



USDA Forest Service

OMB 0596-0217  
FS-1500-19

MODIFICATION OF GRANT OR AGREEMENT			PAGE OF PAGES
			1      31
1. U.S. FOREST SERVICE GRANT AGREEMENT NUMBER: 20-DG-11052021-229	2. RECIPIENT COOPERATOR GRANT OR AGREEMENT NUMBER, IF ANY: 30-2F-311142/3	3. MODIFICATION NUMBER: 002	
4. NAME ADDRESS OF U.S. FOREST SERVICE UNIT ADMINISTERING GRANT AGREEMENT (unit name, street, city, state, and zip + 4):  USDA Forest Service, State and Private Forestry Pacific Southwest Region (Region 5) 1323 Club Drive Vallejo, CA 94592-1110	5. NAME/ADDRESS OF U.S. FOREST SERVICE UNIT ADMINISTERING PROJECT ACTIVITY (unit name, street, city, state, and zip + 4):  Same as Block #4		
6. NAME ADDRESS OF RECIPIENT COOPERATOR (street, city, state, and zip + 4, county):  College of Natural & Applied Sciences University of Guam (UOG) UOG Station 303 University Drive Mangilao, GU 96913-1800	7. RECIPIENT COOPERATOR'S HHS SUB ACCOUNT NUMBER (For HHS payment use only):		
<b>8. PURPOSE OF MODIFICATION</b>			
CHECK ALL THAT APPLY:	This modification is issued pursuant to the modification provision in the grant/agreement referenced in item no. 1, above.		
<input type="checkbox"/>	CHANGE IN PERFORMANCE PERIOD:		
<input checked="" type="checkbox"/>	CHANGE IN FUNDING: Add federal funding \$24,418, cooperator match waived		
<input type="checkbox"/>	ADMINISTRATIVE CHANGES:		
<input type="checkbox"/>	OTHER (Specify type of modification):		
Except as provided herein, all terms and conditions of the Grant/Agreement referenced in 1, above, remain unchanged and in full force and effect.			
9. ADDITIONAL SPACE FOR DESCRIPTION OF MODIFICATION (add additional pages as needed):			
<b>10. ATTACHED DOCUMENTATION (Check all that apply):</b>			
<input type="checkbox"/>	Revised Scope of Work		
<input type="checkbox"/>	Revised Financial Plan		
<input checked="" type="checkbox"/>	Other: SF 424, SF 424A and narrative		
<b>11. SIGNATURES</b>			
AUTHORIZED REPRESENTATIVE: BY SIGNATURE BELOW, THE SIGNING PARTIES CERTIFY THAT THEY ARE THE OFFICIAL REPRESENTATIVES OF THEIR RESPECTIVE PARTIES AND AUTHORIZED TO ACT IN THEIR RESPECTIVE AREAS FOR MATTERS RELATED TO THE ABOVE-REFERENCED GRANT/AGREEMENT.			
11.A. UNIVERSITY OF GUAM SIGNATURE  	11.B. DATE SIGNED  1/14/14	11.C. USDA FOREST SERVICE SIGNATURE  	11.D. DATE SIGNED  
(Signature of Signatory Official)	(Signature of Signatory Official)		
11.E. NAME (type or print): LEE S. YUDIN	11.F. NAME (type or print): SHERRY HAZELHURST		
11.G. TITLE (type or print): Dean/Director, College of Natural & Applied Sciences, University of Guam	11.H. TITLE (type or print): Director, State & Private Forestry, Region 5		



USDA Forest Service

OMB 0596-0217  
FS-1500-19**12. G&A REVIEW****12.A. The authority and format of this modification have been reviewed and approved for signature by:****CONSTANCE ZIPPERER** Digitally signed by CONSTANCE ZIPPERER

Date: 2021.06.09 16:12:25 -07'00'

**CONSTANCE ZIPPERER**

USDA Forest Service Supervisory Grants Management Specialist

**12.B. DATE SIGNED****Burden Statement**

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<b>Application for Federal Assistance SF-424</b>			
*1. Type of Submission: <input type="checkbox"/> Preapplication <input type="checkbox"/> Application <input checked="" type="checkbox"/> Changed/Corrected Application	*2. Type of Application: <input type="checkbox"/> New <input type="checkbox"/> Continuation <input checked="" type="checkbox"/> Revision	* If Revision, select appropriate letter(s): A. Increase Award  *Other (Specify): _____	
* 3. Date Received: May 17, 2021		4. Applicant Identifier:	
5a. Federal Entity Identifier:		*5b. Federal Award Identifier: 20-DG-11052021-229	
<b>State Use Only:</b>			
6. Date Received by State:		7. State Application Identifier:	
<b>8. APPLICANT INFORMATION:</b>			
*a. Legal Name: University of Guam College of Natural and Applied Sciences			
*b. Employer/Taxpayer Identification Number (EIN/TIN): 98-0032933		*c. Organizational DUNS: 7799081510000	
<b>d. Address:</b>			
*Street 1: Agriculture and Life Sciences Building			
Street 2: 303 University Drive			
*City: Mangilao			
County/Parish: Guam			
*State: Guam			
Province: _____			
*Country: USA			
*Zip / Postal Code: 96923			
<b>e. Organizational Unit:</b>			
Department Name: College of Natural and Applied Sciences		Division Name:	
<b>f. Name and contact information of person to be contacted on matters involving this application:</b>			
Prefix: Dr.	*First Name: Aubrey		
Middle Name: _____			
*Last Name: Moore			
Suffix: _____			
Title: Entomologist			
Organizational Affiliation: University of Guam			
*Telephone Number: 1-671-686-5664		Fax Number:	

\*Email: aubrey.moore@triton.uog.edu

**Application for Federal Assistance SF-424**

**9. Type of Applicant 1: Select Applicant Type:**

H. Public/State Controlled Institution of Higher Learning

Type of Applicant 2: Select Applicant Type:

Type of Applicant 3: Select Applicant Type:

\*Other (Specify)

**\*10 Name of Federal Agency:**

USDA Forest Service

**11. Catalog of Federal Domestic Assistance Number:**

10.680

CFDA Title:

Cooperative Forest Health

**\*12 Funding Opportunity Number:**

\_\_\_\_\_

\*Title:

\_\_\_\_\_

**13. Competition Identification Number:**

\_\_\_\_\_

Title:

\_\_\_\_\_

**14. Areas Affected by Project (Cities, Counties, States, etc.):**

**\*15. Descriptive Title of Applicant's Project:**

Control of Little Fire Ant (LFA) and Coconut Rhinoceros Beetle (CRB) on Guam

**Application for Federal Assistance SF-424****16. Congressional Districts Of:**

\*a. Applicant:

\*b. Program/Project:

Attach an additional list of Program/Project Congressional Districts if needed.

**17. Proposed Project:**

\*a. Start Date: 06-01-2020

\*b. End Date: 12-31-2022

**18. Estimated Funding (\$):**

*a. Federal	\$ 215,941
*b. Applicant	\$ 0
*c. State	\$ 0
*d. Local	\$ 0
*e. Other	\$ 0
*f. Program Income	\$ 0
*g. TOTAL	\$ 215,941

**\*19. Is Application Subject to Review By State Under Executive Order 12372 Process?**

- a. This application was made available to the State under the Executive Order 12372 Process for review on \_\_\_\_\_
- b. Program is subject to E.O. 12372 but has not been selected by the State for review.
- c. Program is not covered by E.O. 12372.

**\*20. Is the Applicant Delinquent On Any Federal Debt? (If "Yes," provide explanation in attachment.)** Yes       No

If "Yes", provide explanation and attach.

21. \*By signing this application, I certify (1) to the statements contained in the list of certifications\*\* and (2) that the statements herein are true, complete and accurate to the best of my knowledge. I also provide the required assurances\*\* and agree to comply with any resulting terms if I accept an award. I am aware that any false, fictitious, or fraudulent statements or claims may subject me to criminal, civil, or administrative penalties. (U. S. Code, Title 218, Section 1001)

 \*\* I AGREE

\*\* The list of certifications and assurances, or an internet site where you may obtain this list, is contained in the announcement or agency specific instructions.

**Authorized Representative:**

Prefix: Dr. \*First Name: Lee

Middle Name: \_\_\_\_\_

\*Last Name: Yudin

Suffix:

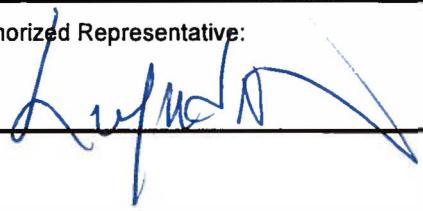
\*Title: Dean/Director

\*Telephone Number: 1-671-735-2000

Fax Number: 1-671-734-4600

\* Email: lyudin@triton.uog.edu

\*Signature of Authorized Representative:

A handwritten signature in blue ink, appearing to read "Linda N.", is written across the top of the box.

\*Date Signed: May 5, 2021

## BUDGET INFORMATION - Non-Construction Programs

OMB Approval No. 0348-0044

### SECTION A - BUDGET SUMMARY

Grant Program Function or Activity (a)	Catalog of Federal Domestic Assistance Number (b)	Estimated Unobligated Funds		New or Revised Budget		
		Federal (c)	Non-Federal (d)	Federal (e)	Non-Federal (f)	Total (g)
1. Forest Health Protection	10-680	\$	\$	\$ 215,941	\$	\$
2.						
3.						
4.						
5. Totals		\$	\$	\$ 215,941	\$	\$

### SECTION B - BUDGET CATEGORIES

6. Object Class Categories	GRANT PROGRAM, FUNCTION OR ACTIVITY				Total (5)
	(1)	(2)	(3)	(4)	
a. Personnel	\$	\$	\$ 156,450	\$	\$
b. Fringe Benefits			24,780		
c. Travel			4,000		
d. Equipment					
e. Supplies			2,545		
f. Contractual					
g. Construction					
h. Other			28,166		
i. Total Direct Charges (sum of 6a-6h)			215,941		
j. Indirect Charges			0		
k. TOTALS (sum of 6i-6j)	\$	\$	\$ 215,941	\$	\$
7. Program Income	\$	\$	\$	\$	\$

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Prescribed by OMB Circular A-102

### SECTION C - NON-FEDERAL RESOURCES

(a) Grant Program	(b) Applicant	(c) State	(d) Other Sources	(e) TOTALS	
8.	\$	\$	\$	\$	
9.					
10.					
11.					
12. TOTAL (sum of lines 8-11)	\$	\$	\$	\$	
<b>SECTION D - FORECASTED CASH NEEDS</b>					
	Total for 1st Year	1st Quarter	2nd Quarter	3rd Quarter	4th Quarter
13. Federal	\$ 100,000	\$ 25,000	\$ 25,000	\$ 25,000	\$ 25,000
14. Non-Federal					
15. TOTAL (sum of lines 13 and 14)	\$	\$	\$	\$	\$
<b>SECTION E - BUDGET ESTIMATES OF FEDERAL FUNDS NEEDED FOR BALANCE OF THE PROJECT</b>					
(a) Grant Program	FUTURE FUNDING PERIODS (Years)				
	(b) First	(c) Second	(d) Third	(e) Fourth	
16.	\$ 115,941	\$	\$	\$	
17.					
18.					
19.					
20. TOTAL (sum of lines 16-19)	\$	\$	\$	\$	
<b>SECTION F - OTHER BUDGET INFORMATION</b>					
21. Direct Charges: 215,941	22. Indirect Charges: 0				
23. Remarks:					

# **Control of Little Fire Ant (LFA) and Coconut Rhinoceros Beetle (CRB) on Guam**

Glenn Dulla PhD, Guam Department of Agriculture (LFA)

Aubrey Moore PhD, University of Guam (CRB)

May 5, 2020

Revised May 4, 2021

# 1. Combined Budget

Item	Cost(UOG)	Cost(GDOA)	Total
Personnel	\$83,463	\$72,987	\$156,450
Benefits	\$19,196	\$5,584	\$24,780
Travel	\$4,000	\$0	\$4,000
Supplies	\$0	\$2,545	\$2,545
<b>SUBTOTAL</b>	<b>\$106,659</b>	<b>\$81,116</b>	<b>\$187,775</b>
Administrative fee	\$15,999	\$12,167	\$28,166
<b>TOTAL</b>	<b>\$122,658</b>	<b>\$93,283</b>	<b>\$215,941</b>

**Personnel (UOG)** includes salary for an insect pathologist (Dr. James Grasela, 1 FTE, \$64,000) and salary for a technician \$19,463. Total=\$83,463.

**Benefits (UOG)** benefits calculated at 23%\*\$83,463=\$19,196.

**Personnel (GDOA)** Research Associate I, (fulltime, \$20.34/hour, 2080hrs)=\$42,307; Research Assistant III (fulltime or 2x halftime, \$14.75/hour, 2080hrs)=\$30,680. Total=\$72,987.

**Benefits (GDOA)** Social Security and Medicare (7.65%\*\$72,987)=\$5,584.

**Travel (UOG)** includes airfare and other relocation expenses for Dr. Grasela who resides in Missouri.

**Supplies (GDOA)** Fuel (7,488miles, work truck, 20MPG, \$4.50/gallon)=\$1,685; Survey Supplies (peanut butter, chopsticks, fluorescent tape/flags, ziplock bags)=\$500; Wrist Garmin GPS units (2x \$180)=\$360. Total=\$2,545.

**Administrative fee (UOG and GDOA)** 15% of direct costs is charged by the Research Corporation of the University of Guam for services provided.

## **2. Little Fire Ant Management**

Please see next page.

## GRANT NARRATIVE FORMAT

1. S&PF PROGRAM – Forest Health Protection
2. STATE AGENCY NAME – University of Guam, College of Applied and Natural Science
3. PROJECT COORDINATOR(S) –

Glenn Dulla, Ph.D.  
Research Affiliate, University of Guam  
Invasive Species Coordinator, Guam Department of Agriculture  
USDA Plant Inspection Station  
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4. STATEMENT OF NEED/PURPOSE –

Little fire ant (LFA), *Wasmannia auropunctata*, was detected in November 2011 in Yigo, a northern village of Guam, at the side of a green waste repository. Subsequent LFA surveys throughout Guam found it to be established at over 35 widely dispersed sites. Previous LFA infestations in the Pacific Basin include those of the five major islands of Hawaii, New Caledonia, French Polynesia and Northern Queensland, Australia. The devastating effects of LFA on agriculture and forest ecosystems observed in these other Pacific jurisdictions are being repeated on Guam and may potentially occur on other Micronesian islands that are LFA-free. LFA's spread into and throughout Guam is due to human transport of plant related material.

The Guam Invasive Species Management Plan identifies LFA as a Priority Invasive Species (p.15) and recommendations “to seek grants and other mechanisms to provide the LFA working group with the needed tools and resources to continue control efforts and subsequently the eventual eradication of the LFA from Guam” (p.35). Additionally, the Regional Biosecurity Plan for Micronesia and Hawaii (Attachment L, Guam, p.L-12) recommends to “Improve Post Border Biosecurity” through the high priority action item to “Determine the extent of Little Fire Ant infestation and then to manage and/or eradicate this species”.

The Forestry Division of Guam Department of Agriculture (GDOAG) maintains a 70-acre conservation acacia forest in Santa Rita, a southern village of Guam. The Cotal Conservation

Forest is a long-term reforestation project to return nutrients to the soil, replace acacia trees with native flora such as the endangered *Serianthes nelsonii* and ultimately reintroduce native fauna such as the endangered Guam Rail. The forest is directly bordered on the west by residential homes. Further southwest is Department of Defense (DoD) land on which the US Naval Base Guam Munitions site is situated. LFA was identified in the residential neighborhood in 2015 and subsequent surveys by the University of Guam (UOG) showed rampant spread throughout the area. The Center for Environmental Management of Military Lands also conducted surveys in 2017 to delineate LFA infestation at the Naval Base Guam Munitions site. LFA were found at 22 sites along the northeast border of the munitions site. A preliminary survey by the DOAG-Biosecurity division in 2017 identified LFA at 33 points along walking trails throughout the forest and along the residential border. Although unconfirmed, LFA infestation is believed to have originated in the residential area from contaminated backfill soil. UOG personnel, lead Dr. Ross Miller, began survey and treatment in 6/2018 and ended in 2/2019 after depletion of funds, Figure 1. DOAG resumed treatment of ~25-acre infestation in 6/2019 as funds and personnel were made available. Survey results from 2/2020 (Figure 2) show significant progress towards eradication, yet a substantial amount of area still infested despite approximately two years of treatment.

This project proposes to continue LFA eradication efforts in the conservation forest and establish and maintain a perimeter/buffer zone to prevent further infestation from surrounding areas. Currently, an ongoing joint effort by DOAG (funded by Department of the Interior) and UOG (funded by DoD) is working to eradicate LFA in the forest. Techniques developed by the University of Hawaii-Hilo Ant Lab, that have proven effective on Hawaii, are transferrable to Guam. We utilize these methods and adapted them for aerial drone use. Application drones allow for faster and safer treatment of technically challenging areas such the forest canopy or dense impassable jungle. Significant resources have been invested in reclamation of this forest and LFA infestation compromises future restoration work. If successful, the eradication will also reduce the threat of DoD land infestation posed by LFA in the neighboring forest.

This project addresses Guam State-Wide Assessment and Resource Strategy (SWARS), Strategy 2: Protect, Conserve and Restore Forests On State, Private, And Other Nonmilitary Lands (p.103). The eradication of LFA will increase forest resiliency and conservation can be achieved by “reduce(ing) stressors to existing forest through enhancement of current stands” (p.108)



Figure 1. Initial delineation of LFA in the Cotal Conservation Forest section south of Cross Island Rd. on 6/2018. Red dots denote presence of LFA and green dots denote no LFA. Broken line represents buffer area proposed in objective 2. Data collected by UOG, edited by G.Dulla.

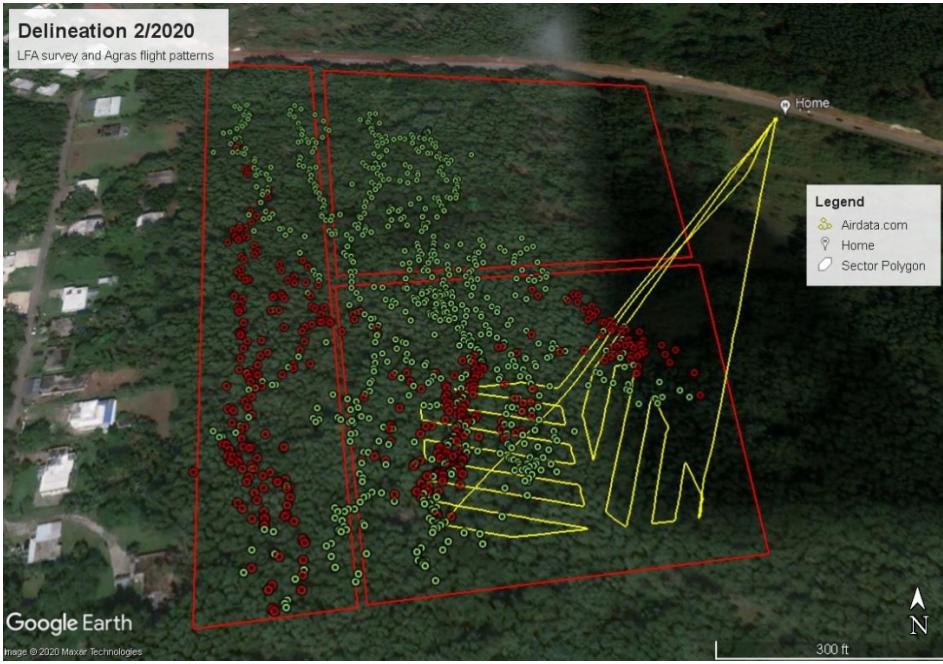


Figure 1. Delineation of LFA in the Cotal Conservation Forest section south of Cross Island Rd. on 2/2020. Red dots denote presence of LFA and green dots denote no LFA. Yellow lines are examples of aerial treatment patterns utilizing drone dispersal. Data collected by DOAG.

## 5. GOALS –

The project's overall goal is to eradicate LFA from the 70-acre conservation forest in Santa Rita, Guam and develop the capacity within DOAG to treat LFA on a large scale with limited

personnel. This is an ongoing project to eradicate LFA from the Cotal conservation forest with working objectives to clear all LFA in the forest and prevent re-infestation through a monitored buffer and/or eradicate LFA from the adjoining residences. It is anticipated that the Biosecurity and Forestry Divisions will maintain these after the end of the funding period.

## 6. OBJECTIVES –

The following objectives address Guam SWARS, Strategy 2: Protect, Conserve and Restore Forests On State, Private, And Other Nonmilitary Lands (p.103). The eradication of LFA will increase forest resiliency (p.108) and conservation can be achieved by “reduce(ing) stressors to existing forest through enhancement of current stands.”

1. Eradication of LFA from forest
2. Establish buffer zone around forest to prevent re-infestation
3. Provide training workshops and direct assistance for neighboring residents on LFA recognition, prevention of infestations, and treatment of LFA.  
-This objective addresses the “Next Step and action: Identify a short list of likely landowners that would be willing to participate in a forest protection program”. (Guam SWARS, p.111)

## 7. SPECIFIC ACTIVITIES –

1. Eradication of LFA from forest

Eradication of LFA from the forest requires a combination of surveying and pesticide treatment. Current routines will be maintained for the extension of this management project which includes monthly surveillance of targeted infested areas and quarterly surveillance of the ~25-acre portion of forest initially delineated and infested with LFA. Manual treatment with Tango (liquid pesticide-bait mixture@ up to 4-gallons/acre) and Siesta/Probait (granular pesticide@ 1.5lb/acre) is focused on the 6.5-acre section bordering the residential homes. Drone pesticide dispersal (granules @1.5lbs/acre: Amdro, MaxForce, Advion, Extinguish/ liquid: Tango) is utilized on the southeastern 7.2-acre sector where terrain make treatment difficult and dangerous. Treatment of the 7.9-acre northeast sector will cease on 7/2020 but continued surveying will occur quarterly.

- Progress will be measured in the reduction of LFA infested survey points or area throughout the project period.

2. Establish buffer zone around forest to prevent re-infestation

The border between the forest and the residential neighborhood is currently heavily infested with LFA. Treatment is ongoing with slow but significant progress. Once this 6.5-acre area is cleared

of LFA, regular monthly surveying will continue along a 380m long by 5m wide path along the border. Treatment will be done as needed.

- Progress will be measured in the reduction of LFA infested survey points or area throughout the project period and regularity of monitoring along buffer.

3. Provide training workshops and direct assistance for neighboring residents on LFA recognition, prevention of infestations, and treatment of LFA.

To treat the root cause of the LFA infestation and potentially eliminate the perpetual need to survey and treat the buffer zone, this project proposes to teach the residents how to treat LFA on their property. Previous work has developed a residential workshop for LFA detection and treatment. There are 16 homes along the forest border and other willing residents will be offered the workshop. DOAG will provide treatment supplies, manage treatment schedules and regularly follow-up with resident households to ensure completion. Surveys will be completed by DOAG personnel.

- Success will be measured in the reduction of positive LFA survey points or area throughout the project period and regularity of resident treatment.

All activities above will be led by Ashley Toves, the current Research Associate I/field supervisor. Treatment, surveys and outreach are performed by the project coordinator, field supervisor and one or two field technicians. Grant administration and funds management will be managed by the Research Corporation of the University of Guam (RCUOG). RCUOG policies align with CRF 200 procurement services regarding micro purchase, small purchase, and competitive bidding thresholds.

#### 8. KEY PERSONNEL –

Ashley Toves, Research Associate I, hired 6/2019.

- B.S. in Biology, Guam EPA Core pesticide certification and FAA small UAS Remote Pilot Certificate

- Current duties include fieldwork (treatment and surveys) supervision, data management, supplies procurement, human resources, website editing and drone operation.

Trevor Boykin, Research Assistant III, hired 11/2019.

- College coursework in Life Sciences, Guam EPA Core pesticide certification

- Current duties include fieldwork (treatment and surveys), outreach and education, and drone operation under supervision of licensed pilots.

#### 9. RESPONSIBILITIES – No special responsibilities are required of the Forest Service or others.

#### 10. MONITORING & EVALUATION –

The project coordinator is responsible for monitoring and reporting project activity. Daily activity progress will be tracked in weekly activity reports by paid employees and reviewed by project coordinator. Quarterly survey results will be mapped and the reduction of LFA positive points and infested area will be quantified. Bi-annual reports, including objective progress narratives and standard federal financial forms, will be provided to USDA-FS 30 days after reporting period.

Consultation and project review by DOAG-Forestry and USDA-FS will determine if goals and Guam SWARS #2 is adequately addressed.

### **3. Coconut Rhinoceros Beetle Biological Control**

Please see next page.

If you want to use active hyperlinks in the attached document, please download from

<https://github.com/aubreymoore/2020-FS-CRB-biocontrol-project/blob/master/combined-proposal.pdf>

Grant Proposal: USDA Forest Service FY2020

# **Establishment of Self-sustaining Biological Control of Coconut Rhinoceros Beetle Biotype G**

Aubrey Moore PhD

University of Guam College of Natural and Applied Sciences

May 5, 2020

Revised May 4, 2021

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# **1. Project Description (revised May 4, 2021)**

This grant proposal is a request for funds to extend employment of an insect pathologist, Dr. James Grasela, for one year to work on the Guam CRB Biological Control Project, a multiyear project partially funded by USDA-APHIS and Department of the Interior Office of Insular Affairs. Additional funds, if approved, will be used to hire a project technician whose primary task will be maintaining a CRB rearing facility to provide beetles for bioassays and auto-dissemination.

The specialized knowledge and skills of an insect pathologist are essential to success of this project. Dr. Grasela has identified two biological control agent candidates for CRB-G (both are isolated of *Oryctes* nudivirus (OrNV)). If funded, the proposed grant will support the insect pathologist's participation in the following activities.

**Objective 1: Survey to Determine Background OrNV Incidence** A pre-release survey will be performed to measure the incidence of OrNV in the Guam CRB-G population using PCR.

**Objective 2: Establish Sustainable CRB-G Biocontrol by Autodissemination of OrNV** OrNV isolates which have been identified as biological control agents will be propagated *in vivo* and introduced into the Guam CRB-G population via auto-dissemination under conditions specified by an existing USDA-APHIS permit.

**Objective 3: Establish Island-wide Monitoring Systems for CRB and Coconut Palm Health** Incidence of OrNV infection in beetles caught by pheromone traps will be monitored using PCR.

## **1.1. Background**

**The coconut rhinoceros beetle, *Oryctes rhinoceros*, is a major pest of coconut palm, oil palm and other palm species.** Palms are damaged when adult beetles bore into the crowns of palms to feed on sap. Tree mortality occurs when beetles destroy the growing tip (meristem). Immature beetles (grubs) do no damage. They feed on dead, decaying vegetation in breeding sites. Preferred breeding sites are dead, standing coconut stems, and piles of decaying vegetation such those left behind by typhoons or after replanting of oil palm plantations. If a CRB population outbreak is not suppressed, it is possible for initiation of a positive feed-back cycle where adult beetles kill massive numbers of palms, thereby generating more food for even more grubs which turn into adults which kill even more palms. An outbreak following this scenario occurred in the Palau Islands during the late 1940s resulting in about 50% coconut palms being killed by CRB throughout the archipelago and 100% mortality on some of the smaller islands (Gressitt 1953). A similar outbreak, initiated by Typhoon Dolphin (2015), is currently impacting Guam.

**Following 40 years of no geographical range expansion, CRB is again “on the move” in the Pacific.** CRB was recently detected for the first time at several Pacific Island locations including Saipan (2006), Guam (2007), Port Moresby, Papua New Guinea (2010), Oahu, Hawaii (2013), Honiara, Solomon Islands (2015), Rota, CNMI (2017), and Aguiguan, CNMI (2019).

Eradication of CRB has been attempted many times but is extremely difficult, having been achieved only once, on Niuatoputapu (formerly known as Keppel Island), a tiny island belonging to Tonga, with an area of only 16 km<sup>2</sup> (3% the area of Guam) (Catley 1969).

Failing eradication, the usual response to CRB infestations during the second half of the 20th century was introduction of *Oryctes* nudivirus (OrNV), the biological control agent of choice for this pest Jackson 2009 . OrNV attacks only CRB, typically reducing damage by up to 90% with population suppression lasting indefinitely (Bedford 2013). OrNV is auto-disseminated, meaning the pathogen is carried between feeding and breeding sites by CRB adults. Like many biocontrol agents, OrNV is density-dependent, working best at high population densities. After release, OrNV sustains itself within the CRB population, limiting damage to very low levels (See Appendix A.1: Self-sustaining biological control of CRB in Fiji using OrNV).

**Current invasions of Pacific Islands by CRB involve a new invasive biotype that has escaped from biological control by OrNV.** Discovery of OrNV nudivirus in the 1960s enabled the successful management of CRB populations in Pacific Island Countries (Huger 2005). Augmentative release of OrNV continues to be an important mechanism for CRB management in both coconut and oil palm growing regions. For about 40 years after adoption of this biocontrol strategy, no new outbreaks of CRB were reported from uninfested palm growing islands in the Pacific ensuring continuity of palm based village economies. However, the situation has recently changed. For the first time in 40 years, CRB invasion into completely new areas has been reported. Additionally, Pacific areas with established CRB populations (e.g. Palau) have reported increased severity and frequency of CRB damage. Common to all these areas is the high incidence of severe palm damage by beetles not seen since the introduction of OrNV.

Initial attempts to introduce OrNV into the Guam CRB population were unexpectedly unsuccessful, raising the possibility that the population that invaded Guam is tolerant or resistant to the commonly applied OrNV isolates. Subsequent DNA analysis showed that the Guam population is genetically different from other populations in the region. On the basis of distinct genetics and tolerance to currently available OrNV isolates, the Guam population has been designated a new biotype, CRB-Guam (CRB-G) (Sean David Goldie Marshall et al. 2015; Sean D. G. Marshall et al. 2017).

DNA analysis from an ongoing survey has detected the CRB-G biotype in Guam, Rota, Hawaii, Palau, Port Moresby (PNG) and Honiara (Solomon Islands). Thus, current invasions in the Pacific involve the CRB-Guam biotype and it is expected that these populations are tolerant isolates of OrNV previously used as biocontrol agents. However, Recent work has identified OrNV isolates which are new biocontrol candidates for CRB-G. (See the **Recent progress** section below [1.1].)

**Uncontrolled CRB-G outbreaks on islands may kill most palms within a few years and risk of accidental spread to other islands is high.** A worse case scenario for a CRB infestation may be triggered by a massive outbreak of adult CRB emerging from abundant breeding sites made by large amounts of decaying vegetation left in the wake of a typhoon, from large scale land clearing or large environmental destruction during a war. The current uncontrolled outbreak on Guam was initiated by Typhoon Dolphin which visited Guam in May, 2015. Massive amounts of decaying vegetation left in the wake of this storm provided abundant CRB breeding sites. Very high feeding activity by adults emerging from these breeding sites killed mature coconut palms, leaving standing dead coconut trunks which became ideal breeding sites for subsequent generations of beetles.

During a severe CRB outbreak, there will be an increased risk of further spread to uninfested islands throughout the Pacific. Palms are important on Pacific Islands for various reasons: as a cash crop for nuts, oil and lumber, as an ornamental tree appreciated by residents and tourists. On some of the smaller, more traditional islands, coconut palm is referred to as *the tree of life*. On these islands, this species is an essential natural resource providing income, housing, food, oil, soap, clothing, mats, baskets, and other containers. The smaller, poorer Pacific islands will suffer the most if spread of CRB-Guam cannot be controlled. If CRB-G infests islands and atolls where the coconut palm as the *tree of life*, islanders may have to migrate to larger population centers.

**Recommended response to CRB-G invasions.** Entomologists working on the CRB-G problem agree that the most feasible way to prevent massive palm mortality during outbreaks is establishment of biological control using an isolate of OrNV which is highly pathogenic to CRB-G (Jackson 2015; Vaqalo et al. 2015; Secretariat of the Pacific Community 2017).

The consensus among Pacific-based entomologists is that the most feasible way to stop massive palm mortality during CRB-G outbreaks is to find a find and release a have met several times to plan a response to CRB-G invasions. In a special meeting on CRB-G at the XXVth International Congress of Entomology , the following strategic plan was suggested:

A coordinated regional project should be organized and adequately staffed and funded to accomplish 3 objectives:

1. Survey CRB populations throughout the Asian/Pacific region to delimit the geographical distribution of CRB-G and identify its centre of origin.

2. Survey CRB-G populations from the centre of origin to find isolate(s) of OrNV (or other pathogens) that are highly pathogenic for the CRB-G biotype.
3. Implement *in vivo* or *in vitro* propagation of selected OrNV isolates for auto-dissemination on islands infested with CRB-G.

The CRB-G problem is not limited to American-affiliated islands. Attempts to find financial support for a well-coordinated Pacific-wide response to this problem have failed. However, there is an *ad hoc* international community of entomologists, the *CRB-G Action Group*, which meets annually (Table 1.1).

Table 1: Meetings of the CRB-G Action Group

2015	Pacific Entomology Conference, Honolulu, HI, USA
2016	International Congress of Entomology, Orlando, USA
2017	Japanese Society for Insect Pathology, Tokyo, Japan
2018	Society for Invertebrate Pathology, Gold Coast, Australia
2019	XIX International Plant Protection Congress, Hyderabad, India
2020	(tentative): Pacific Plant Protection Organization, Guam

**Recent progress.** Recent work at the University of Guam, supported by grants from DOI-OIA and USDA-APHIS, has produced encouraging results (See Appendix B: *DOI-OIA Grant D17AP00107 Progress Report 4* for details):

- Laboratory tests indicate that OrNV from two sources can be considered as potential biocontrol agents CRB-G: OrNV isolate V23B maintained in insect tissue culture by AgResearch New Zealand and OrNV isolate UOGTW from bodies of CRB collected in Taiwan by the University of Guam CRB-G Biocontrol Project. Further laboratory testing of these virus samples is underway.
- PCR tests of recently collected CRB-G adults on Guam indicate presence of OrNV in this population. This virus could be from OrNV autodissemination earlier in the Guam CRB project or from fortuitous introduction.

## 1.2. Statement of Need

This proposal is, in part, a response to an urgent need expressed by Micronesian Island leaders in the 22nd Micronesian Islands Forum Communique (Appendix C):

In recognition of the urgent need for a Pacific-wide project to find an effective biological control agent for the CRB, the Leaders instructed RISC to seek financial support for such a project, to be conducted with partners at the University of Guam, the Secretariat of the Pacific Community, New Zealand, the USDA and others, as appropriate.

In addition to loss of coconut as ornamental plants and an emergency food supply, the uncontrolled CRB-G outbreak on Guam is a major environmental disaster rivaling that caused by the brown treesnake (BTS). BTS killed the birds in Guam's forests. CRB-G is now killing the trees. A 2002 US Forest Survey reported that the three most populous trees in Guam's forests were *Cycas micronesica*, *Cocos nucifera* and *Heterospathe elata* accounting for 16%, 12% and 11% of total trees with a stem diameter of five inches or greater (Moore 2018). These three species, 39% of the trees in Guam's forests, are being attacked by CRB-G<sup>1</sup>. If the Guam CRB-G outbreak is not brought under control, the island's forest health will continue to decline, accidental export of CRB-G to other islands in the American Pacific (in addition to Oahu and Rota) will be inevitable and cascading impacts from loss of forests will cause damage to other systems (erosion leading to reef fouling for example).

Despite the severity of the Guam CRB-G problem, the US federal government has provided relatively little support for response efforts. USDA-APHIS granted Plant Protection Act funding for CRB-G work in Hawaii and Guam for several years (Moore 2020). However, a grant proposal requesting \$331,4904 for Guam from FY2020 PPA funding was unexpectedly rejected. A proposal requesting support (\$3.5M) for a cooperative CRB biocontrol project to be performed by North Carolina State University, Colorado State University and the University of Guam has been submitted to DOD's Strategic Environmental Research and Development Program (SERDP). If granted, this SERDP project will begin in the middle of 2021. Thus, **work towards mitigating the CRB-G problem on Guam during FY2020 is currently unfunded.**

The immediate challenge is to procure bridge funding to retain the Guam Biological Control Project's insect pathologist, Dr. James Grasela, for an additional year so that we can begin propagation and field release of the two OrNV biological control candidates he has identified. Specialized skills of an insect pathologist are essential to implementing successful biological control of CRB-G on Guam and elsewhere.

### 1.3. Goals and Objectives

Recent laboratory bioassays indicate that two recently tested OrNV isolates, V23B and UOGT are potential biocontrol candidates. There is also recent evidence indicating that there is already OrNV actively spreading within the Guam CRB-G population.

#### 1.3.1. Objective 1: Survey to Determine Background OrNV Incidence

CRB adults collected from breeding sites and pheromone traps throughout Guam will be tested for presence of OrNV using PCR. Laboratory bioassays will be performed on OrNV isolated from these beetles to evaluate potential for biological control.

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<sup>1</sup>Attack of *Cycas micronesica* by CRB-G adults is a recent discovery.

### **1.3.2. Objective 2: Establish Sustainable CRB-G Biocontrol by Autodissemination of OrNV**

OrNV biocontrol candidates will be propagated *in vivo* using established methods (Huger 2005) and released into the Guam CRB-G population by autodissemination. Autodissemination involves infecting healthy CRB adults with OrNV. These infected beetles are then released at points dispersed throughout the island where they vector disease to conspecifics. A permit for field release of OrNV on Guam has already been obtained from USDA-APHIS. Field releases on CEMML on DOD land and by UOG on the rest of Guam. All released beetles will be marked by etching unique numbers on their elytra using a computer-controlled laser engraving system already in use for this application at UOG.

Beetles for *in vivo* propagation and autodissemination will be field-collected from breeding sites and pheromone traps because this is far more efficient than rearing beetles in the lab at the current time. Impact of virus releases will be monitored using pheromone traps and a novel roadside video analysis system (see Subsection 1.3.3). A subset of beetles captured in traps will be used to estimate the virus infection rate. Concurrent with virus releases, we will continue to screen OrNV isolates to find candidate biocontrol agents.

### **1.3.3. Objective 3: Establish Island-wide Monitoring Systems for CRB and Coconut Palm Health**

The CRB-G outbreak on Guam is currently unmonitored on an island-wide basis. An island-wide pheromone trapping system, using about 1500 traps, was operated by the University of Guam from 2008 to 2014. This monitoring system was transferred to the Guam Department of Agriculture which abandoned the effort at the end of February, 2016. Currently, many coconut palms are being killed by CRB-G. But, in the absence of a monitoring system, we do not have an estimate of tree mortality or whether or not the damage is increasing or decreasing. Clearly, establishment of a monitoring system is necessary to evaluate success of the proposed biocontrol project, or any other mitigation efforts. We intend to re-establish island-wide trapping and to establish a sustainable roadside video survey which uses artificial intelligence to detect CRB damage in dash-cam videos.

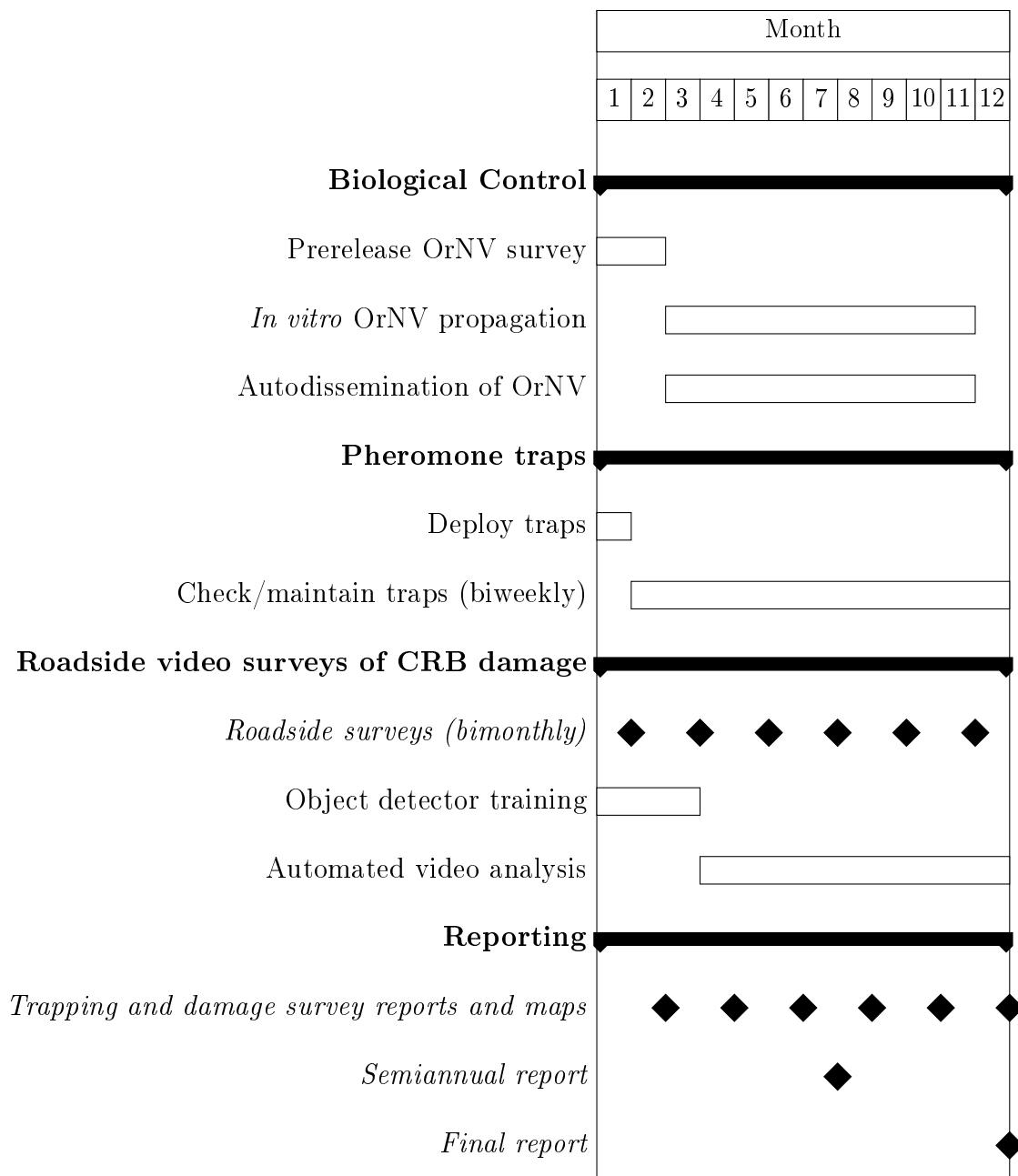
**Pheromone Traps** We plan to installed 150 CRB pheromone monitoring traps. These will be baited with oryzcalure and serviced semimonthly. These traps catch approximately equal numbers of males and females which remain alive in the traps for several weeks. Collected beetles will be used for autodissemination of virus and a subsample will be used for virus detection. Traps will be deployed at least 3 months prior to initiation of autodissemination.

A web database already exists for Guam CRB trap data and it is available for use by this project (URL: [mysql.guaminsects.net](http://mysql.guaminsects.net); database: **oryctes**; user: **readonlyguest**; password: **readonlypassword**; main tables: **trap** (2,265 records) and **trap\_visit** (89,114 records)).

**Roadside Surveys** Damage symptoms such as v-shaped cuts to fronds, bore holes, and dead standing coconut palm stems are readily observed during roadside surveys. Survey data will be collected on a smart-phone dash-cam app which georeferences each image. Initially, images of coconut palm damage by CRB-G will be detected, classified and tagged by a technician. When a large number of images have been tagged, these will be used to train an object detector. This work will result in a fully automated CRB damage detection and monitoring system which generates detection alerts and damage maps. This automated system will be useful as an early detection device for CRB. Roadside surveys on Guam will be performed bimonthly and the system will also be tested on Tinian, an island just north of Guam on which CRB has never been detected.

The envisioned system has already been successfully prototyped (See Supporting Technical Data 2). A custom object detector for CRB damage has been trained using the TensorFlow implementation of the Faster R-CNN Deep Learning model (Moore, unpublished).

## 1.4. Timeline



## 1.5. Potential Benefits

- This project will directly benefit Guam. Without implementation of effective biological control, it is likely that 50% or more coconut palms will be killed by CRB-G. In addition to loss of coconut as ornamental plants and an emergency food supply, the uncontrolled CRB-G outbreak on Guam is a major environmental disaster with 39% the trees in Guam's forests are at risk of attack by CRB-G. If the outbreak is not brought under control, the island's forest health will continue to decline, accidental export of CRB-G to other islands in the American Pacific (in addition to Oahu and Rota) will be inevitable and cascading impacts from loss of forests will cause damage to other systems (erosion leading to reef fouling for example).
- This project will indirectly benefit all other islands in Micronesia and other areas of the Pacific. With very high populations of CRB-G in Gaum risk of accidental introduction to other islands is extremely high. CRB-G has already been intercepted twice on Saipan, it is established on Rota and it has been detected on Aguiguan. If CRB-G infests smaller islands and atolls in the FSM and RMI where the coconut palm as the *tree of life*, islanders may have to migrate to larger population centers. If funded, the proposed project will be run under the *Open Science* concept. All data and analyses will be publicly shared on the internet in near-real time and samples of biocontrol candidates will be also be shared other Pacific islands battling CRB-G.
- It is expected that establishment of OrNV as an effective biological control agent will provide permanent self-sustaining population suppression of CRB-G. This is in contrast to temporary results from invasive species control programs which rely on population suppression using pesticide application, trapping or physical removal.
- Technology developed for automated roadside video surveys may be used for early detection of CRB on islands on which this pest has not yet been detected. Roadside videos may be of use for monitoring spread of invasive weeds such as Mexican creeper, *Antigonon leptopus*.

## A. Supporting Technical Data

### A.1. Self-sustaining Biological Control of CRB in Fiji Using OrNV

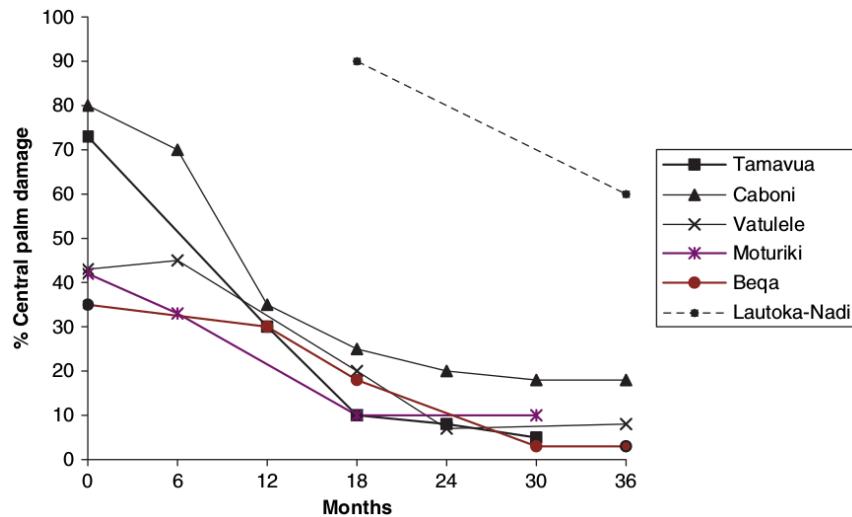


Figure 1: Reduction in coconut palm damage following release of *Oryctes rhinoceros* nudi-virus in Fiji. (Jackson 2009)

Reduction in palm damage recorded over 36 months after release of *Oryctes virus* on five sites in the Fiji Islands from 1970 to 1972. No virus was released in the Lautoka area where damage remained high 18 months after the start of the program but natural incidence of disease was recorded in the area after 36 months coinciding with a decline in visible damage. Population suppression of CRB by OrNV in Fiji was still in effect 35 years after virus introduction (Bedford 2013).

## A.2. Automated Monitoring of CRB Damage Using Roadside Video Surveys



Figure 2: Training an Object Detector to Locate Coconut Palms Damaged or Killed by Coconut Rhinoceros Beetle. <https://youtu.be/zzSorqcmt9U>.

Result of a first attempt to train an object detector (Faster R-CNN) to locate coconut trees killed or damaged by coconut rhinoceros beetle in a video. Dead palms are in red boxes, damaged palms are in green boxes. Not perfect, but it does serve as a proof of concept.