# Horticulture Innovation Australia

## **Final Report**

Controlling multiple heading and transplant shock in lettuce

Dr Gordon Rogers Applied Horticultural Research Pty Ltd

Project Number: VG12017

#### VG12017

This project has been funded by Horticulture Innovation Australia Limited with co-investment from Applied Horticultural Research Pty Ltd and funds from the Australian Government.

Horticulture Innovation Australia Limited (Hort Innovation) makes no representations and expressly disclaims all warranties (to the extent permitted by law) about the accuracy, completeness, or currency of information in *Controlling multiple heading and transplant shock in lettuce*.

Reliance on any information provided by Hort Innovation is entirely at your own risk. Hort Innovation is not responsible for, and will not be liable for, any loss, damage, claim, expense, cost (including legal costs) or other liability arising in any way (including from Hort Innovation or any other person's negligence or otherwise) from your use or non-use of *Controlling multiple heading and transplant shock in lettuce*, or from reliance on information contained in the material or that Hort Innovation provides to you by any other means.

ISBN 0 7341 3787 7

Published and distributed by: Horticulture Innovation Australia Limited Level 8, 1 Chifley Square Sydney NSW 2000

Tel: (02) 8295 2300 Fax: (02) 8295 2399

© Copyright 2016

## **Contents**

Summary	5
Background	5
Blindness	5
Transplant shock	5
Keywords	7
Introduction	8
What is multiple heading in lettuce?	8
Transplant shock	8
Aim of this project	9
Methodology	10
Commercial incidence of lettuce blindness	10
Grower survey	10
Monitoring blindness in nurseries	10
Glasshouse trials	11
1. Effect of seedling mix	11
2. Effect of nutrient mix and concentration	11
3. Effect of increased nutrients and extended day length	12
4. Effect of salinity, physical or chemical damage	12
5. Effect of salinity, chlorine and calcium supplementation	12
Reducing transplant shock	13
Field trial 2014	13
Field trial 2015	13
Outputs	15
Outcomes	16
Commercial incidence of lettuce blindness	16
Glasshouse trials	16
1. Effect of seedling mix	16
2. Effect of nutrient mix and concentration	16
3. Effect of increased nutrients and extended day length	17
4. Effect of salinity, physical or chemical damage	17
5. Effect of salinity, chlorine and calcium supplementation	17
Reducing transplant shock	17
Field trial 2014	17
Field trial 2015	18
Evaluation and Discussion	19
Lettuce blindness	19
Reducing transplant shock	19
Recommendations	20
Lettuce blindness	20
Reducing transplant shock	20
Intellectual Property/Commercialisation	21
Acknowledgements	21

Appendices	22
1. Commercial incidence of lettuce blindness	23
Grower survey	23
Background and Aim	23
Method	23
Results	23
Discussion	25
Monitoring blindness in nurseries	26
Background and Aim	26
Method	26
Results	27
Discussion	30
2. Bosch glasshouse trials	31
Effect of seedling mix	31
Background and Aim	31
Method	31
Results	32
Discussion	37
Effect of nutrient mix and concentration	38
Background and Aim	38
Method	38
Results	40
Effect of high nutrient mix and extended day-length	42
Background and Aim	42
Method	42
Results	43
Discussion	44
Effect of salinity, physical or chemical damage	45
Background and Aim	45
Method	45
Results	48
Discussion	52
Effect of salinity, chlorine and calcium supplementation	53
Background and Aim	53
Method	53
Results	55
Discussion	56
3. Reducing transplant shock	57
Background	
Field trial 2014	
Aim	
Method	
Results	
Discussion	59

5. Reducing transplant shock in lettuce – Fact Sheet	
4. Lettuce Blindness Fact Sheet	67
Discussion	65
Results:	61
Method	60
Aim	60
Field trial 2015-2016	60

## **Summary**

## **Background**

Multiple heading, or 'blindness' is a physiological disorder affecting up to 20% of lettuce crops. It is most prevalent in NSW and Queensland during summer months, but can occur any time. Blindness refers to loss of the apical shoot. The result is a lettuce that is deformed and unsalable. The causes of blindness are unclear, although a range of factors are suspected to increase risk.

Blindness occurs when lettuces are still young seedlings, although the disorder is often not apparent until after planting and development. In some cases the young plants can recover. However, further stress exacerbates the situation.

Transplanting lettuces while they are still small minimises damage to the developing roots and allows the plants to establish quickly. However, young plants are fragile and difficult to handle. Transplanting when lettuces are further developed means they are more robust, but inevitably damages roots and can result in 'transplant shock', where the plants fail to establish well and thrive.

In this project we have examined the two important issues affecting lettuce growers of blindness and transplant shock.

## **Blindness**

Stakeholders were surveyed on factors they considered likely to increase the disorder, and mitigation strategies. High temperatures and humidity are widely believed to be a key factor. The incidence of blindness in seedling nurseries in NSW and Queensland was monitored between November and March and correlated to onsite temperature and humidity records. Blindness was extremely variable, and while incidence increased after some hot spells, after others it did not. No specific relationships were found between blindness and temperature.

A series of trials were conducted in the glasshouse examining some of the stress factors that may contribute to blindness. These included high nutrient concentrations, high/low calcium, high/low boron, applications of insecticide, wetting agents, physical damage from overhead irrigation or brushing, saline irrigation water and chlorinated irrigation water.

Even quadrupling the strength of fertigation solutions did not increase blindness. Blindness was also not induced by physical damage, low calcium or boron, or insecticides which simply 'burned' the plants. However, increasing EC with salt, or adding chlorine to the irrigation water, did induce blindness. There was some evidence that a foliar spray with chelated calcium could protect plants, with this treatment slightly reducing blindness in controls as well as plants irrigated with saline water.

## Transplant shock

Field trials tested the effect of seedling drenches on early growth and final yield of iceberg and cos lettuces. Drenches were applied alone and in combination, and 48 hours before transplanting as well as immediately before transplanting.

Drenching with potassium nitrate increased leaf growth, particularly early in growth, although in the

second trial the effect was lost by the time lettuces reached harvest maturity. This may be due to reduced root development in these plants, which made them more susceptible to hot weather and irrigation stress.

Confidor® and Durivo® may be applied to seedlings as drenches to control lettuce aphid before transplanting. While some growers felt that these products could also enhance yield, the opposite effect was also possible. Our trials did not find any significant effect of either product on lettuce growth or yield, or consistent interaction when they were combined with the potassium nitrate drench.

While the potassium nitrate drench offered some benefits, the largest effect in the second trial was due to position in the field. This was due to changes in soil type, as well as possibly uneven irrigation. It was concluded that although potassium nitrate can reduce transplant shock in lettuce, field effects may be more important in terms of overall yield.

## Keywords

Lettuce, Multiple heading, Blindness, Transplant, Deformed, Disorder, Cos, Iceberg, Drench, Confidor®, Durivo®, Potassium nitrate, Insecticide, Shock

## Introduction

## What is multiple heading in lettuce?

Blindness, or multiple heading, is a significant disorder affecting lettuce crops. Blindness occurs when the main apical shoot of the lettuce fails to develop properly. This usually occurs in the seedling phase, but may go unnoticed until the seedlings have been planted out in the field and are well on the way to maturity.

Lettuce plants affected by the disorder normally respond by producing other vegetative buds, which try to take over the role of the apical shoot. The result is multiple, small and misshapen heads and an unsaleable product.

Ideally, blind seedlings are removed from trays before planting out. However, this is both difficult and costly. However, if blind seedlings are planted in the field, all the costs of production are incurred for an product that cannot be sold.

Blindness is mainly an issue for seedling producers in NSW and Queensland, particularly during summer. It occurs sporadically and unpredictably, so no single cause for the disorder has been identified. However, up to 20% of seedlings may be affected at times. The disorder is estimated to cost seedling producers and growers \$8.8 million annually, making it a major production issue.

Factors that are believed to increase rates of blindness in seedling nurseries include:

- Variety susceptibility
- High temperatures during seed germination
- · Boron deficiency in the seedling media
- Molybdenum deficiency

In some cases, seedlings appear to recover after planting. However, other factors can exacerbate the problem, including:

- Transplant stress, such as due to poor irrigation management or overdeveloped seedlings
- High temperatures and humidity during early growth
- Application of certain fungicides or insecticides, particularly under warm conditions
- Incorrect planting depth

## **Transplant shock**

Even with well designed seedling trays and optimum care, the process of transplanting from trays to soil inevitably damages plant roots and disrupts growth. If seedlings are transplanted while very small they are likely to recover quickly, and maximise subsequent growth. However, small seedlings with immature root systems are difficult to handle. They are easily damaged during transplanting, have little resistance to water stress and so are susceptible to transplant shock.

Seedlings that are grown for longer before transplanting are sturdier and easier to handle. However, they are also more cell-bound, with dense knots of roots that may fail to develop correctly after planting

out.

Seedlings that are either too small or too large when transplanted can fail to develop well in the field. Some may die, further reducing yield, especially if stressed by adverse weather conditions.

There is some evidence that applying nitrogen and potassium to the roots as a potassium nitrate drench just before transplanting can help plants overcome transplant shock. Such a treatment may help the plant adjust to other stresses, such as insecticides used to control lettuce aphid.

## Aim of this project

This project was developed in consultation with industry, particularly seedling producers in NSW and Queensland. The two major issues of concern were;

- 1. Blindness, and the resulting problem of multiple heading and unsalable lettuce
- 2. Transplant shock and the effects on harvestable yield, particularly if plants were also treated with an insecticide (such as Imidacloprid, the active ingredient in Confidor®) for aphid control

The aims of the project were therefore to:

- 1. Examine whether the incidence of blindness is correlated with other environmental factors, such as temperature and humidity
- 2. Test treatments that could potentially induce or prevent blindness in susceptible lettuce varieties
- 3. Trial the use of a potassium nitrate drench for reducing transplant shock in lettuce seedlings and measure the effect on yield
- 4. Test whether drenching lettuce seedlings has a positive or negative effect on growth and yield, and whether such treatments interact with potassium nitrate.

## Methodology

The research conducted during this project involved a range of different activities. These included surveys of growers, weekly counts of blind lettuces at seedling nurseries, glasshouse trials and finally field trials.

## Commercial incidence of lettuce blindness

## **Grower survey**

Around 25 seed suppliers, seedling producers, and lettuce growers were contacted regarding their observations of the incidence of lettuce blindness. Interviews were in person, or by phone. Growers were asked about what lettuce types and specific varieties they had found to be susceptible to the disorder, the time of year when the problem was most likely to be severe, factors they thought contributed to the disorder and measures that may help control.

The results were summarised to find common themes, which could then be prioritised for further research.

## Monitoring blindness in nurseries

Data-loggers were used to record temperature and humidity at Withcott Seedlings in Queensland and Choice Seedlings in NSW from September 2013 to January 2014. Each week, 10 trays of seedlings (cv. Leu Quintus at Withcott and cv. Kireve at Choice) which were minimum 3 weeks post seeding were examined to determine the number of blind plants.

Withcott nursery in Queensland conducted two small additional trials;

- 1. Lettuce cv. 'Arganda' was germinated at either 15°C or 20°C for two days before transfer to the main nursery.
- 2. Lettuce cv. 'Arganda' were given weekly applications of foliar nutrients;
  - a. standard nutrients (control)
  - b. +calcium
  - c. +boron
  - d. +boron and calcium
  - e. +boron, calcium and molybdenum

The number of blind seedlings was recorded 23 days after seeding. Blindness was defined as any seedling where the apical point was either missing or distorted (Figure 1).





Figure 1 - Normal lettuce seedling (left) compared to blind seedling (right) with distortion of leaves at the apical point

Full experimental methods, results and discussion from these trials are included as Appendix 1 to this report.

## Glasshouse trials

A series of five trials were conducted in the Bosch glasshouse at Sydney University. This facility has controlled temperature and irrigation scheduling, and supplementary lighting for use during winter. Room settings provided high temperatures and humidity, particularly during the day. This was most successful in summer, as losses during winter often prevented it attaining the high setpoint.

All trials used standard 128 cell seedling trays filled using a commercial seedling mix. The primary aim was to induce blindness in lettuce seedlings to identify factors that increase this disorder. This would then allow development of mitigation strategies.

## 1. Effect of seedling mix

This trial tested the effect of three different commercial seedling mixes on rates of blindness of two potentially susceptible lettuce varieties – 'Blanes' (cos type) and 'Kireve' (oak leaf type).

Lettuce were seeded into the three different mixes. The trays were then cut in half, with one half receiving fertigation (Aquasol, 16g/10L water) and the other irrigation alone. Plants were examined 21 days after seeding to determine rates of blindness, establishment and plant weight.

#### 2. Effect of nutrient mix and concentration

Several growers had suggested that high EC was a key cause of increased occurrence of blindness. Calcium and boron deficiency have also been linked to lettuce blindness.

A fertigation mix was fabricated using inorganic materials. The objective was to produce a basic mix with a similar nutrient profile to 'Nursery Blend', but to which calcium and boron could be added or subtracted.

Twenty five trays of lettuce cv. Blanes were divided among the five fertigation treatments;

1. Standard

- 2. High calcium
- 3. Low calcium
- 4. High boron
- 5. Double strength standard

Plants were examined 28 days after seeding as previously described.

## 3. Effect of increased nutrients and extended day length

This was a repeat of the previous trial but with more extreme conditions: higher room temperatures, longer daylength and up to 4 x normal strength fertigation. Plants were examined 26 days after seeding as previously described.

## 4. Effect of salinity, physical or chemical damage

This trial tested a wide range of methods of inducing stress in lettuce seedlings. A total of 26 trays seeded with lettuce cv. Kireve were divided among 13 treatments;

- Control
- 2. Mechanical brushing
- 3. Chlorpyrifos
- 4. High fertiliser
- 5. High fertiliser + small growing cells (198 cell tray)
- 6. Wetting agent
- 7. Sodium molybdate
- 8. High boron
- 9. Strong overhead irrigation
- 10. Chlorinated water (200ppm)
- 11. Saline water (2,000 µs.cm<sup>1</sup>)
- 12. Saline water (4,000 µs.cm<sup>1</sup>)
- 13. Saline water  $(2,000 \mu s.cm^{1})$  + chlorine (200ppm)

All trays were also given normal fertigation. Plants were examined 27 days after seeding as previously described.

## 5. Effect of salinity, chlorine and calcium supplementation

Trial 4 identified irrigation water salinity and chlorine as factors that increased blindness in lettuce seedlings. This trial therefore repeated some of these treatments, but with and without the addition of calcium as a remediating factor. A total of 24 trays seeded with lettuce cv. Kireve were divided among 8 treatments;

1. Control

- 2. Control + calcium
- 3. Chlorine (100ppm)
- 4. Chlorine + calcium
- 5. Saline water (2,000 μs.cm<sup>1</sup>)
- 6. Saline water + calcium
- 7. Saline water+ chlorine
- 8. Saline water + calcium + chlorine

All trays were also given normal fertigation. Plants were examined 20 days after seeding as previously described.

Full experimental methods, results and discussion from these trials are included as Appendix 2 to this report.

## Reducing transplant shock

Field trials were conducted in Western Sydney over two seasons (2014 and 2015). Both trials examined the effect of a seedling drench with potassium nitrate alone or in combination with an insecticide.

#### Field trial 2014

Cos (cv. Quintas) and Iceberg (cv. Toscanas) lettuce seedlings were treated with one of seven different drenching solutions;

- 1. Control (water only)
- 2. Potassium nitrate
- 3. Seasol®
- 4. Confidor®
- 5. Durivo ®
- 6. Seasol® + Potassium nitrate
- 7. Duriyo® + Potassium nitrate

The seedlings were then transplanted as a randomised block design with 4 blocks per treatment. Plant weight was recorded 4 weeks and 7 weeks after transplanting.

#### Field trial 2015

Trays of iceberg lettuce seedlings (cv. Bernadenas) were drenched 48 hours before transplanting with Confidor® or Durivo®, each with or without the addition of Potassium nitrate. Immediately before transplanting these drenches were repeated on additional trays. Trays were also drenched with potassium nitrate alone or water only (control), giving a total of ten treatment combinations.

Seedlings were transplanted in a randomised block design with 4 blocks per treatment. Both shoot and root weight were recorded 3 weeks and 7 weeks after transplanting.

Full experimental methods, results and discussion from these trials are included as Appendix 3 to this report.

## **Outputs**

Preliminary results were presented at the Ausveg regional roadshow at Rossmore 18/5/2014. The presentation included a description of different symptoms of blindness and discussion with local lettuce growers as to contributing factors.

An article on the trials was published in Vegetables Australia December 2014 issue. The results from the 2014 transplant shock trial were communicated to growers, explaining that potassium nitrate applied to seedlings prior to transplant significantly improved yield results — up to 20%

Meetings have been held and results communicated directly with commercial lettuce hydroponic growers in the Windsor, Annangrove and Kenthurst areas, as well as with the seedling producers involved in the trials (Choice and Withcott).

Two Fact Sheets have been drafted and will be distributed as part of the Integrated Crop Protection extension project. These fact sheets are currently with the graphic designer and will be printed for distribution as well as made available electronically through the ICP and AHR websites. Draft copies of the Fact Sheet on Lettuce Blindness (Appendix 4) and use of potassium nitrate drenches (Appendix 5) are appended to this report.

## **Outcomes**

## Commercial incidence of lettuce blindness

There were a number of common comments among lettuce seedling suppliers as to factors that increase lettuce blindness. While these were at times contradictory, a number of key themes emerged including;

- High EC in irrigation water
- High air or media temperatures during germination and early development
- High humidity / low soil moisture
- High light levels
- Unbalanced nutrients boron, calcium, magnesium

Managing temperature was seen by many as a key part of reducing the incidence of blindness. Plant nutrition was also seen as very important, as was variety selection, with some varieties consistently reported as susceptible to the disorder.

There seems strong evidence that blindness is more common during summer. However, counts of blind plants combined with temperature and humidity data in NSW and Queensland failed to find any strong or specific relationship between these factors. Even when high rates of blindness were recorded following a hot spell, a similar series of hot days at another time would fail to have any effect.

It was concluded that while hot weather may increase susceptibility, it is only one of a range of factors that induce this disorder.

#### Glasshouse trials

The percentage of blind lettuces was extremely variable across all trials, with some results appearing contradictory.

## 1. Effect of seedling mix

Nearly 10% blindness was observed in lettuces cv. 'Blanes' grown in Mix 2 without added fertigation. However, the opposite effect occurred for cv. 'Kireve' grown under the same conditions, with blindness higher in the lettuces which did receive fertigation.

In this trial most of the lettuces failed to develop properly, mainly due to nutrient stress. The high rates of irrigation increased humidity, but also leached nutrients from the soil. Apart from improved growth in Mix 2, which included fertiliser, no consistent differences were observed between the 3 mixes tested.

#### 2. Effect of nutrient mix and concentration

Day temperatures during this trial were 30-34° combined with moderate to high RH. Only a single blind seedling was found out of 3,200 grown. Doubling the nutrient concentration, adding or subtracting

calcium and increasing boron to high levels all failed to induce blindness.

## 3. Effect of increased nutrients and extended day length

This trial repeated the conditions in trial 2, but with longer daylength, higher temperatures and even higher concentrations of nutrients. Despite this, no blind lettuces were found. This confirmed the previous finding that – in opposition to the common belief held among seedling producers – high rates of fertilisation do not increase lettuce blindness, at least under the conditions in this trial.

## 4. Effect of salinity, physical or chemical damage

Extreme treatments were used in this trial, so many of the lettuces grew with acute stress. These stresses caused a range of symptoms, which included loss of the apical shoot – blindness – but also burning, damage and stunting of the growing point.

The chlorpyrifos treatment, for example, caused immediate and acute burning of the inner leaves, resulting in symptoms very similar to blindness. Irrigating plants with water containing 200ppm chlorine also caused severe damage, with very short or stunted apical shoots. Saline water reliably caused both blind and stunted plants, particularly if water was also chlorinated.

While the overhead irrigation resulted in a very low level of blindness, the brushing treatment did not, suggesting that physical damage is unlikely to be a key factor. Strong nutrient solutions, increased boron, and other treatments also did not result in any blind plants, again confirming the results found in trials 2 and 3.

The results therefore suggest that plant stress is a key factor in induction of this disorder – whether due to saline water, chlorinated water, or chemical challenge.

## 5. Effect of salinity, chlorine and calcium supplementation

This trial confirmed previous results, indicating that saline and/or chlorinated water can induce blindness in 'Kireve' lettuce. This supports observations by growers that blindness is increased if water has high EC.

Calcium reduced the incidence of blindness. This trial was the first in which blindness occurred in the untreated control trays. However, when calcium was also added, no blindness was observed. There was a clear trend to reduced rates of blindness in the chlorine and saline water treatments when plants were also provided with extra calcium – although variability in results means data is not significantly different.

The results confirm the effects of plant stress on blindness, and suggest a protective role for calcium when applied as a foliar spray (1g.L<sup>-1</sup>) twice weekly during early development.

## Reducing transplant shock

Different results were found in 2014 to those in 2015. It seems clear that the effects of soil drenches on lettuce seedlings vary with other environmental factors.

#### Field trial 2014

Yields of both iceberg and cos lettuce were increased when seedlings were drenched with potassium

nitrate prior to harvest. However, adding potassium nitrate to either a Confidor® or Durivo® drench failed to result in a significant difference to Confidor® or Durivo® alone, with a trend to reduced head weights in these plants.

Head weight of iceberg lettuces drenched with Seasol<sup>®</sup>, Confidor<sup>®</sup> or Durivo<sup>®</sup> alone was not significantly different to the untreated controls. In the case of cos lettuce, there was a small but significant increase in harvest weight when lettuces were drenched with Seasol<sup>®</sup> or Confidor<sup>®</sup>, but not Durivo<sup>®</sup>, and not when potassium nitrate was also added to the drenching solution.

The results indicated that drenching lettuce seedlings with Confidor<sup>®</sup>, Durivo<sup>®</sup> or Seasol<sup>®</sup> had little effect – either positive or negative – on final head weight when compared to untreated seedlings.

#### Field trial 2015

Drenching with potassium nitrate before transplanting increased early growth of shoots and decreased root growth in iceberg lettuce. The effect on yield was lost, however, once plants reached harvest maturity, although a small but significant impact on root growth remained. It seems possible that early root growth is reduced in the potassium nitrate drenched seedlings as nitrogen and potassium are readily available within the root zone. However, if water stress occurs – as it did in this trial, due to two separate heat waves – then plants with smaller root systems will be disadvantaged, allowing others to 'catch up'.

This trial confirmed the previous results that Confidor® or Durivo® drenches had no significant effect on lettuce growth and development.

In this trial, the largest effect on lettuce growth was due to position in the field. This was due to changes in soil type across the length of the beds, as well as uneven irrigation patterns.

## **Evaluation and Discussion**

#### Lettuce blindness

This project has identified several factors that increase blindness in lettuce seedlings. These include

- Saline irrigation water
- Chlorinated irrigation water
- Application of chemicals that can result in a chemical burn
- High temperatures and humidity

Many growers felt that high nutrient levels in fertigation solutions were also an issue, with many irrigating after fertigation to remove nutrients from the leaves. In these trials the only effect of increasing the concentration of nutrients was increased growth by the plants.

The results also did not find any increase in blindness due to physical disruption of the growing tip or application of wetting agents or sodium molybdate to young plants. Despite large differences in nutritional profile and composition, there were also no differences in blindness rates between seedling mixes.

One of the difficulties encountered during the trials was that opinions differed as to what constituted a blind plant.

The original definition of 'blindness' was based on it being a physiological disorder. While the lack of an apical shoot is the key symptom, a plant classified as 'blind' may also be one with deformed developing leaves, or which has multiple growing tips with no clear dominance. Moreover, a plant classified as truly blind also usually has thickened and distorted outer leaves, giving the whole plant a 'mongreled' appearance.

However, similar symptoms may be induced by chemical or physical damage. These can occur due to chemical burn or severe physical injury. While such treatments physically destroy the apical shoot, the plant may sometimes recover and still produce a normal heart. Stress can cause stunting of the apical shoot, and the appearance of blindness, but the plant will recover if the stress is removed.

## Reducing transplant shock

Overall, the trial results support the previous observation that potassium nitrate drenches can reduce transplant stress. This can, in some circumstances, result in increased yield or earlier harvest maturity.

However, if plants are stressed, then the benefits of this early adaption may be lost. This is due to reduced early root growth in the potassium nitrate drenched seedlings, which makes them more vulnerable to hot weather events or irrigation deficits.

Both the 2014 and 2015 trials found no effect of drenching with the insecticides Confidor<sup>®</sup> or Durivo<sup>®</sup> on growth of lettuce seedlings. This means that if growers need to drench seedlings to control lettuce aphid, they are able to do so. However, the results also suggest that – contrary to beliefs held by some growers – such products do not enhance growth of lettuce seedlings.

## Recommendations

#### Lettuce blindness

The reduction in incidence of blindness when chelated calcium was applied as a foliar spray is a promising result worthy of further replication. It is interesting to note that calcium had less effect when applied as part of the fertigation solution.

Trials using foliar applications of calcium could be done as a larger glasshouse trial, as was done here, where stressed seedlings are given a foliar application. The method could also be tested under commercial conditions at times of year when growers feel that blindness is more likely to be an issue.

The trials have identified a number of factors that appeared to increase blindness. However, the occurrence of blindness is extremely variable. Repeating such work, preferably on a larger scale and/or under commercial conditions, could confirm whether these factors are key to this disorder in lettuces.

## Reducing transplant shock

The positive results recorded for potassium nitrate drenches on reducing transplant shock appear promising. However, variability with the field trial in 2015 demonstrates the important of good site selection for such trials. Selecting a site with more uniform characteristics may have improved the result.

The effects of potassium nitrate drenching on root development is particularly interesting. It had been expected that root growth would increase, correlating with shoot growth. However the opposite occurred. While it is hypothesised that this is due to greater nutrient availability within the root zone, this also needs confirmation. Studying this effect is particularly important if such a treatment is to become more widespread, as it is likely to make plants less tolerant of drought or other environmental stresses.

## **Intellectual Property/Commercialisation**

No commercial IP generated

## **Acknowledgements**

This project would not have been possible without the generous assistance of a number of different seedling producers and seed supply companies.

Of particular note:

Jeff Borg Choice Seedlings

Graeme Erhardt Withcott Seedlings

**Boomaroo Nursery** 

Phil Ritchie Rijk Zwaan

Harry Turna Rijk Zwaan

Chris Portelli Portelli's Produce

## **Appendices**

## 1. Commercial incidence of lettuce blindness

## **Grower survey**

## **Background and Aim**

Blindness in lettuce is a condition that can develop in the seedling phase. If the condition persists it results in multiple heading in mature plants and the lettuce is unsalable. The condition is generally a problem for seedling producers over summer from October to February in NSW and September to January in Queensland.

Lettuces that have failed to develop normally as seedlings should therefore be removed from trays before sending to growers for planting. This can be a significant cost, not only due to wasted seedlings, but due to the manual labour required to check trays and replace as necessary.

As this issue is a major concern to growers and breeders, many have closely observed factors that appear to increase or decrease incidence of this disorder. The aim of this activity was therefore to 'shortlist' possible contributing factors, which could then be examined further in controlled trials.

#### Method

Around 25 lettuce breeders and seedling producers were contacted about the project. Representatives from major suppliers such as Boomaroo, Withcott, Choice and Speedy Seedlings were interviewed personally. Other producers were contacted by phone and/or letter.

Seedling producers were asked questions including:

- Irrigation method for seedlings
- Cover type (if any)
- Tray size
- Type of potting media
- Germination room or open benches
- Experiences with lettuce blindness how big a problem, what varieties, possible causes and treatments to minimise blindness that should be investigated

The results were summarised to find common themes, which could then be prioritised for further research.

## Results

A number of common themes were identified from the survey as well as individual discussions with seedling producers. Certain varieties were identified as being particularly susceptible to this disorder (Table 1). Factors that many producers felt increased the likelihood of blindness included; high EC in irrigation water, high temperature and humidity during early growth, stress due to availability of water, nutrients or incorrect pH and strong, direct light. A wide range of different control strategies were

proposed for testing, many of which were focused on these issues (Table 2).

Table 1 - Summary of grower responses to survey - lettuce varieties susceptible to blindness, and factors that increase incidence

Susceptible variety	Susceptible lettuce type	Causative factors
Kireve (green oak) Rz	Worse in cos than iceberg	EC (various & contradictory statements)
Ralph (baby cos) Rz	Lollos	Humidity
Arganda (cos)	Butter	Temperature during germination
Goblin (cos)	Some multi leaf types	Low moisture
Blaines (cos)	Red coral	High light levels
Shrek(cos)	Green coral	Nutrient imbalance (boron, calcium,
Snowline (iceberg)	Some oaks	magnesium)
Vintage icberg	Cos (newer types)	High pH
Aquino (multi butter)	Multi-Red Salinova	Temperature of media
Descartes (multi butter)	Multileaf butterheads	Consecutive days of high temperature
Cavernet (red coral)		(above 30°C) product > 18 days old not
Raider (Crisphead)		affected
EU varieties		High temp/low humidity during
		germination
		Confidor <sup>®</sup> application

Table 2 - Control strategies suggested by seedling producers as possible ways to minimise blindness in lettuce.

Broad control area	Suggested techniques
1. Temperature	Reduce temperature during germination/seedling phase.
management	Germinate seeds in chambers of 15°C/98% humidity for 48 hours.
	<ul> <li>Seed and lay out trays late in the day/at night to avoid hot weather.</li> </ul>
	<ul> <li>Keep temperature as low as possible during seedling phase, i.e. by misting/fogging on hot days.</li> </ul>
	Store seed at 4°C before use.
	Place trays initially under whitewashed glass.
Nutrient management and pH	Higher boron levels (during warmer weather, reduce leaching of boron).
	Target pH 5.8-5.9 in media and water.
	Target 0.5-1.0g boron/m <sup>3</sup> of media.
	<ul> <li>Liquid feed with nutrient solution containing 2-4g borax (0.22- 0.44ppm).</li> </ul>
	<ul> <li>Manipulate EC levels and timing (&lt;1.8 in winter and &lt;1.1 in summer).</li> </ul>
	Adjust EC with temperature and age of plant.
	Increased magnesium & iron levels during first 7 days growing.
	<ul> <li>Higher magnesium than nitrogen early growth as help stabilise plants and stop elongating of leaves.</li> </ul>

Irrigation and water management	<ul> <li>Irrigate without nutrients in warm weather, apply nutrients at night with low EC (base EC level on ambient conditions).</li> </ul>	
	Reduce light intensity and transpiration.	
	Maintain adequate moisture levels during hot periods.	
	<ul> <li>Don't irrigate with large droplets or under pressure, keep water gentle</li> </ul>	
Pesticides	Minimise fungicides and insecticides eg Confidor®	

#### **Discussion**

Many producers suggested that high temperatures and humidity were key factors inducing lettuce blindness. During hot periods, some growers reported that up to 30% of seedlings could fail to develop normally and produce proper heads. This represents a major loss of income for seedling producers and growers alike.

Many producers also regarded high EC levels as contributing to blindness. This was thought to be mainly associated with use of strong nutrient solutions. Some growers do not fertilise for the first 1-2 weeks of growth due to concerns about blindness. Others use a fertigation solution, but then immediately irrigate to ensure the solution is thoroughly washed from seedling leaves.

The sporadic nature of this disorder makes it difficult to test treatments that potentially reduce incidence. An alternative strategy is to test ways of inducing blindness. If it can be demonstrated that growing lettuce seedlings with high temperatures and humidity, combined with strong fertigation solutions and/or unbalanced nutrient mixes, increases blindness then mitigation methods can be developed.

## Monitoring blindness in nurseries

## **Background and Aim**

Discussions with stakeholders clearly indicated that lettuce blindness is a sporadic issue that can fluctuate from zero to a high percentage of affected plants from week to week, particularly during summer months.

Recording the incidence of blindness and matching this data to weather and production records (particularly temperature) could provide some indications as to causes of this disorder.

#### Method

Trays of lettuce seedlings at Withcott Seedlings in Queensland (cv. Leu Quintus) and Choice Seedlings in NSW (cv. Kireve) (Figure 2) were monitored weekly from September 2013 to January 2014. All plants were examined 3–4 weeks post seeding, by which time plants have normally developed 4–6 true leaves. At each examination date 10 trays of the same variety and planting date were checked. The number of blind seedlings per tray was recorded to determine percentage blindness.



Figure 2 - Locations of Withcott and Choice Seedling nurseries

Temperature and relative humidity were monitored in the seedling production areas in both nurseries over the assessment period.

Withcott nursery in Queensland conducted two small additional trials;

3. The first compared blindness in lettuce cv. 'Arganda' germinated at either 15°C or 20°C for two days (10 trays per treatment), before all trays were moved to the main nursery area. The number of blind seedlings was counted 11 days after seeding.

- 4. A second trial compared blindness in lettuce cv. 'Arganda' when seedlings were given weekly applications of foliar nutrients (5 trays per treatment). Fertigation mixes applied contained;
  - a. standard nutrients (control)
  - b. calcium
  - c. boron
  - d. boron + calcium
  - e. boron + calcium + molybdenum

### **Results**

While there was a general trend to warmer weather being associated with increased blindness at both locations, incidence was too variable to produce strong conclusions.

#### Correlation between temperature and blindness at Choice Seedlings

Average daily maximum temperatures at Choice Seedlings were generally 25-35°C over the experimental period. Although hot spells lasting several days were observed, there was no clear pattern of an increase in blindness following such events. The highest rate of blindness observed (>10%) was recorded from plants seeded on 12<sup>th</sup> November, a period when temperatures were relatively mild. A high rate (8.6% blind) was also recorded from lettuce seeded on 29<sup>th</sup> January. For several days following, maximum temperatures exceeded 35°C. However, similar or even hotter conditions on 20<sup>th</sup> December and 15<sup>th</sup> January did not result in increased rates of the disorder.

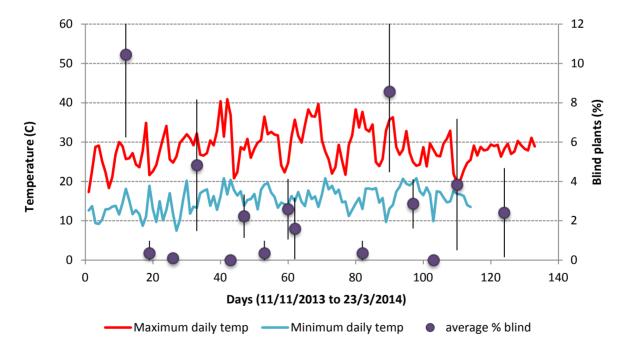


Figure 3 - Daily maximum and minimum temperatures at Choice Seedlings NSW and percentage blind seedlings (cv. Kireve). Blindness was assessed after minimum 3 weeks, so points are located at seeding date + 10 days. Bars indicate the standard deviation of each mean value (n=10)

#### Correlation between temperature and blindness at Withcott Seedlings

Despite their geographic difference, temperatures at Withcott Seedlings were fairly similar to those recorded at Choice, with daily maximums generally ranging between  $25 - 40^{\circ}$ C and minimums around  $15 - 20^{\circ}$ C.

Recorded rates of blindness were lower at this site. This may be due to differences in what the assessors considered to be 'blind', as well as environmental factors such as water quality, plant shading, irrigation method and fertigation procedures. Also, different lettuce varieties were assessed at each site, so it is not possible to compare results.

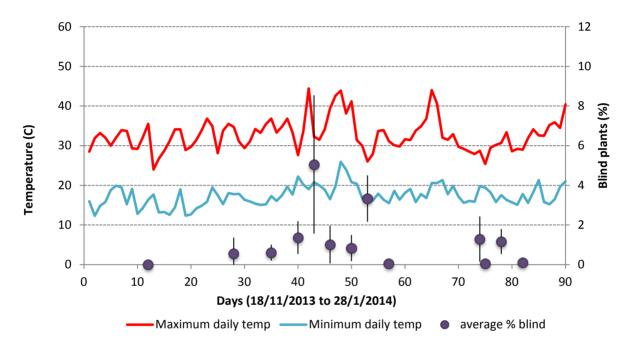


Figure 4 - Daily maximum and minimum temperatures at Withcott Seedlings QUEENSLAND and percentage blind seedlings (cv. Leu Quintus). Blindness was assessed after minimum 3 weeks, so points are located at seeding date + 10 days. Bars indicate the standard deviation of each mean value (n=10)

The highest rates of blindness were observed for lettuces seeded on 20/12 (5.1%) and 30/12 (3.3%). While the former seeding date was followed by several days over 35°C, for the latter date maximum temperatures were 30-33°C. Much higher maximum temperatures on  $2^{nd}$  to  $6^{th}$  January did not result in high rates of blindness.

Results from Withcott are therefore consistent with those from Choice. The data does not support a specific link between a high temperature event at seeding or during early growth, and subsequent development of blindness in lettuce seedlings.

#### Effect of germination temperature on blindness (Withcott Seedlings)

Blind lettuces were generally not observed until 23 days after seeding; recorded numbers of blind seedlings were far lower when the plants were assessed 11 and 17 days after seeding. This does not mean that the symptoms were not present, but that they were less clearly visible. This result indicates that the assessment time may be a critical factor in determining percentages of blind seedlings. Previous trials indicated that some seedlings assessed as 'blind' recovered and developed normally.

In this trial, fewer blind seedlings were observed when trays were stored at 15°C for 2 days post seeding than when trays were held at 20°C. This result suggests that germination temperature may be a critical factor in this disorder. However, the test needs to be repeated to confirm such a strong effect.

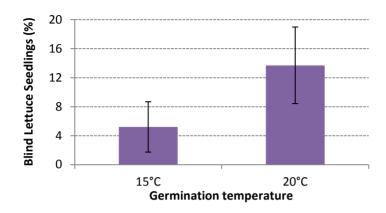


Figure 5 - Effect of placing seeded lettuce trays at 15 or 20°C for two days before moving to outdoor racks on the percentage of blind seedlings cv. Arganda (assessed 23 days after seeding). Bars indicate the standard deviation of each mean value (n=10).

#### Effect of foliar nutrients on blindness (Withcott Seedlings)

Foliar applications of calcium and boron both reduced rates of blindness in lettuce seedlings cv. Arganda compared to the normally fertigated control plants. However, when both boron and calcium were applied the percentage of blind plants was increased to similar levels as observed among control plants.

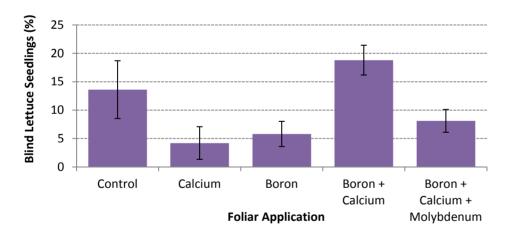


Figure 6 - Effect of foliar nutrients on the percentage of blind seedlings cv. Arganda (assessed 23 days after seeding). Bars indicate the standard deviation of each mean value (n=5).

The observed reduction in blindness when calcium and boron were applied is consistent with previous reports that calcium and boron may have critical roles in development of this disorder. However, the increase when both were applied together was unexpected. The results of trials such as these are highly variable, and need to be repeated for confidence in the results.

#### **Discussion**

There appears to be a general effect of high summer temperatures on increased rates of blindness. However, this study did not find evidence for specific effects of a high temperature event during germination or early growth on increased rates of this disorder. It seems more likely that there is an interaction between high temperatures and other environmental or genetic factors. This could include some of the issues identified by growers, such as high EC in irrigation water, retention of fertigation solution on leaves, high light levels or application of insecticides and fungicides.

The results from the small trials at Withcott certainly provide other avenues for investigation. Low temperature germination and application of calcium or boron appear promising techniques, and should be tested further. The conflicting results when multiple products are applied together may be due to increased EC in the fertigation solution.

The occurrence of blindness in lettuce is highly variable. It seems probable that this disorder relates to combinations of factors; it is unlikely to have a single causal factor, rather various issues that increase stress on plants and cause blindness in susceptible individuals.

## 2. Bosch glasshouse trials

## Effect of seedling mix

## **Background and Aim**

The previous farm surveys and interviews with growers indicated that blindness occurs more in peak summer than during cooler months, and that some varieties appear to be particularly susceptible to the disorder. There was also a suspicion among some growers that the seedling mix itself may play a role. A trial was therefore conducted to test the effect of three different seedling mixes on rates of blindness of two potentially susceptible lettuce varieties – 'Blanes' (cos type) and 'Kireve' (oak leaf type) – grown under high temperatures and humidity.

#### Method

A glasshouse trial was designed to induce blindness by growing lettuce seedlings at 35-40°C. The trial was a randomised complete block design (RCBD) with two lettuce varieties 'Blanes' (Cos type) and 'Kireve' (Oak leaf type). The lettuce were mechanically direct seeded into standard 128 cell seedling trays filled with three different commercial seedling mixes with ingredients as shown in Table 3.

Table 3 - Ingredients in	three different	commercial seedling m	ıixes
--------------------------	-----------------	-----------------------	-------

	MIX 1	MIX 2	MIX 3
Vermiculite	200L	200L	200L
Peat moss	500g (sphagnum)	225L (Netherlands)	225L (Latvian)
Coir peat	140L		
Fine superphosphate	1,000g		1,000g
Blood and bone	1,000g	1,000g	1,000g
Lime	1,000g		
Dolomite		750g	
Banana special		500g	

Samples of each mix were sent to Phosyn Analytical Pty Ltd for analysis of nutrients, pH, EC, air filled porosity, water holding capacity and wettability.

Each tray was then cut in half; one half of each tray received fertigation while the other was only watered. Fertigation treatment consisted of Aquasol with a nutrient content of N23:P4:K18, applied at the recommended rate of 16g/10L of water. Fertigation was applied at 15, 17 and 20 days after sowing, with approximately 1L solution added to each tray.

For the duration of the experiment the daytime temperature was set at 35°C (16 hours) and the night time temperature was set at 21°C (8 hours). Seedlings were irrigated using 180 degree risers for 15 minutes at 10:00am, 12:00 noon, 2:00pm and 4:00pm.

The following assessments were conducted 21 days after sowing:

Plant establishment percentages from 64 cells per tray

- Number of fully expanded leaves and leaf length (cm) of the largest leaf
- Yield in dry weight of 10 whole plants
- Number of seedlings where the apical point was missing or distorted i.e. the plant was 'blind' (Figure 7)





Figure 7 - Normal lettuce seedling (left) compared to blind seedling (right) with distortion of leaves at the apical point

### **Results**

## **Temperature management**

The greenhouse system successfully controlled temperature, with daytime temperatures approaching 32 to  $40^{\circ}$ C and night times around  $22^{\circ}$ C (Figure 8). Relative humidity was maintained at close to 100% throughout the trial. These conditions replicated, or were more extreme than, the hot summer conditions previously associated with blindness development in lettuces.

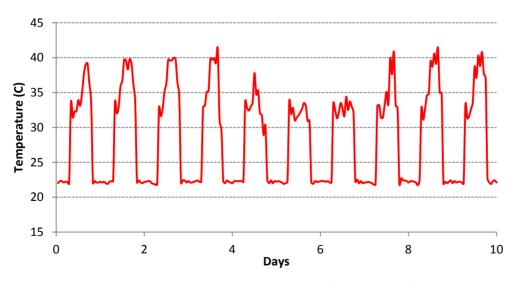


Figure 8 - Greenhouse temperature management during the first 10 days of the trial

#### **Differences between mixes**

EC levels differed significantly between the mixes (P= 0.05). Seedling mix 1 and 3 measured a significantly higher EC compared with mix 2.

The mixes were also significantly different (P <0.001) in pH. Seedling mix 1 had the highest pH compared to mix 2 and 3.

Lettuce variety did not affect EC or pH.

Analysis of the seedling mixes showed that mix 2 had very high levels of exchangeable phosphorus (P) (179.9 mg/L) compared to the other two seedling mixes, in combination with low exchangeable potassium (K) (63 mg/L) (Table 4).

Table 4 - Analysis of nutritional properties and physical characteristics of three different seedling mixes.

Analysis		Mix 1	Mix 2	Mix 3
Air-filled porosity	%	17	12	10
Total water holding capacity	%	58	59	75
Wettability	min	<1	3	<1
рН		7.2	5.1	5.2
EC	dS/m	1.7	1.2	1.9
NH4-N	mg/L	73	52	40
NO3-N	mg/L	2	2	122
Cl	mg/L	151	82	34
Mn (1)	mg/L	<1	1	<1
Ca	mg/L	241	230	408
Mg	mg/L	44	191	40
Р	mg/L	33.7	179.9	93.9
K	mg/L	244	63	118
S	mg/L	310	294	254
Fe	mg/L	2	10	11
Mn	mg/L	<1	9	3
Zn	mg/L	1	1	1
Cu	mg/L	<0.1	2.2	0.7
В	mg/L	<0.01	0.03	0.1
Na	mg/L	76	48	54
K (2)	mg/L	350	120	173

All nutrient values are from DTPA extraction unless otherwise indicated

(1) Water extractable (2) NH4OAc extractable

#### Plant development

Growth of lettuce seedlings was uneven throughout the replicate trays for both lettuce varieties, fertigation treatments and seedling mixes (Figure 9). This is thought to be a result of nutritional stress due to fertigation only occurring weekly and uneven watering from the overhead watering system.

The percentage of seedling establishment was also affected by uneven watering, in which the 'Blanes' variety had 80% to 88% establishment while the 'Kireve' variety had 59% to 80% establishment.

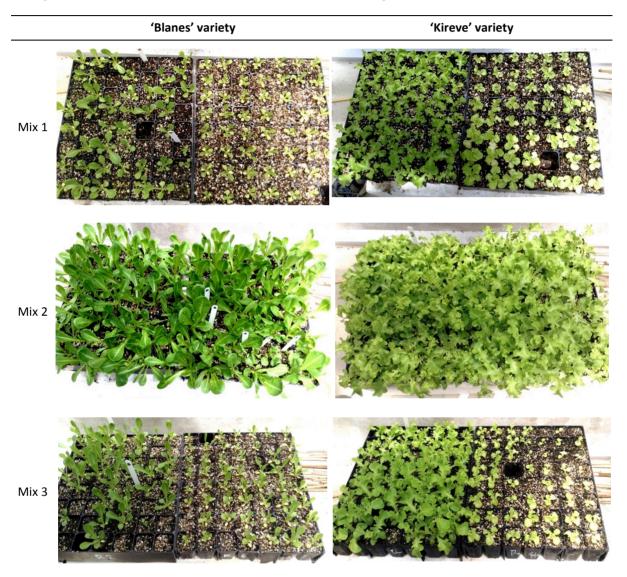


Figure 9 - Lettuce grown in different mixes showing seedling development 21 days after sowing. Plants in the left tray were fertigated three times during the trial, those in the right tray received water only.

The number of fully expanded leaves per plant varied significantly between lettuce varieties and seedling mixes (<0.001). The 'Kireve' variety had 5 fully expanded leaves when grown in mix 2 while the 'Blanes' variety had 5 fully expanded leaves when grown in mix 1. No significant differences were found between fertigation treatments.

The lengths of fully expanded leaves were significantly different between lettuce varieties and seedling

mixes (P<0.001). The 'Blanes' variety measured the longest leaves when grown in mix 2 with an average leaf length of 5cm. The 'Kireve' variety measured the longest leaves when grown in mix 1 with an average leaf length of 5cm.

As shown in Figure 10, the seedlings grown in mix 2 were larger and more vigorous than those grown in the other mixes. This is likely due to the inclusion of more fertiliser in this mix. Fertigation provided during the trial did not fully compensate for this difference between the mixes.

An interaction was found between seedling mixes as well as variety for yield dry weights (P=0.004). Both varieties grown in mix 2 had significantly higher dry weights compared to all other mixes (P<0.001).

Table 5 - Plant dry weights in g/seedling

	'Blanes'		'Kireve'	
Seedling mix	No fertigation	Fertigation	No fertigation	Fertigation
1	0.03	0.05	0.14	0.19
2	0.22	0.22	0.22	0.28
3	0.04	0.1	0.02	0.11

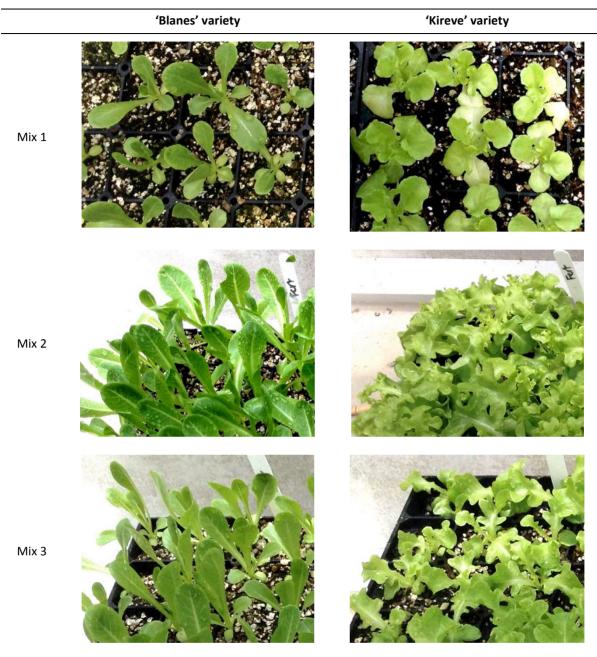


Figure 10 - Lettuce grown in different seedling mixes showing variability in development 21 days after sowing.

## **Incidence of blindness**

The incidence of blindness was significantly higher in the 'Blanes' variety compared to 'Kireve' (P=0.006). The highest percentage of blind seedlings was observed in the 'Blanes' variety grown in mix 2 (P=0.006) without fertigation. The 'Kireve' variety also expressed the highest percentage of blindness in mix 2, however in this case in the fertigated mix.

Table 6 - Percentage of blind 'Kireve' and 'Blanes' lettuce grown in three different seedling mixes with and without fertigation

	'Blar	nes'	'Kire	eve'
Seedling mixes	No fertigation	Fertigation	No fertigation	Fertigation
1	0.8 %	1.8 %	0.0 %	0.0 %
2	9.6 %	1.4 %	0.0 %	1.8 %
3	0.4 %	0.5 %	0.5 %	1.3 %

## **Discussion**

Blindness was induced in 'Blanes' and 'Kireve' lettuce grown in almost all seedling mixes regardless of fertigation (only 'Kireve' grown in seedling mix 1 did not result in any blind plants). Based on this result, lettuce blindness is not likely to be caused by one of these three seedling mixes in particular. Furthermore, in most cases blindness was induced regardless of whether fertigation was used.

The higher rate of blindness (9.6%) found in 'Blanes' lettuce grown in seedling mix 2 without fertigation is unlikely to be related to that particular seedling mix or lack of fertigation, as other levels of blindness in seedlings grown in seedling mix 2 or without fertigation were significantly lower. Therefore the combination of 'Blanes' lettuce grown in seedling mix 2 needs repeating in further work before any conclusions can be drawn about the role of seedling mix, variety or fertigation in lettuce blindness.

Fertigation and irrigation treatments failed to replicate commercial practice and resulted in poor seedling establishment. It is possible that lettuce blindness was induced in this trial due to a stress from not enough irrigation and fertigation or from far too much irrigation.

# Effect of nutrient mix and concentration

# **Background and Aim**

The symptoms of blindness vary, which may be one reason so many explanations are offered for the cause of these symptoms.

The field identification guide to Pests, Diseases and Disorders of lettuce produced by NSW Agriculture (2003) includes a picture of a blind lettuce with thickened inner leaves with boron deficiency. It notes that this can be confused with calcium deficiency – which is often associated with high temperatures and saturated humidity.

Several growers had suggested that high EC was a key cause of increased occurrence of blindness. This was defined as as being a solution with conductivity significantly higher than 1.2 dS/m. This limits the strength of the fertiliser applied, and means that some seedling producers irrigate after fertigation to wash high EC solution off the leaves.

In this trial fertigation was compared to normal commercial practice through fertigation using solutions high or low in calcium, with added boron or with double strength nutrients to increase EC.

### Method

Commercial seedling nurseries commonly fertigate seedlings three times weekly using 'Nursery Blend', with the following analysis:

Nitrogen	12.3% w/v
N as nitrate	7.2%
N as ammonium	5.1%
Phosphorus (P)	3.1%
Potassium (K)	5.2%
Calcium (Ca)	2.0%
Magnesium (Mg)	0.3%
Manganese (Mn)	0.005%
Iron (Fe)	0.01%
Specific gravity @ 20C	1.29 kg/L

A fertigation mix was fabricated using inorganic materials. The objective was to produce a basic mix with a similar nutrient profile to 'Nursery Blend', but to which calcium and boron could be added or subtracted.

To avoid calcium reacting and coming out of solution, the fertigation mix was produced in two parts. The concentration of the standard solution was adjusted to give EC = 1.2 dS/m, equivalent to 768 ppm. The other solutions were similar  $\pm 0.2$  dS/m, with the exception of the double strength solution which was approximately 2.7 dS/m, well above levels believed to cause blindness.

Table 7 - Fertigation solutions produced from inorganic base materials

Solutions	Ingredient	Rate (per litre)		
Standard	Calcium nitrate	34.66	(g)	Solution A
	Potassium nitrate	13.54	(g)	Solution B
	MAP	43.22	(g)	Solution B
	Manganese sulphate	0.022	(g)	Solution B
	Iron sulphate	0.050	(g)	Solution B
High calcium	Calcium nitrate	79.35	(g)	Solution A
	KH2PO4	17.93	(g)	Solution B
	Manganese sulphate	0.022	(g)	Solution B
	Iron sulphate	0.050	(g)	Solution B
High Boron	Calcium nitrate	34.66	(g)	Solution A
	Potassium nitrate	13.54	(g)	Solution B
	MAP	43.22	(g)	Solution B
	Boric acid	0.27	(g)	Solution B
	Manganese sulphate	0.022	(g)	Solution B
	Iron sulphate	0.050	(g)	Solution B
Low calcium	Urea	11.68	(g)	Solution A
	Potassium nitrate	13.54	(g)	Solution B
	MAP	43.22	(g)	Solution B
	Manganese sulphate	0.022	(g)	Solution B
	Iron sulphate	0.050	(g)	Solution B
Double				
strength	Calcium nitrate	69.31	(g)	Solution A
	Potassium nitrate	27.08	(g)	Solution B
	MAP	86.44	(g)	Solution B
	Manganese sulphate	0.044	(g)	Solution B
	Iron sulphate	0.100	(g)	Solution B

Twenty-five seedling trays were filled using a commercial seedling mix (mix 2) and seeded with lettuce cv. 'Blanes'. Trays were placed on racks in the glasshouse with temperature set to 33°C daytime temperature and 21°C night temperature (12h/12h). Five replicate trays were allocated to each of the five fertigation treatments;

- 6. Standard Normal strength fertigation
- 7. High calcium Normal fertigation with double strength calcium
- 8. Low calcium Normal strength fertigation with sources of calcium removed
- 9. High boron Normal strength fertigation with double strength boron
- 10. Double strength fertigation

Trays were randomly distributed around the glasshouse. The fertigation mixes were watered onto trays individually four times weekly from 12 days after seeding until maturity. Approximately 1L/tray was watered over each tray with a watering can. With the exception of the double strength fertigation

solution, the trays were then briefly rinsed with water only to remove salts from the leaves (as per commercial practice) before being replaced in random positions on the benches.

The greenhouse was programmed to provide a 12 hour day / night cycle. Temperatures were set to average 33°C during the day, 21°C at night. Irrigation was scheduled for two cycles per day, each lasting just under 20 minutes. The irrigation system itself was also adjusted from the previous trial so as to provide more even coverage with shortened and more frequent risers, larger droplet spray heads and reduced pressure.

The trays were seeded on 17<sup>th</sup> March and were ready to harvest on 14<sup>th</sup> April. At harvest each tray was examined to determine establishment rate and the number of blind seedlings were counted. A sub sample of 64 lettuces per tray were cut just above ground level and compiled to calculate yield and percentage dry weight from each treatment.

### Results

All of the lettuce seedlings grew well during the trial, with 96-100% establishment in all treatments. There were clear differences in growth rates, particularly noticeable between the standard and the double strength fertigation treatment (Figure 11). No negative effects of the strong fertigation solution were observed. There were also no symptoms of tipburn, even in the low calcium treatment, or other nutritional deficiencies.

The lettuces grown with increased nutrients were also slightly darker green than those grown with standard solution (Figure 12).

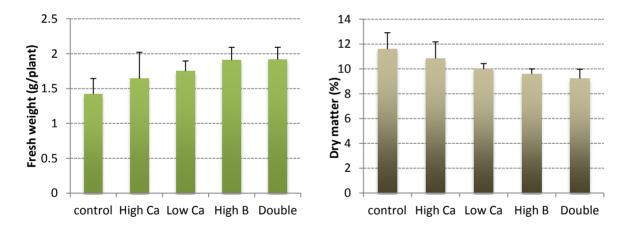


Figure 11 - Fresh weight and dry matter content of lettuces grown with different nutrient solutions. Bars indicate the standard deviation of each mean value (n=5)



Figure 12 - Lettuce seedlings grown with standard nutrient mix (control) compared to double calcium, no calcium, added boron, or standard nutrients at double concentration.

Of the total 3,200 seedlings grown during the trial, only one blind seedling was found.

It was concluded that, under the trial conditions ( $30-34^{\circ}$ C daytime temperature,  $21-22^{\circ}$ C night temperature, 70-100% RH), neither calcium or boron availability or high nutrient concentration induced blindness.

# Effect of high nutrient mix and extended day-length

# **Background and Aim**

The previous trial failed to induce blindness, despite combining several of the factors expected to induce this disorder. A second trial was therefore developed to expose the plants to similar, but more extreme nutrient concentrations, in combination with extended day-length and high temperature.

#### Method

This trial was similar to the previous study and also used 'Blanes' variety lettuce seedlings. However the factors that had been expected to induce blindness were increased to more extreme levels than previously:

- Seedling mix 2 (from trial 1.1) was used, this having produced a significant number of blind plants in the first trial
- Trays were hand seeded rather than mechanically seeded
- The temperature in the greenhouse was increased to an average of 37°C during the day (35 to 40°C range) and 27°C at night
- Day length was increased to 16 hours, with 8 hours night
- Irrigation was reduced to only 8 minutes applied twice daily
- Supplementary lighting was used to extend day length (especially important as this trial was conducted during May). Halogen lights located over the benches were programmed to be on for 14 hours each day
- Plants were not rinsed after treatment, allowing fertigation mixes to remain on the leaves

Four different treatments were applied, using the same fertigation mix formulas as in trial 1.2:

- 1. Control no fertigation applied, only water
- 2. Double strength standard fertigation mix applied at 2x the normal concentration
- 3. Quadruple strength standard fertigation mix applied at 4x the normal concentration
- 4. Low calcium low calcium mix as in trial 1.2



Figure 13 - Trial set up showing germination of lettuces one week after seeding (left) and lights over mature seedlings

## **Results**

The plants grew rapidly and were at harvestable size less than three weeks after seeding. However they were allowed to grow for another week to ensure that symptoms were clear, giving a total growing time of 26 days.

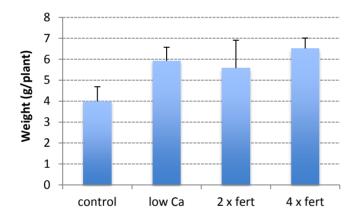


Figure 14 - Fresh weight of lettuces grown with no fertigation (control) or with various concentrations of solution, 26 days after planting. Bars indicate the standard deviation of each mean value (n=3)

All plants appeared very healthy at harvest, including the controls, which had not received supplementary fertigation (Figure 15). Seedling establishment was again high, with an average 97% of cells containing a healthy plant.



Figure 15 - Lettuces grown with no fertigation (top left), double fertigation (top right), quadruple fertigation (bottom left), or fertigation but no calcium (bottom right)

No blind plants were found in any of the treatments from a total of 1,497 seedling examined.

### **Discussion**

The first three trials were developed based on observations that blindness / multiple heading are more often problems during summer and were associated with high fertiliser use. It was unclear whether this was because of high temperatures, high humidity, long day length, fast growth rate or a combination of all of these factors.

Based on the results from these trials, high temperatures and/or long days may contribute to development of lettuce blindness, but cannot induce blindness in the absence of other – unknown - factors. Applying high concentrations of fertiliser also failed to induce this condition. Lettuces grew rapidly in trial 1.3, but again without blindness developing, suggesting growth rate itself is not the issue. Blindness also seems unlikely to be due to a nutritional imbalance, although clearly not all nutrients were tested in these limited studies.

These trials were conducted in a sterile, fully controlled greenhouse. Another theory regarding causes of blindness is that affected seedlings were damaged or 'touched' by an insect or disease during early growth. No causal organism has ever been identified, but it is possible it might have occurred well before symptoms were observed.

A further trial could test this hypothesis by damaging the plant apical shoot during early growth.

# Effect of salinity, physical or chemical damage

# **Background and Aim**

Following a lack of consistent results in inducing lettuce blindness in previous glasshouse trials, a further range of treatments were proposed as possible ways to induce the disorder. Lettuce blindness is more common in the summer. Apart from high temperatures and humidity, other factors more common at this time include:

- · High salinity irrigation water
- Chlorinated irrigation water
- Insecticide spray damage
- Physical damage to the apical shoot from insects
- More frequent watering, and damage to the seedlings when using overhead sprinklers

Treatments were developed that simulated these factors, along with high concentrations of fertiliser, small growing cells, and a wetting agent which growers have had issues with.

### Method

Lettuce cv. Kireve were hand seeded in Choice seedling mix on 29 June 2015. Seedlings were grown in the Bosch Glasshouse at Sydney University under summer simulated growing conditions (Table 8).

**Table 8. Glasshouse growing conditions** 

Growing environment factor	Set or measured average		
Day length*(hours)	12		
Night length (hours)	12		
Day temperature (°C)	32		
Night temperature (°C)	27		
Irrigation	2 cycles of 14 minutes per day		
Humidity (% RH)	60		

<sup>\*</sup>Lights were used for the duration of the day length, although lights on one side of the room did not stay on consistently

Thirteen treatments were applied, with two commercial 112 cell seedling trays of each treatment (except treatment 6). Trays from treatments 1-8 were randomly placed in the irrigated section of the glasshouse, with treatments 9-13 randomly placed in the non-irrigated area. Treatment applications commenced at the fully-expanded cotyledon stage (4 days after sowing) and consisted of:

## **Automatically irrigated treatments:**

- 1) Control
- 2) *Mechanical brushing* –plants gently brushed daily, initially with a paint brush, then with a dust-pan brush
- 3) Chlorpyrifos sprayed weekly to leaf wetness, at a concentration of 1 mL/L

- 4) High fertiliser –1L liquid fertiliser (Diamond Blue\*) at EC of 8000 μs.cm<sup>-1</sup> (7g/L), twice per week
- 5) Wetting agent -sprayed Designer (by Nufarm) twice weekly at 6 mL/L to leaf wetness
- 6) Small growing cells plus high fertiliser 198 cell tray, with high fertiliser treatment
- 7) Sodium molybdate Sprayed twice weekly at 1.2 g/L to leaf wetness
- 8) High concentration boron applied with regular fertiliser, twice weekly at 0.9 g/L Borax

#### **Hand-watered treatments:**

- 9) Overhead irrigation watered daily overhead with hand sprinkler for 20 seconds per tray
- 10) Free chlorine watered each tray daily with 1.5 L of chlorinated water, at 200 ppm chlorine
- 11) NaCl 4000 µs.cm<sup>-1</sup> watered each tray daily with 1.5 L of saline (NaCl) water, at an EC of 4000 µs.cm<sup>-1</sup>
- 12) NaCl 2000 μs.cm<sup>-1</sup> + free chlorine watered each tray daily with 1.5 L of saline (NaCl 2000 μs.cm<sup>-1</sup>) water combined with 200 ppm free chlorine (made up from pool chlorine)
- 13) NaCl 2000  $\mu$ s.cm<sup>-1</sup> watered each tray daily with 1.5 L of saline (NaCl) water, at an EC of 2000  $\mu$ s.cm<sup>-1</sup>

\*Diamond Blue (Campbells Fertilisers Australasia) analysis: 19% N, 2.1% P, 17% K, 1.7% S, 1.5% Mg, 0.05% Fe, 0.025% Mn, 0.01% B, 0.008% Zn, 0.006% Cu, 0.004% Mo.

All treatments without high fertiliser applications were fertilised twice weekly with 1L liquid fertiliser (Diamond Blue – EC 4000  $\mu$ s.cm<sup>-1</sup>) per tray. Treatments were applied over a period of 15 days, following which plants were left to grow for a period of 7 days, during which all trays received automatic overhead sprinkler irrigation and two standard fertigation applications.

Seedlings were harvested 27 days after sowing, when all seedlings were assessed for blindness, stunted apical shoots, commercial marketability and average leaf number. Additionally, fresh and dry weights were measured from 20 plants per tray.

Blindness was defined as seedlings lacking an apical shoot, or with a deformed apical shoot, while a stunted apical shoot was defined as a clearly short apical shoot when compared to control plants (Figure 16).

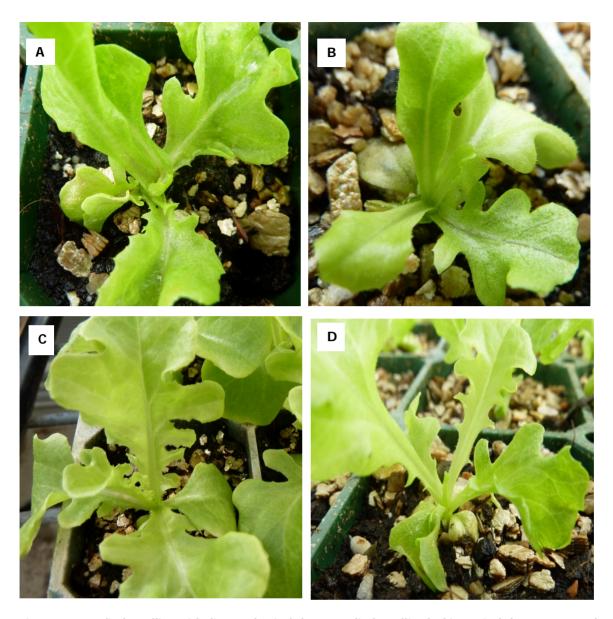


Figure 16 – A. Blind seedling with distorted apical shoot B. Blind seedling lacking apical shoot C. Normal seedling with apical shoot D. Stunted apical shoot.

## **Results**

Final seedling weights were highly variable between treatments (Figure 17), with stunted seedlings observed in the chlorpyrifos, overhead irrigation, chlorine, NaCl 4000  $\mu s.cm^{-1}$  and NaCl 2000  $\mu s.cm^{-1}$  plus chlorine treatments. Dry matter tended to be lower in treatments with high electrical conductivity such as high fertiliser, NaCl 4000  $\mu s.cm^{-1}$  and NaCl 2000  $\mu s.cm^{-1}$  (Figure 18).

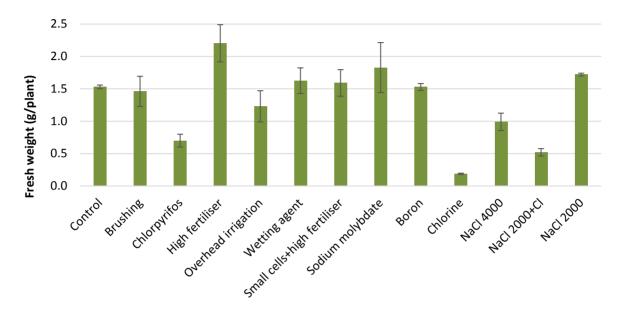


Figure 17. Fresh weight of lettuces grown with different physical or chemical treatments. Bars indicate the standard deviation of each mean value (n=2).

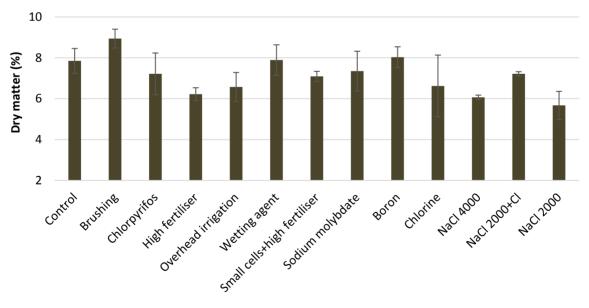


Figure 18. Dry matter content of lettuces grown with different physical or chemical treatments. Bars indicate the standard deviation of each mean value (n=2).

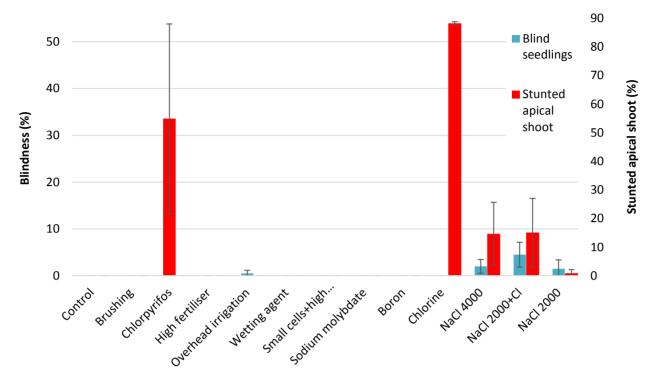


Figure 19. Percentage of blind lettuce seedlings and stunted apical shoots from lettuces grown with different physical or chemical treatments, when assessed 27 days after sowing. Bars indicate the standard deviation of each mean value (n=2).

Blind seedlings were found in five of the treatments (Figure 19), and were first visible at 20 days after sowing, apart from the chlorpyrifos treatment where spray damage to the apical shoot was evident 7 days after the first treatment. Severely stunted apical shoots were evident in four of the treatments, in which only a very short apical shoot was present. Stunted growth from these treatments was visible within 5 days of initial treatments.

Root development was reduced and the tap root was burnt in the chlorine treatment (Figure 20 a,b) and overall root mass was greatly reduced in the NaCl 4000 µs.cm<sup>-1</sup> treatment (Figure 20 c).

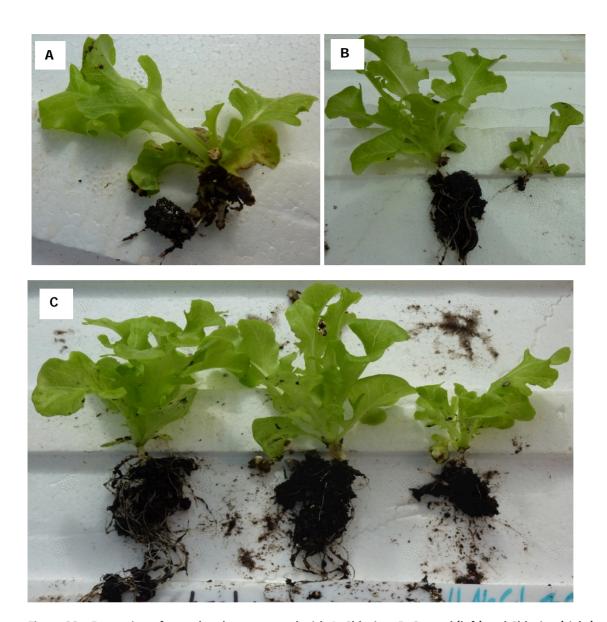


Figure 20 – Formation of roots by plants watered with A. Chlorine; B. Control (left) and Chlorine (right); and C. Control, NaCl 2000 μs.cm<sup>-1</sup> and NaCl 4000 μs.cm<sup>-1</sup> (from left to right).

Some treatments also had an effect on leaf form, with leaf distortion and curling on seedlings treated with the overhead sprinkler, chemical burn and distortion on chlorpyrifos treated seedlings and leaf curling on seedlings treated with chlorine (Figure 21).

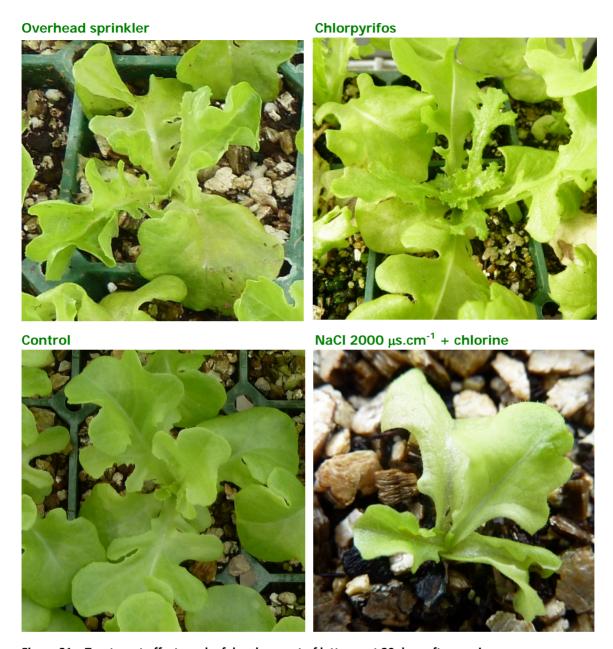


Figure 21 – Treatment effects on leaf development of lettuces at 20 days after sowing.

### **Discussion**

This trial has demonstrated that lettuce blindness can be induced at temperatures below 35°C when plants are stressed in some way.

Lettuce blindness was present to some extent in all treatments irrigated with saline water, suggesting a role for NaCl in lettuce blindness. Growing lettuces under saline conditions could indirectly influence the uptake of a particular nutrient. For example, one study found significantly lower calcium concentrations in young leaves of lettuces grown in saline conditions<sup>1</sup>. Alternatively, the growth of the apical shoot may be inhibited directly as a result of increased Na and Cl concentrations in the growing shoot. Water is more likely to be saline in summer, when water storages are low – which is consistent with observations that most blindness occurs at this time of year.

Application of chlorpyrifos to lettuce seedlings reduced seedling growth and burned the apical tip, resulting in blindness. While it is not common commercial practice to apply chlorpyrifos (T/N Lorsban) to lettuce seedlings, it may be worth testing whether other insecticides can also chemically 'burn' delicate seedling leaves, especially under hot conditions.

Daily brushing and overhead irrigation with a high pressure hand sprinkler failed to induce blindness in this trial. This suggests that physical damage to the growing tip is less likely to be a cause of blindness.

This trial has also verified work from the previous trials in which high concentrations or additional nutrients failed to induce significant levels of blindness.

Further work is required to replicate results in which lettuce blindness was found in this trial. Seedlings in this trial were treated continuously for 15 days. A trial in which treatments are applied at a specific growth stage could determine when lettuce seedlings are most susceptible to damage resulting in blindness or multiple heading disorders.

Seedling nurseries may add chlorine to irrigation water to kill pathogens and eliminate algae. They may also, at times, use irrigation water that has become saline due to factors such as drought, runoff or intrusion into water tables. Correlating these factors with the incidence of blindness would indicate whether these are causal factors at the levels likely to occur under commercial conditions.

<sup>&</sup>lt;sup>1</sup> Lazof, D., & Bernstein, N. (1999). Effects of salinization on nutrient transport to lettuce leaves: consideration of leaf developmental stage. *New phytologist*, *144*(1), 85-94.

# Effect of salinity, chlorine and calcium supplementation

## **Background and Aim**

In the previous trial (1.4), high salinity and chlorine both induced blindness in lettuce seedlings. It was hypothesised that this could be due to reduction of calcium uptake; sodium competes with calcium for plant uptake, while Na+ and Cl- may reduce the uptake of calcium due to reduced osmotic potential in the growing media and root. Such an effect would be consistent with the observation that lettuce blindness increases when temperatures and RH are high.

High temperature and humidity commonly result in the disorders 'blossom end rot' and 'tip-burn'. Both are symptoms of calcium deficiency in rapidly dividing plant tissue. Even if calcium is present in the soil, high humidity reduces transpiration, slowing transport of Ca+ into growing shoots. Lettuce blindness has also been suspected of association with low levels of calcium. Conversely, increased calcium in the soil solution can reduce the likelihood of deficiency and inhibit uptake of CI- or  $Na+^2$ .

This trial therefore aimed to repeat some of the treatments found to increase blindness, as well as add calcium to test whether this would reduce incidence of the disorder.

#### Method

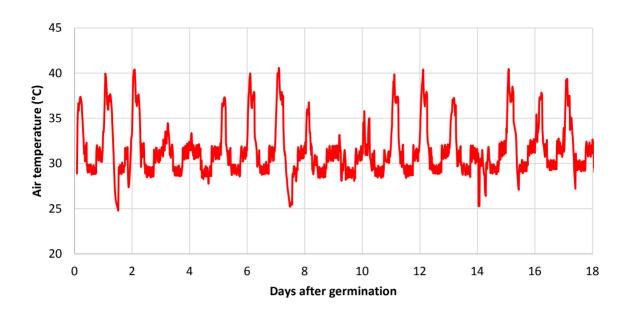
Lettuce cv. Kireve was hand sown into twenty-four 112 cell trays of commercial seedling mix. Trays were laid on racks at the Bosch Glasshouse at Sydney University, with the room programmed to provide high temperatures and humidity (Figure 22).

All seedlings were fertigated twice weekly with Diamond Blue (Campbells Fertilisers Australasia) analysis: 19% N, 2.1% P, 17% K, 1.7% S, 1.5% Mg, 0.05% Fe, 0.025% Mn, 0.01% B, 0.008% Zn, 0.006% Cu, 0.004% Mo.

Additional daily treatments commenced three days after seeding, at first emergence of the cotyledons. Three trays were randomly allocated to each of eight treatments:

1	Control	water only
2	Control + calcium	water daily and chelated calcium (1g/L) twice weekly
3	Chlorine	chlorinated water @ 100 ppm
4	Chlorine + calcium	chlorinated water (100 ppm) daily and chelated calcium (1g/L) twice weekly
5	Saline	Water containing 2000µs.cm <sup>-1</sup> NaCl
6	Saline + Calcium	Water containing 2000µs.cm <sup>-1</sup> NaCl daily and chelated calcium twice weekly
7	Saline + Chlorine	Water containing 2000µs.cm $^{\text{-}1}$ NaCl daily combined with chlorinated water @ 100 ppm Cl
8	Saline + Ca + Cl	Water containing 2000µs.cm <sup>-1</sup> NaCl daily combined with chlorinated water @ 100 ppm Cl. Chelated calcium twice weekly

<sup>&</sup>lt;sup>2</sup> Marschner, H. (1997) Mineral Nutrition of Higher Plants: Second Edition. Academic press



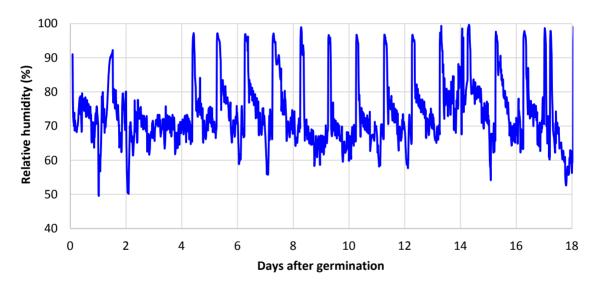


Figure 22. Glasshouse temperature and humidity during the trial.

Treatments were applied for 18 days, after which all seedlings were assessed for blindness. Blindness was defined as seedlings lacking an apical shoot, or with a deformed apical shoot, as previously described in trial 1.4. Shoot fresh weight was recorded for 20 seedlings per tray.

## **Results**

Shoot weights were reduced in all treatments except from salt (NaCl) (Figure 23), with seedling growth stunted where calcium or chlorine was applied (Figure 24). Calcium was initially applied at too high a rate, with seedling shoots burnt after 1 week. The calcium application rate was then halved, which allowed plants to partially recover. Despite this, plants irrigated with calcium were significantly smaller than the control plants when assessed.

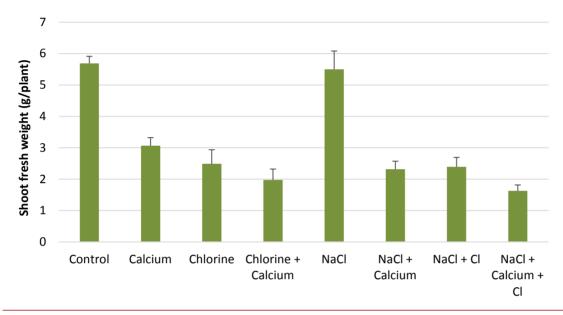


Figure 23 – Fresh shoot weight of lettuce seedlings irrigated with different chemical treatments, measured at commercial maturity (18 days after germination). Bars indicate the standard deviation of each mean value (n=3).

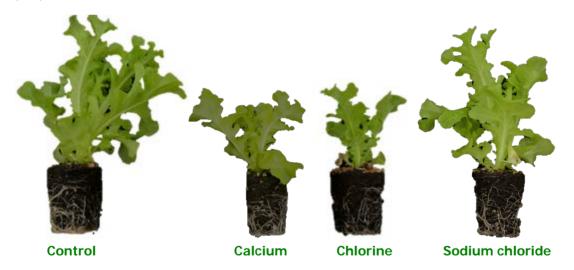


Figure 24 – Size of untreated (control) plants compared to those treated with calcium, chlorine or sodium chloride.

Excluding calcium applied alone, blindness was evident in all treatments and the control (Figure 25). However the incidence of blindness was highly variable across replicates. Some trays within a treatment were free of blindness, while others had over 6% incidence.

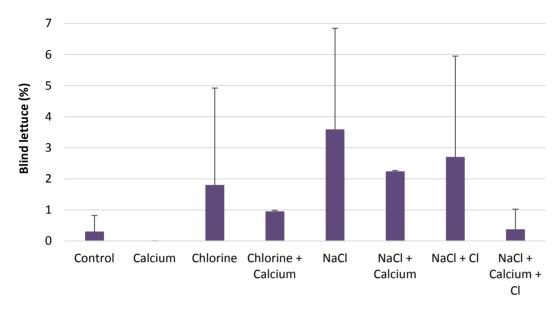


Figure 25 – Percentage of blind lettuce seedlings irrigated with different chemical treatments, assessed at commercial maturity (18 days after germination). Bars indicate the standard deviation of each mean value (n=3).

### **Discussion**

This trial confirmed previous results, indicating that saline and/or chlorinated water can induce blindness in 'Kireve' lettuce. This supports observations by lettuce seedling growers associating high EC readings with increased incidence of blindness. The results suggest that growers should carefully monitor the EC of their irrigation water, particularly during summer.

Saline water could even be used as a tool to screen different lettuce varieties for susceptibility to blindness. Irrigating seedlings with water with sodium chloride added to result in an EC of 2000 µs.cm<sup>-1</sup>, reliably induced blindness in these trials, and may allow ranking of different varieties based on susceptibility to this treatment.

In this trial, blindness was evident in the control (at a low level), which was not the case previously. This may be due to the high temperatures attained, with daily maximums up to 10°C higher than previously. This supports the observation from growers that blindness is more likely to occur if daily maximum temperatures exceed 35-40°C.

No blind plants were observed when the seedlings were treated with calcium. Moreover, on average, adding calcium to NaCl and Cl treatments reduced blindness. While this supports the hypothesis that blindness is caused by calcium deficiency in the apical shoot, this result would need to be repeated. This is especially the case as calcium was initially applied at too high a rate, stunting seedling growth.

# 3. Reducing transplant shock

# **Background**

Transplant shock is a check in growth that can occur when seedlings are transplanted from the seedling tray into the field. Significant transplant shock can result in poor plant stands and a lower percentage cut of good quality lettuce.

The timing at which lettuce seedlings are transplanted is critical to controlling transplant shock. Young seedlings are more delicate than older seedlings and more sensitive to damage during transplanting. Young seedlings can develop stronger root systems if transplant shock is controlled, leading to higher yields.

There is some evidence to suggest that applying nitrogen and potassium in the form of potassium nitrate (KNO<sub>3</sub>)—either just before or just after transplant—can help overcome transplant shock in lettuce (Dennis Phillips, pers. comm).

The trial was conducted to see whether the insecticide drenches Confidor<sup>®</sup> and Durivo<sup>®</sup> (used to control lettuce aphid) and the plant growth drench, Seasol<sup>®</sup>, containing gibberellic acid, exacerbate or reduce transplant shock. The impact of these insecticide drenches on growth and yield was evaluated, and a test was conducted to see if the application of potassium nitrate could minimise the effects of transplant shock. The drenches were applied to the seedlings prior to transplant to determine which nutrient combination and application method was better at overcoming transplant shock.

### Field trial 2014

## Aim

The aim of this trial was to evaluate the growth and yield response to potassium nitrate, Confidor<sup>®</sup>, Durivo<sup>®</sup> and Seasol<sup>®</sup>, individually and combined, applied as a drench to lettuce seedlings at transplanting.

### Method

Cos (cv. Quintas) and iceberg (cv. Toscanas) lettuce seedlings were transplanted into beds to a density of 44,000 plants per hectare on 23 September 2014.

A randomised complete block design (RCBD) was used, with four replicate blocks for each of seven treatments. Treatments were as follows:

1. Potassium nitrate 40 g to 2.5 L water

2. SEASOL® \* 1: 250 solution Seasol®

3. CONFIDOR® \*\* 35 ml/ 1000 seedlings

4. SEASOL® + Potassium nitrate 1:250 Seasol® solution + 40g KNO<sub>3</sub> to 2.5 L water

- 5. DURIVO® \*\*\* 30 ml/ 1000 seedlings
- 6. DURIVO® + Potassium nitrate 30 ml Durivo to 1000 seedlings + 40 g KNO<sub>3</sub> to 2.5 L water
- 7. Control
- \* soluble concentrate containing 100 g/L of gibberellic acid
- \*\* active ingredient 200 g/L Imidacloprid
- \*\*\* active ingredients 100 g/L of Chlorantraniliprole and 200 g/L Thiamethoxam

All treatments were applied as drenches before planting.

The plants were assessed at two stages; first at four weeks post-transplant (on 21 October) and then at harvest (on 10 November) seven weeks after planting.

At each assessment, eight individual plants were sampled from the centre of the rows, with the outer rows and end buffer plants discarded. The plants were trimmed for marketing as fresh market naked lettuce with the roots from the base of the stem removed. Leaf fresh weights were measured at each assessment stage and then recorded in grams per plant.

#### Results

### **Iceberg lettuce**

The potassium nitrate drench resulted in a significant increase in harvest head weight compared to the untreated controls, in the order of approximately 19%. Other values were not significantly different to the controls

This was less evident at four weeks post-transplant. At this stage none of the treatments were significantly different to the controls. However, there was a clear trend to increased weight in the plants drenched with potassium nitrate, whether or not they had also been treated with Seasol® or Durivo®.

Table 9 - Fresh weight of iceberg lettuce (cv. Toscana) recorded 4 weeks after planting and at commercial harvest

Treatment	Head weight (g)				
rreatment	Pre-harvest		At harves	At harvest	
Control	81	bc	857	b	
Potassium nitrate	101	ab	1023	a	
Seasol <sup>®</sup>	91	b	969	ab	
Confidor <sup>®</sup>	81	bc	957	ab	
Durivo <sup>®</sup>	78	С	907	b	
Seasol <sup>®</sup> + Potassium nitrate	105	ab	909	b	
Durivo® + Potassium nitrate	105	ab	899	b	

#### Cos lettuce

Cos lettuces drenched with potassium nitrate were significantly larger than the untreated controls, both

four weeks after transplanting and at commercial maturity. The increase was similar to that observed for iceberg lettuces, being around 19%.

As with the icebergs, there was a non-significant trend to larger lettuces after four weeks when seedlings were given a potassium nitrate drench, whether or not this was combined with other factors. However, this effect had largely disappeared by the time cos lettuces were ready to harvest.

Table 10 - Fresh weight of cos lettuce (cv. Quintas) recorded 4 weeks after planting and at commercial harvest

Treatment	Head weight (g)			
rreatment	Pre-harvest		At harves	st
Control	192	b	960	d
Potassium nitrate	235	a	1142	a
Seasol <sup>®</sup>	183	С	1078	b
Confidor <sup>®</sup>	200	b	1036	bc
Durivo <sup>®</sup>	205	b	1020	bcd
Seasol <sup>®</sup> + Potassium nitrate	215	ab	1022	bcd
Durivo® + Potassium nitrate	211	b	996	cd

## **Discussion**

Lettuce fresh weight was significantly increased when potassium nitrate was applied as a drench at transplanting at a dilution rate of 40 g per 2.5 L of water per 1000 seedlings. Overall, potassium nitrate was observed to reduce transplant shock, resulting in 12–19% higher yields at harvest. Alternatively, this increase in size could allow an earlier harvest, effectively reducing risk, freeing up land and potentially providing a faster return on investment.

The effects of the other drenches were more mixed. There were no negative effects from the insecticide treatments, and in some cases there was a small benefit in terms of head size.

This trial needs to be repeated to confirm the positive effects found.

## Field trial 2015-2016

### Aim

To repeat results from the 2014 lettuce transplant shock trial to confirm growth responses to potassium nitrate alone or in combination with insecticide drenches Confidor® and Durivo®, when applied 48 hours prior to, or immediately before, transplanting.

### Method

Ten trays of commercially grown iceberg lettuce seedlings (cv. Bernadenas) were obtained at normal commercial planting maturity. All seedlings were placed in dark storage for 48 hours to simulate transportation from nursery to farm.

A total of ten different treatments were applied during the trial (1 per tray). Each tray was then split into four replicate blocks for planting, giving a total of 40 treatment units.

Four of the trays were drenched 48 hours before transplanting with Confidor® or Durivo®, each with or without the addition of potassium nitrate (80g/1000 seedlings). This treatment was repeated with four extra trays immediately before transplanting. Two additional trays were used as controls, and drenched with either water or with potassium nitrate (80g/1000 seedlings) only, immediately before transplanting (Table 11).

Table 11 - Pre-transplant drenches

Treatment	Potassium nitrate	Timing	Rate/1000 seedlings
Control	+KNO3	At transplant	5 L water
Control	- KNO3	At transplant	o L water
Confident 40 hr	+KNO3	40 hvo nyo tvononlant	25 mal
Confidor® 48 hr	nfidor® 48 hr 48 hrs pre-transplant – KNO3		35 ml
C	+KNO3	At too a subset	25 ml
Confidor® 0 hr	- KNO3	At transplant	35 ml
D ' @ 40 l	+KNO3	40.1	20
Durivo® 48 hr	- KNO3	48 hrs pre-transplant	30 ml
D : @ 0.1	+KNO3		20 1
Durivo® 0 hr	- KNO3	At transplant	30 ml

All treatments were applied with a watering-can in 5 L water per 1000 seedlings

The seedlings were hand-transplanted into beds at a density of 44,000 plants/ha on 14<sup>th</sup> October 2015 at a commercial vegetable farm in Maroota, NSW. The trial design was a randomised complete block design with four replications.



Figure 26 - Lettuce transplanting on 14 October 2015.

Shoots and roots were weighed 22 and 49 days after transplanting, using 5 or 10 plants per treatment unit respectively. Shoots were weighed fresh. Roots were dried and soil shaken off before weighing on the first sampling date. Unfortunately this resulted in significant breakage of the fine roots. The protocol was therefore changed and roots were washed and weighed fresh on the second sampling date.

### **Results:**

### Mid-harvest assessment

There was a trend towards higher shoot weights in all treatments that included potassium nitrate, although these were not significantly different from the control (Figure 27).

Shoot weights of seedlings treated with Confidor $^{\text{®}}$  and Durivo $^{\text{®}}$  48 hours before transplanting were 15% lower than the control (p=0.000). When potassium nitrate was applied with these treatments, shoot weights were at least 20% higher.

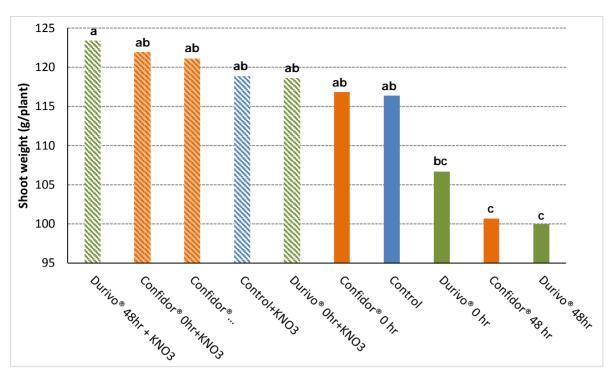


Figure 27 - Shoot weight of iceberg lettuce (cv. Bernadenas) treated with Confidor or Durivo soil drenches 0 or 48 hours before transplanting. Assessed 22 days after transplanting. Columns with different letters are significantly different at p<0.05.

Table 12 - Comparison of individual treatment factors (averaged across combined treatments) at mid-harvest.

Treatment factor	Shoot weight (g/plant)	Root weight (g/plant)
-KNO <sub>3</sub>	108a .	3.25a .
+KNO <sub>3</sub>	120 b	3.09 b
	p=0.000	p=0.0023
Confidor®	115	3.2
Durivo®	112	3.2
	ns	ns
48hr drench delay	111	3.2
No delay	116	3.2
	ns	ns
Position in field (max yield)	141a .	3.5a .
Position in field (min yield)	91 b	2.9 b
	p=0.000	p=0.0036

Root weights in treated seedlings did not significantly differ from the control (Figure 28). Treatments with higher shoot weights tended to have lower root weights, such as those treatments that included potassium nitrate.

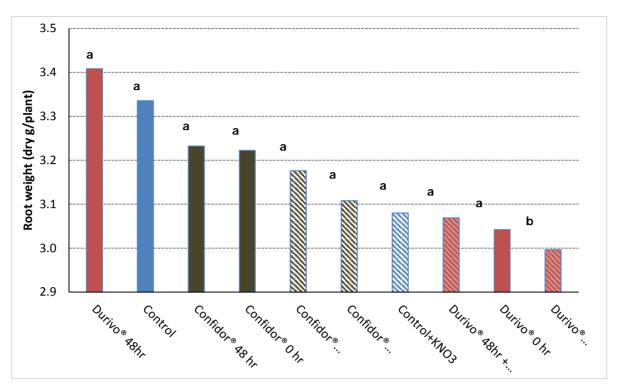


Figure 28. Root weight of iceberg lettuce (cv. Bernadenas) treated with Confidor or Durivo soil drenches 0 or 48 hours before transplanting. Assessed 22 days after transplanting. Columns with different letters are significantly different at p<0.05.

## Final harvest assessment

By final harvest, the trend seen at mid-harvest of higher shoot weights in potassium nitrate treated plants was absent, as were the lower shoot weights of seedlings treated with Durivo® and Confidor® 48 hours before transplanting.

Shoot weights in all treatments did not significantly differ from the control, although there were some significant differences between insecticide treatments (Figure 29). Applying Confidor® at transplanting rather than 48hrs before-hand, resulted in 11% higher shoot weight (p=0.0013).

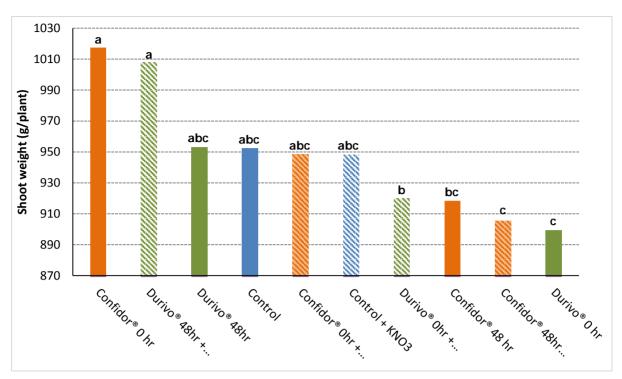


Figure 29 - Shoot weight of iceberg lettuce (cv. Bernadenas) treated with Confidor or Durivo soil drenches 0 or 48 hours before transplanting. Assessed at commercial maturity. Columns with different letters are significantly different at p<0.05.

Table 13. Comparison of individual treatment factors (averaged across combined treatments) at final harvest.

Treatment factor	Shoot weight (g/plant)	Root weight (g/plant)
-KNO <sub>3</sub>	948	32a .
+KNO <sub>3</sub>	946	31 b
	n.s.	p=0.0095
Confidor®	947	32a .
Durivo®	945	30 b
	n.s.	p=0.0003
48hr drench delay	946	31
No delay	946	30
	n.s.	n.s.
Position in field (max yield)	999a .	37a .
Position in field (min yield)	835 b	25 b
	p=0.000	p=0.000

Effects of treatments on root weight at harvest did not significantly differ from the control. Similar to the mid-harvest, there was a tendency for plants treated with potassium nitrate to still have lower root weights than other treatments, although differences were not significant.

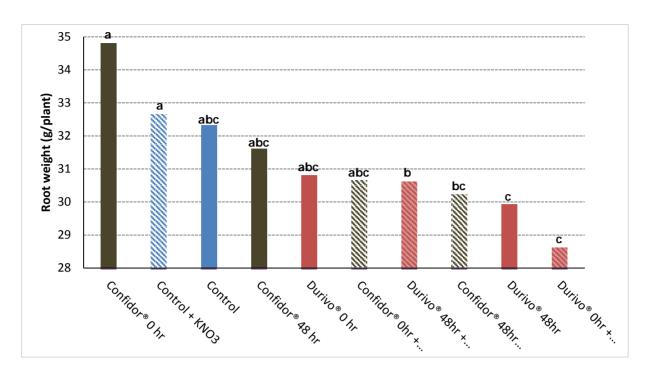


Figure 30. Root weight of iceberg lettuce (cv. Bernadenas) treated with Confidor or Durivo soil drenches 0 or 48 hours before transplanting. Assessed at commercial maturity. Columns with different letters are significantly different at p<0.05.

#### **Discussion**

Significant differences in root and shoot growth were found during this trial. However, in most cases treatments were not significantly different to the untreated controls. Moreover, many of the trends that were apparent early during the development of the crop had disappeared by the time the lettuces reached commercial maturity, 49 days after transplanting.

For example, in this trial there was an initial significant reduction in shoot growth when Durivo® and Confidor® were applied 48 hours prior to transplant. However, this reduction was not evident at final harvest, when there were no significant differences between the treated and control plants, regardless of when the drench was applied. This contrasts with the previous trial, which found a small but significant increase in final head weight when the Confidor® or Durivo® drenches were applied. Overall, however, the results suggest that drenching lettuce seedlings with Confidor® or Durivo® is likely to have minimal effect on yield.

The results provide evidence that drenches with potassium nitrate can reduce transplant shock. Shoot weight was increased early during growth when potassium nitrate was used. However, by the time plants grew to commercial maturity, differences in yield were no longer significant. This is different to the previous seasons data, which indicated that early differences in leaf development were retained to maturity.

The significant decrease overall in root weight when plants received potassium nitrate may provide some clue to why this occurred. It is possible root growth was initially less because available nitrogen

was immediately available in the root zone after planting, so the roots did not need to grow as far. A smaller root system can mean plants are less tolerant to water stress. Two heatwaves occurred during the 2015-16 trial. Also, soil type and irrigation pattern was not uniform across the site. Such stresses may have affected the potassium nitrate treated plants (with smaller root systems) than control plants, allowing growth of the latter to 'catch up'.

In this trial, position in the field had a far greater effect on lettuce size and yield, as well as root growth, than any of the treatments applied. This was due to the above noted changes in soil type across the site, compounded by uneven irrigation patterns.

In summary, potassium nitrate drenches can reduce transplant shock and have a positive effect on growth of iceberg lettuce. However, the effects are likely to depend on age of seedlings at transplanting, weather and other factors. Moreover, the effects of potassium nitrate drenches may be less than effects due to soil and water. Getting these right, and focussing on growing only in productive areas, may provide a better return on investment than potassium nitrate drenching.

## 4. Lettuce Blindness Fact Sheet



#### What is blindness?

Blindness occurs when the main apical shoot or growing tip of the lettuce is lost during the seedling's early growth. It is also sometimes called multiple heading or apical meristem decline. A similar disorder also occurs in brassicas such as cabbage, and in tomato seedlings. Lettuces seldom recover from blindness, developing into a distorted, unmarketable plant with no proper heart.

#### How much of a problem is this disorder?

In bad cases, blindness can affect more than 30% of seedlings. Ideally, affected lettuce should be removed from trays and replaced with healthy plants by the seedling producer. However, this is labour intensive and expensive. Moreover, blind plants are not easy to spot, especially when plants are still small. However, if blind seedlings are planted out, then all the costs of planting and growing are incurred without any saleable crop.



Blind lettuce with deformed developing leaves
The developing or older leaves appear thickened, stunted and
distorted

Following transplanting, this seedling is likely to continue to grow. However, it will fail to develop normally and will probably be unmarketable.

These lettuces are sometimes referred to as 'mongreled'.



Blind lettuce lacking apical shoot
This is the most typical symptom of blindness.
Seedlings lacking an apical shoot are unlikely to recover and grownormally.



Blind lettuce with multiple growing tips
A number of growing tips develop with no clear dominant shoot
Seedlings with this symptomoan develop multiple heads. In
varieties such as iceberg or cost his prevents development of a
proper heart. Again, the plant is unmarketable.

This project has been funded by Horticulture Innovation Australia Limited using the vegetable levy and









# 5. Reducing transplant shock in lettuce - Fact Sheet



#### What is transplant shock?

Transplant shock is a check in growth that can occur when seedlings are transplanted from the seedling tray into the field. Stresses due to root damage, changed environment or water stress can all contribute to transplant shock. Significant transplant shock can result in poor plant stands and a lower percentage cut of good quality lettuce.

#### How old should transplants be?

The timing at which lettuce seedlings are transplanted is critical to controlling transplant shock. Transplanting lettuce seedlings while they are small minimises injury to the developing roots and potentially allows the plants to establish quickly. However, young plants are fragile and are therefore difficult to handle without damage.

Transplanting when lettuces are further developed means they are more robust. However, if the roots are compacted



or have started to circle the cell in the tray they may fail to develop a good structure after planting in the field. They are also likely to be damaged during transplanting.

Ideally, lettuce seedlings should be grown in an environment similar to that in which they are to be planted. For example, transplant shock is more likely if seedlings grown under heavy shading are planted in a hot field with strong sunlight. Similarly, seedlings that have been grown rapidly using strong nutrient solutions are more likely to develop transplant shock than those grown more slowly.

One of the key factors for good establishment of transplants is a high root shoot ratio. That is, the roots need to be large relative to the upper part of the plant. Nitrogen causes seedlings to grow fast, but this is often at the expense of the roots, resulting in a small root shoot ratio. Small root systems leave the plant vulnerable to transplant shock, especially if conditions are hot, dry and/or windy.

Lettuce seedlings therefore need to be transplanted when they are not too young, or too old, but just right.

#### What else can I do to avoid transplant shock?

Good soil preparation, proper planting depth and thorough irrigation in the days following transplanting can all reduce risk of transplant shock.

One new technique that has been reported as reducing transplant shock is drenching seedling trays with a solution of potassium nitrate. This can be done just before planting.

Trials in 2014 and 2015 examined growth of iceberg and cos lettuces drenched with a solution of potassium nitrate, applied at a rate of 80g per 1,000 seedlings compared to those left untreated. Three weeks after transplanting, the potassium nitrate drenched lettuces were larger than those left untreated in both 2014 and 2015 (Figure 1).

However, the root systems of the drenched plants were smaller than the untreated seedlings (Figure 2). This is likely due to the potassium nitrate increasing availability of nitrogen and potassium in the root zone. This favoured

This project has been funded by Harriculture Innovation Australia Limited using the vegetable levy and funds from the Australian Government.







