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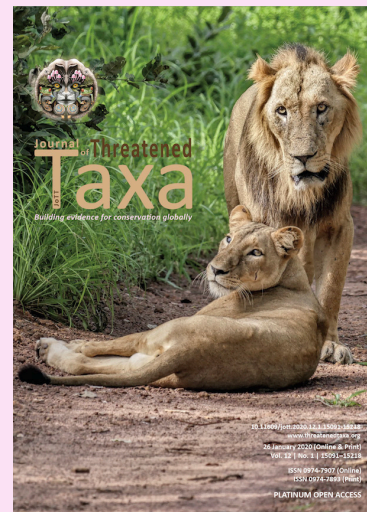
## COMMUNICATION

### DIGITAL IMAGE POST PROCESSING TECHNIQUES FOR TAXONOMIC PUBLICATIONS WITH REFERENCE TO INSECTS

Nikhil Joshi, Hemant Ghate & Sameer Padhye

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## Digital image post processing techniques for taxonomic publications with reference to insects

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**Abstract:** There exists substantial literature for capturing digital images of insect specimens for taxonomy purposes but very few papers are available on post processing of these images. We present a few techniques for editing digital images of insects using Adobe® Photoshop® which can be performed in a relatively short amount of time. The results clearly show that techniques using a combination of options like Curves, Dodge/Burn, Hue/Saturation and Lab Color mode in the software, enhance the quality of the original image without changing any taxonomic information. These methods applied in different combinations can be used for taxonomy of any insect taxon. We also caution the readers of the abuse of such techniques in context of taxonomy.

**Keywords:** Adobe® Photoshop®, beetles, insects, lab color, purple fringing.

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**Author contribution:** SP and HG conceptualized the idea. HG and SP carried out photography. NJ and SP carried out the post-processing of images. All authors contributed to writing the manuscript.

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## INTRODUCTION

Insect taxonomy using some/any kind of illustrations to support descriptions has always made it easier for the reader to comprehend those descriptions. The form of illustrations has evolved from simple drawings used in early studies to the use of digital imaging via confocal microscopy, scanning electron microscopy, dual beam scanning electron microscopy, and micro-CT (among others) for studying and describing specific aspects of their morphology (Wipfler et al. 2016).

By and large though, images (and drawings) taken by digital cameras with/without use of stereomicroscopes and/or bright field compound microscopes are widely used in insect taxonomy currently (Buffington et al. 2005; Holzenthal 2008). Digital imaging systems and techniques such as the 'Auto Montage' (hardware and software) (Azorsa & Sosa-Calvo 2008; Jansen & Halbert 2016; Otto 2016), 'Dome lighting' (hardware) (Kerr et al. 2008) and 'Natural color 3D models' (hardware and software) (Nguyen et al. 2014) are now utilized for documentation and taxonomic studies of various insects.

Subsequently, many softwares are available to cater to the processing of such captured images (e.g., Adobe® Photoshop® and GIMP). These provide a plethora of tools and techniques for different aspects of image editing. Using these tools, Jakubec et al. (2018) have provided an excellent and less time consuming method which is used for background isolation of the entomological digital illustrations. While literature exists detailing various methods of photo documentation for different insect groups (Häuser et al. 2005; Riedel 2005; Buffington & Gates 2008), to our knowledge, not much literature exists for the image processing details in context of taxonomy. The best example we know of are the image editing procedures explained on the journal, 'Zootaxa' website (<http://mapress.com/zootaxa/imaging/index.html>).

With this background, we present a few digital image processing techniques by using Adobe® Photoshop® which can be done in a relatively less amount of time using Coleoptera (images) as a model system.

## MATERIALS AND METHODS

### Material studied

Species from four families of Coleoptera, viz., Cerambycidae, Chrysomelidae, Dytiscidae, and Endomychidae were used for standardizing the different image editing processes. Selection of the insect group

was based solely on the availability of specimens, no other selection criterion was used.

### Methods

Multiple images were taken either via 1) Canon 400D SLR camera with a 100mm macro lens and/or 2) Stereo Binocular Microscope (Leica MZ6 with attached Canon PowerShot S50). Multiple images were taken and digitally stacked using COMBINE ZP (<http://www.hadleyweb.pwp.blueyonder.co.uk/>), a freeware. The photo processing techniques were standardized on Adobe® Photoshop® CS5 student version on Windows 10.

We have explained some of the basics before the actual procedures (given below), though, absolute essentials of Adobe® Photoshop® are beyond the scope of this work and hence, not covered here. Readers who wish to learn about it can visit the official site for help. Terms and terminologies are as per Adobe® Photoshop® CS5 software.

We have used only a single representative image of a cerambycid beetle while describing the processes for consistency.

### Some pre-requisite basics are first explained below before the actual methods.

#### 1) New Layer

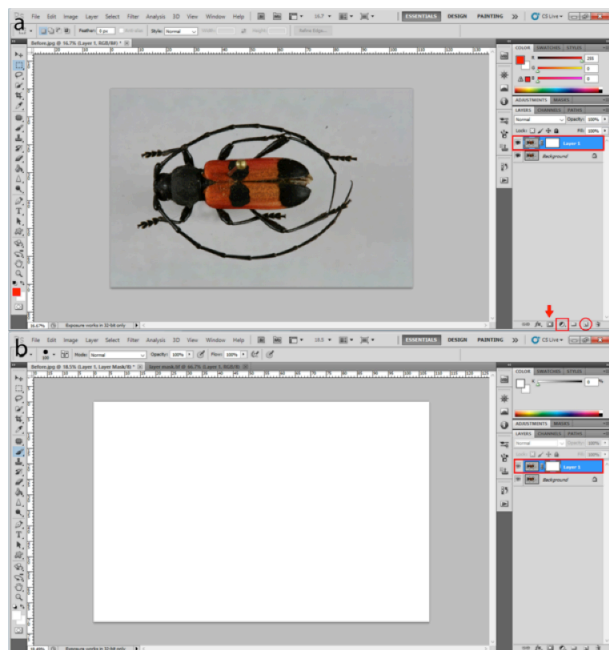
New layer creates a blank space where in additional colors and vectors can be put and later merged/blended with the image to be processed. New layer can be created by pressing the 'Create New Layer' icon on the bottom right of the main window (Image 1a 'circle') or by going to the 'Layers' drop down menu, selecting 'New' followed by 'Layer'. New layer can also be created by pressing Shift+Ctrl+N.

#### 2) Duplicate Layer

Duplicate Layer makes a copy of the original image/Layer. This is made so that the original image is not changed or processed in any way. A Duplicate Layer can be created by going to the 'Layers' drop down menu and selecting 'Duplicate Layer'. Duplicate Layer can also be made by pressing Ctrl + J.

#### 3) Adjustment Layer

Adjustment Layers are used to edit the images and using them is more advantageous as they can be switched on/off and/or modified later. Adjustment Layers can be created by pressing the 'Create Adjustment Layer' icon located on the bottom right of the main window (Image 1a 'square') or going to the 'Layers' drop down menu and selecting 'New Adjustment Layer'.



**Image 1.** Display window showing main window of Adobe® Photoshop® and its Layer Mask options. a—Layer Mask option – thick arrow; New layer – circle; Adjustment layer – square; Layer Mask applied – rectangle | b—Layer mask specifically selected – rectangle.

#### 4) Layer Mask

A layer mask is created in Adobe® Photoshop® to either reveal or hide all the details of the image. A white layer implies all the details are revealed while a black layer means that all the details are hidden (Image 1b 'rectangle'). Layer mask can be made by selecting the 'Create Layer Mask' icon on the bottom right of the main window (Image 1a 'thick arrow') or going to the 'Layers' drop down menu, selecting 'Layer Mask' and further selecting either 'Reveal All' or 'Hide All'. Pressing Alt on the mask displays the actual layer mask (Image 1a & b).

### Image processing techniques

#### 1) Aligning the image (Image 2a)

Generally, while making scientific illustrations, it is important to have well aligned images in 90° or 180°. Hence, when the captured image is not at a desirable position, it can be aligned using the Image Rotation option.

The image can be aligned as per user specification.

- ⌚ Duplicate the layer
- ⌚ Go to Image > Image Rotation (Image 2a 'square') (Extent of rotation is determined by the user).

#### 2) Lightening or darkening parts of the image (Image 2b).

Overexposure and/or under exposure in portions of the images is edited by two tools namely, 'Burn' (darkens overexposed parts of the image) and 'Dodge' (lightens the dark parts of the image) respectively (Image 2b 'Rectangular box').

- ⌚ Select the option as per the image exposure
- ⌚ Select the 'Shadows' part in the dropdown menu after selecting 'Dodge' (Image 2b 'thick arrow') so that only the darkest parts of the image are highlighted and mid tones are left in their natural state. Similarly, select the 'Highlights' parts in the drop down menu after selecting Burn so that only the white parts (overexposed) are darkened.
- ⌚ Brush size is selected as per the area of the image which needs either of the two tools (Image 2b 'circle')
- ⌚ Exposure (intensity) of the brush is selected as per the requirement for the image (Image 2b 'thin arrow'). A value between fifty to seventy percent usually works.

#### 3) Adjusting the Levels (Image 3a & b)

The lighting levels of the photo can be quickly edited by using 'Curves' in the 'Adjustment Layer' menu. Levels can also be adjusted by using the option 'Levels' in the 'Adjustment Layer' (not explained here).

- ⌚ Select the 'Curves' option (Image 3a 'arrow') in the 'Adjustment Layer'.
- ⌚ This will open a graph of the composition of the image (Image 3b 'square box')
- ⌚ Adjust by moving the slider (via mouse) either in the X or Y axis as required (Image 3b thin and thick arrows, respectively)

#### 4) Sharpening (Image 4a & b; Image 5a & b)

This tool is used when the details within the image come out soft and need to be emphasized more. Basic sharpening includes using Sharpen tools in Filter menu (not explained here).

One way of doing effective sharpening of the image is by a combination of a) Lab color mode and b) High pass filter

- ⌚ Duplicate the layer
- ⌚ Go to Image pull down menu and select 'Mode' followed by 'Lab color' (Image 4a 'square box & arrow'). A message will follow this selection for which Don't flatten should be chosen (Image 4b)
- ⌚ Duplicate this layer again (and this layer should be selected)
- ⌚ Go to 'Filter' and select 'Other' followed by 'High Pass' (Image 5a 'square box')

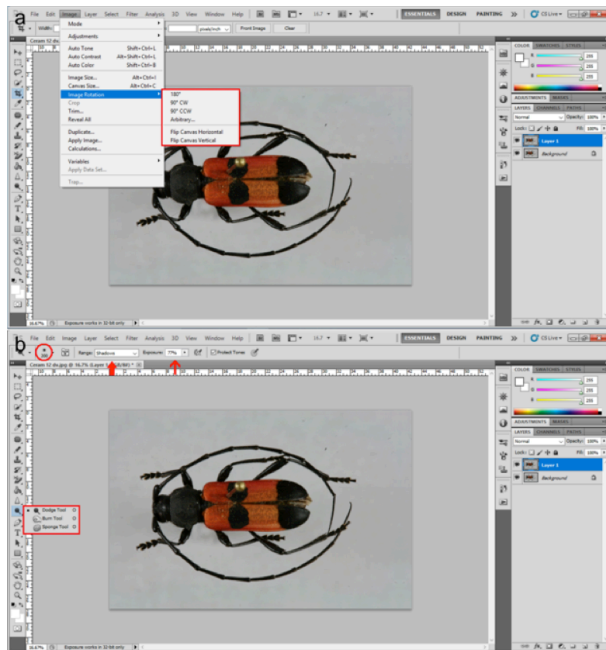


Image 2. Display windows showing 'Image Alignment' and 'Dodge/Burn' tools. a—Image Alignment option – rectangle | b—Dodge/Burn tool – rectangle; Brush size – circle; Range selection (for either Shadows, Highlights or Midtones) – thick arrow; Exposure (intensity) – thin arrow.

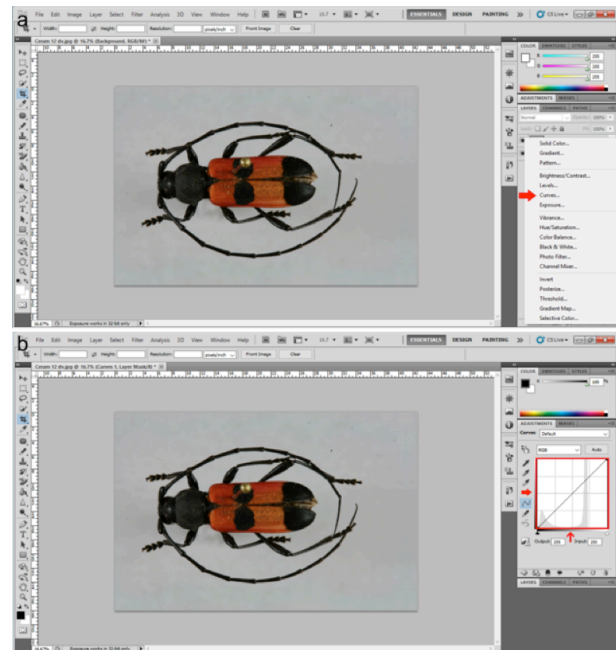


Image 3. Display window showing process for altering the 'Curves'. a—Curves option in Adjustment Layer - thick arrow | b—a graph for changing the Curves settings - square; The Y and X axes respectively - thick and thin arrows.

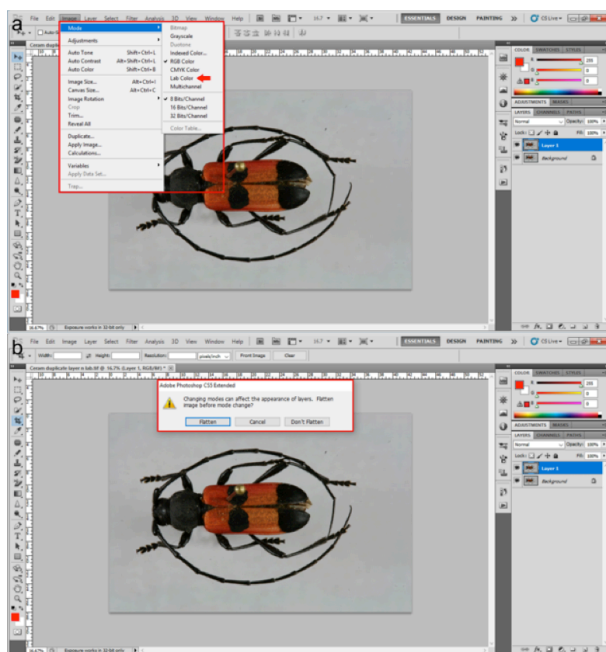


Image 4. Display window showing the process selecting the Lab color mode. a—panel for selecting Lab color option - square; Lab color option - thick arrow | b—window showing the option of 'Flatten Image' – rectangle.

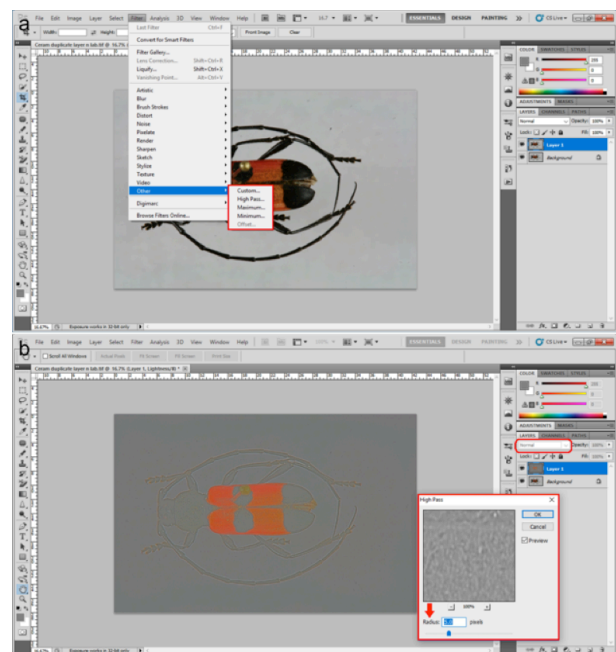


Image 5. Display window showing the process of image sharpening by using Lab color mode. a—High Pass option – rectangle | b—High Pass option window - square; Blend mode drop down menu - rectangle; Slider for the radius - thick arrow.

⌚ In the 'High Pass' window (Image 5b 'square'), select the radius; optimum value ranges between 2 and 6 (Image 5b 'arrow'). Exact value has to be selected as per the image requirement. Click OK.

⌚ Select the 'Soft Light' or 'Overlay' Blending mode (In the drop down menu below Layers on the Right Hand) (Image 5b 'round edged rectangle')

⌚ The two layers should then be merged (this can be done by selecting the two layers and then pressing Ctrl + E).

#### 5) Color artifacts (Image 6 a,b)

The captured image sometimes contains color artifacts which alters its original color. Many times, images also have purple fringing (PF), a chromatic aberration occurring at the edges of the image (especially when the microscope does not have achromatic lenses). These can be edited by making use of 'Hue/Saturation' option in the Adjustment layer in Adobe® Photoshop®

⌚ Image must be in RGB mode (if it has been earlier converted to Lab color)

⌚ Duplicate the layer (Ctrl + J)

⌚ Go to the 'Adjustments Layer' on the bottom right of the main window (Image 1 'square') and select 'Hue/Saturation' (Image 6a 'rectangle' & 'thick arrow')

⌚ In the 'Adjustment Layer' window click on the second drop down menu (Image 6b 'circle') and select Magenta/Blue (for PF) (or the color of the aberration/artifact)

⌚ After selecting the color, a Dropper Tool icon will be active located below the 'Lightness' slider (Image 6b 'thick arrow'). Select the dropper tool and move it to the part on the image which has the artifact/s. After selecting it, Photoshop will give a color range of that color (located below the dropper tool) (Image 6b 'thin arrow') (Re-check if it is the right shade).

⌚ Drag the Saturation slider to the left-hand side till the point the color artifact is not seen anymore (Image 6b 'rectangle')

In many cases, it also affects the natural coloration of animals

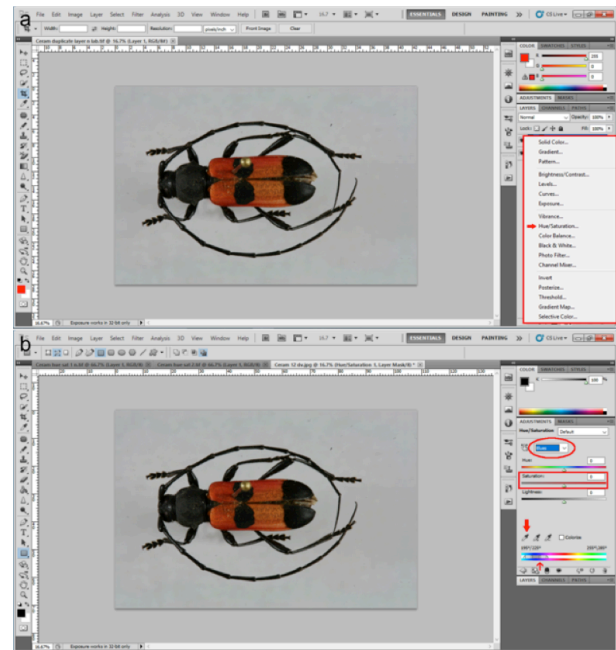
What can be done for this?

⌚ Make a 'Layer Mask' (Image 1a)

⌚ Hold the 'Alt' key and left click on the Layer mask to select it (Image 1b)

⌚ Press Ctrl + I to convert the white layer to black (thus hiding all the desaturation done)

⌚ Select the Paint brush tool with white color followed by stroking on the parts which have purple fringing so that only those parts are edited and show up while the original color is retained for the rest of the



**Image 6.** Display window showing the process of removing color artifacts. a—Adjustment Layer - rectangle; Hue/Saturation option - thick arrow | b—Hue/Saturation slider - rectangle; Color shade option - circle; Dropper tool for selecting the specific type of color (to be altered) - thick arrow ; color range provided by Photoshop for selected color - thin arrow.

image.

#### 6) Background color (Image 7 a,b)

This tool is used to get a uniform background color of choice for any image. It is ideal for photos taken with any uniform background.

Please note: For using this tool, the original background should have fewer colors to begin with and it is not recommended when the image contains complex backgrounds (e.g., Live animal in its natural habitat). Chroma+ method can also be used for unifying background if you have images with chroma background and neutral background. This method is surely less time consuming when you have images with both chroma+ and neutral backgrounds. If time for capturing image is also considered, then both chroma+ and below mentioned method consumes equal amount of time with similar final results.

⌚ Create an empty Layer (Image 7a 'square') below your image and fill it with White (or the background color of your choice) (Take care that the color selected should not be the same shade of the new intended color for this method to work. E.g., If the background color of the original image is green, refrain using any shades of green for the new intended background) (Image 7a 'thin

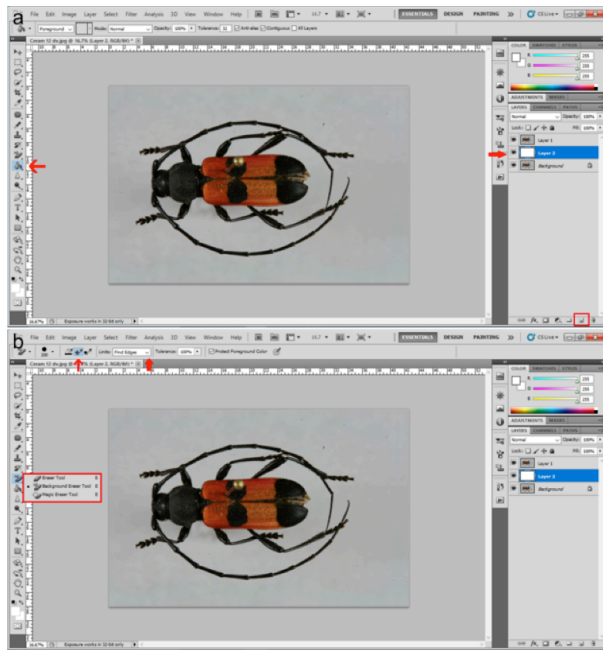


Image 7. Display window showing the process of removing the background color. a—New Layer option - rectangle; New Layer in the Layers menu - thick arrow; Paint Bucket tool - thin arrow | b—Background eraser tool option - rectangle; Limits option - thick arrow; Type of background erasing - thin arrow.

and thick arrows')

⌚ Right click on the 'Eraser' tool on the left-hand side panel of Photoshop and select 'Background Eraser Tool' (Image 7a 'rectangle')

⌚ Select 'sampling once' from the sampling tool bar, which is placed next to the 'brush preset picker' (Dropper icon with a bullseye mark) (Image 7b 'thin arrow')

⌚ Select the 'Limits' (for selecting type of background erasing) as 'Find Edges' from the drop down and set the tolerance between 50–70 % (this value will change as per the image) (Image 7b 'thick arrow')

⌚ Start erasing the background. Just make sure the plus mark seen in the Brush pointer should be always placed on the background while clicking not on the image (otherwise any colors resembling the background in the specimen will also be erased).

## RESULTS AND DISCUSSION

The processed images showed a marked improvement without any loss of taxonomic information. Details which were hidden due to insufficient light were highlighted clearly using Curves (Image 8a & b) and Dodge tools (Image 8c & d). The soft parts of the images

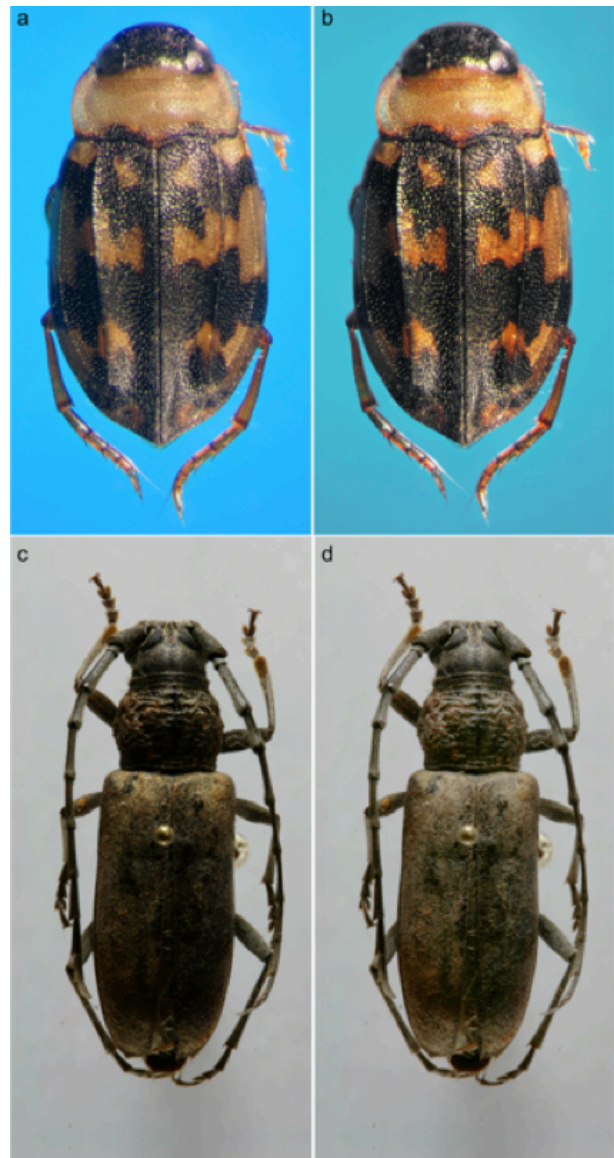
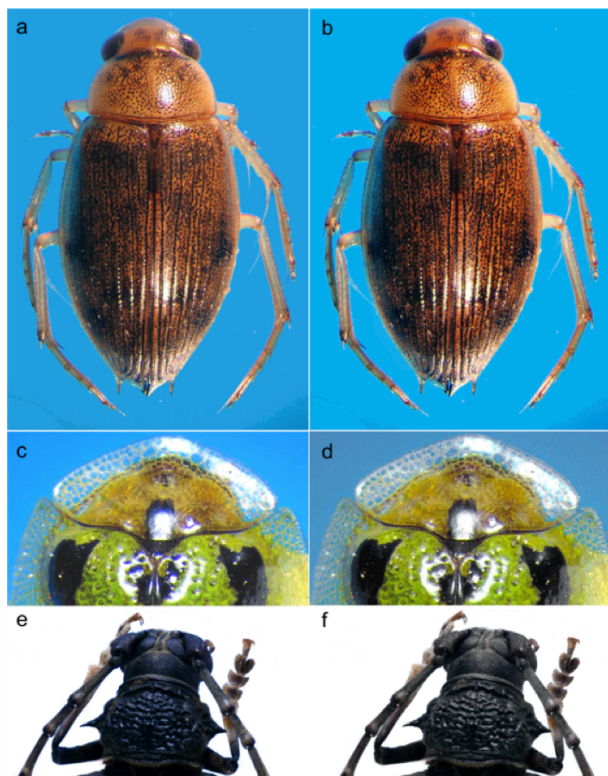


Image 8. Images showing a change in contrast due to altering Curves. a—(Dytiscidae) before | b—after | c—image showing a change in highlights using Dodge tool (Cerambycidae) – before | d—after.

were refined noticeably after sharpening, (Image 9a & b) while the color artifacts were completely nullified thereby revealing the true color of the specimen (Image 9c-f). The background of the image was completely changed bringing more contrast to the image (Image 10a & b). The photos then become very suitable for taxonomy publications as shown here.

The aforementioned techniques can be used singly or in combination (E.g., Image 10c & d) as per the researcher's requirement for any insect taxon (with slight alterations), though, image capture techniques need to be selected appropriately beforehand given the taxa under consideration; for example, the number of images

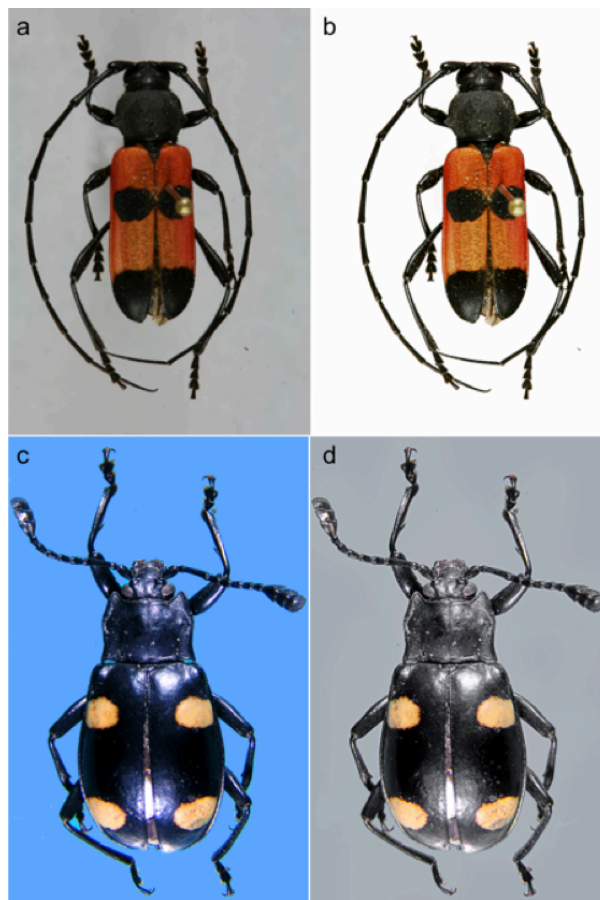


**Image 9.** Image showing a change in sharpness using Lab color mode and High Pass filter: a—(Hydrophilidae) before | b—after | Image showing removal of Purple fringing due to altering Magenta color using Hue/Saturation tool – (Cassidinae): c—before | d—after | (Cerambycidae): e—before | f—after.

required for stacking for a beetle would be different than that for a butterfly given their body convexity (Riedel 2005).

An ideal image is the one which does not require any or very little processing but that does not happen in many cases. Many laboratories do not have the necessary infrastructure due to financial and/or logistical constraints. Capturing high resolution images optimally in an affordable way poses a challenge which needs to be tackled (Buffington & Gates 2008). Still, good images can be taken by adjusting conditions such as correct and/or additional lighting, use of correct lenses (if using SLR or micro 4/3<sup>rd</sup>), finest use of manual Mode in digital cameras and making adequate (not excessive) use of photo processing tools (as is shown through this study). Simple tools such as 'Unsharp Mask' can help sharpening the image in Adobe Photoshop (R) as suggested by Zootaxa (see guidelines for preparing images).

Images or line drawings considerably improve the contents of the taxonomy papers and providing such illustrations gives valuable information while describing and/or revising new species, genera or families; e.g.,



**Image 10.** Image showing a change in background color using Background Eraser tool: a—(Cerambycidae) before | b—after | Image showing a composite editing (using Curves, Sharpening and Color artifacts removal) (Endomychidae): c—before | d—after.

assassin bugs (Weirauch et al. 2014), tiger beetles (Moravec 2016), and scarab beetles (Rossini & Vaz-de-Mello 2017). In spite of the availability of the latest technology people are still using poor quality images in biology papers in many instances. Preparing good images or drawings is an important issue in taxonomy and our paper assists in this issue to a certain extent. This article focuses on post processing techniques of already existing image while earlier work cited here, mainly describes procedures for obtaining good quality images. We understand that software would be updated frequently but all the protocols provided here are basic and would be functional in the updated versions. There is a high chance that, all these editing processes will be automated with the progress in the technology.

We would like to caution the readers that our aim here was to present ways to process only properly taken digital images where in altering few aspects such as orientation, background color and exposure better the

already good quality of the image. These techniques are not meant for enhancing or editing poorly captured images. We would also like to point out that, even though the use of good photographs are extremely beneficial and could be used as substitutes for the type specimens in some cases, they should never replace actual type specimens (Rogers et al. 2017). Actual specimens act as replicable datasets and a single image would not be able to capture this entire data contained in an actual specimen (Ceriaco et al. 2016; Rogers et al. 2017).

## CONCLUSION

Our work shows that digital images used for insect taxonomy can be edited to an extent which doesn't alter the image properties and thereby morphological characters altogether but, enhances it enough so that it can be used in taxonomical research. These methods are quite easy to perform as well. We also stress on the fact that a poor image with heavy editing is no substitute for a properly taken one with less editing or no editing.

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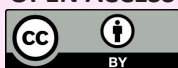
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