

Digital Imaging Requirements Review:

Report by Dr Les Walkling



ATLAS OF **LIVING**
AUSTRALIA
sharing biodiversity knowledge



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1 Introduction

1.1 About this document

Dr Les Walkling was commissioned by the Atlas of Living Australia (ALA) to advise on the imaging needs of Australia's natural history collections, notably museums and herbaria.

Between 29 March and 6 July 2010, Les conducted interviews with directors,

collection managers, IT personnel, scientists and scholars at three museums, four herbaria and two universities.

This report documents Les's findings from those consultations. In particular, it identifies practices and makes recommendations that categorise the current digitisation requirements of these institutions.

2 Overview

In 2010 digital imaging is a well established practice. Cultural institutions such as the National Gallery of Victoria have been digitising their collection for fourteen years. But while there are parallels between the imaging requirements of cultural and biological institutions, there are also significant differences. Existing digital imaging standards and practices will have to be adapted to the diverse requirements of biological institutions and biodiversity collections.

There are already examples of outstanding digitising practices taking place in Australian biological institutions that could be emulated by other collections. But the current overall level of biological digitisation is significantly less than what is occurring in cultural institutions.

Cultural collections tend to be more homogenous than biological collections,

both in scale and application. While cultural digitisation falls into one of three categories, curatorial, conservation, and publication, biological collections encompass additional considerations such as endangered species, biosecurity, and agriculture. Biological specimens are also more variable. They might range from a blue whale to a microscopic fungi spore. In both cases the same principles of digitisation apply, and guidelines and specifications can be proposed, tested, and evaluated. In general though a broader range of digitisation approaches will be required than currently exist in cultural institutions. This includes how the specimen is prepared and arranged for digitisation, and the technologies employed.

I have therefore divided biological digitisation requirements into two categories and three levels:

	General Publication ¹	Research & Classification
Micro ²	✓	✓
Macro ³	✓	✓
Terrestrial ⁴	✓	✓

1. General publication includes web pages and field guides
2. Micro digitisation utilises compound microscopes to X1000M
3. Macro digitisation utilises stereo or dissecting microscopes to x40M
4. Terrestrial views do not require magnification

The digitisation specifications and/or technology required in each category and level can be quite different. A comprehensive digitisation strategy will also need to consider related tasks, such as

the digitisation of labels and other identification markers, with or without the use of OCR (optical character recognition) and ICR (intelligent character recognition) techniques. Some technology requires specialists, both in its operation, application and maintenance, while other systems can be operated by relatively untrained/unspecialised personnel.

Access technology such as Zoomify for on screen magnification without loss of image quality will facilitate levels of interaction for multiple purposes. However images need to be initially captured and then prepared for such extended access. Focus

stacking ,where multiple captures of the same specimen are montaged into one image, usually in 2D but can also be in 3D, will enhance interaction with and interpretation of the specimen potentially across all three levels, micro, macro and terrestrial. But techniques like focus stacking require operator experience and skill. Programs such as Helicon Focus can

automate this with Canon EOS digital cameras, but generally such automated solutions are based on integrated propriety systems such as the Smart Drive SatScan, the Visionary BK Lab System or the Leica LSA microscopy system that require less operator training and interpretative skill.

3 Issues confronting institutions

While collective support was expressed by the interviewees for the general principles and advantages of digitising biodiversity collections, varying requirements, existing facilities and future projections exist among the institutions.

Space for installing digitising workstations while needing consideration, does not appear as significant as access to and the transportation of specimens. Therefore portable workstations, in proportion to the diversity of collection locations, may need to be considered, though quality control will be harder to maintain.

Sufficient skilled operators will be a significant hurdle. While non-specialists can digitise most terrestrial specimens, and with basic identification training also some macro and possibly micro specimens, specialised handling and interpretative skills will still be required in most cases.

The speed of digitisation will also be a significant issue, especially given the size of many collections. That is, how long it takes to digitise a single specimen or group of specimens. For example, three views of a bee can take a considerable amount of time because of the handling and restaging required for each view. On the other hand the digitisation of a specimen in a single orientation, such as a lateral or dorsal view can be accomplished without skilled or interpretative handling. A drawer filled with multiple specimens can be digitised as a single image. Most interviewees believed the general availability of data was initially more important than focused data. These factors will also have a bearing on the institutional support for a digitising program.

In the case of large collections of specimens, typically invertebrates, different methods and timelines may need to be applied than to smaller collections.

Maintenance of equipment is another consideration. If a system fails, who will fix it? Sophisticated systems with significant purchase costs, have high replacement costs and high maintenance fees. The quality and efficiency of a sophisticated-specialised system may need to be traded off against the generic advantages of prosumer systems. At a significantly lower purchase cost, multiple prosumer systems might significantly increase throughput without sacrificing quality, as long as there are enough operators to run them.

While satisfactory workstations can be currently purchased and guidelines and workflows established, unless there is also an investment in training the workstations will possibly be under utilised and in a worse case scenario will produce invalid results. Training can also include proactive measures to minimise maintenance and downtime.

It is unlikely that all biological data can be satisfactorily auto digitised into RGB (red, green, blue) images. At least some interpretation will be required at different stages for many images. Batch processing of similar images, such as captured for focus stacking, will still require initial interpretation, and/or post-processing adaptation to an output device.

The archiving of data is also going to be an issue for all the institutions I visited, because their existing IT infrastructure is not setup to handle a deluge large files day in and day out. Local multi level RAID storage devices may have to be included in initial equipment installations.

The location and association of metadata with images, and their permissions will be another challenge to standardise and organise.

4 Differences between institutions

The differences between institutions is not just a reflection of their collections, but also their collecting policies and priorities which can be as individual as the individuals who manage them.

The overall focus of biological collections is biodiversity and specimen classification. This is an on-going and evolving task accelerated by new classification methods such as DNA sequencing. Current digitisation of these collections varies from significant to non-existent. Parallel applications such as field guides require extensive on-site documentation of specimens, often including photographic records in addition to or in place of original field notes and sketches. A metric such as specimen colour that variably fades after collection can be digitised in the field before collection. Vascular plants for example lose more or less of their colour depending on how they are collected, pressed and transported. While a metric such as colour is less significant for classification, it is dominant in field guides. Individual curators place more or less emphasis on field guides relative to taxonomy.

The scale of specimens in herbaria vary between microscopic fungi spores ($\times 1000M$) to A3 vascular plant specimen sheets. The vascular plants were out numbered by the non-vascular plants, and specimens such as lichens and the vascular plant specimen sheets were somewhat standardised, other specimens such as seeds and fruits range over a much larger scale.

The scale in museums is even larger, right up to the blue whale. There are also different levels of observational data that can be important.

There were also different levels of confidence in different data sets between institutions, where some could be released immediately, and others were thought to contain errors that would first have to be corrected before public release, and this was not a simple job in every case. There are parallels in existing digitisation practices where little or no colour management practices are in place, therefore the visual relevance of the images is compromised.

No one agreed that there was a single set of standards or guidelines for the presentation of specimens.

Specimens that can be seen by the unaided eye, such as the vascular plants or vertebrates are generally already well represented online.

There are differences between the descriptions of different specimens, for example fungi are complex.

Existing film based image collections have depended on an individual or a few individuals' initiative, and so has their digitisation.

The standards and codes of practice in the printing and publishing industry are vastly more evolved than in biological institutes.

5 Technical issues in biodiversity imaging

Digitisation transforms continuous resolution and brightness into steps of resolution (pixels) and steps of brightness (levels). The question that arises is how many pixels and levels are required for the digitisation of various specimens.

There are eight components that must be configured into any digitisation facility:

- Optical resolution
- Bit depth
- Dynamic range
- Depth of field
- Lighting
- Angle of view
- File size
- Storage and access.

5.1 Optical resolution (pixels)

At 100 pixels/inch the average human observer will have difficulty distinguishing individual pixels at a distance of 40 cm. Tied to digital resolution (in pixels) is the optical resolution or resolving power of the optical system (camera, lens and/or microscope). A high pixel resolution is wasted if the optical resolution is not equal to or greater. Nyquist's theorem requires two line pairs or pixels to create the equivalent of a physical boundary. Therefore the digitising resolution should be at least twice the required optical resolution. For example, in calculating depth of field for a digital sensor and optical system, twice the circle of confusion (the pixel pitch in the sensor) is used. A Hasselblad H4D with a pixel pitch of 0.006 mm therefore determines a circle of confusion of $2 \times 0.006 = 0.012$ mm.

5.2 Bit depth (levels)

The appropriate bit depth is proportional to the brightness range of the specimen,

including its lighting and captured perspective or angle of view. Dark current noise (non-image) that is present in all capture and amplifier circuits begins to overtake and compromise the visual data when the signal strength falls too low.

Bit depth is a function of the capturing device and the range of brightnesses that it can capture.

An 8-bit per channel image provides up to 256 (0-255) levels of brightness per channel. A 16-bit per channel image provides up to 65,536 (0-65535) levels. But in both cases the bit depth may describe a broad or narrow range of brightness. Not all the bit planes will be used in all images. For example a camera that digitises in 12-bits per channel when stored in a 16-bit file will not contain any data (0, 1) in the four least significant bit planes. But all of the original 12-bit data will be preserved.

Bit depth can also be related to the specimen's brightness range. The greater the difference between the brightest and darkest values in the specimen, the greater the required bit depth to faithfully capture those values. It is generally thought that when the signal strength falls below 100 levels that we will begin to detect in smooth toned areas the individual steps of brightness (levels).

So there are really two criteria here – bit depth as a function of signal strength, which is related to noise limitation of the available dynamic range, and bit depth as a function of posterization – that is, not enough levels of tonality to promote the illusion of continuous tone.

Most specimens I observed and measured had a very low brightness range less than 4 EV or $2^4 = 16$ levels of brightness. Therefore many specimens could be captured in 8-bits per channel and not suffer from either noise or posterization limitations. Herbarium sheets for example,

if digitised in soft enveloping light could be adequately represented in 8-bits per channel. However interpretative lighting (modelling) and other presentation considerations or enhancements can easily push the brightness range beyond these limits, where 16-bits per channel should be considered the norm.

5.3 Depth of field

Depth of field is an illusion. Only the plane of focus is critically sharp. Smaller lens apertures result in a reduced circle of confusion on the sensor. When the out-of-focus circle of confusion is less than our visual threshold, we do not distinguish it as being 'out of focus' and the apparent depth of field is increased. An acceptable circle of confusion depends on the depth of the specimen, its image magnification, and viewing perspective. Infinite depth of field is not possible because of diffraction limitation in lenses, where smaller apertures result in a loss of critical sharpness and therefore poor focus across the entire field of view.

The depth of field can be extended in post-processing by focus stacking multiple images of the specimen taken at different planes of critical focus throughout the specimen, but without moving either camera or specimen. This allows an optimum lens aperture to be used for highest optical performance, resulting in the equal resolution of different identification keys within the specimen.

It is expected that some form of focus stacking will be required in the digitisation of many specimens.

5.4 Lighting

A light source can be described by its brightness level, the angle of illumination, its colour temperature (spectrum), how broad (diffuse) or focused (specular) it is, and its brightness difference (ratio) relative to other light sources.

Lighting can be flat with minimal interpretation, or modelled to influence our perception of shape, volume and texture. Many specimens such as insects and herbarium sheets will benefit from flat (soft enveloping) lighting to minimise reflections and specularities, while allowing the specimen's 'inherent structure' to dominate the image. In other cases, values within the specimen will need to be enhanced (modelled) so as to clearly distinguish aspects of their morphology that are important as identification keys or a visual aid.

The capture of reflected light also incorporates the management of colour appearance. However most interviewees remarked that colour while useful as a screening aid, especially in live specimens or field guides, is not useful as an identification key because of its variability in preserved specimens and changes over time, even in dark storage. Therefore colour management is a less critical capture criteria than resolution and lighting to preserve or enhance a specimen's identification.

5.5 Angle of view

There appear to be no common standards of presentation of a specimen across all collections and departments. Common sense implies that lateral, dorsal and ventricle views will be helpful as general screening aids, however which way should the head be facing in a lateral view? Critical keys will require specialised knowledge to be clearly revealed in an image, along with skill in handling the specimen to achieve this angle of view.

Parallax errors can also result from single captures of large specimens, or drawers of specimens. An X-Y table that moves the specimen(s) under the camera, or the camera over the specimens as in the Smart Drive SatScan, taking multiple exposures that are assembled (stitched) in post-processing into a single image, will eliminate most parallax errors and

improve the overall coherence of the image.

5.6 File size

The file size should be proportional to the required resolution and final use. In the printing industry this is easily specified because the required resolution of a printing press is known. The uses of a digitised collection can only be guessed at. Therefore capturing at the highest possible resolution is a consideration, though it impacts on productivity, storage, transfer and access requirements. While the optical resolution of a lens has well known theoretical limits, the limits of digital resolution (pixilation) though relative to the resolving power of the lens, are still being investigated. Capturing at the highest available pixel resolution doesn't prevent the manual or automated production of derivative images fit for specific purposes, such as web graphics, nor for future requirements yet to be discovered. The danger would be not capturing at a sufficiently high pixel (and optical) resolution. This effect on productivity and throughput will moderate over time as faster processing becomes possible, but can also be moderated in the short term by increasing the number of digitisation workstations and operators.

5.7 Storage and access

The method of cultural institutions is to record an 'archival master' at a sufficiently high bit depth and optical resolution that it equals and/or exceeds all possible uses, commensurate with contemporary and projected applications.

The issue for biological collections is that specimens are often inspected at varying resolutions (magnifications), including macro and micro resolutions in order to determine the significant identifier keys. Therefore in many cases the higher the optical resolution captured in the archival master the more it can be inspected at a various magnifications including through technology such as Zoomify. Derivatives including croppings of the original scan can be easily obtained from high resolution archival masters without the specimen having to be re-digitised. This method will not be possible for all specimens. For example some specimens will need to be imaged at significantly different resolutions that can not be captured by a single camera setup or point of view, and there are also cases where dissection is required, as in insect genitalia, in order to fully classify the specimen.

There are also non-optical criteria that are becoming increasingly important, including nucleotide sequencing and cultural definitions that fall outside of the visual criteria for digitisation.

Until centralised high speed data repositories become available, local collection digitisation and management may also require local storage solutions. The National Library of Australia and the National Archives of Australia recommend at least three copies of any file, with one copy stored off-site in case of fire and/or theft, and preferably using different media to store different copies. High level RAID systems, both propriety (Drobo) and non-propriety are available with robust parity checking of data integrity and accuracy.

6 Technical solutions

The technical solutions can be divided into three types:

1. Custom built systems, such as SmartDrive SatScan
2. Propriety systems, such as BK Lab System
3. Adapted systems, that utilise professional/consumer technology and techniques.

The difference between the custom and propriety systems can be subtle. Custom built does utilise ready made components, such as lenses and CCD sensors, but they tend to be fixed in their use for a particular application, such as the SatScan at the Australian National Insect Collection which has been configured and precisely adjusted for the scanning of their specimen drawers, one at a time. Propriety systems tend to use common camera systems, such as Nikon or Cannon, with customised hardware and/or software, and can be adapted to capture a wider range of specimens under different conditions without a major refit and realignment of the system as is the case with the SatScan. Adapted systems are cobbled together by individuals from commonly available components, such as commercial lighting systems and camera systems, and can be quick to set up and adapt for different work.

There is a sliding scale in the design, implementation and application of digitising systems that trades off productivity against quality. Fit for purpose systems such as the SatScan do not necessarily produce superior efficiency for the same image quality. Real world testing needs to be undertaken, such as setting up competitive systems side by side, e.g. the SatScan versus a Hasselblad H4D multi-shot camera covering the same area.

The Smart Drive SatScan system takes 20 minutes to scan a whole drawer (490mm x

490mm) to produce a 327 MB RGB tiff file. A Hasselblad H4D50_MS camera takes 20 seconds to capture the same area in a single image in six shots resulting in a 600 MB RGB tiff file (or 400 MB when cropped to the square of the insect drawer). But is the resolution the same or different, better or worse, and does the parallax in the single capture of an entire drawer compromise its value?

The Smart Drive SatScan did not have a ready made Zoomify add on to the installation at the NICS. It will have to be developed, or the images – tiles if you like – will have to be combined into a single image that is then Zoomified. Zoomify has a Photoshop export module that would simplify this workflow. It can support extremely large files – certainly files many times bigger than what we are currently capable of capturing.

Many images will have to be post-processed in an image editing program like Photoshop in order to enhance critical specimen features and distinctions, and potentially Zoomify the image as well.

Of the propriety systems for micro-digitisation, Leica has developed a highly integrated system of microscopes, cameras and software purpose designed for biological illustration. For example:

- Leica MZ16 (x11M) and M205C (16xM) dissecting microscopes with diffused light adapter
- Leica DFC500 camera 12mpx 14-bpc = 42-bit RGB in 1 shot (1360 x 1024), 4 shot (1360 x 1024), 16 shot (2720 x 2048) and 36 shot modes (4080 x 3072 pixels) with an encapsulated CCD sensor to prevent condensation with difficult specimens
- LAS Software with z-stepper for focus stacking

High quality graphics monitors, such as the Eizo Colour Edge series will be critical

to the accurate capture, editing and presentation of specimens. Their colour critical performance though important will be less critical than their digital

equalisation technology that ensures even brightness, contrast and resolution across the screen.

7 More information

More information on the equipment mentioned in this report is available on the following web sites:

Canon cameras and accessories
<http://canon.com.au>

Ezio monitors <http://www.eizo.com.au/>

Hasselblad cameras and accessories
<http://www.hasselblad.com.au/hb/>

Helicon Focus software
<http://www.heliconsoft.com/heliconfocus.html>

Leica microscopes and accessories
<http://www.leica-microsystems.com/home/>

Nikon cameras and accessories
<http://nikon.com.au/>

Photoshop software
<http://www.adobe.com/ap/products/photoshop/compare/>

SmartDrive SatScan
<http://www.smartdrive.co.uk/>

Visionary Digital imaging solutions
<http://www.visionarydigital.com/>

Zoomify software
<http://www.zoomify.com/>