Capturing natural-colour 3D models of insects for species
discovery and diagnostics

Chuong V. Nguyen1*, David R. Lovell1, Matt Adcock1, John La Salle2,3
1 CSIRO Computational Informatics, Canberra, ACT, Australia
2 CSIRO Ecosystem Sciences, Canberra, ACT, Australia
3 Atlas of Living Australia, Canberra, ACT, Australia
* E-mail: chuong.nguyen@csiro.au

Abstract
Collections of biological specimens are fundamental to scientific understanding and characterization of
natural diversity—past, present and future. This paper presents a system for liberating useful information
from physical collections by bringing specimens into the digital domain so they can be more readily
shared, analyzed, annotated and compared. It focuses on insects and is strongly motivated by the
desire to accelerate and augment current practices in insect taxonomy which predominantly use text, 2D
diagrams and images to describe and characterize species. While these traditional kinds of descriptions
are informative and useful, they cannot cover insect specimens “from all angles” and precious specimens
are still exchanged between researchers and collections for this reason. Furthermore, insects can be
complex in structure and pose many challenges to computer vision systems. We present a new prototype
for a practical, cost-effective system of off-the-shelf components to acquire natural-colour 3D models of
insects from around 3mm to 30mm in length. (“Natural-colour” is used to contrast with “false-colour”,
i.e., colour generated from, or applied to, gray-scale data post-acquisition.) Colour images are captured
from different angles and focal depths using a digital single lens reflex (DSLR) camera rig and two-axis
turntable. These 2D images are processed into 3D reconstructions using software based on a visual hull
algorithm. The resulting models are compact (around 10 megabytes), afford excellent optical resolution,
and can be readily embedded into documents and web pages, as well as viewed on mobile devices. The
system is portable, safe, relatively affordable, and complements the sort of volumetric data that can be
acquired by computed tomography. This system provides a new way to augment the description and
documentation of insect species holotypes, reducing the need to handle or ship specimens. It opens up
new opportunities to collect data for research, education, art, entertainment, biodiversity assessment and
biosecurity control.

Introduction
Technology has a critical role to play in accelerating the understanding of biological diversity and, for
decades, scientists have strived to create accurate 3D duplicates of plants and animal specimens [1]. This
paper describes a novel method of using technology to liberate information about physical specimens by
bringing them into the digital domain as natural-colour 3D models—consistent with ideas and directions
articulated by several other authors [2–10]. In particular, the proof of concept system we present fits well
with the suggestion of Wheeler et al. [11] to “engineer and deploy a network of automated instruments
capable of rapidly creating 3D images of type specimens” as part of a larger strategy of dealing with the
massive backlog of insect types that are not yet digitized in any form. High resolution 3D scans, as well as
being useful as versatile replicas, also have the potential to act as a common frame of reference for other
data relating to the original insect such as annotations, auxiliary image collections, and measurements.
These additional aspects are vital for the ways taxonomists convey the various morphological characters
that distinguish a new species from those previously discovered.

Our work is focused on the digitization of insect species, building on research and development at
the Australian National Insect Collection (ANIC) which currently holds over 12 million specimens, and
is growing by around 100,000 specimens every year. Our mission is to enable high-quality 3D models of insects to be acquired quickly and cheaply, for ANIC to use as a component of its digitization strategy. Like many Natural History collections around the globe, the ANIC maintains many (thousands) Holotypes - each the single specimen of a species that is used to define the characteristic features of that species. Holotypes exist as a physical object carefully protected from damage through handling. Digital colour 3D models of sufficient detail will enable collections managers to liberate these precious specimens for the research work they are intended to fulfill.

Micro Computed Tomography (Micro CT) is currently a key method [12,13], able to create micron-accurate volumetric models of millimeter-scale objects and their internal structure. However, like recent 3D reconstructions from scanning electron microscope (SEM) micrographs [14,15]. Micro CT is unable to capture important information about the surface of the object: its natural colour. Exposure and reconstruction times can be long (tens of hours) and, as an X-ray imaging method, Micro CT generally demands special safety equipment. Current systems cost in the hundred-thousand dollar range and, while more compact desktop models are available, these are still not especially portable.

The inability of X-ray based methods for insect digitization to capture colour led us to consider image-based 3D reconstruction techniques as reviewed in [16,17]. These methods have been successfully applied to the reconstruction of 3D cityscapes and other (generally fairly simple) objects [18,20]. Some small biological specimens have been digitized [21,23] but the methods used do not specifically cater for the complex structures and challenging surface optical properties of insects. Human-in-the-loop approaches have been proposed for insect modeling [24] as have methods (limited to simple insect geometries) for inferring 3D insect shape from a single 2D image [25]. Experiments [26,27] with laser scanning systems like [28] have suggested that this approach has difficulties with the fine structures and the small scale of many insects, as well as reflective, transparent or iridescent surfaces.

One way to avoid these difficulties is to steer clear of 3D reconstruction altogether and simply present 2D images obtained from different viewing angles [29]. While this method of 3D visualization is popular for museum collections it does not provide the quantitative information (e.g., 3D morphology) needed to analyze and compare insect specimens. Furthermore large amounts of data are involved: many high-resolution images are needed to give a convincing illusion of looking at an actual 3D object. This makes smooth, realistic interaction difficult and precludes straightforward email exchange or embedding of the object data.

In summary, there is a lack of existing systems that could capture the 3D structure and surface optical properties of small, intricate insect specimens at sufficient resolution for ANIC and other collections to digitize, share, analyze and compare their holdings. The rest of the paper describes our prototype system and its operation, and how it has achieved these design objectives.

Materials and methods

Here we provide overviews of the digitization process and equipment. A video [30] as depicted in Figure 1 shows the main components of the system and the digitization process in action.

Process overview

In high-level terms, our system and work-flow involve three main steps (Figure 2):

Mounting: the physical specimen is pinned onto a pre-printed mat used later by the reconstruction software to estimate camera pose (viewing angle and position).

Acquisition: 2D images of the specimen are automatically acquired from different orientations (and focal depths for small insects). This step marks the transition from the physical to the digital domain.
Reconstruction: in which a 3D model is inferred from multiple 2D images. For small insects, this involves multi-focus image stacking before the general steps of extracting camera pose, shape and colour.

The system has two modes of acquisition, depending on the specimen size. Insects larger than 10mm are captured in normal-mode in which the depth of focus of the normal DSLR camera lens is enough to keep the whole specimen in focus at any viewing angle. Insects smaller than 10mm are captured in macro-mode using a high-magnification lens. Because of the shallow depth of focus of this lens, multiple images are captured at different distances from the specimen and processed into a single in-focus image.

Equipment overview
Figures 3 and 4 show normal- and macro-mode setups. The main hardware components of the system are:

- A two-axis turntable to present views of the specimen from different angles of rotation
- A macro-rail to vary the distance between the camera and specimen in macro-mode
- A camera and flash.
- Two laser pointers for specimen alignment
- A computer for 2D image processing and 3D reconstruction.

It is noted that in macro-mode our system uses a macro-rail to capture multi-focus images exactly at predefined depths, as opposed to refocusing the camera lens. A camera flash is needed to eliminate motion blur due to camera shutter’s vibration when capturing at high magnification.

To minimize cost and development time we sought to use off-the-shelf components wherever practicable. These are described in detail in the Supplementary Information.

Step 1: Mounting
Collections usually store and display insects larger than ~10mm by pinning them so that the insect’s long axis is horizontal and the pin vertical. Insects smaller than ~10mm are usually either pinned or glued in cards. This paper however focuses on pinned insects and issues arising from this mounting method. Pinning insects horizontally allows many insects to be stored in wide, flat display drawers but creates a few problems for our system:

- The pin becomes part of the 3D model and must be edited or segmented out in post-reconstruction
- Editing can often not fully remove evidence of the pin
- Images of the underside of the specimen can be difficult or impossible to capture, leading to an incomplete 3D model.

Re-pinning the insect so its long axis is vertical helps with image acquisition but risks damaging the specimen, including parts, such as genitalia, that are important for the identification of some species. For some specimens, these affected parts can be isolated through dissection and scanned separately.

After the specimen is pinned, the pin is glued to a small magnet (Figure 5C) that will hold the pin in position on the turntable. Next, a specially patterned mat (Figure 5B), required by the reconstruction software (3DSOM™), is attached to provide information about camera pose and position relative to the specimen. Generally the suitable size of the pattern is about one to two times the length of the insect to be scanned. Scanning smaller insects requires smaller patterns to be printed. Currently,
modern laser printers with 1200 dpi printing resolution can produce patterned mats as small as 5mm in diameter. Printing smaller patterns that are sharp enough to be recognised by the reconstruction software is currently a technical challenge.

Finally, the whole assembly is placed on the two-axis turntable and positioned (with the assistance of horizontal and vertical laser pointers) so the specimen is centered on the intersection of the axes of tilt and rotation. The lasers are aligned to the rotation axes of the turntable. A specimen is manually aligned to each of the laser beams such that each beam hits the centre of the insect’s body.

**Step 2: Acquisition**

This is the point at which the physical specimen enters the digital domain.

In essence, the acquisition process is about automatically obtaining 2D images of the specimen in different poses. As far as the relationship between the camera and specimen goes, this system has three degrees of freedom: pan, tilt and (in macro-mode) distance along the specimen-camera axis. With the specimen mounted at the intersection of the pan and tilt axes of the turntable, this amounts to rotating the turntable through a range of pan and tilt angles, capturing an image at each step (Figure 6A). In macro-mode there is an additional “inner loop” of translating the camera to acquire partially focused images at different distances from the specimen for later processing into a single image with all parts of the specimen fully in focus (Figure 6B).

There are many ways to automate the acquisition process. The desire to use off-the-shelf components led us to use the GigaPan™ Panorama Robot EPIC 100 [32] for mounting the specimen. The GigaPan™ is designed for mounting and controlling a camera—and this led to the GigaPan™ robot also acting as the acquisition controller. In other words, it is the turntable that triggers the macro-rail. The macro-rail moves and triggers the camera which triggers its flash and takes an image. The Supplementary Information contains more detail about this set-up.

In normal-mode, using rotation and axis tilt, the set-up captures 144 individual images. In macro-mode, the additional up to 31 images required at each step mean that the system can capture up to 4,464 separate images per specimen. Capturing more images is also possible.

**Step 3: Reconstruction**

The third and final step of the digitization process is where the 2D digital information acquired from a physical specimen is manipulated to produce a 3D digital model (Figure 7).

In macro-mode, the stack of partially focused images acquired at different specimen-camera distances must be combined into a single in-focus image for a given viewing angle. We used Helicon Focus [33] for this because of its ability to exploit multiple CPU cores. Single core open-source alternatives are available [34,35].

Armed with a set of in-focus 2D images of an object from different viewing angles, there are two main 3D reconstruction techniques that could be applied:

Visual hull (also known as volume carving) algorithms [36,37] project the silhouette of the object into a virtual volume at each viewing angle, carving away the volume outside the silhouette to leave a 3D visual hull which approximates the shape of the actual object. This approach does not recover concave surfaces, but photo-consistency can be used to correct this to an extent [38]. The extent of improvement by photo-consistency is limited for some insects due to strong specular reflections on the outer-surface and fine body structures such as legs, antennae, spikes and hairs.

Multi-view stereo algorithms generally rely on photo-consistency measures to identify the location of common features seen in different views [39,40] and can also incorporate silhouette information [41].
Both strategies are computationally intensive and the computational demands increase with reconstruction resolution. Image clustering and improved feature descriptors have been previously proposed to enable reconstructions to better exploit the very high image resolution produced by professional photography cameras.

Our initial investigations indicated that the visual-hull-based method could more accurately reconstruct some of the thin structures found in insects (e.g., legs, antennae, wings) and insect surfaces with strong specular reflections. 3DSOM™ was used to provide off-the-shelf visual-hull-based reconstruction as it produced the best quality output of the different approaches.

Figure 7 sets out the detail of the reconstruction process, including the extraction of the camera pose in each input image. 3DSOM™ initially estimates this information from the target pattern captured in the image and further refines these estimates during 3D reconstruction. Specimen silhouettes are extracted from input images. Once the 3D geometry of the specimen’s surface is reconstructed, texture colour is extracted from the images and added to the model. The resulting 3D model can then be exported to different formats—including HTML (with WebGL, Flash or Java), X3D, 3DS (AutoDesk), and STL (STereoLithography)—for subsequent viewing, analysis or embedding into documents. X3D is a convenient format as it is supported by popular 3D visualisation software, and a X3D file can include an embedded object or as XML inline in an HTML5 file for 3D web visualisation. InstantReality’s tool “aopt” can perform this conversion X3D to 3D-supported HTML automatically.

Results and discussion

Figure 8 shows high-resolution natural-colour 3D models of insects ranging from 3mm to 30mm in length. These 3D insect models are also available for interactive viewing at and can be downloaded at.

The smallest of these—the 3mm granary weevil—proved challenging to resolve due to an out-of-focus problem when its images were captured at 2× magnification. The 3D model of granary weevil was obtained from images captured in macro-mode, while 3D models of larger insects were obtained from images captured in normal-mode. The 3D visualisation of insect models is based on the open-source X3DOM framework which uses WebGL for plug-in-less display within a web browser (such as Firefox and Chrome). The file size of models, including 3D mesh and texture, depends on the desired visualisation quality and the complexity of the geometry and colour of the actual specimen. For the 3D models shown at, the file size ranges from 5 to 24 megabytes, with number of vertices from 80,000 to 130,000 and texture resolution from 4 to 16 megapixels.

Figure 9 illustrates the effectiveness of macro-mode image acquisition as compared to normal-mode image acquisition when applied to very small insects such as the granary-weevil. A Canon EF-65mm macro lens was employed in both cases. In normal-mode, a stencil with a 2mm hole had to be attached immediately in front of the camera (Figure 9A) to reduce the effective aperture and increase the depth of focus. In both cases a flash was used to mitigate the effects of wobble due to the camera shutter movement. With a flash, the exposure time of an image is effectively the very short duration of the flash when it triggers, and therefore it minimizes any motion blur. Flash energy in macro mode was of full power and in normal-mode (for the 2mm aperture) it was of full power. The results shown in Figure 9 clearly illustrate the improvements of macro-mode. The macro-mode model was reconstructed with multi-focus stacking of 31 images from each view, each captured with an F/8 lens aperture at increments of 0.25mm along the specimen-camera axis.

Figure 10 provides a qualitative comparison of a natural-colour 3D model obtained using our system and a Micro CT model of a different specimen of the same species. While the 5.7µm resolution Micro CT clearly captures more details of the surface geometry than our optical approach (including the missing antenna socket in inset A), there are features that it cannot resolve at these resolutions because they are to do with variation in the colour of the specimen (e.g., the compound eye in inset B). One option could be to develop ways to combine the strengths of both approaches: fertile ground for further research.
By convention, insect specimens are often mounted horizontally. However, this mounting orientation may not be ideal for 3D reconstruction. To investigate the effect of mounting orientation on reconstruction quality, we acquired images of a specimen mounted horizontally, then vertically (Figure 11). For the structure of that particular specimen, vertical mounting gave markedly better reconstruction of both geometry and colour, avoiding occlusions and capturing texture in more detail. Increasing the number and variety of poses by acquiring images at different tilt angles improved the reconstructions of both vertically and horizontally mounted insects (Figure 12). Even in this case, vertical mounting afforded more detail in geometry and colour. We therefore note that the best mounting orientation is specimen-dependent: visual hull reconstruction of geometry improves the more surface normals are captured in silhouette, while colour and texture improve the more surface normals are captured parallel to the camera viewing axis.

Further surface geometry issues arise as the structures of specimens become more complex. Wings, for example, can be especially challenging as shown in Figure 13(A-C) where self-occlusion causes poor reconstruction of the wings. Fortunately, additional informative views can be obtained to alleviate this problem (Figure 13D-F). Ideally, some of these additional views will be captured tangentially to the wing surface to ensure the reconstructed wings have the correct thickness.

We explored ways to achieve an informative mounting orientation even when the specimen cannot be re-pinned (e.g., when the specimen is too precious to handle, or the pin too firmly embedded to remove without certain damage). Previously, we mentioned that vertical orientation provides better quality than the horizontal orientation. However, re-pinning the specimen to have a vertical orientation causes damage, while keeping the horizontal orientation produces a lower-quality 3D model. To avoid this trade-off, the normally-pinned insect can be attached to a second pin (in this case using yellow Blu-Tack) so that the specimen is rotated on its long axis (Figure 14A). Then, the pins and the Blu-Tack need to be removed digitally to produce a clean final 3D model of the specimen. There are two methods to do this. The first method involves editing the Blu-Tack and mounting pins out of the set of 2D images (Figure 14B) during background removal prior to reconstruction. However, this method does not work well with image views where the pins and Blu-Tack occlude parts of the insect and the resulting reconstruction shows contaminated texture colour (Figure 14C). The second method is to keep the pins and Blu-Tack with the specimen during 3D reconstruction (Figure 14D and E) then remove them from the 3D model using a mesh editor. Overall, this second strategy produces the better result (Figure 14F).

In this paper, we have shown that high resolution, natural-colour 3D digitization system for insects and other small specimens can be implemented using readily available components with hardware and software cost under AUD8000. As well as being cost effective, the system produces digital 3D models that are fairly efficient in terms of the ratio of information to data. The file size of the 3D granary weevil model shown in Figure 9H is around 10 megabytes. It was reconstructed from 18 megapixel 2D JPEG images (2 – 4 megabytes/image) taken at 144 different angles and 31 different distances creating 10 – 17 gigabytes of 2D image data in all for a single specimen. By stacking each set of 31 multi-focus images into a single in-focus one, the image data is reduced approximately 20 times. By transforming this 2D data into a 3D model, the system further achieves a 30:1 compression of data. This level of compression enables useful information about the specimen to be exchanged via email, presented in web pages and embedded in 3D PDF documents.

This work raises a number of research challenges and opportunities for further improvement, including:

- Eliminating the need for the printed mat: 3DSOM™ requires this mat to estimate the camera pose of individual images. We have reached the lower size limit of what we can straightforwardly print and attach to specimens. Furthermore, the range of poses is limited to those in which the mat is viewable. There are reconstruction methods that do not need this kind of pattern to estimate camera pose (e.g., [56, 57]), relying instead on feature matching and bundle adjustment. However, the accuracy of these estimates depend strongly on the geometry of the specimen and other objects captured in the images.
• Detailed features, such as hairs and surface roughness, demand higher 2D image and 3D model resolution and a concomitant increase in the memory and computation needed to store and visualize the model. Our strategy is to leverage the high resolution 2D image corresponding to a particular pose of interest, reminiscent of the approach used in [29].

• Concave surfaces: current photo-consistency based methods to resolve concavities can be challenged by the specular reflective properties of many insects.

• Transparent wings and membranes pose challenges for acquisition, reconstruction, and for representation and rendering of the resulting 3D model.

• View- and lighting-dependent appearance such as iridescence or sub-surface light scattering is also difficult to capture, represent and render.

• 3D annotation standards, strategies and software are not yet as developed as 2D approaches. The ability to augment 3D models with additional information is important for taxonomy and other scientific ends, as well as engaging a broader range of end users.

Despite these future challenges, we believe that the proof-of-concept prototype presented in this paper demonstrates that natural-colour 3D model digitization is feasible and affordable enough for insect collections to implement and apply right now.

An initial investigation of the usefulness of 3D insect models, as described in the Supplementary Information section, showed that the quality of 3D insect models were good enough to provide sufficient information for species identification, and allow for easier specimen examination than the actual specimen being viewed under a microscope.

The specific usage scenarios for wider communities such as quarantine officer or educator. A quarantine officer can use 3D models of invasive insects while on duty to improve the speed and the accuracy of identification process. The challenges and possible solutions by using 3D models in quarantine control have been discussed in [58]. For educators, 3D models of insects can be used as rich education materials, allowing students to interact with insects without the need to access to fragile specimens.

Supplementary Information

Traditional insect mounting

Figure 15 shows insects mounted horizontally with a pin going through the body from the back. This mounting technique gives a strong hold on the insect body and facilitate specimen handling but provides limited access for 3D scanning.

Image processing and 3D reconstruction software

Figure 7 shows the overview of image processing and 3D reconstruction pipeline. Software used in this process are:

• Helicon Focus [33] for multi-focus image stacking to extend depth of field. There are alternative open-source software such as CombineZP [34] and Huggins & Enfuse [35]. However we found Helicon easier to use and able to exploit multi-core processing.

• 3DSOM™ software [31] for 3D reconstruction from multiple view images based on visual hull technique. The 3D reconstruction pipeline used by 3DSOM™ is described in [59]. There are other 3D reconstruction software including commercial software Agisoft Photo Scan [56]; and open-source softwares Bundler [43] and Patch-based Multi-view Stereo [42,43], and visual hull mesh software [44]. These two off-the-shelf software packages, together, cost around AUD1700 based on 2013 prices.
**Image acquisition equipment**

We use the following off-the-shelf components:

- **A GigaPan™ Panorama Robot EPIC 100**. Normally this device is used to control the tilt and pan of a camera to capture panorama images. In this project, we turn it side-ways and use it as a two-axis turntable to control the tilt and pan of the specimen to be imaged. The GigaPan™ Panorama Robot connects to a camera via a cable and triggers a camera to capture one or more images per rotation angle.

- **A Canon™ 600D camera or better**. It provides low-noise, high resolution images (18MP). It is highly customizable with several ports for external trigger input, flash trigger output, and a USB port for remote tethering. One problem with many DSLR cameras is that they rely on a mechanical shutter which can cause mechanical vibration and image blur. The image blur due to mechanical shutter vibration can be alleviated by using a camera flash triggered between shutter movement.

- **A Canon™ EF 100mm macro lens for normal-mode and Canon™ MP-65mm for macro mode**. The EF-100mm lens provides magnification of $1 \times$ or smaller, while the MP-E 65mm lens provides magnification from $1 \times$ to $5 \times$. These macro lenses also have much less optical distortion than some other lenses. A low-cost solution to make normal lens achieve higher magnification is to add a macro extension tube between the camera and the lens. However this often increases optical distortions. The depth of field of MP-65mm lens as function of magnification and aperture can be obtained from its user manual.

- **A Viltrox™ JY-670 macro ring flash**. This flash provides illumination and reduces exposure time. Short exposure time is critical for macro photography to reduce image blurring due to vibration from the camera’s mechanical shutter. Additionally, a Tronix SpeedFire flash power supply is used for fast flash charging.

- **A StackShot™ macro-rail**. This device enables us to capture high-magnification partially-focused images at predetermined depth intervals with high position accuracy. This macro-rail can run automatically to capture images along a single direction. The process of movement and image acquisition using macro-rail starts with a press of “Up” button on the rail’s control panel. To synchronize StackShot™ macro-rail and GigaPan™ robot, we built a circuit interface, shown in Figure 16, to convert the robot’s trigger signal to a press-button effect. The step size of the rail is set to be approximately 70% of the depth of field of the macro lens to allow for adjacent overlapping required for stacking multi-focus images.

- **Two 1mW laser pointers** for specimen alignment. The laser beams are used to locate the specimen at the center of rotation of the two-axis turntable so that the specimen stays at the same position while being rotated and imaged.

- **Aluminium frames** to hold the turntable, laser pointers, camera, and macro-rails in position.

- **Button magnets**, **plastic spacers**, **epoxy glue**, and **pins** to mount insect specimens.

The system does not need any special software for acquiring images. The EOS Utility software accompanying the camera is needed to remote-tether the camera and transfer images to a computer during image acquisition. Based on 2013 prices, we estimate the total hardware to cost around AUD3200 to AUD5600.

Flow charts of how the image acquisition system works in normal and macro-modes are shown in Figure 6. These charts are generalized so that they are hardware independent. “Pan rotation” refers to rotation around the vertical axis of the specimen and “tilt rotation” refers to rotation around horizontal axis (perpendicular to both the vertical axis and the camera-specimen axis).
Estimation of processing time

The reconstruction process described in this paper is a proof of concept and not yet optimized for speed and for large-scale digitization. A time estimation for each stage of the 3D reconstruction procedure for the insects shown in Figure 8 is provided in Table 1. The longhorn beetle, Christmas beetle and Amycterine ground weevil take relatively similar amount of time to obtain a 3D model. The sand wasp has increased reconstruction time due to the extra time required to manually correct the errors of background removal around the wings, and to refine camera pose during reconstruction (hairs significantly increase the time for pose refinement). The granary weevil, due to its small size, takes much more time to mount using a microscope, acquire multi-focus images and perform 3D reconstruction. It is often the case that the reconstruction stage is repeated several times to find and fix errors in background removal, or to iterate the refinement process until the result is acceptable. Therefore, the times provided in Table 1 should only be considered as indicative.

Informal feedback on using a 3D insect model for species identification

One possible use case for 3D insect models is species identification. In drafting this paper, we needed to fully identify the Christmas beetle shown in Figure 8. We asked CSIRO entomologist Mr Tom Weir to try to do so from the photograph, the 3D model, and finally the actual specimen. In addition to identifying the specimen as a male *Anoplognathus viriditarsus* from the 3D model alone, Mr Weir provided feedback which we summarize as:

- The single top-view photograph does not capture the key features needed to identify the species, just enough information to determine its genus. The features are located at different parts of the insects such as abdomen, head, mouth, claws, rear end, etc. Multiple images captured at particular angles are required to show all these features.

- The 3D model provides useful extra information and allows views from any angle. There are still missing features such as the mouth area and hairy surface on the head/nose due to resolution of geometries and texture colours. However the available features are roughly enough to identify its species. Examination and species identification would be facilitated if the texture information was provided more clearly, or high-resolution images of the features were attached to the model.

- The actual specimen obviously provides all the information needed but needs examination under a microscope for some features such as the mouth area and hair surface on the head. However, out-of-focus effect and other physical restrictions makes the use of microscope to view the actual specimen more cumbersome than to view the 3D model of specimen on a mobile device (such as iPad).

- Identification involves matching the given specimen to its corresponding specimen in the Australian National Insect Collection and comparing it to closely related species.

- For identification purposes, the natural-colour 3D model is much more helpful than the Micro CT 3D model. The better geometry accuracy of the Micro CT model does not assist in this instance.

- Higher resolution of the natural-colour 3D model is desirable to provide details of key identification features. This is particularly important for features, such as hairs, that never get included in the 3D models. One solution would be to attach high resolution photographs to locations of the key features.

- To make the 3D model useful for species identification, it is important to know in advance all key features and the identification procedure and level. Ideally, this information would be arranged in a way that directly supports fast, correct identification. 3D visualization and annotation could make
the identification procedure more obvious that the instructions currently provided in text and 2D illustrations.

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Author Contributions

Created, guided and informed the project: DRL JLS MA. Conceived and designed the methods and the experiments: CVN. Performed the experiments and analyzed the data: CVN. Wrote the paper: CVN DRL JLS MA.

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Figure Legends

Tables

Table 1. Time consumption estimation

| Insect                          | Mounting (min) | Acquisition (min) | Reconstruction (min) |
|---------------------------------|----------------|-------------------|----------------------|
| Longhorn beetle, Christmas beetle and ground weevil | 5              | 15                | 60                   |
| Sand Wasp                       | 5              | 15                | 120                  |
| Granary Weevil                  | 30             | 180               | 120                  |
Figure 1. 3D visualisation of a granary weevil on web as part of a video showing an overview of the 3D scanning process. Click on the figure, or go the link at [30] to view the video.

Figure 2. The three main steps to create a natural-colour 3D model of specimen. The steps are mounting the insect onto a pin, acquisition of 2D images of the specimen at different poses, then reconstruction of a single 3D model from those multiple images.
Figure 3. Connections (A) and hardware (B) for normal-mode image acquisition. The green sphere marks the center of rotation and mounting location of specimens. The turntable is the master device that triggers the camera after rotating to predetermined pan and tilt angles. Images can be stored in camera memory or transferred directly to the computer as they are acquired.

Figure 4. Connections (A) and hardware (B) for macro-mode image acquisition. The macro lens, macro ring flash and macro-rail are needed for capturing high-magnification and depth-extended images of small insects. At each rotation step, the turntable triggers the control box of macro-rail. The macro-rail then moves to a set of predetermined positions. At each position, the control box triggers the camera to capture an image.
Figure 5. Preparing insect specimen for scanning. A) Steps to prepare insect specimens for image capturing. B) A special mat target needs to be attached to a scanned specimen for 3DSOM\textsuperscript{TM} software to estimate of camera viewing position and angle. C) For a large insect such as this 30mm long Christmas beetle, the pin is glued to a $\varnothing 10$mm rare-earth disk magnet which is in turn attached to a $\varnothing 50$mm mat target. D) For a small insect such as this 3mm long granary weevil, the micro pin is glued to a $\varnothing 5$mm mat target. E) shows comparison in size of the two specimens.

Figure 6. Automated image acquisition process. A) Normal-mode. B) Macro-mode.
Figure 7. Image processing pipeline for normal-mode and macro-mode images. Macro-mode images require an extra step to stack each set of multi-focus images captured from the same viewing angle (but at different depth distances) into a single in-focus image.

Figure 8. Various 3D insect models. Click on individual figure or go to the link at [46] to interact with the 3D models or [47][54] to download. Top: 3D models of the insects with natural-colour texture, scaled to have similar sizes. They are A) a granary weevil (*Sitophilus granarius*), B) a sand wasp (*Bembix sp.*), C) a longhorn beetle (*Aridaeus thoracicus*), D) a Christmas beetle (*Anoplognathus viriditarsis*) and E) a mycterine ground weevil (*Gagatophorus draco*). Bottom: F) A photograph of the real insect specimens of the 3D models captured.
Figure 9. Comparison of natural-colour 3D reconstructions using (A) a small aperture and (B) a F/8 aperture with multi-focus image stacking. A) shows an extra mask with a 2mm hole put in front of the lens to extend depth of focus as compared to B) an F/8 lens aperture. C) the resulting images captured at the same angle by small aperture. D) multi-focus image stacking from 31 partial-focus images captured at distances 0.25mm apart. E)-H) show screen shots of resulting 3D models without and with texture colour.
Figure 10. Comparison of a natural-colour 3D model, a Micro CT reconstruction and 2D image at a similar angle. The surface geometry of the natural-colour 3D model (A) is less detailed than the Micro CT model (C) and missed concavities such as the antenna socket shown in the enlarged inset of C. However, the natural-colour 3D model can capture useful surface information such as the compound eye in the enlarged insect of B. False-colour Micro CT model (D) and a 2D image (E) are shown for comparison.
Figure 11. The impact of mounting orientation on reconstruction quality. Traditional horizontal mounting (A-C) produces inferior results to vertical mounting (D-F) for this specimen.

Figure 12. Impacts of mounting orientation and tilt on reconstruction quality. While additional images at tilting angles of $10^\circ$, $20^\circ$, $30^\circ$, and $40^\circ$ improve reconstruction quality in both horizontal and vertical mounting (in comparison with Figure 11), vertical mounting leads to sharper model with more vivid colours and textures.
Figure 13. **Additional camera poses can improve wing reconstruction.** A) A typical set of camera poses cannot resolve the occlusion created by the wings of this insect, leading to inaccurate reconstruction between its wings (B-C). D) Additional images taken from camera poses looking along the insect body and wing surfaces dramatically improves reconstruction accuracy (E-F).
Figure 14. Two methods to deal with an insect whose pin cannot be removed. A) The raw image shows the pinned specimen attached to a second vertical pin so the long-axis of the insect is vertical. B) An image of the specimen after all other parts of the image are masked to some extent. C) Ventral view of the 3D reconstruction from masked images shows a splotch of contaminated texture colour. D) An image of the specimen and pins retained. E) 3D reconstruction of insect and pins. F) Ventral view of E with pins edited out of the 3D model.

Figure 15. Traditional insect mounting. A metal pin pierces the body of an insect from the back providing stronghold to the specimen. However this mounting can be inconvenient for 3D scanning due to the specimen’s horizontal body axis.
Figure 16. Modification of StackShot™ macro-rail’s control box to accept external trigger. A) Circuit diagram to provide interface between trigger output of GigaPan™ robot and StackShot™ control box. The resistor and the opto-coupler convert the trigger signal to a “press button” action. B) the resistor and opto-coupler are soldered to a connector which can be mounted to the case of the control box. C) Finished the control box with the extra input connector on the left. Note that the connector pins of GigaPan™ trigger cable have to be swapped such that red cable goes to position 2 and white cable to position 3.

Figure 17. Examples of actual set-up in normal-mode (A) and macro-mode (B). The GigaPan™ Panorama Robot is mounted sideways to act as a two-axis turntable. A) camera is turned 90° around optical axis so that it can capture images that better fit the specimen and the mat target. Video of the system at work are available at [30].