

# A Protocol for Basic Post-Mortem Examination and Sampling of the Cardiovascular System of Great Apes

Strong *et al.* (2015)

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**Ape Heart Project (EAZA Great Ape TAG Endorsed)**

Twycross Zoo, Burton Road, Atherstone, Warwickshire, UK, CV9 3PX

Email: [heartproject@twycrosszoo.org](mailto:heartproject@twycrosszoo.org)

Website: [twycrosszoo.org/conservation/research-at-twycross-zoo/ape-heart-project/](https://twycrosszoo.org/conservation/research-at-twycross-zoo/ape-heart-project/)

# GUIDELINES

## 1. Introduction

This protocol has been created as part of the EAZA Great Ape TAG endorsed Ape Heart Project. Based at Twycross Zoo (UK), the project is a Europe wide collaborative initiative striving to achieve a better understanding of great ape cardiovascular disease through a combination of epidemiological study, clinical and pathological investigation.

The Ape Heart Project is now supported by the EAZA Biobank, which facilitates international sample transfers from European countries. The EAZA Biobank is a vital initiative for supporting species population management and conservation research.

This protocol aims to provide a guideline for performing a systematic and comprehensive approach to the post-mortem examination and sampling of the cardiovascular system in great apes.

Cardiovascular system examination should form part of a whole-body gross examination and histopathology; this protocol is therefore intended to be supplementary to the general Great Ape TAG Veterinary Guidelines for performing post-mortem examination.

The purpose of this protocol is to:

- Promote consistency and quality in post-mortem examination of the cardiovascular system in great apes
- Standardise and maximise information gathering
- Facilitate comparative study between post-mortem findings and relevant samples
- Permit consistent sampling of the heart for subsequent examination by a designated pathologist

It is our ambition that all veterinarians and pathologists will follow these guidelines when performing post-mortem examination of great apes within EAZA collections.

If you still wish to use your own pathologist for examination of the heart, please refer them to us for a copy of our full cardiac post-mortem examination protocol.

When post-mortem examination of the cardiovascular system is performed, it is requested that photographs are taken at all stages of the process, and in particular of any abnormalities.

If the abnormal accumulation of fluid is noted at any stage, it should be quantified (in ml, or weighed if clotted), characterised (colour, consistency, specific gravity) and where possible a sample stored.

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## 2. Identifying Information

All information requested in the sample submission form should be provided. All photos, paperwork and samples must be clearly labelled with patient identifying information, including the following:

- Studbook number
- Species (and subspecies if known)
- Individual institution ID (name/number)
- Zoological collection
- Date of birth
- Date of death

## 3. Supporting Documents/Files

A copy of the full post-mortem report or summary of the findings elsewhere in the body should also be sent, where possible. Other documents of use are;

- Copy of the animal's clinical history
- Photographs taken during the post-mortem examination;
  - Ideally photographs should be taken at all stages of the examination
  - Photographs of any abnormalities found are of particular use

## 4. Sending Samples

For full guidance, please refer to the document entitled “Important information for sending samples” on our [website](#).

We are pleased to be supported by the EAZA Biobank, whose Copenhagen hub now acts as a collection point for Ape Heart Project samples within Europe. If you are outside the UK and EU, CITES permits may be needed – please contact the project coordinator on the email address below to discuss.

Once the heart is fully fixed (4-12 days minimum, depending on size), remove it from the formalin (this can be re-used) and wrap it in saline soaked gauze swabs or similar to prevent desiccation. Double bag the sample and place it into a sealed container that is big enough to ensure the heart is not squashed. Serum is very valuable to the research we are conducting on vitamin D and cardiac biomarkers. If available, please send serum (0.5–2ml) from the animal, even if this was taken months/years prior to death. Formalin-fixed hearts should be stored and sent at room temperature. Any other samples should be stored frozen and sent chilled with an ice pack/cooler where possible. See **Figure 1** for a summary of useful samples.

## 5. Report of Findings

We aim to provide a full written report of macroscopic and histopathologic findings (by email) to the submitting zoo/vet within 8 working weeks of receipt of samples at Twycross Zoo. Please note that samples sent via the EAZA Biobank will likely be received in batches therefore it may take longer to perform analyses. We are happy to update you on the progress in the meantime – just get in touch via email!

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## Protocol for Post-Mortem Examination and Sampling of the Cardiovascular System of Great Apes

STEP	ACTION	EXPLANATORY NOTE
1	<b>Weigh the animal</b> <b>Body condition score</b> <b>Measure crown-rump length</b>	Record the weight (kg) BCS scale 1-5 From top of head (crown) to the bottom of the buttocks (rump)
2	<b>Open the chest and examine the thorax</b>	Note appearance of lungs, pleural cavity etc. Assess for presence of lesions or fluid. Take photos of the heart 'in situ'
3	<b>Examine the pericardium</b>	Assess for lesions, thickening or fluid (if present, quantify, characterise and sample). If any changes detected, formalin-fix the pericardium and submit
4	<b>Remove the pluck</b>  <b>Remove heart from the pluck</b>	Check the anatomy of the great vessels before sectioning (especially in young animals). Cut the pulmonary trunk transversely 3cm above the pulmonary valve. Assess the lumen for thrombi. Transect all vessels as far from the heart as possible
5	<b>Take photos of the heart</b>	Wash/rinse the heart before taking photos of the heart from all sides
6	<b>Open the ventricles</b>	Make a single transverse incision through the lower third of the apex, perpendicular to the long axis of the heart – see <b>Figure 2</b> for clarification. This will ensure adequate formalin uptake into the myocardium
7	<b>Weigh the heart</b>	Remove any clots and rinse the heart before weighing. Record the weight (in grams)
8	<b>Sample the apical myocardium</b>	If sending via the EAZA Biobank, please take a portion ( $\leq 1 \times 1 \times 1$ cm) of the sectioned apical myocardium and immerse in non-methylated ethanol (70-99%). Store frozen until shipment is possible
9	<b>Fix the heart</b>	Fully submerge the heart (including the large portion of sectioned apex) in 10% neutrally buffered formalin, ensuring all surfaces are covered and there is sufficient formalin around the heart. Leave to fix depending on size for 4-12 days minimum
10	<b>Perform gross post-mortem examination of rest of carcass</b>	Open the entire aorta along its length to the level of the iliac bifurcation; sample and formalin fix any lesions. Examine the remaining major body organs as per the Great Ape TAG protocol and take relevant samples for histopathology. Take special note of the lungs, liver and kidney and where possible, also provide us with a formalin fixed sample ( $1 \times 1 \times 1$ cm) of these
11	<b>Contact us</b>	Email the project coordinator at <a href="mailto:heartproject@twycrosszoo.org">heartproject@twycrosszoo.org</a>
12	<b>Send the samples</b>	Refer to point 4 entitled "Sending Samples" above



**Don't forget to take photos at all stages of the cardiac post-mortem examination (include a scale marker)**



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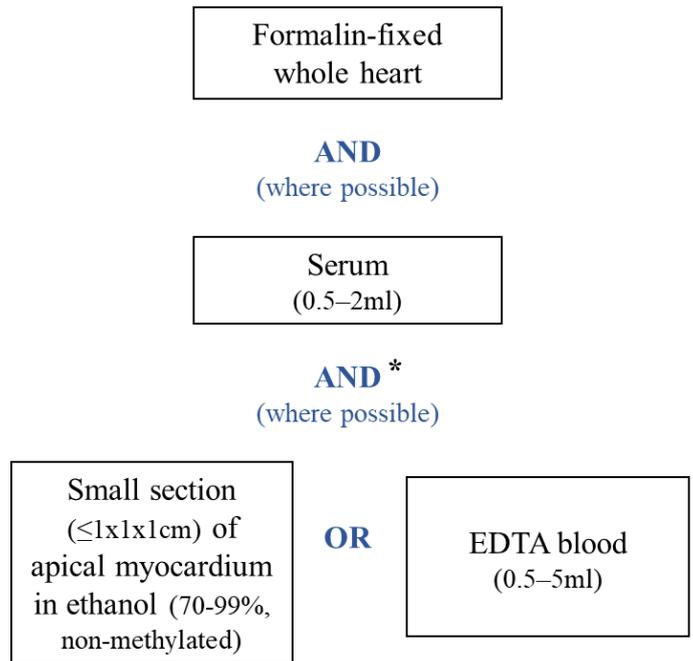
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**Figure 1:**

Shows the samples most beneficial to the Ape Heart Project and/or EAZA Biobank.

For full guidance, please refer to the document entitled “Important information for sending samples” on our website.



\* If sending via the EAZA Biobank

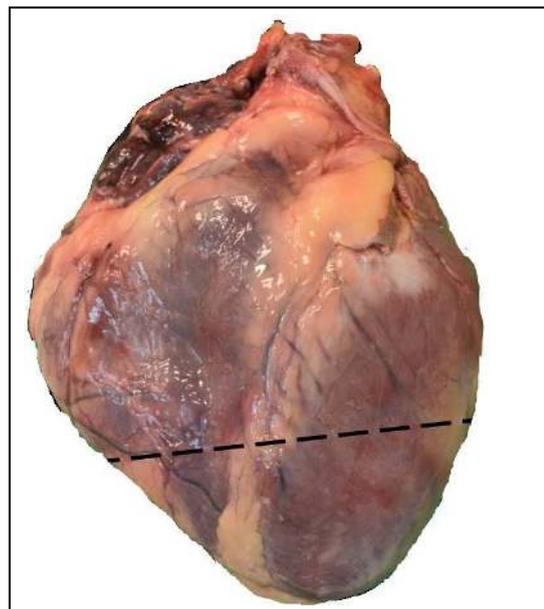
**Figure 2:**

Shows the approximate location of the transverse cut to be made across the lower third of the apex.

The cut should be perpendicular to the long axis of the heart exposing the chambers of both ventricles.

Please remove major post-mortem blood clots which may hinder adequate formalin fixation.

Please also fix and submit the sectioned apex of the heart.



**Don't forget to take photos at all stages of the cardiac post-mortem examination (include a scale marker)**



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