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Field Evaluation of green LEDS for Culicoides brevitarsis

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ABSTRACT

This study was carried out to demonstrate the improved efficiency of green light-emitting diodes (LEDs) over incandescent globes currently used in light traps to monitor the arbovirus vector, *Culicoides brevitarsis* and a range of other *Culicoides* species. Trapping was conducted over a wide range of situations in New South Wales, the Northern Territory, Western Australia and East Timor. Light traps with green LEDs were superior to those with incandescent globes for *C. brevitarsis* at all locations. The green LED traps were also superior for most other species of *Culicoides* which included pests of humans and potential vectors of viruses (e.g. bluetongue and Akabane) affecting livestock and native animals in Australia and in the Indonesian region. It was proposed that MLA support a recommendation to the National Arbovirus Monitoring Program (NAMP) Organising Committee that green LEDs be adopted for the better detection of new and potentially dangerous exotic species and for the improved detection of vectors of livestock viruses outside of the vectors endemic areas and at the margins of their seasonal movements. Improved monitoring will benefit the livestock industry by increasing confidence in vector and virus free areas presented by the NAMP to define limits to disease outbreaks and for the export of livestock to arbovirus sensitive countries.

EXECUTIVE SUMMARY

Project Objectives

The objectives of this project were:

- to improve the trapping efficiency of the light traps currently used to provide information to the National Arbovirus Monitoring Program (NAMP) (Kirkland et al. 1995) for defining vector distributions and virus free areas within Australia,
- to demonstrate the effectiveness of green LEDs for trapping a wide range of Culicoides spp (particularly Culicoides brevitarsis) at different locations and under a range of vector densities.

Methodology

Collections of Culicoides species were made using standard light traps. A single green LED trap and an incandescent light trap were to be located at six sites in NSW, the Northern Territory and Western Australia. Four extra traps were located in NSW at sites marginal for C. brevitarsis and Akabane virus making a total of 22 proposed sampling sites. An additional and limited data set was taken in East Timor using incandescent globes and green and blue LEDS. Locations were selected at random from current National Arbovirus Monitoring Program (NAMP) sites and covered a wide range of geographical, climatic and vector density situations. Traps were placed 20 – 30 m apart at a height of 2 m in paddocks containing cattle. Collections were made over two nights, once per month for five months (January to May 2004). The data were all forwarded to the Gosford Horticultural Institute for analysis.

Results

New South Wales

Twelve species of *Culicoides* were recorded from NSW. Counts of seven species were significantly higher in traps with green LEDs. The other five species also exhibited a higher response to green LEDs but this could not be confirmed. There were significantly more *C. brevitarsis* in the endemic zone and the superiority of the green LEDs was constant inside and outside of the endemic zone.

Northern Territory

Twenty-five species of *Culicoides* were recorded in the NT. Counts of 12 species were significantly higher in traps with green LEDs. Four species had no significant differences recorded. No analysis was possible on nine species. Of these, four were caught in green LED traps only, one in the incandescent trap only and four in both traps.

East Timor

Although not part of the original objectives, data were also collected from East Timor. This enabled us to test trap efficiency on several exotic species likely to enter Australia. Nineteen species of *Culicoides* were recorded in East Timor. Counts of 2 species were significantly higher in the traps with the green LEDs. No significant differences were recorded for 8 other species mainly due to statistical problems with the limited data sets. However, counts for all species were numerically higher with green LEDs than with incandescent globes and the blue LEDs. Nine of these species are not currently present in Australia.

Western Australia

Only four sites could be established in WA. Insufficient data were received from WA due to low insect densities and this data set could not be analysed.

Conclusions

Light traps with green LEDs were superior to the currently used incandescent globes for most Culicoides species in a wide range of environments and for a range of insect densities. Larger catches of these insects would be important for:

● the detection of newly arrived and potentially dangerous exotic species.

● the improved detection of known vectors of livestock viruses outside of endemic areas and at the margins of their seasonal movements.

● obtaining large numbers of individuals for research.

The current system of monitoring could be enhanced by green LEDs in the following ways:

● trapping efficiency for C. brevitarsis would be improved 3 to 9 times.

● the current traps could be used by the simple replacement of the incandescent globes.

● LEDs are relatively inexpensive.

● LEDs have extremely long operational lives

● LEDs require considerably less power than incandescent globes and the traps could therefore be operated for longer periods.

The current system of monitoring could be disadvantaged in the following ways:

● extra insects being caught in high abundance areas could make counting more difficult.

● long term comparisons of numbers obtained using incandescent traps would be jeopardised by the change to the more efficient green LEDs. This should not be a major problem as many other factors can also make comparisons unreliable.

● catches of many insects from other orders were also enhanced. This made counting of Culicoides spp difficult but could help the collection and study of a range of other insects.

Recommendations and Application of Outcomes

There are several advantages to the adoption of green LEDs for trapping *C. brevitarsis* and other potential vectors of bluetongue and Akabane viruses in Australia. The few disadvantages that occur should not prevent their complete adoption. It was concluded that greatest advantages could be obtained where:

● it is necessary to detect exotic invasions of species from the Asian region.

● vector distributions are seasonally and climatically marginal

● virus transmission is occurring in the apparent absence of known vectors.

It is therefore recommended that:

● MLA support a recommendation to the NAMP Organising Committee that green LEDs be adopted for use, at least under the circumstances above.

● data collection continue so as to allow confirmation of the apparent preferences of a large number of species for green LEDs over incandescent globes.

● research into the attraction of *Culicoides* spp. to LEDs be continued in an effort to:

  ● document the responses of the full range of Australian *Culicoides* species to LEDs

  ● determine why such clearly defined groups have arisen within a single genus of insects based on their responses to different colours in the visible and non-visible spectrum.

  ● understand the physiological reasons for the behavioural responses.

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1. Project Background

Biting midges from the genus *Culicoides* (Diptera: Ceratopogonidae) are important because of their effects on both human and animal hosts. The Australian fauna is extensive with distributions primarily dependent on breeding habitats and climate. Several species are vectors of viruses affecting native animals (Standfast *et al*. 1984) while others transmit viruses to livestock. In Australia, *Culicoides brevitarsis* Kieffer is the main species responsible for the transmission of the bluetongue and Akabane viruses (Muller *et al*. 1982) which impact on animal health and impede trade to arbovirus sensitive trading countries. Other potential vector species exist in northern-Australia, while another, *Culicoides wadai* Kitaoka has extended its previously northern distribution into northern-coastal New South Wales (NSW).

The distribution of *C. brevitarsis* extends from the Pilbra region of Western Australia (WA), across the Northern Territory (NT) (Muller *et al*. 1981), through Queensland and down the coastal plains of NSW (Bishop *et al*. 1995). The extent of its distribution is dependent on its seasonal movements which are limited by temperature, moisture and host availability. Its distribution encompasses those of the other potential vectors such that it has been accepted that virus transmission usually occurs within its dispersive range. However, occasionally Akabane activity (and therefore potentially bluetongue activity) has been detected in sentinel cattle in the apparent absence of *C. brevitarsis* (PD Kirkland, personal communication). This makes it difficult to understand the epidemiology of the viruses and to describe acceptable virus free areas.

Adult *Culicoides* species commonly enter light traps and these are used to monitor or study the distributions, behaviours, ecologies and importance of numerous vector and nuisance species (Braverman & Phelps 1981, Zimmerman & Turner 1883, Greiner & Rawlins 1987, Bhatnager *et al*. 1994). In Australia, these light traps have incandescent globes, are designed for use in isolated locations and have photoelectric cells to trigger their operation at night. Most trapping overseas is conducted using ultraviolet (UV) (blacklight) traps. Work completed in 2003 using different coloured light-emitting diodes (LEDs) (Bishop *et al*. 2004, Appendix 1) showed that some species (4 of 8), including *C. brevitarsis*, are attracted to green light and that this could improve trapping efficiency by 3 to 9 times. The other four species were more attracted to blue light. Work completed in 2004 (AL Bishop *et al*., unpublished data, presented and prepared for initial publication in the proceedings of the 9th Arbovirus Research in Australia Conference, Appendix 2) confirmed the previous responses to green light and showed that those species responding to blue light in 2003 had a higher preference for UV light when this was offered as an alternative source of attraction.

2. Objectives

The objectives of this study were:
- to improve the trapping efficiency of the light traps currently used to provide information to the National Arbovirus Monitoring Program (NAMP) (Kirkland *et al*. 1995) for defining vector distributions and virus free areas within Australia,
- to demonstrate the effectiveness of green LEDs for trapping a wide range of *Culicoides* spp (particularly *C. brevitarsis*) at different locations and under a range of vector densities.

3. Methodology

3.1. Light Traps

Collections were made using standard light traps as described by Dyce *et al*. (1971), initially modified by HA Standfast (personal communication) and recently modified be AL Bishop and HJ McKenzie (unpublished data). The traps are powered by three 1.5V alkaline 'D' cell batteries which are replaced after two nights of operation. Insects responding to the light source are drawn into plastic storage bottles containing 70% alcohol with a downwardly directed fan. Collections are returned to the laboratory where they are sorted and identified to species primarily by their wing patterns under x 10 magnification.

3.2. Light Sources

Light sources were 3.5 V incandescent globes and green LED lights standardised with the same quantum output as the incandescent globes using the method described in Bishop *et al*. (2004) (Appendix 1).

3.3. Field Evaluation

A single green LED trap and an incandescent light trap were to be run together at six sites in NSW, the Northern Territory and NW Western Australia. Four extra traps were located in NSW at sites suspected as being marginal for *C. brevitarsis* and Akabane virus making a total of 22 proposed trapping sites. Locations were selected at random from current NAMP sites and covered a wide range of geographical, climatic and vector density situations. Traps were placed 20 – 30 m apart at a height of 2 m in paddocks containing cattle. Collections were made over two nights, once per month for five months (January to May 2004).

Although not part of the original objectives, an additional and limited data set was taken in East Timor using incandescent, green and blue LEDS. These data were taken to gain preliminary information on species that may be a future threat in Australia. Data were also obtained on several species that are currently found in both the Indonesian and Australian regions.

The data were all forwarded to the Gosford Horticultural Institute for analysis.

3.4. Analysis of Data.

Data from NSW, NT, WA and East Timor were considered separately. Insufficient data were received from WA and this data set could not be analysed.

The data for NSW and NT were regarded as repeated measurements over the 5 months and a linear mixed model which accounted for correlation over time and variance heterogeneity was used (Verbyla *et al*. 1999). The data from East Timor were only taken on four sampling occasions at one site. A factor called zone was created from the NSW data only with each of the 10 sites classified as being located either inside or outside the *C. brevitarsis* endemic zone. Month was modelled as a fixed linear trend. Curvature around this line was modelled as a cubic smoothing spline. Tests of whether the linear time trend is the same for both treatments and both zones were also made. The fixed effects of treatment, zone, the linear trend with time and their interactions on *C.brevitarsis* count ($\log_e$ transformed) were tested using ASReml (Gilmour *et al*. 1998). The following additional sources of variation were included in the model as random effects: spline(month), month, treatment x spline(month), zone x spline(month), site and site x month.

4. Results

New South Wales

Counts of seven of the twelve species recorded from NSW were significantly higher in traps with green LEDs (Table 1). A suggestion that the other five species also exhibited a higher response to green LEDs could not be confirmed due to their low and inconsistent numbers. The analysis showed that there were significantly more *C. brevitarsis* in the endemic zone and that the superiority of the green LEDs was constant inside and outside of the endemic zone.

Table 1. Table of back-transformed treatment means per trap for 12 species of *Culicoides* sampled at sites at different locations and with different densities of insects throughout NSW. Means in rows with different letters are significantly different at $P < 0.05$. * Collected at one site only.

Species	Incandescent	Green LEDs
C. austropalpalis	4.5 b	46.5 a
C. brevitarsis	83.4 b	565.5 a
C. bundyensis	0.9 b	11.4 a
C. bunrooensis	0.0	8.0
C. dycei	1.8 b	13.2 a
*C. henryi**	16.7	151.3
C. marksi	5.9 b	61.8 a
*C. marmoratus**	7.7	18.5
*C. narrabeenensis**	0.0	0.6
C. nattaiensis	0.1 b	2.8 a
C.victoriae	2.2 b	8.8 a
*C. wadai**	0.0	1.3

Northern Territory

Counts of 12 of the 25 species recorded in the NT were significantly higher in traps with green LEDs (Table 2). Four species were analysed with no significant differences recorded. These species were only found in low numbers and were recorded infrequently. No analysis was possible on nine species. Of these, four were caught in green LED traps only and one in the incandescent traps only. Four other species were caught in both traps.

Table 2. Table of back-transformed treatment means per trap for 25 species of *Culicoides* sampled at sites at different locations and with different densities of insects throughout the Northern Territory. Means in rows with different letters are significantly different at $P < 0.05$. * Insufficient numbers to analyse.

Species	Incandescent	Green LED
C. actoni	2.5 a	2.9 a
C. austropalpalis	13.9 b	27.9 a
C. brevipalpis	1.1 b	6.2 a
C. brevitarsis	8.8 b	17.6 a
C. bundyensis	6.4 b	17.6 a
C. calcaratus *	0.0	0.2
*C. clavipalpis**	0.0	0.9
*C. cuniculus**	0.0	2.7
*C. dumdumi**	0.4	0.6
C. dycei	0.7 a	0.9a
C. fulvus	2.7 b	11.0 a
C. guttifer *	0.2	0.0
C. histrio *	0.2	0.6
C. marksi	1.8 b	3.7 a
C. narrabeenensis	0.3 b	3.2 a
C. nattaiensis *	0.3	0.2
C. ogoweri *	0.0	0.2
C. ornatus	1.5 b	6.3 a
C. ornatus#6	1.3 b	3.9 a
C. oxystoma	4.3 b	12.6 a
C. pallidothorax	2.9 a	2.7 a
C. peregrinus	6.5 b	12.8 a
C. shermani *	0.2	0.1
C. victoriae	1.6 a	0.9 a
C. wadai	0.6 b	5.5 a

East Timor

Counts of 2 of the 19 species recorded were significantly higher in the traps with the green LEDs (Table 3). No significant differences were recorded for 8 other species. Counts for all species were numerically higher with green LEDs than with incandescent globes and the blue LEDs. The detection of statistical differences for most species was difficult due to the limited data sets.

Table 3. Table of back-transformed treatment means per trap for 19 species of *Culicoides* sampled at sites in East Timor. Means in rows with different letters are significantly different at $P < 0.05$. * Insufficient data to be analysed. # Not currently present in Australia.

Species	Incandescent	Green LED	Blue LED
C. actoni *	0.75	1.00	0.0
C. arakawe * #	0.0	3.30	0.50
C. brevipalpis	2.33 a	15.43 a	2.86 a
C. brevitarsis	8.94 c	77.81 a	15.02 b
C. clavipalpis *	0.0	0.25	0.0
C. effusus	1.97 a	7.98 a	1.24 a
C. flavipunctatus * #	0.0	0.75	0.0
C. fulvus	0.19 b	4.38 a	0.46 b
C. geminus * #	0.25	1.50	1.00
C. guttifer	0.68 a	1.82 a	0.81 a
C. histrio *	0.0	2.00	0.25
C. innoxius * #	4.50	6.50	0.0
C. maculatus * #	0.0	1.50	0.0
C. nudipalpis #	10.53 a	30.43 a	7.34 a
C. orientalis #	17.66 a	38.98 a	11.47 a
C. oxystoma	4.51 a	17.96 a	4.73 a
C. peregrinus	0.97 a	7.63 a	1.05 a
C. pseudopalpis *	0.25	1.75	0.50
C. wadai	5.38 a	7.41 a	4.47 a

Western Australia

Only four sites could be established in WA. Insufficient data were received from WA due to low insect densities and this data set could not be analysed.

5. Discussion

The results confirmed that green LED light traps have a distinct advantage over incandescent traps for all species, even those species whose ultimate preference is for UV. Optimum trapping procedures could be applied to any species once its colour preference has been determined. *C. brevitarsis* is the main species affecting the Australian livestock industry. It is strongly attracted to green light in light traps. The efficiency of green LEDs for *C. brevitarsis* has been shown experimentally and its superiority over the currently used incandescent globes demonstrated under a range of field conditions. The importance of this superiority was highlighted in NSW by *C. brevitarsis* being caught in green LED traps and not in incandescent traps on 20% of sampling occasions. There were two locations where *C. brevitarsis* was caught in green LED traps and never caught in incandescent traps over a five month sampling period. These locations included one site (Tenterfield) where the transmission of Akabane virus had been detected in a sentinel herd and there was no official occurrence of the vector (PD Kirkland, personal communication). The other (Denman) was close to the margin of Akabane activity recorded for 2003-2004 in the Hunter Valley.

Until now, information available on the interaction between *Culicoides* spp and coloured LEDs had only been derived in NSW. The NT and East Timor data greatly enhanced our current level of knowledge both in terms of total species and of potential vector species. In addition to *C. brevitarsis*, five potential vectors of bluetongue virus in northern-Australia (*Culicoides brevipalpis, C. fulvus, C. oxystoma, C. peregrinus* and *C. wadai*) were shown to have a significantly greater response to green LEDs than to incandescent lights. *Culicoides actoni* was the only species in this potential group not to show an increased response. This could be the result of *C. actoni* tending to feed during the day before the light traps became operational (G Bellis *et al*. in press). The most important species affecting humans (*C. ornatus*) also had a greater response to the green light.

Data from East Timor further confirmed the preference of *C. brevitarsis* and *C. fulvus* for green LEDs. The East Timor data strongly indicated that 8 species (Table 3) not yet found in Australia could be detected better with green LEDs. These included potential bluetongue and Akabane vectors (*Culicoides nudipalpis, C. orientalis, C. flavipunctata* and *C. maculatus*) and *Culicoides arakawe*, the main vector of bird malaria in the Asian region. The green LED light traps could therefore be important assets in the detection of incursions of new and potentially dangerous *Culicoides* species into Australia.

Most predictions of the activity and spread of *C. brevitarsis* are based on population monitoring with light traps and are more dependent on the species occurrence than its density (Bishop *et al*. 1995). Larger catches in endemic or established areas where the occurrence of *C. brevitarsis* is not in question may therefore be of little value and the extra time taken to count increased numbers be unnecessary. Greatest benefit for monitoring use would be derived in areas and at times marginal for *C. brevitarsis*, ie. for first occurrences outside of endemic areas and at sites with low density. Further benefits could be derived where larger catches may be required for virus isolation from vectors, for experimental use of vectors with animals or for detecting vectors at key locations involved in the export of livestock (staging areas and ports). Colours with higher attraction could possibly be combined with other stimuli currently being investigated or used alone in trapping systems designed to control the insects (eg. with insecticides, electrified grids or large collection chambers), particularly where important livestock (stud or show animals) are in confined areas such as stables and stalls. Most of these arguments apply to any *Culicoides* species and at any location.

6. Conclusions

Light traps with green LEDs were shown to be superior to the currently used incandescent globes for most *Culicoides* species in a wide range of environments and for a range of insect densities. Larger catches of insects could be important for a number of reasons, the most important being for:
● the detection of newly arrived and potentially dangerous exotic species.
● the improved detection of known vectors of livestock viruses outside of endemic areas and at the margins of their seasonal movements.
● obtaining large numbers of individuals for research.

The current system of monitoring would be enhanced in the following ways:
● trapping efficiency for C. brevitarsis would be improved 3 to 9 times by the inclusion of green LEDs in traps.
● the current traps can be used by the simple replacement of the incandescent globes with green LEDs.
● major alterations to traps would be unnecessary. This could occur if changes were made to systems using chemical stimuli or UV light.
● LEDs are relatively inexpensive.
● LEDs have extremely long operational lives.
● LEDs require considerably less power than incandescent globes and the traps could therefore be operated for longer periods.

The current system of monitoring could be disadvantaged in the following ways:
● extra insects being caught in endemic or high abundance areas could make counting more difficult.
● long term comparisons of numbers obtained using incandescent traps would be jeopardised. This should not be a major problem as many factors can influence numbers recorded in traps at different times and at different locations.
● catches of many insects from other orders were also enhanced. While this also made counting of *Culicoides* spp more difficult, it could be an advantage for the collection and study of a range of other insects.

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8. Achievement of Objectives

1. **"To improve the trapping efficiency of the light traps currently used to provide information to the National Arbovirus Monitoring Program (NAMP) (Kirkland *et al*. 1995) for defining vector distributions and virus free areas within Australia".**

Trapping efficiency for the main bluetongue and Akabane vector, *Culicoides brevitarsis* was improved up to ≈ 10 times by replacing the incandescent globes in standard light traps with green LEDs. Improved trapping efficacy was also confirmed for 16 other species. This report will be used in combination with a recommendation to the NAMP Organising Committee. This recommendation will propose the adoption of green LED light traps in areas where:
- effective monitoring is necessary to detect exotic invasions of species from the genus *Culicoides* from the Asian region.
- vector distributions are marginal.
- it is advantageous to detect initial movements of vectors away from endemic areas.
- vectors may have survived winter in normally non-endemic areas.
- virus transmission is occurring in the apparent absence of known vectors.

2**. "To demonstrate the effectiveness of green LEDs for trapping a wide range of *Culicoides* spp (particularly *C. brevitarsis*) at different locations and under a range of vector densities".**

The superiority of green LEDs over incandescent globes was demonstrated continuously for most species in NSW, NT and East Timor. Superiority was maintained over a wide range of geographic and environmental conditions, both inside and outside of endemic areas. It would be advantageous to continue to record these differences to provide more data for species where their low density or inconsistent presence prevented apparent differences from being validated statistically.

9. Intellectual Property

New information important to the livestock industries in Australia and possibly around the world has been gained during the course of this study. None can probably be regarded as commercially sensitive and needing protection. However, it is possible that the information could be adapted for use in situations where midge problems are of human concern (household protection) or where there is a need to protect valuable (breeding, show or racing) livestock in localised areas. Otherwise, all data from this project could be considered to be in the "public domain".

10. Financial Statement

The receipt and dispersal of funds from MLA during the course of this project are as follows.

INCOME	$
TOTAL FUNDS	12225.00
EXPENDITURE	
Sorting costs	2500.00
Laboratory costs	5250.00
Farmer allowance	1500.0
Consumables	1875.00
Freight for traps	600.00
Supply of LEDs	500.00
Total for MLA	12225.00
SUMMARY	
Initial payment (1/07/2004) (Not claimed) *	6113.00
Final payment (30/09/2004) **	12225.00

* Although milestone requirements were completed by the due date (1/07/2004) a milestone report was not submitted due to medical problems experienced by the Project Leader.

** It is requested that the total amount be paid on receipt and acceptance of this final report.

During the period that the project was undertaken, the direct contribution by the NSW Department was estimated to be $17046.00.

The significant extra contributions the co-operators from the NT Department, AQIS (NT and WA) has not been included and should also be recognized.

11. Impact of Research

The results of this research will significantly increase confidence in the proposed limits to distributions of vectors responsible for the transmission of the bluetongue and Akabane viruses to livestock in Australia. This will impact on questions of animal health and more importantly on the export of livestock to arbovirus sensitive trading countries. It will also aid the early detection of exotic *Culicoides* species entering Australia. Decisions to implement the use of the green LEDs will follow the submission of a recommendation to be submitted to the NAMP Organising Committee in July 2005.

Other benefits to the livestock industry could be derived by combining the improved trapping technology with control strategies on a limited area basis. The same may be applicable to species of human importance. This would require further research.

Scientifically, the research has opened new concepts of insect vision. The division of the genus into two groups based on their responses to green or UV LEDs is unique and is worthy of further consideration.

12. Acknowledgements

This project was carried out in co-operation with:

Dr L Melville, Department of Business, Industry & Resource Development, Berrimah Farm, NT; and

Mr G Bellis, Australian Quarantine & Inspection Service, Northern Australian Quarantine Strategy, Darwin, NT.

We are indebted to the producers / co-operators who maintained and operated the light traps on their properties. Our thanks go to Mrs L Spohr for her biometrical assistance, T Postle who carried out the project in WA and to H McKenzie (NSWDPI) and N Hunt (NTDBIRD) who carried out the technical components of the project.

13. Appendices

Appendix 1

Response of Culicoides spp. to light-emitting diodes

Alan L Bishop, Ross Worrall, Lorraine J Spohr, Harry J McKenzie and Idris M Barchia. 2004. Response of* Culicoides *spp. to light-emitting diodes. Australian Journal of Entomology 43, 184-188.

Response of Culicoides spp. to light-emitting diodes

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Running title Culicoides response to LEDs

Abstract

Light traps with incandescent globes are used in a national monitoring program to detect the presence of *Culicoides* spp responsible for the transmission of viruses to livestock and native animals. Recent events have suggested that the efficiency of these traps should be reconsidered and possibly improved. Subsequently, the response of eight species of *Culicoides* to light-emitting diodes (LEDs) was determined at two locations in New South Wales. *Culicoides austropalpalis* Lee & Reye, *C. bunrooiensis* Lee & Reye and *C. marksi* Lee & Reye were attracted to blue light. Responses to blue and green light could not be separated for *C. bundyensis* Lee & Reye, *C dycei* Lee & Reye, *C. nattiensis* Lee & Reye and *C. victoriae* Macfie. *C. brevitarsis* Kieffer was significantly attracted to green light. This species is the major vector of Akabane and bluetongue viruses in Australia. These responses were all significantly greater than the responses to the incandescent lights currently used in the light traps. The response to red light was less than the response to incandescent light for all species. Catches of *C. brevitarsis* were also related to the intensity of the green LEDs. These were more effective than the currently used incandescent globes at intensities between 46% and 142% of the incandescent intensity.

Introduction

Many species from the genus *Culicoides* (Diptera: Ceratopogonidae) are of medical or veterinary importance with effects on hosts ranging from mild annoyance to the transmission of viruses. Larvae live in a variety of habitats. Adults exhibit crepuscular activity and commonly enter light traps that are used to study their distribution, behaviour, ecology and importance as vectors (Braverman & Phelps 1981; Zimmerman & Turner 1983; Greiner & Rawlins 1987; Bhatnager *et al*. 1994).

The Australian *Culicoides* fauna is extensive and diverse, and, in many cases, distributions are restricted by geography, weather and habitat availability. Several species from the genus are vectors of viruses affecting native animals. As an example, at least eight individual viruses have been isolated from *Culicoides marksi* Lee & Reye (Standfast *et al.* 1984). *Culicoides brevitarsis* Kieffer is the main species responsible for the transmission of bluetongue and

Akabane viruses to livestock (Muller *et al.* 1982). Its distribution is chiefly coastal and it is endemic from the Pilbra region in Western Australia, across the Northern Territory (Muller *et al*. 1981) and down the coastal plains of Queensland to the northern / mid-northern coast of New South Wales (NSW) (Bishop *et al*. 1995b, 1996). Light traps with incandescent globes are used to monitor the presence of the *Culicoides* species and to compare their relative abundances (Dyce *et al*. 1971; Murray 1991; Bishop *et al*. 1995b). The traps are lightweight, designed for easy use in isolated locations and are triggered at sunset by photoelectric cells. However, there have been instances at the margins of the distribution of *C. brevitarsis* in NSW where Akabane activity has been detected by the serological-testing of sentinel cattle herds in the apparent absence of *C. brevitarsis* in light traps (PD Kirkland, Personal communication). It is possible that these traps fail to record low numbers of infective individuals and that this anomaly could be overcome if the efficiency of the traps could be improved.

The light source in traps for mosquitoes has been investigated by determining mosquito responses to the colour and intensity of conventional light sources and light-emitting diodes (LEDs) (Das & Reuben 1978; Browne & Bennet 1981: Ali *et al.* 1990; Burkett *et al.* 1998). There have been no similar studies on *Culicoides spp.* Olfactory and chemical stimuli have also been added to light traps to improve their efficiency for collecting mosquitoes (Takken & Kline 1989; Kemme *et al.* 1993). Similar responses have been reported for some *Culicoides spp* (Ritchie *et al.* 1994) and we are considering these effects on *C. brevitarsis* separately.

Insects can generally perceive and respond to light in the 350-700 nm range and their relative response can vary considerably over this range (Land 1997). The standard incandescent light sources used in our light traps generally have a maximum output at 700 nm within this range with little or no output below 400 nm. The response of insect eyes to light depends on both photon flux density (PFD) and wavelength. Peak wavelength sensitivities often occur where the output of the incandescent light is low. Recent advances in LEDs in the last 5 to 10 years have produced LEDs that are energy efficient, often producing a greater total photon flux (TPF) than incandescent globes in the 400-700 nm range for the same power input making them suitable for battery operation. LEDs also have the added advantage that they can provide closely defined outputs across narrow or wide spectral ranges, giving much higher TPF than incandescent globes over certain spectral ranges.

The aim of this study was to determine the response of a range of Australian species of *Culicoides* capable of transmitting viruses to livestock and native animals to different colours in the visible spectrum by the use of LEDs. Improved trapping efficiency, particularly for *C. brevitarsis,* was a major objective.

Materials and Methods

Experiments were carried out at Tocal (32.38S; 151.35E) and Denman (32.20S; 150.11E) in 2002 and 2003 in the Hunter Valley, NSW.

The light source (incandescent globe) in standard light traps was replaced in treatment traps with a range of light-emitting diode (LEDs) treatments and compared to the incandescent lights (see Table 1 for specifications). The traps were powered by three 1.5V alkaline ‘D’ cells, which were replaced after two nights of operation. All experiments were carried out when the effects of rainfall, wind speed and moon phase on trap catches were minimal.

The incandescent globes were placed initially in a 110 mm spherical chamber lined with high reflectance white paint. The quantum output was measured at 20^{o}C with a LI-COR Model LI-250 Light Meter with a LI-COR quantum sensor (approximately linear over 400-700 nm) through a port the same size as the active area of the sensor. The voltage supplied to the globes was 3.9 V to take into account the 0.6 V drop from the 4.5 V (total batteries) caused in the traps by the switching bipolar silicon transistor. The PFD of the trap with the incandescent

globe installed was 0.32 μ mol $m^{-2}s^{-1}$ measured with the light meter at 120 mm from the light source in a horizontal plane.

The LEDs (three for blue, green, white and red; and five for yellow) were placed in the same chamber and the current adjusted until the quantum output was the same as the incandescent light source as measured by the quantum sensor. Readings are presented in Table 1. The LEDs other than yellow were mounted in polycarbonate plastic diffusers (120^o apart) to ensure more even distribution of light. The yellow LEDs were mounted facing directly outwards on the same plastic caps at 72^o apart due to their lower quantum output per current input compared to other LEDS used, possible absorption by the diffusers and restrictions on the power available from the batteries. Current to the LEDs in the traps was controlled by a regulated constant current source.

Trial 1 – This trial was conducted at Tocal in March and April 2002 using six light-frequency treatments: blue, green, white, yellow and red LEDs and the standard incandescent globes. The traps were hung from 2 m "L" shaped frames and placed at 20 m intervals 3 m from one side of the common boundary of two adjacent ≈ 30 ha paddocks containing cattle. Four experiments were conducted 3 to 7 days apart. Yellow was not included in the first two experiments and replaced red in the next two experiments. The five treatments were arranged in five randomised blocks. The positioning of replicates remained constant while treatments were re-randomised for each experiment. Collections were made into bottles containing 70% alcohol over two nights. *C. brevitarsis* was identified from its wing pattern under x10 magnification and its total numbers counted and recorded.

Trial 2 - Four experiments were conducted, each 2 days apart at Denman in early-February 2003. Denman was chosen because it frequently has the greatest diversity of species at sites monitored in coastal NSW (AL Bishop, Unpublished data). It is also marginal for *C. brevitarsis* in most years. Five light frequency treatments (blue, green, yellow, red and incandescent) were used in this trial. White was omitted because it crossed the ranges of each of the other treatments. The treatments were arranged in five randomised blocks which were re-randomised at the start of each experiment. Traps were hung on 2 m high "L" shaped frames placed at 12 m intervals on two sides of a ≈ 10 ha paddock containing cattle. Collections were made over one night, the samples sorted and numbers of *C. brevitarsis, C. austropalpalis* Lee & Reye, *C. bundyensis* Lee & Reye, *C. bunrooensis* Lee & Reye, *C. dycei* Lee & Reye, *C. marksi , C. nattaiensis* Lee & Reye, *C. victoriae* Macfie identified from their wing patterns and counted.

Trial 3 - Four experiments were conducted at Tocal, 2 days apart in late-February 2003. Green LEDs at four intensities relative to the intensity of incandescent globes were compared with the incandescent light against *C. brevitasis*. The intensities were varied by adjusting the current to the LEDs. The five treatments were arranged in five randomised blocks in a 36 ha paddock containing cattle. The treatments were re-randomised for each experiment. Collections were made over one night and *C. brevitarsis* numbers counted as before.

Statistical methods

The influence of light frequency or intensity on counts of *Culicoides* spp. was modelled using a mixed linear regression approach (Searle 1971) which allowed the separation of variance components into fixed and random effects. To reduce heterogeneity of variances insect counts were $\log_e$ transformed for Trial 1, Trial 3 and for *C. austropalpalis* in Trial 2. The square root transformation was used for counts of all other species in Trial 2 due to their low numbers.

For each trial, analysis of the transformed counts was conducted using the REML directive in Genstat 5.4.1, Release 3. Treatment effects were examined for significance using Wald tests while treatment means were compared using the least significant difference (LSD) technique at the 5% level and then back-transformed to the original units. The model is given by

y = treatment + experiment + *block* + *block.experiment* + *experiment.treatment* + *block.plot* + *error*

where y = transformed count and the italicised terms are included in the model as random effects.
In Trial 2, where the recorded counts for *C. brevitarsis* and *C. nattiensis* were very low, the counts for the four experiments were pooled for each block and an Analysis of Variance performed.

Results

Trial 1 - The treatment effect was highly significant [Wald Statistic (WS) = 178.1, 5 df, P < 0.001]. Catches of *C. brevitarsis* were highest with green LEDs, lowest with red LEDs and with significant differences between each single-band treatment (Table 2). White was similar to the blue and green treatments but included all wavelengths with peak emissions in the blue and yellow ranges (Table 1).

Trial 2 - Significant treatment effects were recorded for each of the eight species at Denman (Table 2). Catches of *C. brevitarsis* (Variance Ratio = 7.62, 3,12 df, P < 0.01) were again highest with the green LEDs. Numbers of *C. brevitarsis* were low at Denman and were caught in the green treatment on each sampling occasion. They were first caught in the incandescent treatment in the third experiment. *C. austropalpalis* (WS = 375.2, 4 df, P < 0.001), *C. bunrooiensis* (WS = 38.7, 4 df, P < 0.001) and *C. marksi* (WS = 247.9, 4 df, P < 0.001) each exhibited highest responses to blue LEDs but green LEDs were also more effective than the incandescent light. Significantly higher responses to the blue and green LEDs relative to the incandescent could not be separated for *C. bundyensis (*WS = 40.9, 4 df, P < 0.001), *C. dycei* (WS = 16.3, 4 df, P < 0.01), *C. nattiensis* (Variance Ratio = 4.86, 4,16 df, P < 0.01) and C*. victoriae* (WS=44.8,4 df, P < 0.01).

Trial 3 - The overall treatment effect of different intensities of green LEDs was significant (WS = 118.5, 4 df, P < 0.001). Catches increased with intensity but were not significantly different at the two highest intensities. Significantly more *C. brevitarsis* were caught at all intensities tested than in the incandescent traps. These were between 46% and 142% of the incandescent intensity.

Discussion

Light trapping of *C. brevitarsis* was more efficient when incandescent globes were replaced with green LEDs. Attraction was also more effective as the intensity of the green light was increased, with catches at four intensities significantly greater than those with the incandescent light. An upper threshold of intensity suggested by the two highest intensities requires confirmation. Higher PFD's are possible utilising green LEDs than with the incandescent light, given the same power limitations (see Table 1). While trapping of *C. brevitarsis* was the major aim, trapping of seven other species would also be improved with the green LEDs. Specific trapping of some of these species could be maximised with blue LEDs.

Most predictions of the activity and spread of *C. brevitarsis* in NSW are based on population monitoring with light traps and are more dependent on the species occurrence than its density (Bishop *et al*. 1995a; Bishop *et al*. 2000). Larger catches in endemic or established areas where the occurrence of *C. brevitarsis* is not in question may therefore be of little value and the extra time taken to count increased numbers be unnecessary. Greatest benefit for monitoring use would be derived in areas and at times marginal for *C. brevitarsis,* ie. for first occurrences outside of endemic areas and at sites with low density. For example, Denman is used as part of the National Arbovirus Monitoring Program (Kirkland *et al*. 1995) which defines the distributions of the bluetongue and Akabane viruses and their vectors for overseas trade requirements. A normal monitoring period is two nights per month at this site. No *C. brevitarsis* were recorded by the incandescent trap in the first two experiments at Denman and a negative report was possible for that month. A trap with green LEDs would

have generated a positive report at the site at any time for that month and was 9 times more effective overall. Traps with green LEDs would therefore have a decided advantage for detecting *C. brevitarsis* in marginal areas and at apparently negative sites where virus has been detected serologically.

Further benefits could be derived where larger catches may be required for virus isolation from vectors, for experimental use of vectors with animals or for detecting vectors at key locations involved in the export of livestock (staging areas and ports). Colours with higher attraction could possibly be combined with other stimuli currently being investigated or used alone in trapping systems designed to control the insects (eg. with insecticides, electrified grids or large collection chambers), particularly where important livestock (stud or show animals) are in confined areas such as stables and stalls. Conversely, colours at the other end of the spectrum (red) could possibly provide sufficient light to allow work to continue with animals at night without increasing the attraction of *C. brevitarsis* and the chance that animals would be bitten and infected.

Only eight *Culicoides* species were trapped in these experiments. Other *Culicoides* vectors of the Akabane and bluetongue viruses also exist. These are mainly found in Australia's far north but *C. wadai* Kitaoka has now reached the northern coastal plains of NSW (AL Bishop, Unpublished data). Along with the viruses, these species currently occur within the recorded dispersive limits of *C. brevitarsis* but this may not always be the case. Coastal and estuarine species are often of human importance and improved trapping efficiency may aid in their study and control. Determination of responses to colour in a wider range of *Culicoides* species throughout Australia and overseas could therefore be an important adjunct to the understanding and control of these pest species and this could easily be carried out with LEDs in currently used light traps. The variations in response to coloured LEDs we observed may eventually give greater understanding of insect vision and species behaviour.

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Table 1. Specifications and characteristics of light sources tested for attracting species from the Genus *Culicoides* in Australia

Source	Nominal colour	Peak emission (400 – 700nm)	Light sources per trap	Rated maximum (mA actual per trap)	Total Actual Current* (mA per trap)	Material	Catalogue Number
Incandescent	White	700	1	150 (at 3.5V)	149 (at 3.9V)	Tungsten wire	VCH International G191 539752 3.5V, 0.15A
LED 'white'	White	460 (main) 570 (Secondary)	3	30	69.8	GalnN + fluorescent dye	Z 3981**
LED 'blue'	Blue	475	3	30	33.4	GalnN	Z 3905**
LED 'green'	Green	520	3	30	20.6	GalnN	Z 4013**
LED 'amber'	Yellow	595	5	50	156.0	GaAsP:N	Z 4033**
LED 'sunset red'	Red	640	3	50	43.0	AlGalnP	Z 4031**

* To give the same TFD as the incandescent source
** Dick Smith Electronics

Table 2. Predicted (back-transformed) means of *Culicoides* species in response to coloured light-emitting diodes (LEDs) arranged in spectral order in Trial 1 (**) and Trial 2 (*) in the Hunter Valley in 2002 and 2003 respectively and in relation to the standard incandescent globes. Means in columns with the same letter are not significantly different (P < 0.05).

Treatment	Species *C. brevitarsis***	*C.brevitarsis**	*C.austropalpalis**	*C.bundyensis**	*C.bunrooiensis**	*C.dycei**	*C.marksi**	*C.nattiensis**	*C.victoriae**
Red LED	17.0 e	0	5.1 d	0.1 b	0.5 b	2.2 c	1.4 d	0.3 b	0.3 d
Yellow LED	53.5 d	0.7 b	22.4 c	1.0 b	0.4 b	4.1 bc	2.5 cd	0.5 b	3.2 bc
Green LED	279.5 a	4.7 a	52.4 b	5.4 a	0.8 b	8.9 ab	6.4 b	3.1 a	10.6 a
Blue LED	173.5 b	1.3 b	119.0 a	3.9 a	2.2 a	12.7 a	22.2 a	4.9 a	6.3 ab
Incandescent	104.8 c	0.5 b	25.1 c	0.7 b	0.7 b	4.9 bc	3.3 c	0.5 b	1.5 cd
White LED	206.2 ab								

Table 3. Predicted (back-transformed) means of *Culicoides brevitarsis* responding to different intensities of green light-emitting diodes (LEDs) in relation to the intensity of standard incandescent globes. Means with the same letter are not significantly different (P < 0.05).

LED : Incandescent TFD Ratio (%)	*C. brevitarsis*
46%	152.4 c
96%	208.4 b
115%	301.1 a
142%	326.3 a
100% (Incandescent)	115.1 d

Appendix 2

Improving light trapping of *Culicoides* spp. in marginal vector and arbovirus distribution areas with LEDs

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Improving light trapping of *Culicoides* spp. in marginal vector and arbovirus distribution areas with LEDs

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Introduction

Several species from the genus *Culicoides* (Diptera: Ceratopogonidae) are vectors of viruses affecting native Australian animals (Standfast et al. 1984). *Culicoides brevitarsis* Kieffer is the main species responsible for the transmission of bluetongue and Akabane viruses to livestock (Muller et al. 1982). In Australia, light traps with incandescent globes (Dyce et al. 1971) are used to monitor the presence of *Culicoides* spp. as part of the National Arbovirus Monitoring Program (NAMP) (Kirkland et al. 1995). However, most trapping overseas is conducted using Ultraviolet (UV) (blacklight) traps. The Australian monitoring results designate areas free of vectors (and theoretically viruses) for the export of livestock to arbovirus sensitive trading countries. In most instances, the viruses that *C. brevitarsis* transmits are only detected within the dispersive range of the vector. However, occasionally Akabane activity has been detected in sentinel cattle herds in the apparent absence of the vector (PD Kirkland, personal communication). Questionable occurrences of virus, particularly at the margins of distribution, not only makes it difficult to understand the epidemiology of the virus but also makes it difficult to define acceptable virus free areas.

The objective of this study was to improve the trapping efficiency of light traps for *C. brevitarsis* and other *Culicoides* spp to overcome the problem above. The aim was to determine the responses of *Culicoides* spp. prevalent in New South Wales (NSW) to different colours in and beyond the visible spectrum by the use of light-emitting diodes (LEDs). Work completed in 2003 (Bishop et al. 2004) showed that several species, including *C. brevitarsis*, were attracted to green light from LEDs in standard traps and this improved trapping efficiency for *C. brevitarsis* by three to nine times. Catches of *C. brevitarsis* were also related to the intensity of the green LEDs. Traps with UV LEDs were included in experiments carried out in 2004 since several other species were more attracted to blue than to green light (Bishop et al. 2004) and in view of the common use of blacklight traps elsewhere. The effectiveness of the green LEDs was subsequently proposed for testing in different locations and under different ranges of vector density.

Methods

Responses to LEDs

The incandescent globes in standard light traps were replaced in treatment traps with different coloured LEDs and compared with the incandescent lights. The LEDs were standardised with the same quantum output as the incandescent lights using the method described in Bishop et al. (2004). The experiments were carried out at Tocal (32°38'S, 151°35'E) and Denman (32°20'S, 150°11'E) in the Hunter Valley, NSW in 2003 and 2004.
Traps were hung from 2m high "L" shaped frames ≈ 20m apart along the borders of ≈ 30ha paddocks containing cattle. Five treatments were used in each year. Red, yellow, green and blue LEDs and incandescent globes were used in 2003. The red LEDs were replaced by UV LEDs in 2004. The treatments were arranged in five randomised blocks. In each year, two experiments were carried out at each site and treatments were rearranged after each experiment. Collections were made into bottles containing 70% alcohol over one night. All *Culicoides* present were separated from other insects and identified to species by their wing patterns under x 10 magnification, counted and recorded.
Analysis of data.

The effect of treatment on counts of *Culicoides* spp. was modelled using a mixed linear regression approach (Searle 1971) which allowed for the separation of variance into fixed and random effects. Insect counts were $\log_e$ transformed to reduce heterogeneity. Analysis of the transformed counts was conducted using the REML directive in Genstat 5.4.1, Release 3. Treatment effects were examined for significance using Wald tests and treatment means compared using the least significant difference (LSD) technique at the 5% level and then back transformed to the original units. This was done separately for 2003 and 2004 data.

Field evaluation

A single green LED trap and an incandescent light trap were located at six sites in NSW, Northern Territory and NW Western Australia. Locations were selected at random from current NAMP sites and covered a wide range of geographical, climatic and vector density situations. Traps were placed 20 – 30m apart in paddocks containing cattle. Collections were made into bottles containing 70% alcohol. Samples were taken over two nights, once per month for five months (January to May 2004). Trap catches were sorted, identified, counted and recorded in their respective states/ territories and the data forwarded to the Gosford Horticultural Institute for analysis.

Only data from NSW will be presented here. Four extra traps were located in NSW at sites suspected as being marginal for *C. brevitarsis* and Akabane virus.
Analysis of data.

The data were regarded as repeated measurements over the 5 months and a linear mixed model which accounted for correlation over time and variance heterogeneity was used (Verbyla et al. 1999). A factor called zone was created with each of the10 sites classified as being located either inside or outside the *C.brevitarsis* endemic zone. Month was modelled as a fixed linear trend. Curvature around this line was modelled as a cubic smoothing spline.Tests of whether the linear time trend is the same for both treatments and both zones was also made. The fixed effects of treatment, zone, the linear trend with time and their interactions on *C.brevitarsis* count ($\log_e$ transformed) were tested using ASReml (Gilmour et al*.* 1998). The following additional sources of variationwere included in the model as random effects: spline(month), month, treatment x spline(month), zone x spline(month), site and site x month.

Results

Responses to LEDs

Treatment effects were highly significant for most of the species recorded. Comparisons of the results for *C. brevitarsis* in 2003 (Bishop et al. 2004) and with the inclusion of the UV LED in 2004 are made in Fig, 1. Response to the green LED was consistently higher ($P < 0.05$) than for the other treatments. Similar responses were recorded for *C. bundyensis* Lee & Reye, *C. nattaiensis* Lee & Reye and *C. victoriae* Macfie.

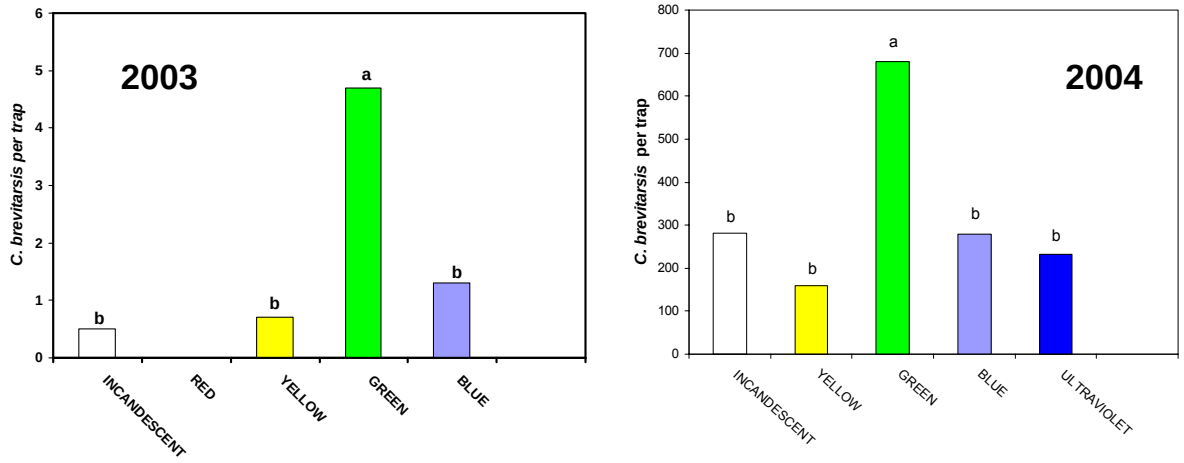

Figure 1. Response of *Culicoides brevitarsis* to different coloured LEDs. Columns with the same letter in a particular year are not significantly different (P < 0.05).

A comparisons of the results for *C.marksi* Lee & Reye in 2003 (Bishop et al. 2004) and with the inclusion of the UV LED in 2004 are made in Fig, 2. Response was to the blue LED in 2003. The main response was to UV in 2004 although more individuals were still found in the blue LED treatment than in the other treatments. Similar responses were recorded for *C.austropalpalis* Lee & Reye, *C.bunrooensis* Lee & Reye and *C. dycei* Lee & Reye.

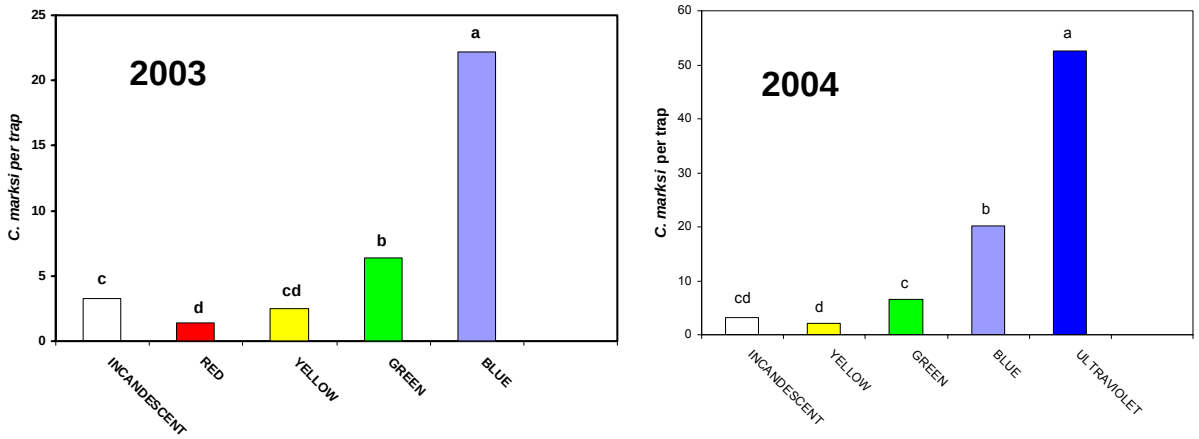

Figure 2. Response of *Culicoides marksi* to different coloured LEDs. Columns with the same letter in a particular year are not significantly different (P < 0.05).

Field evaluation

Counts of seven of the twelve species recorded were significantly higher in traps with green LEDs (Table 1). A suggestion that the other five species also exhibited a higher response to green LEDs could not be confirmed due to their low numbers. The analysis showed that there was significantly more *C. brevitarsis* in the endemic zone and that the superiority of the green LEDs was constant inside and outside of the endemic zone.

Table 1. Table of back-transformed treatment means for 12 species of *Culicoides* sampled at sites at different locations and with different densities of insects throughout NSW. Means in rows with different letters are significantly different at P < 0.05. * Collected at one site only.

Species	Incandescent	Green LEDs
C. austropalpalis	4.5b	46.5a
C. brevitarsis	83.4b	565.5a
C. bundyensis	0.9b	11.4a
C. bunrooensis	0	8.0
C. dycei	1.8b	13.2a
*C. henryi**	16.7	151.3
C. marksi	5.9b	61.8a
*C. marmoratus**	7.7	18.5
*C. narrabeenensis**	0	0.6
C. nattaiensis	0.1b	2.8a
C.victoriae	2.2b	8.8a
*C. wadai**	0	1.3

Discussion

It appears that *Culicoides* spp. can be divided into two distinct groups by their responses to different wavelengths in the visual and UV spectral ranges. The reasons for these differences are unclear. However, it seems logical that species should be examined individually to ensure that their trapping procedures are the most appropriate. *C. brevitarsis* is the main species affecting the Australian livestock industry. It is strongly attracted to green light in light traps. The efficiency of green LEDs for *C. brevitarsis* has been shown experimentally and its superiority over the currently used incandescent globes demonstrated under a range of field conditions. The importance of this superiority was highlighted in NSW by *C. brevitarsis* being caught in green LED traps and not in incandescent traps on 20% of sampling occasions. There were two locations where *C. brevitarsis* was caught in green LED traps and never caught in incandescent traps over a five month sampling period. These locations included one site (Tenterfield) where the transmission of Akabane virus had been detected in sentinel cattle and there was no official occurrence of the vector (PD Kirkland, personal communication). The other (Denman) was at the margin of Akabane activity recorded for 2003-2004 in the Hunter Valley.

All indications are that green LED traps have a distinct advantage over incandescent light traps for all species, even those which have a distinct preference for UV light. In practice, greatest advantage would be at the margins of distributions where the more efficient traps would provide better support for the designation of vector free areas. It could also be an advantage to have larger catches when these are required for research purposes (e.g. for virus isolation). More efficient traps could be a disadvantage where an increase in already large catches in endemic areas would be more difficult to sort and count. It was observed that many other insects from a range of orders were also caught with green LED traps. The increase in biomass was in the order of the increased efficiency of trapping for *C. brevitarsis*. While this made sorting of *Culicoides* more difficult, the response of these insects to specific ranges of wavelength in the visual spectrum could enhance their study. Ultraviolet traps were more efficient for some species but they attracted 14 times greater numbers of Lepidoptera than did the green LEDs making counting difficult and suggesting a needed for improved screening of the traps.

This study is part of a program that is evaluating the appropriateness of green LEDs prior to a recommendation for their adoption as part of the NAMP. It is hoped that the response of an even wider range of *Culicoides* spp. to LEDs can be determined in coastal and northern environments. This could be an important adjunct to the understanding and control of vector and nuisance species and the data from a greater diversity of species may help to clarify why such marked differences in response to wavelength exist within the genus.

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