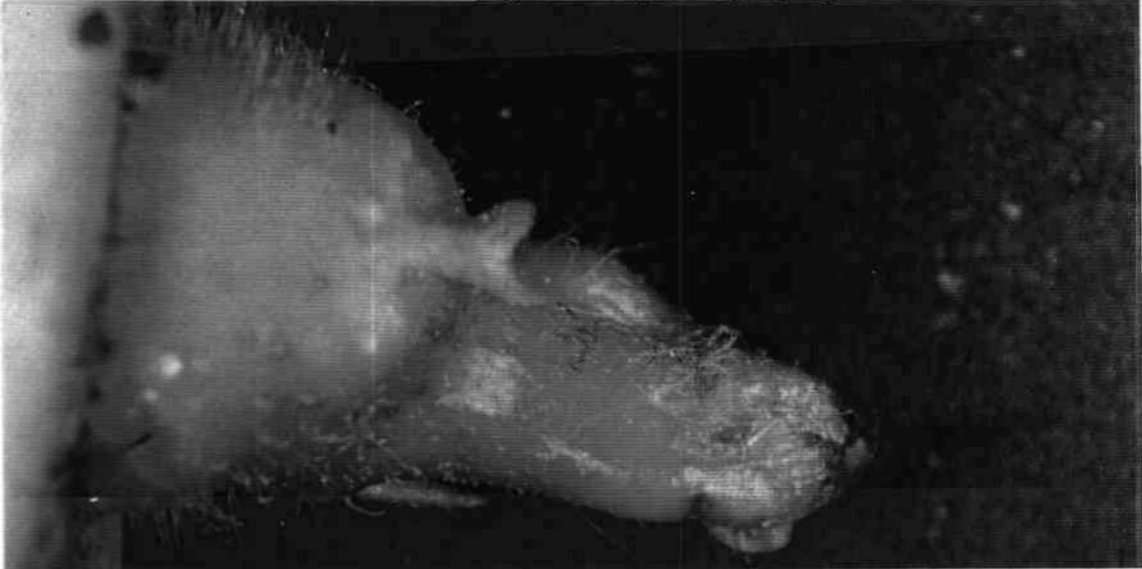


Figure 4. Sprouting untreated potato tuber

In contrast, the application of Talent® has severely damaged the apical sprout (figure 5). Note that the apical sprout is no longer functional and the proliferation of auxiliary sprouting from the base of this sprout. The treatment with Talent® has also stimulated the growth of lateral buds which, with the emerging auxiliary buds, have been damaged by subsequent applications of Talent®.

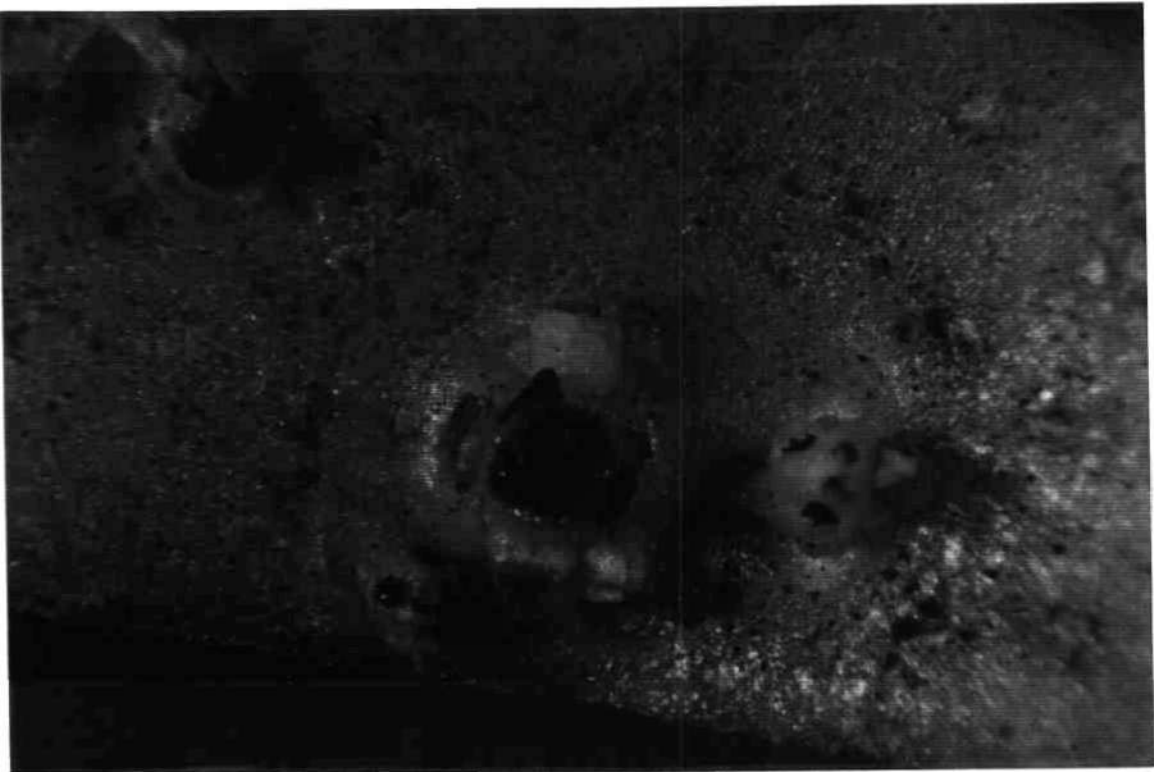


Figure 5. Sprouting Talent treated potato tuber

The section through the apical bud of a Tato-Vapo® treated potato tuber (figure 6) demonstrates the different mode of action of this sprout suppressant. These sprouts generally were less than 2 mm long and with the swollen tissues being undifferentiated. As with Talent®, apical dominance was lost as there were buds at each eye.

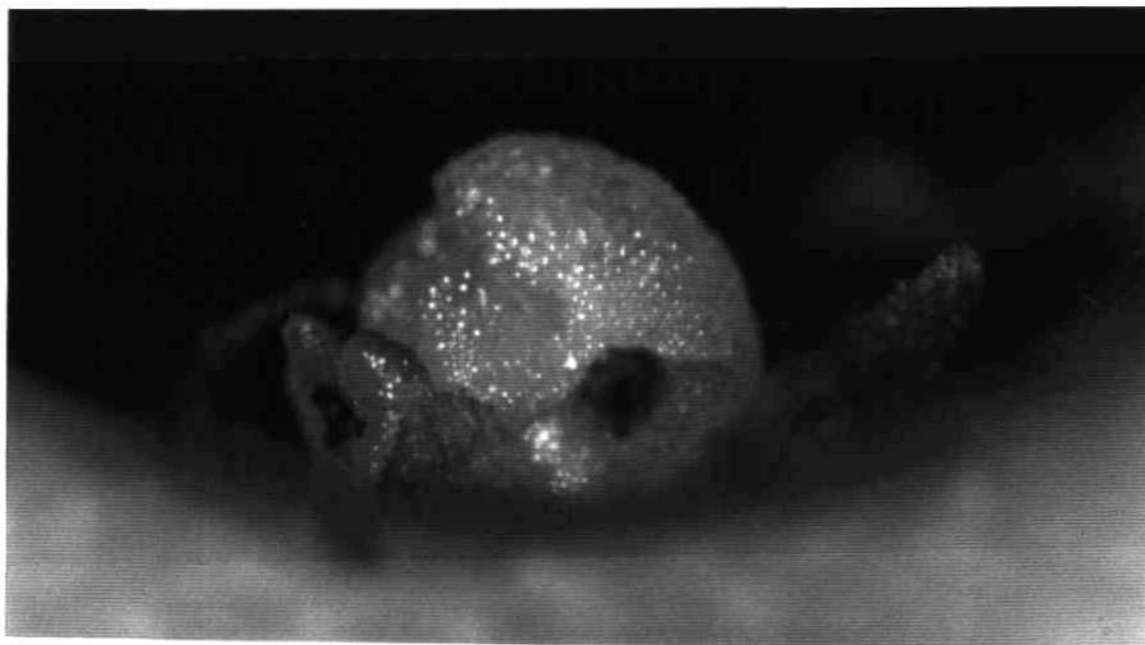


Figure 6. Sprouting Tato-Vapo® treated potato tuber

Microscopic changes

The resin models allowed microscopic examination of apical buds and gave an excellent “snap shot” in time of sprout development prior to visible macroscopic sprout elongation. Figure 7a. shows untreated potato apices at the beginning of the trial and clearly shows an undamaged developing sprout with rapidly differentiating leaf primordia. This model was taken at the beginning of the storage trial two weeks prior to visible sprouting being observed.

see over for photographs



Figure 7a. Apical bud untreated



Figure 7b. Talent treated bud

In contrast figure 7b. shows a Talent® treated tuber at the beginning of the storage trial. This model was taken 1 week after storage some 4 weeks before visible sprout activity could be observed. In comparison with the untreated tuber damage to the leaf primordia is extensive. This photo however suggests that the apical meristem is still undamaged and protected beneath the leaf primordia.

These observations have demonstrated that there is significant microscopic development occurring prior to visual sprouting and the end of “dormancy”. The difference in efficacy between the 1994 and 1995 trials could be attributed to the different stages of microscopic development when Talent® was applied

2.4 Sugar analysis

Periderm core samples

The periderm samples were taken below the apical bud. Plotted below are the concentrations of glucose, fructose and sucrose found in this tissue during storage. These results clearly show that the use of sprout suppressants interfere with the carbohydrate metabolism and these changes occur as the sprouting process begins.

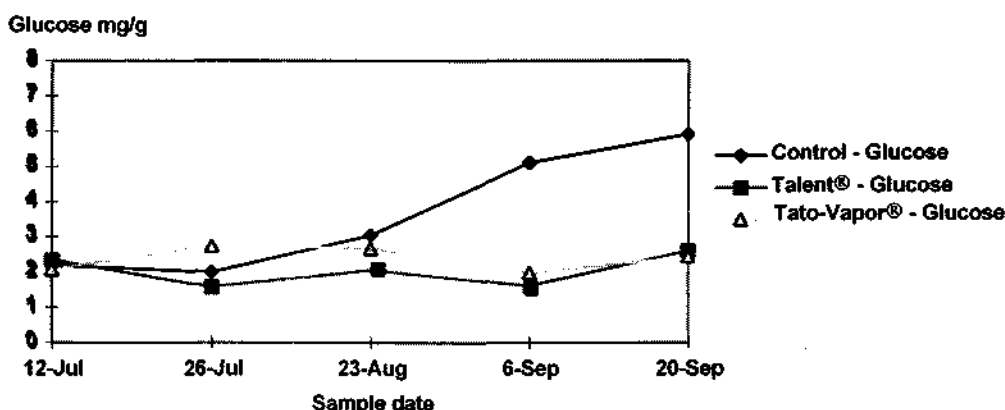


Figure 8. Glucose profile of the periderm sample

The glucose levels in both the Talent® and Tato-Vapo® treated potatoes followed a similar profile with glucose maintaining a steady concentration throughout the storage period. In contrast the untreated control showed a sharp increase from the 23rd August, which coincides with rapid elongation of the apical sprout.

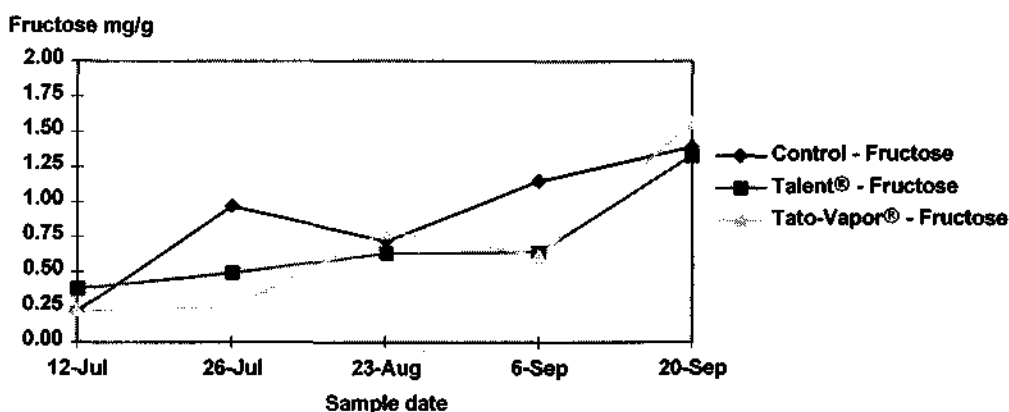


Figure 9. Fructose profile of the periderm sample

The fructose levels for all treatments were variable and, at the end of the storage period, were similar. The untreated control showed the largest initial accumulation and then gradually increased throughout the storage period. In contrast the Talent® and Tato-Vapo® treated potatoes gradually increased to the 6th September and then rapidly increased to a level similar to the untreated control.

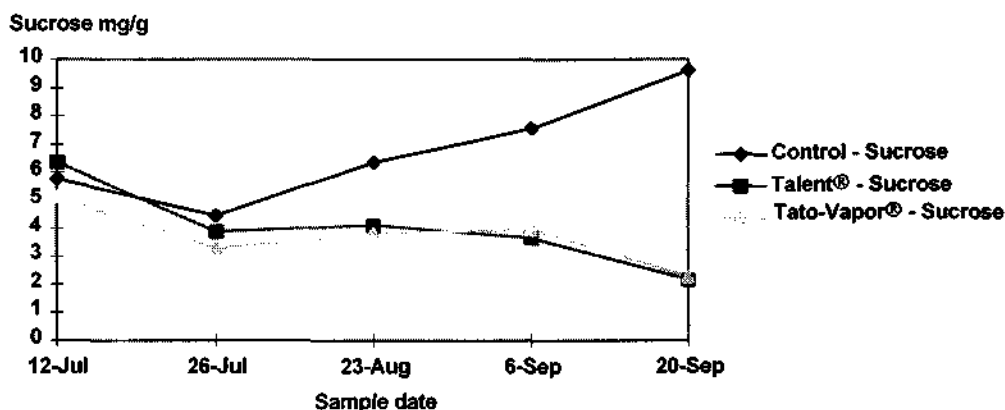


Figure 10. Sucrose profile of the periderm sample

The sucrose profile sharply contrasts the untreated control and those potatoes treated with sprout suppressants. All treatments had a reduction in sucrose in the first two weeks of storage but, from there, the untreated control increased gradually until the end of the storage period, whereas the treated potatoes showed a gradual decline. The rapid accumulation of sucrose in the untreated control appears to have occurred after the increase in fructose and indicated a change in carbohydrate metabolism as sprouting commences.

Cortex core samples

The cortex samples were taken directly below the periderm samples and show in comparison to the periderm tissues different biochemical processes are occurring. Similar to the periderm samples the following plots show the concentrations of glucose, fructose and sucrose through the storage period.

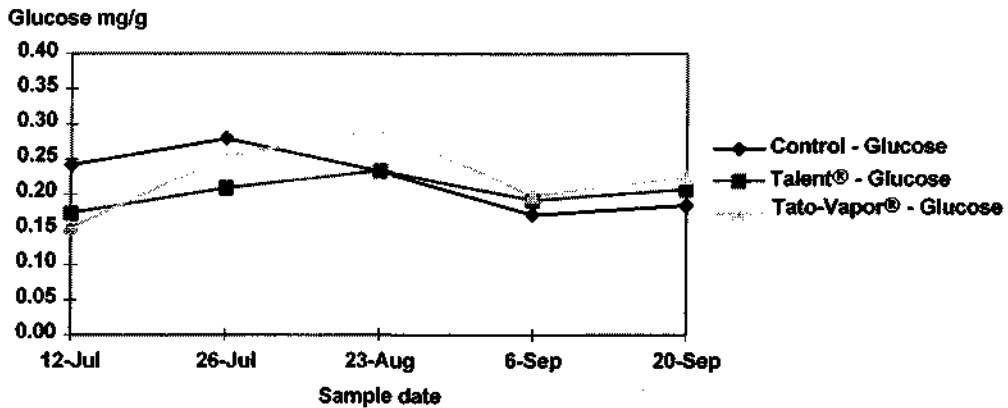


Figure 11. Glucose profile of the cortex sample

The glucose profile for all treatments is similar with a gradual rise over the first weeks of storage and then declining to a level near the starting concentration. In contrast to the periderm samples the sprout suppressant treatments have not affected glucose concentrations in these tissues.

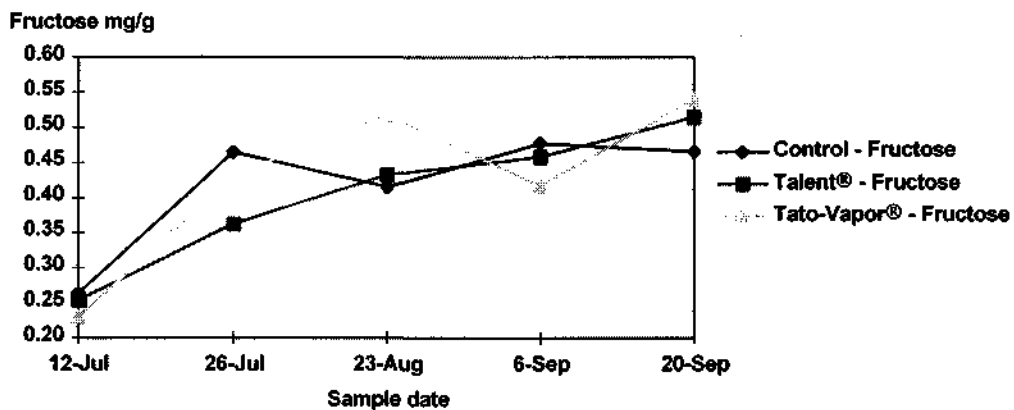


Figure 12. Fructose profile of the cortex sample

The fructose levels for all treatments sharply increased over the first two weeks of storage. The Talent® treated potatoes continued to increase gradually throughout the remainder of the storage period. The profiles for the control and Tato-Vapo® treated potatoes are more erratic. The fructose level in the control sample after the initial increase decreases and only gradually increases throughout the remainder of the trial. In contrast the fructose level in the CIPC treated sample increases over the first six weeks of the trial before sharply decreasing and then increasing over the last four weeks of the trial.

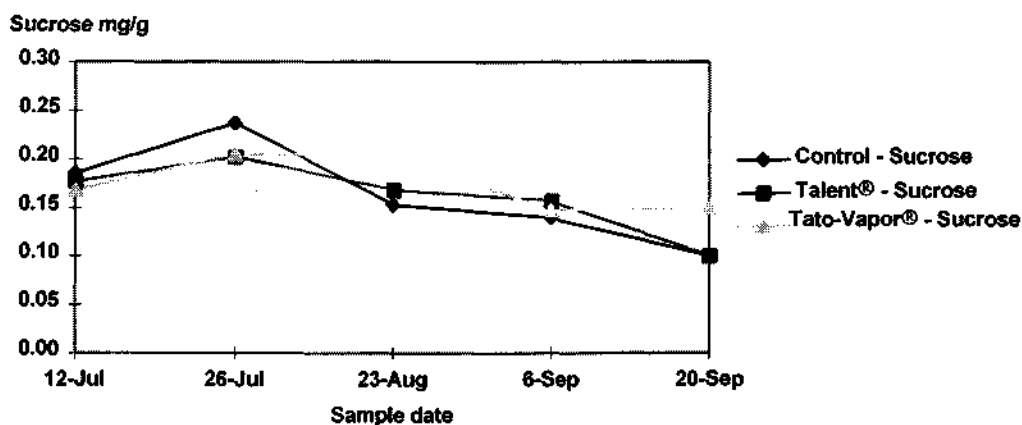


Figure 13. Sucrose profile of the cortex sample

The sucrose profile is similar to the glucose profile and shows that after an initial increase the levels decline over the remainder of the storage period which indicate that sucrose is not being accumulated in these tissues. In contrast to the periderm sample all treatments had a similar level through out the storage period.

2.5 Disease Assessment

Talent® strongly inhibited the development of silver scurf (*Helminthosporium solani*) in storage. Of the 21 tubers examined, 12 tubers showed the presence of silver scurf but all infections covered less than 5% of the tuber's surface area.

In contrast the 23 tubers from the control treatment were all infected; 15 tubers had over 5% of the surface area covered; in some cases this was greater than 50%. Similarly the 24 Tato-Vapo® treated tubers were all infected and 19 tubers had over 5% of the surface area covered.

IV. Discussion

The efficacy of alternative potato sprout suppressants

Talent®

Talent® has been shown to be an effective sprout inhibitor; although Talent® treated potatoes had a higher sprouting index than the Tato-Vapo® treated potatoes this was acceptable by industry standards.

Potatoes treated with Talent® began sprouting later than untreated potatoes and lost apical dominance due to a combination of factors. Among these are residues of Talent® in the periderm layer, the physical "burning off" of sprouts when subsequent applications are made and the partial reduction of assimilate as multi-site sprouting occurs. These observations are evidenced by the appearance of tubers after treatment and as the sprouts begin to re-develop. Only auxillary buds begin to develop at the apical site and all lateral buds begin to develop evenly.

The bulk storage of potato tubers in the 1995/96 trial improved the efficacy of this sprout inhibitor. It appeared that the volume of potatoes in the store and the volume of the head space is important in the distribution of chemical within the store and the concentration of the suppressant available to "burn-off" emerging sprouts. This agrees with the work conducted by Hartmans and Buitelaar (1993) which demonstrated that, for the same amount of chemical applied, repetitive small doses were less effective than fewer large doses.

These researchers have suggested that the head space concentration of Talent® throughout the storage period was important to the efficacy of this product. The trials reported here confirm that the initial head space concentration is critical to efficacy, but concentration in the long term is not important. The monitoring in 1994/95 showed that the head space concentrations reduced to below detectable levels three weeks after application and would have little physical activity on remaining sprouts. Oosterhaven 1993 confirmed that the cellular concentration of Talent® was responsible for sprout suppression after treatment rather than the remaining head space concentration. In the Australian trials, it was observed that the resumption of sprout growth after treatment was longer after each subsequent application. This may indicate that the build up of the Talent® in the potato is a controlling factor in sprout suppression and would agree with Oosterhaven's conclusions.

The timing of application of this sprout suppressant appears to be critical to its efficacy. In the 1994/95 trial, applications were made as soon as potatoes had been stored after a brief curing period, two subsequent applications were made at six weekly intervals. At the first application, no sign of sprouting was apparent but minor sprouting was apparent when the two subsequent applications were made. In comparison, the 1995/96 trials waited until visible signs of sprouting occurred before application. This resulted in physical damage of emerging sprouts but subsequent applications were required within two weeks, and it was not until the third application that acceptable sprout control was achieved.

This provides some evidence that the early presence of Talent® may slow down the sprouting process by both causing damage at the microscopic level and interfering with some of the biochemical changes that occur during sprout initiation and growth.

The efficacy of Talent® on the cultivar Denali was poor and the quality of the potatoes after storage was unacceptable due to excess sprouting and rising sugar levels. The modified refrigerated containers lacked adequate humidity control and Denali which was observed to have a thin periderm layer, lost more condition in comparison to Russet Burbank. This may have stressed the potatoes during early storage and strongly promoted the sprouting process.

The efficacy data collected over the past two years is sufficient for registration of Talent® and will be submitted to the National Registration Authority with the residue studies.

1-4 Sight®

1-4 Sight was shown to be an equally effective sprout suppressant in the 1995/96 trials. 1-4 Sight® controlled sprouting better than Talent® under the given storage conditions and application methods used. This was indicated by the low sprouting index for this treatment.

Potatoes treated with 1-4 Sight® began sprouting later than untreated potatoes and at a similar time to Talent® treated potatoes, but a major difference was that the emerging sprouts did not appear to be physically damaged by the application of the sprout suppressant. As with Talent®, all apical dominance was lost and sprouts that emerged from the apical and lateral buds were singular and undeveloped. 1-4 Sight® appeared to retard the development of emerging sprouts and subsequent application appeared to prevent further sprout development. Unlike Talent®, applying when macroscopic development is observed provides acceptable sprout control.

(d)-fenchone and Spinnaker®

Both these products failed to achieve acceptable sprout control in the 1994/95 and 1995/96 storage trials.

(d)-fenchone is a waste product of the Tasmanian essential oil industry and was thought to have had similar sprout suppression activity as many other essential oils. Whilst the full study of the efficacy of (d)-fenchone was not conducted, its effect at an application rate similar to Talent® proved to be unacceptable. As with Talent®, emerging sprouts were burnt and the tubers lost apical dominance. In contrast to Talent®, sprout emergence after treatment was rapid; this product did not appear to slow down the sprouting process. These observations agree with many reports that detail the differential efficacy of several essential oils. Whilst Talent® like other essential oil based products control sprouting through physical damage, Talent® stands alone as a product that effects the subsequent rate of regrowth. The biochemical reasons for this activity are yet to be discovered.

The active ingredient of Spinnaker®, *imazethapyr* has been successfully trialed overseas and registered in the United Kingdom as a potato sprout suppressant. Whilst short term sprout control was achieved the sprout indices at the end of the trial were unacceptable. This product prevents sprout development through the suppression of cell division in the meristem which was apparent shortly after application. Emerging sprouts began normal growth then stopped. They remained in this state until eight weeks after storage then resumed normal sprout growth. Further work is required to understand this product and factors that affect its efficacy.

Quality Aspects

Tuber quality

It is recognised that, coinciding with the breaking of potato “dormancy” and sprout growth is the mobilisation of starch within the tubers (Bailey 1978). Starch is hydrolysed to simple sugars which include sucrose, glucose and fructose. It is the presence of the hexoses glucose and fructose (reducing sugars) that is chiefly responsible for poor quality at the end of storage. Potatoes containing high levels of these hexoses tend to darken when cooked and produce unacceptable products.

There are a number of factors which can contribute to the accumulation of reducing sugars of which sprout emergence is one. Most Australian stores treat potatoes with CIPC which very effectively controls sprouting and delays the increase of reducing sugars. Generally potato storage managers monitor their stores for sugar accumulation by conducting regular fry tests. Cooked potatoes are compared to a colour charts which are used to describe the quality of the fry produced. It is therefore very important that any new sprout control chemical not only controls sprouting but does not contribute to the accumulation of reducing sugars.

Russet Burbank potatoes treated with either Talent® or 1-4 Sight® produced acceptable fries after storage. This differs to the findings by the Potato Marketing Board Research Group, UK (Briddon 1994) that Talent® caused darkening in some cultivars, including Russet Burbank. However in present trials Russet Burbank performed well.

The cultivar Denali performed poorly in the crisp assessment conducted by Smiths Snackfood Company. In comparison to Russet Burbank, Denali has a very thin periderm which appeared poorly set at the beginning of the storage trial. The modified refrigerated containers had poor humidity control, and this is thought to be a major factor in the deterioration of quality rather than the failure of the sprout suppressants to control sprouting and subsequent sugar accumulation. The major potato crisp producers in Australia are now reducing their reliance on stored potatoes as it is difficult to maintain these cultivars in an acceptable condition during long term storage.

Organoleptic assessment

An organoleptic assessment that was conducted to ensure that no flavour taints were detectable after processing. Talent®, in comparison to a number of essential oils known to inhibit sprouting did not leave any flavour taints. Oils such as 1-8 cineole (1995 Study tour, appendix 2) and (l)-carvone have been shown to leaves detectable residues after storage and processing. Further work to assess flavour taints in potatoes prepared for the fresh market is needed.

Mode of action of Talent®

The macroscopic examination of the tubers between treatments revealed that both Tato-Vapo® and Talent® inhibit sprouting via different mechanisms. Whilst both treatments delayed the onset of sprouting, Talent® treated samples produced near normal sprouts that resumed growth as the affect of the suppressant subsided. In comparison the Tato-Vapo® treated tubers were permanently damaged with sprouts appearing as undifferentiated callus tissue.

It was apparent sprouts not visible at a macroscopic level were damaged by the first application of Talent®. This was evidenced by the appearance of the apical or axiliary apical sprouts when they emerged and the proliferation of lateral shooting due to the loss of apical dominance. The scanning electron micrograph shows the damage to the apical bud soon after application (figure 7a & 7b) and provides some evidence that the early treatment of potatoes maybe important to the efficacy of this product. Further studies are required to quantify this damage, and the extent to which this affects sprout growth.

The direct affect of Talent® and/or its metabolites on sprout growth have been discussed; but growth may also be restricted by the numerous sprouts that are produced after apical dominance is lost. This may be due to the dilution of available carbohydrate which now has to be transported to a number of sprouting sites.

The sugar analysis of the potato tubers through the storage period showed that the process of sprouting and sugar accumulation were linked. The untreated control tubers which broke dormancy early showed a rapid accumulation of sugars in the tissues adjacent to the growing sprout. In contrast, both inhibitor treatments showed similar profiles, with little increase in either glucose or sucrose throughout the storage period.

This study does not indicate whether starch was mobilised from these adjacent tissues or transported from other storage tissues. However, Fishwick and Wright(1980) and Yada et al (1990) have shown, through electron microscopy that the amyloplasts adjacent to sprouting tissues remain intact during initial sprout growth and that assimilate must be transported from more distant storage tissues. It is well known that a condition referred to as “sugary end” is the result of starch mobilisation in the stem end of potato tubers which often occurs after potatoes have been stored for long periods.

The present sugar profile studies have shown that the use of Talent® as a potato sprout suppressant produced similar quality products to Tato-Vapo®. These results conflict with suggestions reported by the Potato Marketing Board after their trials during 1993/94. They concluded that the cultivar Russet Burbank may accumulate sugars more than other cultivars after treatment with Talent®.

Sprout reversibility and fungistatic properties

Talent® and 1-4 Sight®, in contrast to Tato-Vapo®, appear to be fully reversible sprout inhibitors. Talent® caused physical damage to emerging sprouts and as discussed in the efficacy of these products; it is postulated that the amount of suppressant absorbed by the periderm appears to affect the re-emergence of sprouts and growth rate of the sprout. All apical dominance is lost after the first application. 1-4 Sight® achieves a similar result without the physical damage. In contrast to Talent®, whilst apical dominance is also lost, the sprouts remain singular and dormant. The mode of action of this compound should be subject to further research.

The reversible action of these inhibitors may have applications in the seed and fresh market potato industry. For the seed potato industry, these compounds could be used as tools to store seed at higher temperatures, manipulate physiological age and reduce apical dominance in problem cultivars such as Atlantic, Shepody and Coliban. Growing on of treated material demonstrated no yield penalty and a more even tuber size distribution. This will be further studied during 1996/97. For the fresh potato market the use of natural sprout inhibitors may be a marketing advantage.

Oosterhaven (1993) first reported that (d)-carvone, the active ingredient of Talent®, inhibited the growth of *Fusarium solani* var. *coeruleum* and *Fusarium sulphureum*. These studies indicated that Talent® prevented fungal growth by inhibiting the metabolism of glucose. This observation may explain why these pathogens cause diseases during the later part of storage as sugar mobilisation begins. It was shown that this inhibition was not permanent as (d)-carvone was metabolised by the fungi to less effective compounds. During the current mode of action studies, Talent® effectively inhibited *Helminthosporium solani* (silver scurf). Further research is needed to assess the efficacy of this compound against the range of storage diseases found in Australia as it could provide valuable control..

V. Conclusions

When this project was initiated in 1994 the further use of CIPC was uncertain as the toxicology data held by CSU (now part of the NRA) was being reviewed. This data has now become available and a decision on the future of CIPC is imminent.

This project has achieved its goal by collecting the residue and efficacy data for a natural sprout suppressant Talent®. This product has been shown to be a suitable alternative to CIPC from the point of view of efficacy and public safety. Commercially however the cost of this product is inhibitory to its development whilst CIPC is still available. New application technology which is being trialed during 1996/97 has the potential to halve the application rate and thus make Talent® a viable commercial alternative.

The sprouting reversibility and fungistatic properties of Talent® are important findings of interest to the seed potato and fresh potato industries. These properties will be further explored.

During the second year of this project 1-4 Sight® was trialed and shown to be equally effective to Tato-Vapo® as a potato sprout suppressant. A further year of efficacy and residue studies have to be completed before registration can be sought. This product is applied at an extremely low dose rate and is a commercially viable option to Tato-Vapo®. As with Talent®, this product is fully reversible and has similar potential in the seed and fresh potato industries.

Through the potato industry levy and the HRDC further storage trials are being conducted to complete the residue and efficacy studies for 1-4 Sight® and to trial new application technology for Talent® during 1996/97. Sunrise Agriculture Pty Ltd with the University of Tasmania and TIAR will continue to research potato sprout physiology and the use of these sprout suppressants in the seed and fresh market potato industries.

Acknowledgments

Horticultural Research & Development Corporation.

McCain Foods (Aust) Pty Ltd

Simplot Australia Pty Ltd

Smiths Snackfood Company

Dr. Philip Brown, Alistair Gracie and Noel Davies, University of Tasmania

John Fennell and Rowland Laurence, DPIF

Technical assistance; Leon Hingston and Neil Drake, DPIF

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VII. Appendix 1 - Study tour report 6 -27 February 1995

Purpose of tour

During the past twelve months the DPIF has been testing a carvone containing formulation from BV Luxan as a potential natural sprout inhibitors for stored potatoes. The specific purpose of this study tour was to make contact with the major research centers investigating alternatives to CIPC (chloroprotham) as a sprout inhibitor. This objective was achieved with a complete picture of the developments worldwide with respect to sprout inhibitors and the ongoing investigation into CIPC.

General comments

This study included the following centers:

University of Idaho, Kimberley Research Station, Dr. Gale Kleinkopf
Pin/Nip Inc, Boise, Idaho, John Forsythe
McCain Foods Ltd, Florenceville, New Brunswick, John Walsh
Potato Marketing Board, Lincolnshire, UK, Adrian Briddon
KP Agriculture, Riseholme, Lincolnshire, UK, John Vessy
ATO-DLO, Wageningen, Netherlands, Dr. Klaasje Hartmans
BV Luxan, Elst Netherlands, Dr. Peter Diepenhorst

The following sprout inhibitors were investigated and will be reported by title in the following pages. Carvone, dimethylnaphthalene (DMN), diisopropylnaphthalene (DIPN), 1-8 cineole, PSS-25, maleic hydrazide, tecnazine and CIPC

Carvone

A carvone formulation called "Talent" was registered as potato sprout inhibitor in the Netherlands during 1994. Trials by the DPIF, Potato Marketing Board and ATO-DLO have shown this product to be an effective sprout suppressant that does not permanently inhibit the potato from sprouting. Commercial use of this product in the Netherlands has also been successful, however due to the large volumes of product that have to be applied (upto 100 mls/tonne) the current thermal foggers are unsuitable as several hours would be required to fog a large store. Technology is being developed to mist the product into the stores at the air inlets.

Currently this chemical formulation is sold for 100 Dfl / litre which at current label recommendations of 300 to 450 mls per tonne equates to a cost of \$20-30 per tonne. Clearly at this price this product could not be considered for general storage of ware and process potatoes. It may have a place in the seed potato industry and with high priced niche potato products. For example Japan is now asking for CIPC free potato fries and is willing to pay a higher price. Currently this is being achieved by processing large volumes of potatoes early prior to chemical treatment and servicing this market from long term frozen products which would cost more than \$20/tonne

The Potato Marketing Board has completed its second year of trials which show carvone to be an effective sprout suppressant but not equivalent to CIPC/IPC for some cultivars. The efficacy of carvone improved after multiple applications and the results for Russet Burbank were shown to be equivalent to CIPC/IPC. There are still some questions to the colour deterioration of some cultivars stored with carvone, Russet Burbank showed no colour deterioration during the trial. I have a copy of the final report from the PMB if further information is required.

BV Luxan has asked the DPIF to carry out a feasibility study to produce carvone in Tasmania for their markets. BV Luxan is also developing methods to convert waste oils from the citrus industry to carvone, methods to increase the oil content of caraway seed and the extraction efficiency of carvone from caraway seed.

Carvone has been shown to be active against storage diseases such as fusarium dry rot, black scurf, silver scurf, gangrene and early results from the DPIF show activity against common scab at a relatively high concentration. These benefits combined with the non-permanent sprout inhibition may make this product useful in the seed potato industry, particularly in light of the recent export enquiries.

Given the results achieved in Australia during 1994, BV Luxan agree that there is some latitude in the timing and interval between applications of carvone, contrary to their label recommendations of 50-100mls/tonne at six weekly intervals.

Application technology may have to be developed in view of the large volume of chemical that has to be applied.

Recommendation:

- The final large scale trial should be completed by the DPIF to collect efficacy and residue data. The cost of registration or further development will have to be met by BV Luxan or their agents in Australia.
- The fungistatic properties of carvone should be further investigated to check the activity against local strains of these pathogens.
- The correct application rate and application interval need to be established for Australian storage conditions.
- The use of carvone in the seed potato industry as a reversible sprout suppressant should be investigated.
- A full feasibility study of the production of carvone should be carried out to investigate the opportunity of supplying carvone to BV Luxan.

Dimethylnaphthalene (DMN) and diisopropylnaphthalene (DIPN)

This group of compounds were discovered as the potato's own sprout suppressants by British researchers in the late 1970's. Currently these compounds are by-products of the petroleum industry and are used as degreasers.

The University of Idaho has been investigating these products as sprout suppressants and has found them to be effective. The major problem of these industrial grade products is that DMN contains some eight different isomers and each individual isomer is known to have different physical and chemical properties. This is also a complication for registration as this product would be considered a mixture of different products and as such would require toxicological data for each isomer. Currently the production of individual isomers is not possible in the USA.

DMN was shown to be effective at headspace concentrations of 200 ppm but was not as effective as DIPN. Also an industrial grade DMN was shown by the Washington State University to be unsuitable due to flavour taints in stored potatoes after processing. The University of Idaho will discontinue work with DMN and continue to research DIPN which leaves no flavour taints and is effective at lower concentrations. This research will include the generation of residue and efficacy data and the development of breakdown chemistry which is required for registration. The DPIF will be collaborating with Dr. Gale Kleinkopf in this development.

Both DMN and DIPN are applied using a hot plate applicator which, involves placing the chemical on a hot plate and using heated air to fog into potato stores.

Whilst in the USA the registration of a single isomer of DMN was achieved by D-I-1-4, inc. This was achieved through a partnership with a Japanese chemical company that has developed the technology to either purify or specifically produce single isomers of DMN. Pin/Nip inc has registered 1-4 dimethylnaphthalene, Trade name 1,4Sight (EPA number 67727-1) as a potato sprout suppressant with a full toxicological package. 1-4 DMN is misted in and allowed to recirculate with closed vents for 24 hours, it is effective at 20 ppm and three applications prevent sprouting for up to 11 months.

I have a summary of the toxicological package and arrangements with Pin/Nip and their Japanese partners are being made to trial this product during 1995. The production of this isomer is only in small quantities at present but the Japanese company is developing mass production methods. There is no indication of price at the present time, but the principal of Pin/Nip John Forsythe estimates the cost of treating potatoes with 1,4Sight will be 1.5 to 2 times greater than the current costs of treating with CIPC.

Preliminary research shows that this compound aids skin set and wound healing of potatoes and may offer opportunities beyond ware and processing potatoes.

Recommendations

•In collaboration with the University of Idaho DIPN should be investigated as it is commonly available organic solvent. Basic efficacy and residue data can be generated by the DPIF but further development regarding toxicological packages and registration will require input from the suppliers and/or their agents in Australia. Information regarding the purity of industrial grade DIPN is being sought at present.

•A secrecy agreement with Pin/Nip inc will be entered into to develop 1,4 Sight in Australia. Initial indications are that Pin/Nip inc. or their Japanese partners are willing to support registration in Australia.

1 - 8 cineole

This chemical is the major constituent of eucalyptus oil and is widely available. It was identified as a potato sprout suppressant in the USA in 1991. Subsequent work by the Washington State University and McCain Foods Limited, Canada have shown it to be unsuitable due to unacceptable flavour taints and colour deterioration during storage.

Recommendation

•No further investigation into 1-8 cineole is warranted as it is clearly unsuitable.

PSS-25

This chemical was originally developed as a pre-emergent and post emergent herbicide for soybeans. The active ingredient *imazethapyr* was developed by the American Cyanamid Company and its development as a potato sprout suppressant has been developed by Cyanamid-UK and the Potato Marketing Board.

The efficacy data shows this chemical to be effective after one application. The chemical is applied at store loading as a mist (1 litre/tonne containing 1 gram ai / tonne). The mode of action is systemic, cell division is prevented in the meristems by inhibition of amino acid biosynthesis.

A full toxicological package is available and the advantages are that this chemical has been shown to have low mammalian toxicity and low environmental risks due to safe breakdown chemistry. I have a summary of the toxicological package which supports these claims.

An extension of this chemical as a potato sprout suppressant in Australia is likely given that this active ingredient is used in the soybean industry under the trade name "Spinnaker". The only data required would be efficacy and residue data, this would enable an appropriate MRL to be set for stored potatoes. Most of this information will be available through Cyanamid-UK when and if this chemical is registered in the UK.

This chemical is not thermo-stable and as such would not be suitable for fogging by conventional methods used in Australia. This chemical would have to be used as a mist applied to potatoes at harvest or during store loading. New technology being developed in Idaho may assist the development of this product.

This chemical suits the pre-grading and bin storage system peculiar to the United Kingdom and registration is set to occur in 1996/97 after further large scale demonstrations. Contact with Cyanamid-Aust Pty Ltd is in progress and development in Australia will be pursued.

Recommendations

- Small scale trials should be conducted by the DPIF to test efficacy of this product.
- Development will be dependent on developing suitable application technology and Cyanamid-Aust Pty Ltd's attitude to registration in Australia based on the volume of expected sales.

Maleic hydrazide

The University of Idaho is currently investigating better methods of timing the application of maleic hydrazide. This work involves developing a model in which the most appropriate time for application is determined and avoids damage to yield if applied too early or differential performance depending on tuber size if applied too late.

The DPIF will keep in touch with this work and perhaps collaborate on some of these trials as maleic hydrazide is of interest regarding the control of 'ground keepers' in Australia.

Tecnazine

This compound is used by 17% of the potato stores in the United Kingdom and is often used in conjunction with CIPC. Tecnazine is applied at store loading and is particularly useful for treating cultivars with no or little dormancy period.

This sprout suppressant is unlikely to be of any use in Australia, but confidential research is being conducted by the manufacturers and the Potato Marketing Board. I have the contact name in the UK of the company controlling this research and will attempt to contact them in the near future.

CIPC (chloroprotham)

USA

There seems little doubt that re-registration of CIPC will occur in the USA, data that has been submitted to the FDA is due to be reviewed in April 1995. Feedlot and poultry data is still outstanding but the amount of evidence which has been generated

may override this requirement in the short term. Provisional approval may be granted after this review. Full approval is expected to take two years.

Aceto has put together a data package from many sources including the large residue and metabolite studies done by the University of Idaho. Aceto failed to obtain the feedlot data from one of their competitors in the European market.

New application technology which involves the use of CIPC as a pure compound has increased the efficacy at lower rates and reduced the amount of residue found. The industry hopes to lower the MRL from 50 ppm to 20 ppm (still four times higher than the European Standard). The company Pin/Nip inc has developed and holds the patent for "puff" technology and has built three machines that treat 600 000 tonnes of potatoes in the USA annually.

United Kingdom

Registration in the UK will follow the European Union, but at present UK is not supporting the reduction of the MRL to European Standards. The major reason for this is that their bin storage system and poor air circulation lead to very high residues in the tops of bins, sometimes in excess of 100 ppm.

CIPC formulated in methanol as a solvent has been banned due to recent explosions within stores due to incorrectly applied chemical. The major CIPC formulation is in dichloromethane (marketed by BV Luxan) which is non-combustible and cheaper than the methanol formulation as sold in Australia and is claimed to be a better carrier.

European Union

There are three companies that have notified the European authorities that they wish to support the continued registration of CIPC as a potato sprout suppressant. These are Aceto, BV Luxan and Kermira, all of which claim to have sufficient evidence to support the use of CIPC.

In reality none of the companies have a complete data package and both the European companies will not deal with Aceto under any circumstance. They will deal with each other as long as the data they hold individually remains hidden from each other respectively and only viewed by the registration authority in a collective form. The data to be reviewed also include a large study conducted by the European potato industry headed by Dr. Harry Duncan. The NRA may be able to retrieve this document, which like the USA study shows that residues of CIPC, in particular chloro-aniline (a known carcinogen) can not be found in stored potatoes. This is only an issue where potato peels are broken down by soil bacteria, chloro-aniline is found to accumulate in the soil.

The review of these data packages is expected to take two to three years, which is the limit set by Austria which has threatened to ban its use after this time.

Recommendations

- There appear to be two sources of data to support the registration of CIPC in Australia and there is no need to tie the supply of CIPC to either source.

Conclusions

- CIPC is most likely to be retained world-wide and is unlikely to be rivalled as a cost effective method of sprout inhibition. The NRA should be briefed as soon as possible on the developments in the USA and within the European Union to set a realistic approach for the Australian Industry.

- The most promising new sprout suppressant is the single isomer of DMN which has recently been registered by the FDA. This compound should be progressed towards registration during 1995.

- "Talent", formulated by BV Luxan, will continue to be a minor use product in Europe. The cost of treatment with "Talent" will prevent wide-scale adoption in Australia, the main benefits will be to the seed potato industry as a reversible sprout inhibitor with broad fungistatic activity.

- Both DIPN and PSS-25 should be tested to gather basic efficacy and residue data. Further progress will be dependant on the supplying companies and the development of technology that suits storage conditions in Australia.

- New developments in the application technology for CIPC should be pursued in Australia. These methods have the capacity to reduce chemical residue, improve chemical distribution and further lower the cost of application.