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Lesions found in the post-mortem reports of the Asian (*Elephas maximus*) and African (*Loxodonta africana*) elephants of the European Association of Zoos and Aquaria

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List of abbreviations

DIC	Disseminated intravascular coagulation
DNA	Deoxyribonucleic acid
EAZA	European Association of Zoos and Aquaria
EEHV	Elephant endotheliotropic herpes virus
EEHV-HD	Elephant endotheliotropic herpes virus haemorrhagic disease
ELISA	Enzyme-linked immunosorbent assay
PCR	Polymerase Chain Reaction
TAG	Taxon Advisory Group
TB	Tuberculosis

Introduction

Asian (*Elephas maximus*) and African (*Loxodonta africana*) elephants are the largest terrestrial mammals found in the European Association of Zoos and Aquaria (EAZA). A thorough study of the post-mortem reports, highlighting the lesions found, was conducted. The study compromises reports from 1990 until 2018, inclusively. In the past, most scientific papers focused on the necropsy reports of either a single elephant with a specific disease or a group with similar symptoms in the same zoo. One study conducted in the United Kingdom focused on the causes in the United Kingdom between 1995 and 2015. A comparable study was not found in North America.

Separating lesions into age groups, as well as organ systems, can help understand the pathogenesis of diseases, as well as improve the already high-quality care in a specific age group, gearing it towards their common problems. Although common problems that come with age, such as arthritis and a general decrease of the immune system or the cardiovascular system, are to be expected, some of these problems may be overlooked, or, if caught early enough, they could even be slowed.

As elephants are not free from diseases, infectious agents also need to be considered. In recent years, the topic of conversation in the Asian elephant community is the elephant endotheliotropic herpes virus (EEHV), and more specifically the acute EEHV- haemorrhagic disease (EEHV-HD). The first reported case of EEHV was in a circus in Switzerland in 1988 (Ossen *et al*.,1990). Another important infectious problem in the elephant community is tuberculosis (TB) as well as clostridial diseases. The *Mycobacterium tuberculosis* complex in elephants is most often caused by *Mycobacterium tuberculosis* and to a lesser degree by *M. bovis*.

The musculoskeletal system is also an important organ system to discuss, especially when taking into consideration the animals' management factors such as space, diet, flooring and footcare. Common lesions indicative of arthropathy, arthritis, osteoarthritis, as well as osteoporosis are found in all aging animals. The most recent extensive study on the foot health of Asian elephants in the European Zoos highlighted the most common pathological lesions with detailed pictures for specific grading (Wendler *et al*., 2019). Their reasoning for foot and pad lesions included unnatural pressures on the nail, hard surfaces, overgrowth as

well as trauma, obesity, repetitive stereotypic movement and leg malalignment. Other research found that pressure on the whole foot increases with body mass where the front legs have a higher-pressure magnitude than the back legs; most pathological lesions were situated on the lateral toes of the front feet (Panagiotopoulou *et al*., 2012). As elephants grow, the region impacting the floor shifts from a larger contact area of the entire plantar and palmar area to a shift cranially. In the past, digits 3 to 5 had the most common occurrence of pathologies. The same conclusions were reached by a more recent study, adding that most of the pathological lesions occurred on the lateral nail on both front feet but not on the rear feet (Wendler *et al*., 2019).

Increasing with age are also the number of lesions found in the cardiovascular system. These include, but are not limited to cardiovascular failure, atherosclerosis, or an arrhythmia. In the digestive tract, lesions are most often due to an alimentary reason, but a rupture, ulcer or torsion can also be a source of infection.

The elephants' increased health and immune system will ensure a positive breeding program and survival of the species. Conservation breeding programs, needed to avoid extinction of the species, prove to be very important. These programs ensure the survival of a certain species, by selective breeding.

Aims

The aim of this study is to identify the most common lesions in the post-mortem reports of the African and Asian elephants of the EAZA, group them into age based as well as organ system-based lesions and compare within. The objective is to increase our understanding of the lesions and subsequent mortality causes within the age groups, as well as organ systems, to be able to gear the medical care more specifically. Understanding the lesions, and therefore further understanding the pathogenesis of the diseases, would provide more insight to the veterinary medical community. These conclusions will then further improve the quality medical care of the elephants in captivity.

Materials and Methods

For this retrospective study, the main tools used consisted of E-Mails, Microsoft Word and Microsoft Excel Spreadsheets. First, the relevant list of Asian and African elephants, which are part of the EAZA breeding programs, was collected with the help of the elephant Taxon Advisory Group (TAG) advisors. From this, elephants that died between the years 1990 and 2018 were extracted and matched with the relevant zoos. From the existing database of the post-mortem reports, the relevant reports were taken, and the missing ones identified. E-mails were sent out to each zoo asking for the missing necropsy report.

After receiving the missing reports, the information was added to the Excel sheet, extrapolating the lesions deemed most significant by the veterinarian performing the autopsy from the post-mortem examination. Asian and African elephants were kept separate; merely comparing on a species level. The study looked at the overall demographic of each species, placing the elephants into different age groups and defining the most common lesions which were then grouped into organ systems or fatal events.

Results

First, the different age groups will be discussed and, consequently, a classification based on the organ systems will follow. Infectious causes, although not a specific lesion, was also created as an organ group. The neonatal and organ specific results will be explained in detail in Chapter 3 for both the Asian and African elephants together.

Chapter 1: *Elephas Maximus:* Asian elephants' Post-Mortem lesions

Of the 378 Asian elephant reported deaths in the European Association of Zoos and Aquaria, 226 (59.78%) individuals were included in this study.

1.1 Asian elephant Sex Distribution

Asian elephants consisted of 64.60% females (n=146), 30.00% males n=68) and 5.30% unknown (n=12).

1.2 Asian elephant Age Groups

The group of Asian elephants was then split into neonatal, which included any individual under one, as well as any abortion, and subsequent five and ten-year increment age ranges, to identify the most common lesions based on their age. Figure 1 shows the distribution of deaths based on ages.

Figure 1: Asian elephants classified into Age Groups

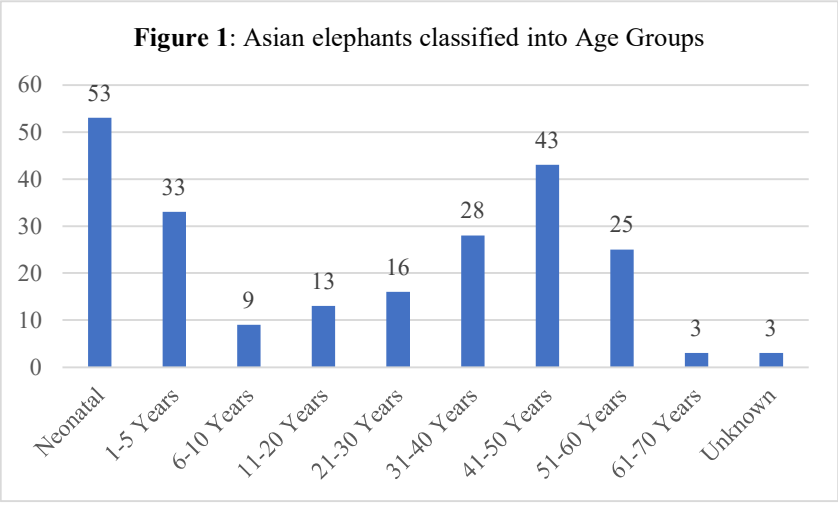

Figure 1 Asian elephant deaths classified first showing neonatal deaths, then five-year increments, and then subsequent ten-year increments.

These findings were then put into Table 1 that lists the organ systems in which the predominant lesions were found, in decreasing order, without repeating one individual for one organ system. If the lesions were due to a known infectious agent, only the infectious agent

was listed, not the associated lesions. For each age group the organ systems were selected that had the most lesions, including the single incidences only where space allowed. The last row, multiple organs in each age class, indicates how many individuals had lesions in more than one organ system.

Table 1: Asian elephant lesions and fatal events classified by Age

Neonatal	**53**	**1 Year – 5 Years**	**33**	**6 Years – 10 Years**	**9**
Stillbirth	13	Infectious	28	Infectious	3
Abortion	10	Circulatory System	4	Circulatory System / Anaesthesia	2
Killed by Dam / Not accepted	6	Digestive System	3	Digestive / Respiratory	1
Asphyxia in birth canal	4	Trauma	2	Musculoskeletal System	1
Multiple Organs	2	Multiple Organs	4	Multiple Organs	1
11 Years – 20 Years	**13**	**21 Years – 30 Years**	**16**	**31 Years – 40 Years**	**28**
Infectious	9	Urinary System	4	Circulatory System	9
Undetermined	2	Musculoskeletal System	4	Musculoskeletal System	8
Digestive / Circulatory System	1	Female Reproductive	3	Infectious	5
Female Reproductive System	1	Infectious / Respiratory	2	Female Reproductive	5
Musculoskeletal System	1	Neurological / Digestive / Anaesthesia	2	Respiratory / Digestive System	3
Multiple Organs	1	Multiple Organs	7	Multiple Organs	10
41 Years – 50 Years	**43**	**51 Years – 60 Years**	**25**	**61 Years – 70 Years**	**3**
Musculoskeletal System	18	Musculoskeletal System	8	Circulatory System	2
Respiratory System	8	Female Reproductive	7	Infectious	1
Circulatory System	7	Urinary	6		
Urinary System	6	Circulatory	6		
Infectious	5	Digestive / Respiratory System	5		
Multiple Organs	13	Multiple Organs	13	Multiple Organs	0
Unknown Age	**3**				
Infectious	2				
Musculoskeletal System	1				
Multiple Organs	0				

Table 1 For each age group the lesions and fatal events were grouped based on organ systems or if an infectious agent was found this was deemed the cause. Multiple organs indicates if an individual had significant lesions in more than one organ. Some of the categories had singular individual instances that could not be included in the graph due to space.

The details of the neonatal group will follow in Chapter 3.1 together with the African elephants.

1.2.1 Asian elephant Age 1-5

The third biggest number of Asian elephant deaths was in the age group of 1-5, totalling 14.60% of the deaths (33 out of 226). The most common cause of death was attributed to infectious diseases, more specifically EEHV-HD, responsible for 84.84% of deaths (n=28). This will be discussed in detail in Chapter 3.2.1. Four individuals showed circulatory lesions including large areas of multifocal petechial haemorrhages, reactive spleen as well as splenomegaly and eosinophilic intranuclear inclusion bodies in myocardial and endothelial cells. The liver lesions included acute hepatitis and hepatomegaly in the same individuals. These lesions are also seen in EEHV-HD but cannot be counted in this category, as some were not tested, or if tested no virus was found.

Two individuals had leg fractures, one also a spine fracture both cases leading to euthanasia. The last individual had an undetermined cause of death, but it was presumed to be a hypersecretory colibacillosis possibly in combination with meningitis.

1.2.2 Asian elephant Age 6-10

3.98% died between the age of 6 to 10 (n=9). Most of the lesions were due to infections, two due to EEHV and one due to Streptococcus agalactiae leading to multiple organ lesions including purulent endometritis and exudative fibrinous peritonitis. Circulatory lesions followed, including one individual with a dilation of the right ventricle, leading to cor pulmonale and subsequent cardiovascular failure. The other cardiovascular lesion was pericarditis with a peritonitis, chronic pneumonia and blockage in the small intestine. Two deaths were attributed to anaesthesia.

1.2.3 Asian elephant Age 11-20

Between 11 and 20 years of age 5.75% of Asian elephants died (n=13). Nine of these cases were due to infections caused by *Clostridium spp*. Five of these mortalities were due to *C. botulinum*, which occurred at the same facility within a 1-week period, one of which displayed the typical botulism signs: systemic vascular disorder with oedema, congestion, haemorrhages, thrombi as well hypoxic degeneration and necrosis of the affected tissues

Others included *C. perfringens*, *C. difficile* as well as *C. septicum*. Two causes of death were undetermined, one possibly due to septicaemia and subsequent shock with low bacterial count found. One of the individuals had osteoarthritis and chronic deforming arthritis

of the elbow and was therefore euthanized. Another individual had lesions in multiple organs which included peritonitis with a disseminated microthrombosis, possibly secondary to infarction and rupture of the uterus.

1.2.4 Asian elephant Age 21-30

16 individuals (7.08%) died between the ages of 21 to 30. Here, the most common lesions were found in the urinary and musculoskeletal system, followed by the female reproductive system. Respiratory, neurological, digestive systems as well as infections and anaesthesia all had the same number of lesions. Within the urinary system, one case of renal insufficiency with hypercalcemia, leading to mineralisation of all organs, led to the decision to euthanize this elephant. Other lesions here included a cyst in the kidney, nephrosclerosis and atrophy of the kidneys, all of which were found in individuals with multiple organs affected.

Musculoskeletal lesions included pododermatitis, severe arthrosis as well as chronic osteoarthritis and subluxation of the femur. Third most common lesions were found in the female reproductive system; two attributed to dystocia, with one case resulting in a fetotomy and a ruptured uterus. One case of leiomyoma was also found in this age group. In the digestive tract, there was one individual with blood in the stomach, a tumour in the pancreas, degeneration of the liver as well as inflammation of the mucous membrane of the intestinal tract and the other exhibiting ulcerative oesophagitis and gastritis.

Infectious agents included *Streptococcus agalactiae* resulting in vaginitis while another elephant was infected with *Clostridium* (unspecified) which led to euthanasia. One case of flaccid paralysis was observed, but was negative for toxins produced by *C. botulinum*; therefore, the cause of death was unknown. Two neurological lesions found were a cerebrospinal haemorrhage during mating and the other traumatic nerve changes, including pododermatitis and inflammation due to assumed immunosuppression. One case of euthanasia was due to multifocal haemorrhage, DIC, pulmonary fibrosis and oedema. Another animal being treated for foot problems died under anaesthesia.

1.2.5 Asian elephant Age 31-40

12.39% of all Asian elephants died between 31 to 40 (n=28). Most of the lesions were found in the circulatory system, followed by the musculoskeletal system and infectious agents. Other lesions were found in the female reproductive, as well as digestive and respiratory

system. Splenomegaly and subcapsular bleeding, as well as necrosis of the gastric fundus mucosa, with catarrhal inflammation of the jejunum in combination with circulatory failure led to a cardiac and subsequent circulatory failure. Other circulatory failures were attributed to trauma caused by herd mates, falls into moats and subsequent drowning, extreme heat as well as anaesthetic and drug related intolerance. All five of the female reproductive lesions were leiomyomas; one had a concurrent adenomyosis.

Musculoskeletal lesions included arthritis, pododermatitis, arthrosis and general foot problems. One elephant had a front foot ulceration. Other musculoskeletal lesions included synovitis of the right front foreleg as well as one elephant with synovitis in most of the main joints of the limbs. Another elephant showed multiple pelvic fractures including a femoral head fracture. One elephant had chronic weight loss after falling on the left knee, causing a permanent patella luxation and arthrosis. The last case of euthanasia was due to septicaemia. Three out of the five infectious agents were due to *Clostridium sp*. while two cases of cow pox were also present.

1.2.6 Asian elephant Age 41-50

For the Asian elephant, the second highest numbers of death cases occurred in the age group between 41 to 50 years, accounting for 19.03% (43 out of 226), with the most common lesions in decreasing order being: the musculoskeletal, respiratory, circulatory, urinary system as well as infectious.

In this age group, the most common lesions were associated with arthrosis, arthritis, osteoarthritis and pododermatitis. As the musculoskeletal system had the second most frequent found lesions in all the Asian elephants, it will be discussed in Chapter 3.3. Respiratory lesions included pulmonary oedema as well as pneumonia while circulatory lesions included cardiac failure and endocarditis. As far as infectious agents were concerned, enterotoxaemia secondary to enteritis as well as enterocolitis secondary to overfeeding were attributed to *Clostridium spp*. while a granulomatous pneumonia was attributed to *Mycobacterium tuberculosis*. Urinary system lesions included cystitis, interstitial nephritis, cysts in the kidney as well as one case of a missing kidney, although the implications of this were not mentioned. One case of kidney adenocarcinoma was indicated.

1.2.7 Asian elephant Age 51-60

11.06% of the Asian elephants died between 51 to 60 (n=25). Most of the lesions were found in the female reproductive system, musculoskeletal system, and urinary system. The circulatory system along with digestive and respiratory system also presented lesions.

Leiomyoma, leiomyosarcoma as well as fibropapilloma were found in the female reproductive system. In the musculoskeletal system lesions included ulcerative arthrosis, pododermatitis, panaritium, as well as osteoarthritis were found. Some were simply listed as locomotive problems or chronic foot and leg problems. Urinary lesions were common in this age group, including chronic interstitial nephritis, multiple cysts, together with intratubular concrements of varying sizes. Chronic renal interstitial fibrosis, chronic nephropathy and nephritis were also found. One case of necrotizing nephritis was due to an Escherichia coli infection. One case of cystic adenomas of the kidneys was found but was deemed not relevant.

Circulatory lesions included myocardosis and coronary thrombosis with a pulmonary embolism as well as coronary atherosclerosis. One case of cardiopulmonary failure was attributed to age related vascular injury and interstitial fibrosis. Pronounced atherosclerosis, myocardial interstitial fibrosis along with a hyperplasia of the tunica media were found leading to circulatory failure. Hydropericardium, cardiac changes due to Selenium and Vitamin E deficiency, together with a cirrhotic spleen and hepatic protozoal cysts were all determined in one individual.

Other digestive lesions included chronic diffuse haemorrhagic ulcerative to fibrinous necrotizing inflammation in the small intestine inflammation, acute, erosive-ulcerative inflammation of the stomach, with isolation of *Clostridium perfringens* and *E. coli*. Metastatic carcinoma of the duodenum was also found, with one case of multifocal impaction of food material leading to a sub-serosal haemorrhage and compensatory hypertrophy of muscular wall. One case of diminished molars led to a decrease in food intake. Respiratory lesions often accompanied cardiovascular lesions including pulmonary embolism, granulomatous pneumonia, as well as lung fibrosis and sclerosis.

1.2.8 Asian elephant Age 61 to 70

Three individuals (1.33%) died between 61 and 70 years. Two of the individuals died due to cardiovascular failure while the last case was due to *Mycobacterium tuberculosis*.

1.2.9 Asian elephant Unknown Age

Within the Asian elephants, 1.33% of the age was undetermined (n=3). Two deaths were attributed to a *Clostridium* infection; the other individual in this age group displayed hoof cancer on both front feet.

1.3 Asian elephant Organ Systems

To represent the Asian elephants as a group, the lesions were grouped based on their organ systems. Figure 2 shows the prevalence of lesions found in each organ system, each animal only counting once per organ system. Infectious was used here as an organ system and if this was the cause of death, other lesions were not included in another organ system. Anaesthesia was also added as a group, as there was always a reason for the procedure. Anaesthetic death is described in detail in Chapter 3.4 with the African elephants. "Other" includes lesions that did not fit into an organ system.

Figure 2: Asian elephant lesions categorized by Organ System

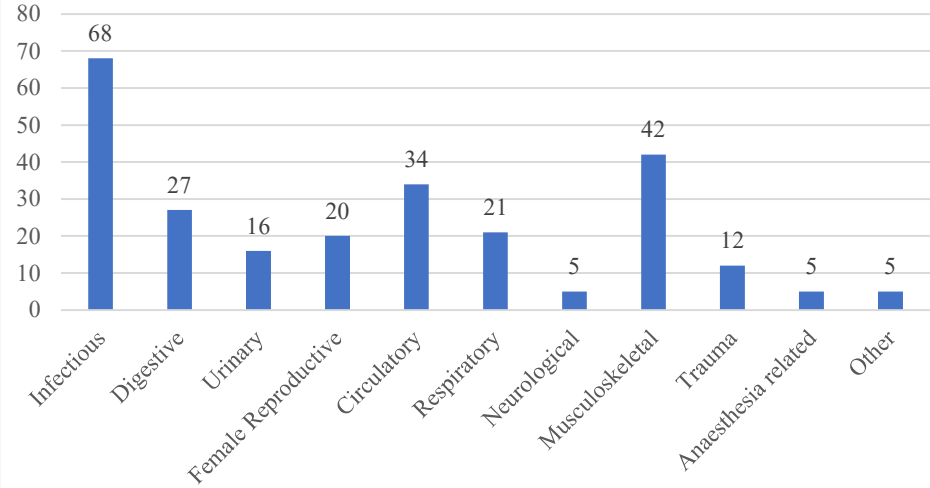

Figure 2 The lesions were classified based on organ system to show the prevalence. Infectious was included if an infectious agent was deemed responsible. The last category "other" includes any lesion that did not fit into an organ system.

For each organ system, the top 5 common findings were chosen and placed into Table 2. For some of these, there were many singular events and as these would not fit, they were left out of this chart. The details on the organ system are listed in Chapter 3.2, alongside the African elephants. Within the organ system, some lesions were simply recorded as the inflammation of an organ, without a specific agent, so they were included here.

Table 2: Common findings in Asian elephants in the Organ Systems

Digestive System	**27**	**Urinary System**	**14**	**Female Reproductive**	**17**
Hepatopathy	11	Renal Cyst	6	Leiomyoma	11
Inflammation	6	Nephritis	4	Endometrial Hyperplasia	2
Gastric Ulcer	3	Renal failure	2	Dystocia	2
Small intestine block / torsion	2	Fibrosis	1	Rupture of uterus	2
Petechiae / Peritonitis	2	Sclerosis	1	Cyst in uterus	1
Circulatory System	**34**	**Respiratory System**	**21**	**Neurological System**	**5**
Cardiovascular failure	13	Pneumonia	6	Craniocerebral Trauma	1
Atherosclerosis	4	Pulmonary Oedema	5	Epileptic Shock	1
Petechiae	4	Pulmonary Fibrosis	4	Meningoencephalitis	1
Miscellaneous	4			Miscellaneous	1
				Traumatic Nerve Damage	1
Musculoskeletal System	**42**	**Trauma**	**12**		
Arthropathy	19	Fracture	7		
Arthrosis	12	Attacked by Dam	4		
Pododermatitis	10	Attacked by Herd Mate	1		
Foot Problems	5	Miscellaneous	1		
Osteomyelitis / Synovitis	2				

Table 2 The table shows the top 5 most common lesions found within the organ system. Due to space not all lesions, especially singular events, were able to be included in this chart. Inflammation of an organ, without a specific agent, was often a finding and was therefore also included here.

1.3.1 Asian elephant Infectious Agents

Since infectious causes constitute one of the most common findings, these were placed in Figure 3. Some of the infectious agents occurred only once, and were therefore grouped together as bacterial or viral. Infectious agents will be explained in more detail in Chapter 3.2.1, together with the African elephants.

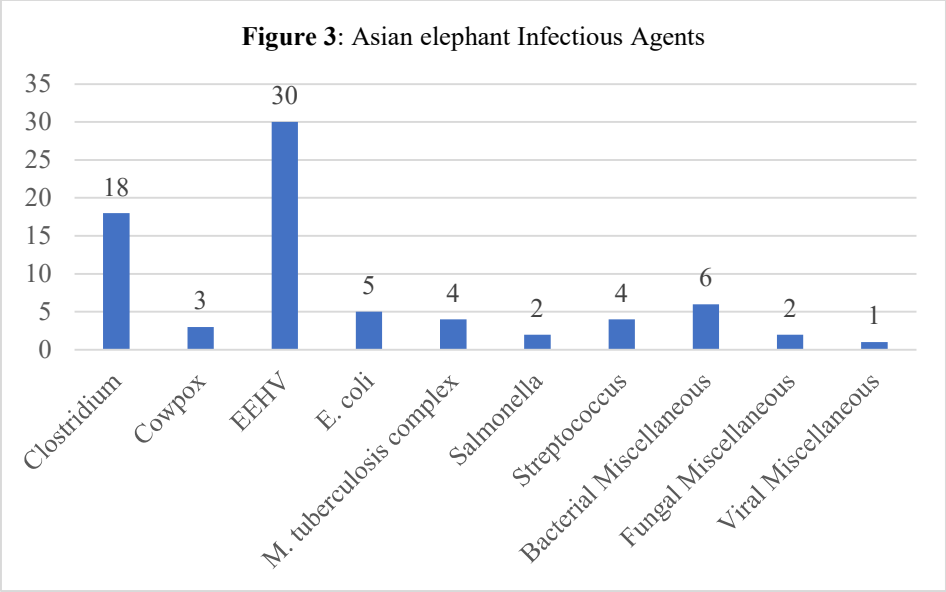

Figure 3 The infectious agents were identified and the most numerous were chosen, classifying the singular cases into broader categories.

1.4 Asian elephant Euthanasia

As some of the elephants were euthanised due to various reasons, Figures 4 and 5 were created to show the reasons. The undetermined category was designed to include anyone that did not have a good quality of life and on post-mortem examination not one specific reason could be picked.

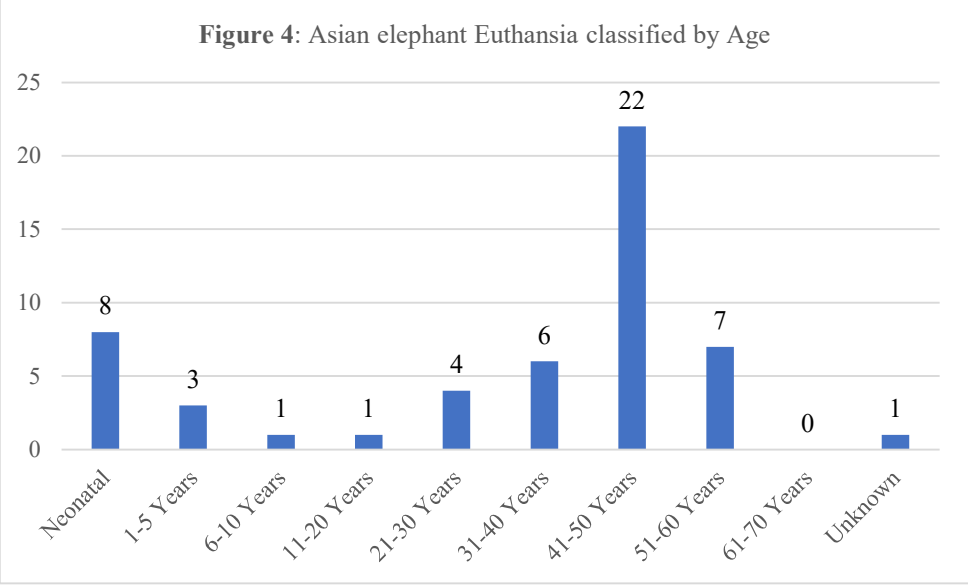

Figure 4 Euthanasia cases classified by age, first by neonatal, then five-year increments and subsequent ten-year increments.

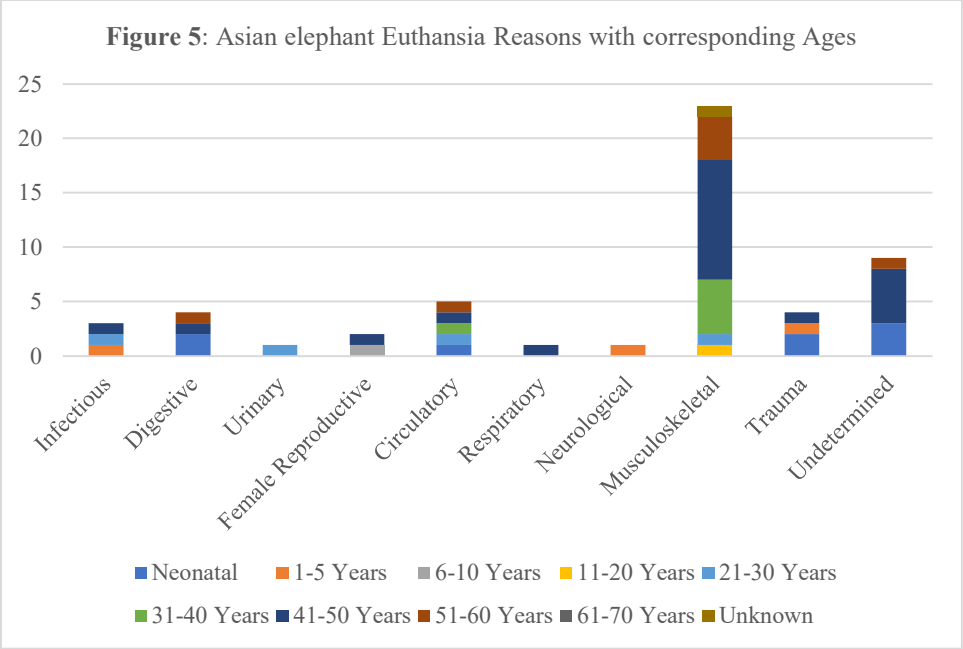

Figure 5 The reasons for euthanasia were classified based on organ system and distinguished by age

Chapter 2: *Loxodonta Africana*: African elephant Post-Mortem Lesions

For the African elephants, the same type of classification system was used. In total 110 post-mortem reports were included in this study; 59.14% of all African deaths between 1990 and 2018 (186 total).

2.1 African elephant Sex Distribution

In this study, 41.94% were females (n=78), 23.63% were males (n=26) and 5.45% (n=6) were unknown.

2.2 African elephant Age Groups

The distribution of the number of elephants in each age group is shown in Figure 6.

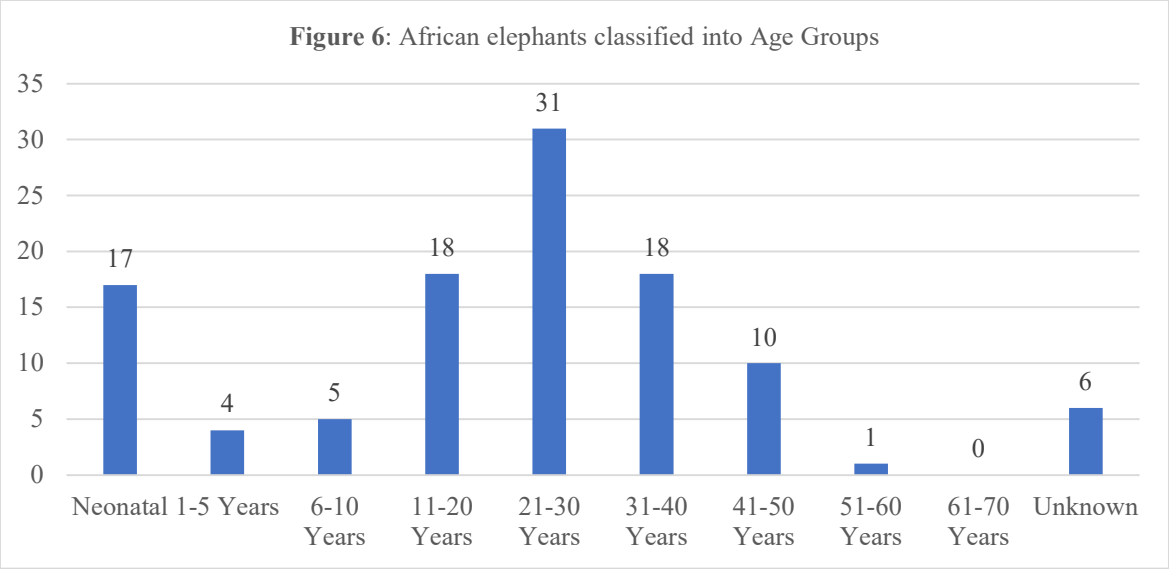

Figure 6 African elephant deaths classified first showing neonatal deaths, then five-year increments, and then subsequent ten-year increments.

In each age group, the most common lesions in the organ systems were collected and displayed in Table 3, just like with the Asian elephants. For each age group the organ systems were selected that had the most lesions, including the single incidences only where space allowed.

Table 3: African elephant lesions and fatal events classified by Age

Neonatal	**17**	**1 Year – 5 Years**	**4**	**6 Years – 10 Years**	**5**
Stillbirth	6	Circulatory System	2	Infectious	3
Abortion	5	Digestive System	2	Digestive System	1
Not accepted / attacked by dam	5	Infectious	1	Musculoskeletal System	1
Pleuritis	1			Circulatory System	1
Multiple Organs	0	Multiple Organs	1	Multiple Organs	2
11 Years – 20 Years	**18**	**21 Years – 30 Years**	**31**	**31 Years – 40 Years**	**18**
Digestive System	6	Circulatory System	8	Digestive System	7
Circulatory System	4	Infectious	8	Circulatory System	5
Musculoskeletal System	4	Trauma System	6	Infectious	4
Infectious System	3	Other	5	Trauma / Respiratory	2
Urinary / Trauma / Neurological	2	Digestive System	4	Musculoskeletal System	1
Multiple Organs	5	Multiple Organs	6	Multiple Organs	4
41 Years – 50 Years	**10**	**51 Years – 60 Years**	**1**	**61 Years – 70 Years**	**0**
Digestive System	4	Circulatory System	1		
Circulatory System	4				
Respiratory / Other	3				
Infectious / Urinary	2				
Musculoskeletal System	2				
Multiple Organs	4	Multiple Organs	0		
Unknown Age	**6**				
Female Reproductive System	2				
Urinary System	1				
Digestive / Infectious	1				
Other	1				
Trauma	1				
Multiple Organs	0				

Table 3 For each age group the lesions and fatal events were grouped based on organ systems or if an infectious agent was found this was deemed the cause. Multiple organs indicates if an individual had significant lesions in more than one organ. Some of the categories had singular individual instances that could not be included in the graph due to space.

The neonatal age group will be discussed in Chapter 3.1 with the Asian elephants.

2.2.1 African elephant Age 1 to 5

Four individuals died between the ages of 1 to 5 (3.63%). The main lesions found were in the circulatory and the digestive system as well as one infectious agent. Digestive lesions included haemorrhagic enteritis, while another showed gastritis leading to an extended anorexia and subsequent undernutrition as well as subendocardial petechiae on the left ventricle. One individual had multiple malignant lymphoma while another individual had a viral infection with lymphocytic tropism: lymphocytic adenitis, hepatitis and myocarditis. The exact viral agent was not determined.

2.2.2 African elephant Age 6-10

Five individuals (4.54%) died between the ages of 6 and 10. Infectious reasons accounted for 60% (n=3), two due to *Mycobacterium tuberculosis* leading to granulomatous pneumonia along with a necrosis in the liver and subsequent right sided heart failure, and one due to *Salmonella typhimurium type B,* which lead to enteritis and septicaemia. Two elephants were euthanized, one due to chronic arthritis and the other due to cholangiohepatitis, centrilobular hepatic congestion as well as splenic congestion. Chronic arthritis at such young age was an outlier here.

2.3.3 African elephant Age 11 to 20

16.36% deaths were in the age group 11 to 20 years (n=18). The digestive, circulatory, musculoskeletal system as well as infections were found to have the most lesions attributing to death. In the digestive system, one case of a mesenteric torsion of the caecum and colon lead existed. Another individual had marked haemorrhages, oedema together with multifocal necrosis and thrombosis in the caecum. One elephant had erosive gastritis, duodenitis, ileitis, faecal impaction along with underlying pleuritis and sero-haemorrhagic pneumonia. This led to pulmonary oedema and acute heart failure. Another elephant showed signs of intestinal perforation. One individual showed an acute and degenerative hepatitis, associated with intrahepatic cholestasis as well as nephrosis and extensive myopathy.

In the musculoskeletal system, one case of a fracture of the humerus, leading to lesions in the nervus radialis and subsequent muscle atrophy of the musculus extensor carpi radialis. Another individual fractured her tibia. The third individual ruptured the left lateral ligament in the elbow joint, had a total detachment of the bone fragment from the humerus and dislocated

the other elbow as well. All three cases lead to euthanasia. Another individual had degenerative arthritis, especially in the stifle joint.

Infectious agents included *Mycobacterium tuberculosis*, leading to granulomatous pneumonia as well as one case of acute myocardial necrosis, multifocal haemorrhages and a septicaemia due to *Citrobacter freundii*. One elephant was infected with the Encephalomyocarditis virus, causing abdominal and pericardiac effusion, pulmonary oedema plus systemic microthrombi with haemorrhages.

2.2.4 African elephant Age 21 to 30

28% occurred between 21 to 30 years (n=31). The most common lesions were found in the circulatory system, due to an infectious agent, trauma or in the digestive system. Within the circulatory system, myocardial infarction was the most common reason, followed by cardiovascular failure and septicaemia. Tuberculosis constituted the most common infection, compromising five cases. Two of these cases, showing lymphadenitis and generalized tuberculosis, were confirmed *Mycobacterium tuberculosis*. One was a presumed *M. tuberculosis* case and two cases of macrofocal pulmonary tuberculosis did not have a specific agent listed. One of these elephants died during anaesthesia. The underlying macrofocal pulmonary tuberculosis most likely contributed to its death. Tuberculosis will be further discussed in the infectious diseases section, Chapter 3.2.1.

As far as trauma was concerned, three cases of trauma occurred due to attack by a conspecific and a further three due to other trauma. Cachexia, sinusitis and otitis were some of the other nonspecific lesions found.

2.2.5 African elephant Age 31 to 40

16.36% of the deaths occurred between 31 to 40 (n=18). Most of the lesions were either in the digestive system, followed by the circulatory system and infections. Trauma, respiratory and musculoskeletal lesions were least common. Digestive lesions included one case of chronic erosive gastritis and colitis, with mesenteric lymph node thrombosis as well as underlying aorta atherosclerosis leading to acute pulmonary oedema. Circulatory lesions included acute cardiac haemorrhages, possibly due to vascular abnormalities. There was one case of hepatic congestion, myocardial fibrosis as well as pulmonary alveolar hemosiderosis. Extensive petechiae on the endocardium were found in one individual, while another was

determined to have had a cardiac arrest. One case of cardiac and subsequent circulatory failure, showing the classical lesions of shock, was due to the infectious agent *Streptococcus agalactiae*. *Pseudomonas spp.* was discovered in a case of a caecal perforation and subsequent peritonitis. Another case of peritonitis was due to a rupture of a colon ulcer. Intussusception followed by necrosis of the intestines were other significant lesions found.

Two cases of *M. tuberculosis* were reported, one leading to euthanasia due to extensive lesions in the lung and a generalized oedema. Musculoskeletal lesions included proliferative osteoarthropathy, with periarticular osteophytes, joint mouse formation, subchondral bone cysts and villous synovial proliferation were detected, being consistent with degenerative joint disease. Trauma included a head trauma with a cervical fracture. This age group also had two cases of conspecific attacks.

2.2.6 African elephant Age 41 to 50

10 individuals (9.10%) died between the ages of 41 to 50. Most of the lesions in this age group were in the digestive and circulatory system, closely followed by the respiratory system, infectious agents as well as musculoskeletal and urinary system.

Within the digestive system, fibrinosuppurative typhlitis, with necrotic and severe multifocal suppurative periodontitis as well as acute and chronic gastric haemorrhages, leading to marked multicentric congestion and pulmonary oedema were seen. Other lesions included a chronic granulomatous necrotizing colitis as well as chronic proliferative fibrous peritonitis. One elephant had a volvulus greater than 360 degrees in the small intestine, with haemorrhages and evidence of *Clostridium perfringens*.

Circulatory lesions were comprised of myocardial fibrosis as well as hemosiderosis of the spleen, atrophy of the lymphatic tissue as well as a generalized micro-thrombosis and haemorrhages. One elephant displayed an acute hypoxic myocardial degeneration, vasculitis as well as thrombosis and acute systemic vascular injury. These lesions were deemed to have a possible connection to the epidermal necrosis with inflammation. Pulmonary oedema represented itself with severe congestion on the left lungs. *Mycobacterium avium subspecies hominisuis* was found in one individual. In the musculoskeletal system osteoarthritis was most common while the urinary system had adrenal necrosis and renal thrombosis.

2.2.7 African elephant Age 51 to 60

One individual died in this age group: hypoxia due to a cardiovascular collapse.

2.2.8 African elephant Age 61 to 70

There were no African elephants between the ages of 61 to 70.

2.2.9 African elephant Unknown Age

Six individuals died of unknown age, accounting for 5.45%. Most of the lesions were found in the female reproductive system, while other organ systems included: the urinary system, digestive system, infectious or due to trauma. One elephant had a uterine tumour, while another had a uterine leiomyoma with effusion in the abdominal and thoracic cavities, as well as hepatomegaly and renomegaly. These lesions were attributed to gaseous gangrene, most likely due to *Clostridium*. One other infectious agent was determined: *M. tuberculosis*. One individual had severe femoral and pelvic fractures, while another died from cardiac arrest during sedation for lameness.

2.3 African elephant Organ Systems

Figure 7 shows the most common lesions based on the organ systems. This was created the same way the Asian Organ System chart was.

Fgure 7: African elephant lesions categorized by Organ System

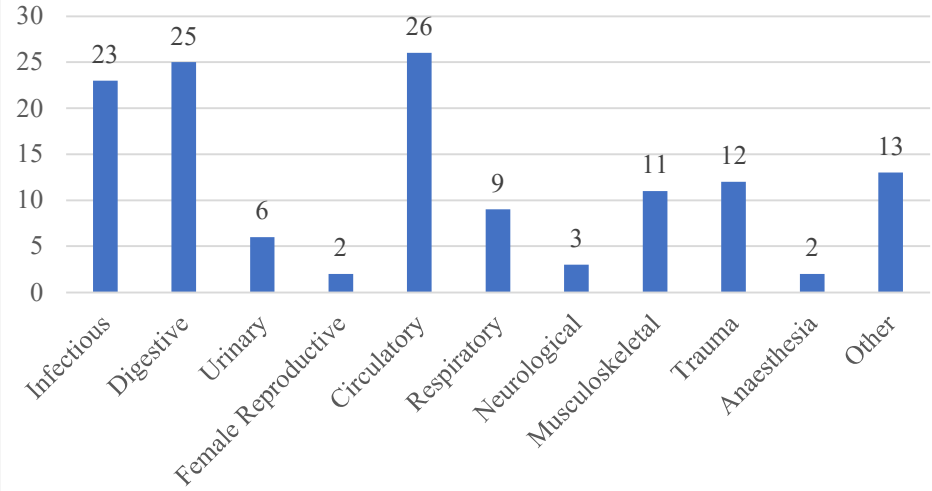

Figure 7 The lesions were classified based on organ system to show the prevalence. Infectious was included if an infectious agent was deemed responsible. The last category "other" includes any lesion that did not fit into an organ system.

Table 4 outlines the 5 most common lesions in each organ system, in order of decreasing incidence. The details will be discussed in Chapter 3.2 with the Asian elephants.

Table 4: Common lesions in African elephant in each Organ System

Digestive System	**25**	**Urinary System**	**6**	**Female Reproductive**	**2**
Inflammation	11	Adrenal Necrosis	1	Leiomyoma	1
Hepatopathy	7	Glomerulonephritis	1	Uterine Tumour	1
Haemorrhages	3	Renal Cyst	1		
Ulcers	2	Renal Thrombosis	1		
Perforation	2	Nephromegaly / Nephrosis	1		
Circulatory System	**26**	**Respiratory System**	**9**	**Neurological System**	**3**
Myocardial Infarct	4	Pulmonary Oedema	4	Brain congestion	1
Splenopathy	4	Pleuritis	2	Leukoencephalomalacia	1
Cardiac Petechiae	4	Pulmonary Haemorrhages	1	Dilated aquaductus mesencephalus	1
Septicaemia	3	Pneumonia	1	Traumatic nerve changes	1
Circulatory Failure	2	Congestion	1		
Musculoskeletal System	**11**	**Trauma**	**12**		
Athropathy	7	Fracture	7		
Athrosis	2	Attacked by Herd Mate	5		
Elbow Luxation	1	Miscellaneous	1		
Ligament Rupture	1				
Myopathy	1				

Table 4 The table shows the top 5 most common lesions found within the organ system. Due to space not all lesions, especially singular events, were able to be included in this chart. Inflammation of an organ, without a specific agent, was often a finding and was therefore also included here.

2.3.1 African elephant Infectious Agents

Out of the 23 infectious agents for the African elephants, 52.17% (n=12) were attributed to *M. tuberculosis* complex, 30.43% (n=7) due to bacterial infection, 8.69% (n=2) due to *Salmonella spp*. infection and 8.69% (n=2) due to a viral agent.

2.4 African elephant Euthanasia

Just as with the Asian elephants, the euthanasia reasons are highlighted below in Figures 8 and 9.

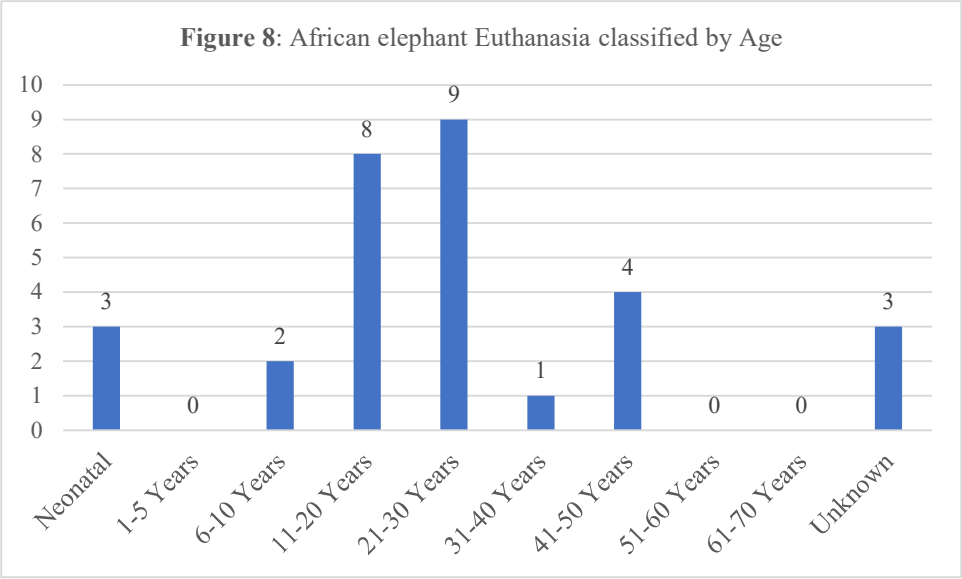

Figure 8 The euthanasia deaths were classified based on age, first distinguished by neonatal age, then five-year increments and then ten-year increments.

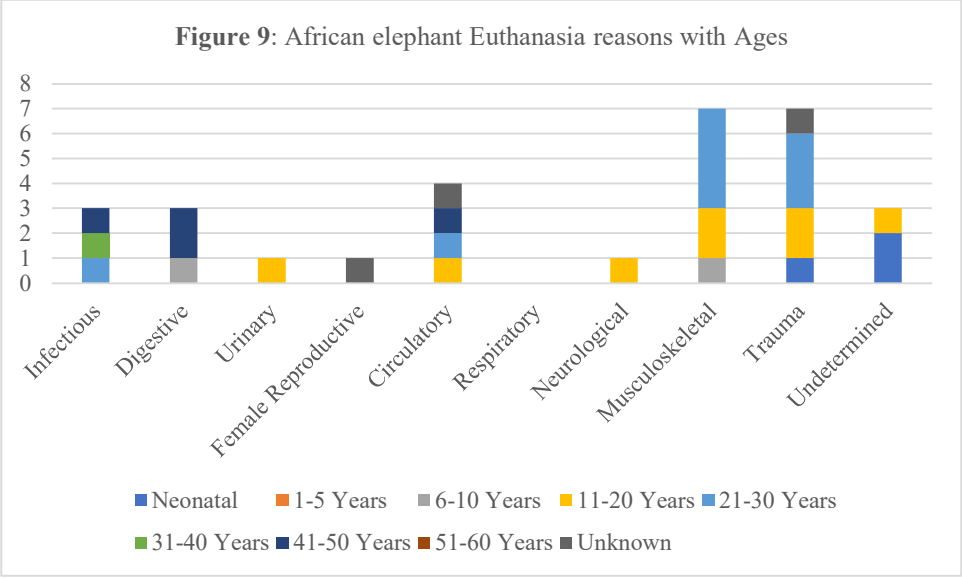

Figure 9 The reasons for euthanasia were classified based on organ system and a distinction was made corresponding with age

Chapter 3: Asian and African elephants

The neonatal age group, as well as organ system lesions, neoplasia, anaesthesia and euthanasia will be discussed together for the species.

3.1 Neonatal Age

The neonatal age group includes any individual under one, as well as abortions and stillbirths. 23.45% of the Asian elephants (n=53) were found in this age group, while for the African elephants this accounted for 15.45% (n=17). For the Asian elephants, this was the largest group whereas for the African elephants it was tied for the third most common age group.

Stillbirth, born in the correct gestation frame but showing no observed vital signs, accounted for 24.52% of the Asian neonatal deaths (13 / 53) corresponding with 5.75% of all Asian elephants' deaths (13 / 226). While for the African elephants, these percentages were 35.29% (6 /17) and 5.45% (6 out of 110) respectively.

Any death occurring before the gestation time of 18-22 months for Asian elephants was marked as abortion, accounting for 18.86% (10 out of 53) of the Asian neonatal deaths (Britannica, 2021). Four abortions occurred due to co-infections of *E. coli hemolytica* and *Mannheimia hemolytica*, and of *Aspergillus* and *Listeria*. One Orthopox (Cowpox) virus infection of the foetus resulted in pustules dispersed over the entire body. Another individual showed an intravascular bacterial proliferation. In one abortion case, where the foetus was expected to be about five months, only a blood clot was found in the uterus. The causes of abortion in the other cases were undetermined. For the African elephants, with an average gestation of 22 months (Britannica, 2021), this percentage was slightly higher at 35.29% (6 /17); one being due to a *Salmonella* infection. The rest were unknown.

Five new-born Asian elephants (14.28%) died right after birth because the mother, grandmother or another herd mate killed the calf. For the African elephants, one case of aggression by the mother was listed, resulting in euthanasia due to extensive trauma (5.88%). One of the Asian elephant calves was not accepted by the dam and hand reared but died after about four months due to critical fluid loss as a result of a *Salmonella spp.* infection. Two African elephant calves were also not accepted by the dam and subsequently hand reared. They died within the first year.

3.2 Organ Systems

In the Asian elephants, infectious diseases were the most common causes of lesions, followed by musculoskeletal lesions and circulatory lesions. In the African elephants, most lesions were seen in the circulatory system, followed by the digestive system and then lesions caused by infectious agents.

3.2.1 Infectious causes

Infectious causes accounted for 25.66% (68 / 265) of the lesions in the Asian elephants, the most common, while in the African elephants this was 16.79% (23 / 137), the third most common. The most common infectious diseases were EEHV in Asian elephants, while tuberculosis and clostridiosis was found in both species

3.2.1.1 Elephant Endotheliotropic Herpes Virus

EEHV-HD comprised the most common infectious agent in the Asian elephants. There were a total of 30 EEHV-HD cases, EEHV1a accounted for 73.33% of the cases (n=22) while EEHV1b, non-specified EEHV and EEHV4 each accounted for 6.67% (n=2). EEHV1 and EEHV5 accounted for 3.33% each (n=1).

3.2.1.2 Tuberculosis

In the Asian elephants, three cases of tuberculosis were found, two due to *M. tuberculosis* while one case of *M. bovis* was determined. 12 cases of *Mycobacterium sp.* were discovered in the African elephants. Out of the 12 cases, 91.67% (11/12) were described as tuberculosis, and one as *Mycobacterium avium hominisuis*. Out of the tuberculosis cases, 72.72% (8/ 11) were caused by *M. tuberculosis*, the other three had no specific agent listed; one was presumed as *M. tuberculosis*.

3.2.1.3 Other Infectious Agents

For the Asian elephants, Clostridial diseases were the second most common infection. 10 out of the 18 cases were classified, which included 5 cases of *C. botulinum* (27.78%), 4 cases of *C. perfringens* (22.22%), one of these being Type A, and one cases of *C. septicum* (4.54%). The African elephants showed one incidence of *Clostridium perfringens* while one case of gaseous gangrene was mentioned, without the exact causative agent. In the Asian elephants, the third most common infectious agent was *Escherichia coli*. Salmonellosis attributed to death in 2 African: one stillbirth and one gastroenteritis along with septicaemia.

In 2 Asian elephants, salmonellosis was found in a case of sepsis and another with severe diarrhoea.

3.2.2 Musculoskeletal Lesions

In our study for the Asian elephants, musculoskeletal lesions were the second most common lesions seen at 15.84% (42 / 265). In decreasing order, these consisted of arthropathy, arthritis and pododermatitis. In African elephants, musculoskeletal lesions were not as common, accounting for almost 8.03% (11 / 137) with arthropathy and arthrosis being the most common lesion found, but no reports of pododermatitis.

Arthropathy for the Asian elephants included 12 findings of arthritis, 5 of osteoarthritis with one individual having both and one being reported as arthritis and arthrosis. There were 12 individuals with arthrosis findings. In the African elephants, there were 4 findings of arthritis, 3 of osteoarthritis, with one individual having arthritis and arthrosis. Overall, two individuals had signs for arthrosis.

Within this group, taking only the limbs into consideration, 40.00% (16 / 40) of the Asian elephants had lesions in front and back legs, while only the front or just the back legs were affected equally in 27.5% (11 / 40) of the cases. Two individuals did not have a specific place noted where the lesions were found. In the African elephants, 54.54% (6/11) showed lesions in front and back limbs, while 27.27% (3/11) showed lesions in the back legs and 9.09% (1/11) in the front legs only. Although the specific digit injuries were not documented in each post-mortem report, in our study most lesions on the digits were laterally moving medially: in decreasing order D4, D3 and D2. Some of the reports simply listed lateral digits or all.

3.2.3 Circulatory

In our study, the most common lesions in African elephants were seen in the circulatory system, accounting for 18.98% (26/137) of the lesions. In the Asian elephants, these lesions were third most common and accounting for 12.83% (34/265). Myocardial infarct, circulatory failure, splenopathy and cardiac petechiae were most common amongst the African elephants, while for the Asian elephant a general cardiovascular failure, atherosclerosis and petechiae were the most common lesions. Circulatory failures in the African elephant were caused by an underlying infection and one caused by a left sided

thrombus formation. Petechiae, especially on the subendo- and subepicardial as well as on the left ventricle, and septicaemia were other common findings. There was one case of malignant lymphoma.

Cardiovascular failure in the Asian elephant was attributed to vascular injury, right sided heart failure as well as ischemia. There was one outlier who showed these symptoms at only 8 years of age. As in other organ systems, many lesions were caused by infectious agents that were not mentioned but simply stated as an inflammatory reaction.

3.2.4 Digestive Tract

Digestive tract lesions accounted for 18.25% (25/137) of the lesions for the African elephants. Common lesions found included inflammation of the digestive tract, hepatopathies as well as haemorrhages. Gastritis and peritonitis were discovered most often, peritonitis being the result of a perforation or a colitis. One case of fibrinosuppurative typhlitis was determined concurrently with a multifocal suppurative periodontitis. There were also instances of intussception along with volvulus of the small and large intestines. Hepatopathies were grouped into one to show the prevalence here, which included cholestasis, hepatomegaly, congestion, hepatitis and cholangiohepatitis.

3.3 Neoplasia

10.18% (n=23) Asian elephants had some type of neoplasia. Most of the neoplasia found were uterine leiomyomas (n=13). Within these, one individual also had a concurrent adenomyosis and one also showed a concurrent endometrial hyperplasia. Another leiomyoma case additionally displayed metastases to the kidneys and adrenal glands while another had a metastatic carcinoma of the duodenum as well as a parathyroid adenoma. A uterine leiomyosarcoma was found in one individual, with a solid focal adenoma in the kidneys. One case of uterine adenocarcinoma was determined. There was one case of fibropapilloma on the vulva and one solitary case of endometrial hyperplasia. A malignancy to parenchymal organs was discovered, with no stated origin along with a multifocal metastatic carcinoma in the lungs of unknown origin. In one instance, a cortical adenoma in the adrenal gland was discovered, while another found a bifocal renal adenocarcinoma. One incidence of a tumour in the pancreas was reported, while another found a neoformation in the stomach, spleen as well as lung metastases. Hoof cancer was another finding.

In the African elephants 2.73% (n=3) had neoplasia signs. There was one finding of uterine leiomyoma, one uterine tumour and one case of multiple malignant lymphoma.

3.4 Anaesthetic Death

For the Asian elephants, five individuals (2.21%) died while under anaesthesia, all of them for various veterinary procedures. One individual was undergoing a dental examination, while two were being treated for musculoskeletal reasons. One of the deaths found lesions all pointing to shock. Two African elephants died while under sedation, one was transport related while the other was for an x-ray examination for lameness, of unknown age.

3.5 Euthanasia

23.45% (n=53) of the Asian elephants were euthanised, most often due to musculoskeletal lesions. The next biggest reason was a result of undetermined reasons followed by circulatory lesions. Musculoskeletal concerns included arthrosis, arthritis as well as foot and pad problems. Undetermined included instances where the animal was found either agonizing, was not getting enough milk from the dam or not accepted. Diarrhoea leading to critical fluid loss led to the decision to euthanise. Circulatory reasons included a case of severe haemorrhage, oedema and DIC as well as atrophy and core formation in the myocardium.

In the African elephants, 27.27% (n=30) of individuals were euthanised, most common reasons were due to musculoskeletal reasons and trauma, followed by circulatory reasons and digestive and infectious agents. Musculoskeletal causes included arthritis, osteoarthritis while trauma was mainly due to fractures or attacks by conspecific. Infectious agents were predominantly due to *M. tuberculosis* while digestive issues included a volvulus as well as a case of colitis.

Discussion

For the first time on such a large scale, we have summarised the most important lesions found in the post-mortem reports of the Asian and African elephants, part of the EAZA. The aforementioned study in the United Kingdom did have some overlap with our study, as some of those individuals were also included in our study. The discussion will follow the layout of the results.

For both species about 60% of the reports were submitted. Reasons for the absence of reports can be attributed to the current pandemic and therefore shortage of staff and hours, increasing the project length significantly. The study dates back to 1990 and therefore some of the reports are not accessible, have gone missing or a change of management has caused delay here, or they simply cannot be found anymore. Reluctance to participate was also encountered.

There was a skewed proportion of females, compared to males, in both species. This was attributed to the fact that there are more females than males in the captive population. This unequal ratio is also to avoid the aggression between multiple bulls and females. The undetermined genders are attributed to abortions where the sex could not be established.

1.1 Asian Age Based Classification

For the Asian elephants, the greatest number of deaths occurred before age one; the second biggest number of deaths occurred between the ages of 41 to 50, while between 1 to 5 the third highest deaths occurred. Between 41 to 50, 30.23% of the elephants had lesions in multiple organs (n=13). Taking their age into consideration, this is not as surprising as one would expect there to be health concerns in multiple organs. Comparable to the age group of 1 to 5, only 12.13% (n=4) had lesions in multiple organs. Here, most of the lesions were attributed to EEHV. EEHV will be discussed in detail in Section 1.5.1 but considering that EEHV is such a current problem, this number was sadly not surprising.

Based on the age distribution from the Asian elephants, one could conclude that most number of deaths would occur in the neonatal age as well as between 41 to and 50, followed by age 1 to 5. With optimal conditions, these expectations would change, decreasing the overall number. Excluding reasons such as infectious causes and reproductive problems, for example, would eliminate most of the neonatal age group as well as individuals between 1 to

5. Based on the findings, most Asian elephants above 50 years old would die of musculoskeletal lesions.

1.2 African Age Based Classification

Within the African elephants, most deaths occurred between 21 to 30, followed by the same number between 11 to 20 and 31 to 40, and then neonatal. Between 21 to 30, 19.35% had lesions in multiple organs, but most numerous lesions were found in the circulatory system, specifically myocardial infarction. Given that the oldest African elephant was between 51 and 60, myocardial infarct being common in this age group was not expected. The digestive system lesions were most common between 31 to 40 as well as 11 to 20, and circulatory followed closely for both. Interestingly, musculoskeletal lesions were only in the top five for ages 31 to 40, never the most common lesion found in one age group. Infectious reasons were mainly due to *M. tuberculosis.* Both 31 to 40 and 11 to 20 had the most lesions in the digestive system, followed by the circulatory system. Musculoskeletal and infectious followed.

One could conclude that most African elephants would die between 21 to 30, with the neonatal age also having a majority of cases. With optimal conditions, these numbers could be decreased. Solving reproductive problems would exclude most of the neonatal ages, while infectious would exclude one third of the elephants between 21 to 30. For the African elephants, most common were circulatory and digestive lesions, which included inflammation of the digestive tract. Solving this inflammation would also exclude a majority of the. One could conclude that above 50 years of age circulatory lesions would be a major point of concern.

1.3 Asian and African neonatal elephants

The higher number of deaths in the neonatal group for the Asian elephants (n=53), compared with the African elephants (n=17), was attributed to the fact that between 1990 and 2018 there were 170 total Asian elephant births while for the African elephants this number was lower at 128 individuals only.

Neonatal deaths included stillbirth, reasons for which were not always known. Attributing factors were dystocia or weak calves. Insufficient elasticity of the skin around the long vertical part of the birth canal of older primiparous females exposes the calf to a risk of

asphyxia, compression or even tear of the umbilical cord (Personal communication with Dr. Willem Schaftenaar). Dystocia has been a major cause for stillbirths (Murray *et al*.,1996). Foetal malpresentation, leading to dystocia, is associated with an increase in perinatal morbidity and mortality (Kaur *et al*., 2011). Physiological and psychological factors, the intensified breeding program, older nulliparous females, infectious reasons, and inadequate nutrition leading to an oversized calf or obese dam all contribute to dystocia (Hermes *et al.*, 2008). Fortunately, the EAZA breeding program today is not an intensified breeding program anymore, excluding this factor. In our study, the birth process was not always listed, leading to a possible underrepresentation of dystocia, but we did find that out of the Asian stillbirth cases, five could be attributed to dystocia, while another two cases were possibly related to dystocia. Reasons for the dystocia included larger calf size as well as malpresentation, supporting above mentioned findings of Kaur *et al*., (2011).

Foetal anterior presentation is normal for most herd animals, while posterior presentation is much more common in elephants (Holland *et al.,* 1993, Hermes *et al.,* 2008). Anterior presentation can lead to foetal neck flexion and subsequent head impaction in the pelvic canal, resulting in dystocia (Ilic *et al.,* 2021). Dystocia was associated with anterior presentation (Hartley and Stanley, 2016). Another study from 2015 to 2018 showed in 10 out of 34 (29.41%) dystocia cases, the foetuses were in anterior presentation (Ilic *et al*., 2021). In our study, three Asian elephant dystocia cases reported the position of the foetus, two of them being in anterior position. For the African elephant, two cases of dystocia were documented with no information on the position of the foetus. Finding more dystocia cases in Asian elephants (n=5, possible dystocia n=7) than compared to the African elephants (n=2), supported the findings that the occurrence of dystocia cases was more prevalent in Asian elephants than African elephants (Hildebrandt *et al*., 2003).

The birth weight was reported in 12 Asian elephant cases, the two heaviest being 133.4 kg and 162 kilograms - one being a definite dystocia case, with anterior presentation, and the second dying of asphyxia inside the birth canal, possibly due to dystocia. The average weight of the Asian elephant calf in dystocia cases was 132 kg while in the natural birth process the average weight was 108 kg (Hartley and Stanley, 2016). Two cases of episiotomy were

reported as well as three confirmed fetotomy cases. Fetotomy, as a last resort to save the dam, is a relatively new way of birth management (Hildebrandt *et al.,* 2003; Schaftenaar, 2013).

Other reasons for neonatal death included attack by a herd mate as well as rejection by the mother. Traditionally, the herds at zoos are made of unrelated females, which could restrict the ability of previous generations to impart their knowledge of behaviour and maternal instinct, attributing to the attacks on the neonatal (EAZA Elephant Best Practice Guidelines, 2020).

Hand rearing did not prove to be adequate support for the calf, therefore every attempt must be made to avoid hand rearing. The exact reason why hand rearing is unsuccessful is unclear, as it has been shown that even though elephants have an endotheliochorial placentation with modest maternal to foetal transplacental transfer, the antibody levels in the neonates are as high as in the mother and that majority of the transfer is transplacental (Nofs *et al.,* 2013).

1.4 Organ Systems

The organ systems with the most common lesions will now be discussed.

1.4.1 Infectious Agents

For the Asian elephants, EEHV-HD was the leading cause of the infectious agents, as well as between the ages of 1-5. This was mainly caused by EEHV1a, supporting previous findings that most of the known causes of EEHV-HD in the Asian population have been caused by EEHV1a, and to a lesser extent the chimeric counterpart EEHV1b, while EEHV4 and EEHV5a and EEHV5b were rarely lethal (Long *et al.,* 2015). Important to add is that it was EEHV-HD, the haemorrhagic disease, where even adults as carriers do not die from the EEHV infection. In our study, as well as past findings, the most susceptible age has been between 1 and 5 years of age.

Tuberculosis was the most common infectious agent amongst the African elephants. Histopathology and gross pathology cannot distinguish between *M. tuberculosis* and *M. bovis* and therefore acid-fast stain, polymerase chain reaction (PCR) and deoxyribonucleic acid (DNA) sequencing in tissues are mandatory for a proper diagnose of TB (Miller *et al.,* 2021). Two African elephants were positive for *M. bovis*, a rarity, arguing the paucity of *M. bovis*

cases found in African elephants could be due to their sporadic exposure, innate resistance of the species as well as limited surveillance (Miller *et al.,* 2021). In our study, there was only one case of *M.bovis* in the Asian elephant community, while the African elephant community showed none. Overall, all tuberculosis cases in the Asian elephants were identified, while in the African elephants most were confirmed as *M. tuberculosis*, three were unknown cases and one was presumed to be tuberculosis.

Clostridiosis was common amongst the Asian elephants, although the exact agent was not always known. *E.coli*, which usually causes enterotoxaemia, was associated with a co-infection in all cases, being present possibly only secondarily, and in our study was never solely responsible. *Salmonella spp*. was found in both species, and has been associated with gastroenteritis, intermittent shedding, and sometimes causing septicaemia. In one case, it was found to be an attributing factor to diarrhoea. Both species displayed *Salmonella spp.* septicaemia cases.

For all infectious agents, the exact strain of each agent should be identified. As there is still discussion on the transmission and status of the animals for EEHV, serum should be collected to measure the antibody levels in recently developed cases. ELISAs should be carried out to determine the influence of maternal antibodies on the development of EEHV-HD (Fuery *et al*., 2020, Hoornweg *et al*., 2021). Subtype-specific ELISAs are currently being tested with promising results (Pers. comm. Schaftenaar 2021).

Regular testing, screening as well as vaccination, where possible, should be implemented. Implementing a vaccination protocol for each elephant, across all zoos, could also be another idea. A vaccination recommendation was implemented by the Elephant TAG group on September 25th, 2021, hopefully helping a decrease here.

1.4.2 Musculoskeletal

Arthritis was the most common finding amongst both species, followed by osteoarthritis and arthrosis. As arthritis can be a contributing factor to arthrosis, one could argue that the arthrosis could be the result of previous arthritis. In our study, comparable with the aforementioned studies by Panagiotopoulou *et al.,* (2012), most of the elephants had the same amount of lesions on the front and back feet, rather than one specific region. Our finding did support the hypothesis that most, and more significant, pathological lesions occurred on

the lateral nails, rather than the medial ones. Routine foot care is physically demanding, especially trimming the nails, where most start medially and move laterally, strength decreasing as you move laterally, possibly a contributing factor that lesions on the nail were seen most often laterally (Wendler *et al.*, 2019).

Most of the musculoskeletal lesions were lesions that come with age, rather than a preventable disease. As arthrosis is a regressive change, a result of wear and tear, not a 100% preventable lesion, supportive care can be provided here. Slowing the progression could be achieved by improving the flooring to provide more padding or providing joint supplements. Increased inspection of the musculoskeletal system, including general movement practices, could also decrease these lesions. As the nails and pads are prone to cracks or lesions, they often provide a port of entry for bacteria, which could then spread to the whole body. Improving foot care therefore can also help decrease this incidence.

1.4.3 Circulatory

Circulatory lesions were the most numerous lesions for the African elephants, and third in the Asian elephants. African elephants experienced a high number of myocardial infarcts, which results most often after a blockage, causing oxygen loss to the heart. Other lesions included calcification of the tunica media of the major vessels, which can lead to coronary heart disease or even rupture of the aorta. One thrombus was recorded in an African elephant in the left side of the heart, partially on the atrioventricular valve and the endocardium. Further investigation showed it to be white fibrin with haemorrhagic foci, most likely a mixed thrombus. This could be due any predisposing factors listed in Virchow's triangle, such as an alteration in the vascular endothelium, such as damage due to an infection, or immune mediated; it could also be an alteration in blood flow because of turbulence, shock or even a cardiomyopathy; or it could be an alteration in blood constituents. In this case, there were reports of intermittent cardiac arrhythmia, which could contribute to an alteration of flow. The fact that it was mixed also suggests that there was a change in the constituents of the blood, leading to a hypercoagulability. These factors together probably lead to the formation.

For the African elephant, cardiac petechiae were included and these were either sub-endothelial or sub-pericardial petechiae. For two of these elephants, they were the only lesion found and therefore included as a lesion. Suffocation or septicaemia can cause these lesions,

and one of the individuals was a new-born, who had only taken a few breaths. Another individual showed pulmonary oedema as well as congestion in the brain, also suggesting suffocation.

In the Asian elephants, it seemed that most cardiovascular failures were attributed to age and as aging cannot be prevented, routine health checks on the cardiovascular system, including blood work and ultrasound, could be beneficial. This includes but is not limited to performing an echocardiogram or checking pulse on the tail vein. Possibly increasing the daily activity of the elephants could help strengthen the heart muscle.

1.4.4 Digestive

Inflammation of the digestive tract, such as gastritis, is often due to alimentary issues. One instance of inflammation of almost the entire gastrointestinal tract led to pulmonary oedema and an acute heart failure. Interestingly, many of the inflammations were stated as erosive, highlighting the severity of the inflammation. Stress, acids as well as certain medications can contribute to this. Molar and gingiva problems in other animals have been known to cause issues in the gastrointestinal tract, which could also be the case in elephants. For instance, one individual was found with suppurative periodontitis, and possibly consequentially a fibrinosuppurative typhlitis. Other cases such as volvulus and intussusception, as in other animals, do not have one specific reason for occurring.

Although haemorrhages can be attributed to various reasons, it was deemed an important lesion in the digestive tract in the African elephant since in one example the case was described as colic, but the actual lesion included haemorrhages. There are times when haemorrhages alone are not significant, but in the whole picture, they were deemed important. A couple cases were described as colic, without listing the actual lesions. Colic can be caused by many factors and sometimes specific lesions simply cannot be found. In one instance, an elephant was attacked by a herd mate, which led to colic. These were classified under miscellaneous, and no further conclusions can be made.

Hepatopathies included hepatitis as well as cholangiohepatitis, leading to cholestasis and congestion. Hepatomegaly was found to cause effusion in the abdomen. Even though the liver was classified under the digestive system, it is also connected to the circulatory system, for example myocardial fibrosis led to a passive congestion hepatic congestion.

Surprisingly, sand impaction was not listed as a common lesion, possibly being underreported, or not seen as a significant lesion. The longer the sand impaction is in the intestines, the heavier the impaction gets and as a result increased muscular layer development around the impaction. It may have therefore been a previous reason for treatment, which then lead to an enteritis.

The digestive tract is one where as far as management goes, improving the already high-quality feed that the elephants receive, could potentially make a difference. As there were many lesions associated with the liver, increased care of the liver with protectants, such as silymarin, which is used in other species, could be a suggestion for the future. In other species, decreasing the acidity of the intestines helps decrease formation of ulcers and erosions; therefore, it could maybe also work in elephants. Decreasing stress, by providing more enrichment and stimulation, could also help here. Providing a well-balanced diet, to support a healthy gut, would aid in decreasing the prevalence of infectious agents.

1.5 Neoplasia

Although neoplasia is a rare due to the LIF6 zombie gene, our study did find some instances (Wei-Haas, 2016). The youngest Asian elephant to display neoplasia was 21, while for the African elephant this was two years of age. Most of the other Asian elephant cases were above 30, with the highest prevalence between 51 to 60 years of age. Majority of the neoplastic lesions were benign growths, mainly uterine leiomyomas. Three individuals (23.08%) with uterine leiomyomas were pregnant some time before. Two out of the three African elephant individuals had unknown ages, most likely adults. The significantly lower number in the African elephants is interesting; it could be attributed to no findings of a significant lesion or the incidence was lower. This could be a research point for the future.

1.6 Anaesthetic Death

All the anaesthetic procedures were necessary for medical interventions and can be attributed to underlying conditions, age, as well as human error. The age range for the Asian elephant was between 7 and 58 years old, the latter age most likely a contributing factor. One of these individuals had existing lesions that almost certainly contributed to the anaesthetic death. For the African elephants, there were two cases of sedation; one of the sedations was

for transport, where the elephant fell, compressing the trachea and dying of subsequent asphyxia.

1.7 Euthanasia

Euthanasia, being the last resort medical decision in an animals' life, was also chosen in some of the elephants in our study. Most of these were elder elephants, where medicine could not help anymore. The authors decided to highlight these as it shows that most were elderly elephants with musculoskeletal or circulatory lesions. The undetermined subcategory was created as some did not have a black and white reason on paper, but the agonizing and suffering was enough of a reason.

2.0 Suggestions for Future Post-Mortem Reports

The current post-mortem report for the elephants of the EAZA, was not used by every institution (Elephant Necropsy Protocol EEP). Increasing the usage of this document would help create a better overview. A general suggestion is to provide a detailed but separate organ system sheet for each organ system, more details decreasing the discrepancies found. For the neonatal ages, information about the dam such as previous pregnancies, progress of the current pregnancy, age of the dam as well as weight of both the dam and the calf should be included. Information about the birth should include the weight of the calf as well as the presentation, when possible. In the case of abortion and stillbirth infectious agent testing should take place and be included in the report.

In the musculoskeletal system description of the feet and pad, including toes that were affected, the weight of the elephant, the flooring system as well as their typical daily behaviour should be included. If there are different flooring inside and outside, time spent on each floor and preferences would be preferred. A detailed grading list of nail and pad lesions to discern the severity of the lesions would be one way to make it easier for the people filling out the forms. Another consideration to take into account is that although these numbers are lower than expected, these lesions may have existed but deemed not significant, therefore underreported. Including a separate mandatory musculoskeletal report would help shed some light here.

Understanding that this will also take more time, the overall scheme of the additions should be checklists and / or boxes for them to check. Each organ system could have the most

common diseases listed, with spaces for more details to be added, while also leaving space for new findings. This system would also make it easier for the elephant community and the TAG group to draw conclusions from them.

This work was endorsed and supported by the TAG team and the authors were therefore happy to provide and share these findings with the professional community, so that we can all learn from the past and use the data for prevention.

Abstract

Post-mortem reports are a way to understanding diseases, and their processes, to help the future generations. This retrospective study looked at the reports of 226 Asian (*Elephas maximus*) and 110 African elephants (*Loxodonta africana*) in the European Association of Zoos and Aquaria. The reports were based between 1990 until, inclusively, 2018, and the most important lesions were summarised. For the Asian elephants, most deaths occurred before the age of one, followed by 41 to 50 and then the ages of 1 to 5. The most common lesions for these elephants consisted of infectious diseases, specifically the elephant endotheliotropic herpesvirus and C*lostridium spp.*, followed by musculoskeletal and then circulatory lesions. Musculoskeletal lesions were most commonly a kind of arthropathy, arthrosis or pododermatitis. Circulatory lesions included cardiovascular failure as well as atherosclerosis. Most of the African elephant mortalities occurred between 21 to 30, followed by 11 to 20 and 31-40, followed by neonatal. Most lesions in the African elephants were seen in the circulatory system, which included myocardial infarct, circulatory failure, splenopathy and cardiac petechiae. The digestive system followed including inflammation of the digestive tract, hepatopathies as well as haemorrhages. Lesions caused by infectious diseases were the third most common, including clostridiosis and tuberculosis.

References

EAZA Best Practice Guidelines for Elephants (2020). Veterinary Advisors to the EAZA elephant TAG.

Elephant (2021) Encyclopaedia Britannica. URL: https://www.britannica.com/animal/elephant-mammal Accessed: September 7, 2021.

ELEPHANT NECROPSY PROTOCOL EEP (2018). Veterinary Advisors to the EAZA elephant TAG.

Fuery A., Pursell T., Tan J., Peng R., Burbeo P., Hayward G.S., Ling P.D., 2020: Lethal Hemorrhagic Disease and Clinical Illness Associated with Elephant Endotheliotropic Herpesvirus 1 Are Caused by Primary Infection: Implications for the Detection of Diagnostic Proteins. *Journal of Virology*. 94. 3. [Journal Article]. URL: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7000966/ Accessed: August 28, 2021.

Hartley M., Stanley C.R., 2016: Survey of reproduction and calf rearing in Asian and African elephants in European zoos. *Journal of Zoo and Aquarium Research*. 4.3. [Journal Article.] URL: https://doi.org/10.19227/jzar.v4i3.161 Accessed: August 28, 2021.

Hermes R., Saragust J., Schaftenaar W., Göritz F., Schmitt D.L., Hildebrandt T.B., 2008: Obstetrics in elephants. *Theriogenology.* 70. 2. pp. 131-144. [Journal Article.] URL: https://doi.org/10.1016/j.theriogenology.2008.04.003 Accessed: August 28, 2021.

Hildebrandt T.B., Strike T., Flach E., Sambrook L., Dodds J., Lindsay N., Furley C.F.,Glatzel P.S., McGowan M., 2003: Fetotomy in the elephant. *Erkrankungen Zoo*. 41. pp. 315–318.

Holland M.D., Speer N.C., LeFever D.G., Taylor R.E., Field T.G., Odde K.G., 1993: Factors contributing to dystocia due to fetal malpresentation in beef cattle. *Theriogenology*. 39. 4. pp. 899-908.

Hoornweg T.E., Schaftenaar W., Maurer G., van der Doel P.B., Molenaar F., Chamour-Galante A., Vercammen F., Rutten V., de Haan C.A.M., 2021: Elephant Endotheliotropic Herpesvirus Is Omnipresent in Elephants in European Zoos and an Asian Elephant Range Country. *Viruses 2021*. 13. 2. 283. [Journal Article.] URL: https://doi.org/10.3390/v13020283 Accessed: September 1, 2021

Ilic D., Ilic A., Jakovljevic B., Dzigurski J., Dragic N., Simic S., Petkovic B., Sekulic S., 2021: The Incidence of Anterior and Posterior Presentation at Birth in Asian (*Elephas maximus)* and African Elephants (*Loxodonta Africana*): A Review Study. *Indian Journal of Animal Research.* [Journal Article.] URL: http://arccarticles.s3.amazonaws.com/arcc/Final-article-attachemnt-with-doi-B-1319.pdf Accessed: August 28, 2021.

Kaur I., Dhindsa S.S., Harpreet K., Prabhjot S., 2011: Various constraints of dairy farming in central zone of Punjab. *Journal of Dairying Foods & Home Services*. 30. 4. pp. 242-245.

Long S.Y., Latimer E.M., Hayward G.S., 2015: Review of Elephant Endotheliotropic Herpesvirus and Acute Hemorrhagic Disease. *Institute for Laboratory Animal Research Journal*. 56. 3. pp. 283-296. URL: https://academic.oup.com/ilarjournal/article/56/3/283/2732573 Accessed: September 1, 2021.

Miller M. A., Kerr T.J., de Waal C.R., Goosen W. J., Steicher E. M., Hausler G., Rossouw L., Manamela T., van Schalkwy L., Kleynhans L., Warren R., van Helden P., Buss P.E., 2021: *Mycobacterium bovis* Infection in Free-Ranging African Elephants. *Emerging Infectious Diseases Journal.* 27. 3. pp. 990-992. URL: https://wwwnc.cdc.gov/eid/article/27/3/20-4729-t1 Accessed: September 2, 2021.

Murray, S., Bush, M., Tell, L.A., 1996: Medical management of postpartum problems in an Asian elephant (*Elephas maximus)* cow and calf. *Journal of Zoo and Wildlife Medicine*. 27. 2. pp. 255-259.

Nofs S.A., Atmar R.L., Keitel W.A., Hanlon C., Stanton J.J., Tan J., Flanagan J.P., Howard L., Ling P.D., 2013: Prenatal passive transfer of maternal immunity in Asian elephants (Elephas maximus). *Veterinary Immunology and Immunopathology*. 153. pp. 308-311. [Journal Article.] URL: https://pubmed.ncbi.nlm.nih.gov/23582266/ Accessed: August 28, 2021.

Panagiotopoulou O., Pataky T.C., Hill Z., Hutchinson J.R., 2012: Statistical parametric mapping of the regional distribution and ontogenetic scaling of foot pressures during walking in Asian elephants (*Elephas maximus*). *The Journal of Experimental Biology*. 215. pp. 1584-1593. URL: https://journals.biologists.com/jeb/article/215/9/1584/11342/Statistical-parametric-mapping-of-the-regional Accessed: September 2, 2021.

Schaftenaar W., 2013: DELAYED POSTPARTUM FETOTOMY IN AN ASIAN ELEPHANT (*ELEPHAS MAXIMUS). Journal of Zoo and Wildlife Medicine*. 44. 1. pp. 130-135. [Journal Article.] URL: https://doi.org/10.1638/1042-7260-44.1.130 Accessed: August 28, 2021.

Wei-Haas, M., 2018: Cancer Rarely Strikes Elephants. New Clues Suggest Why. *National Geographic.* URL: https://www.nationalgeographic.com/science/article/news-cancer-elephants-genes-dna-new-research Accessed: September 7, 2021.

Wendler, P., Ertl N., Flügger M., Sos E., Schiffmann C., Clauss M., Hatt, J.M., 2019: Foot health of Asian Elephants (*Elephas maximus*) in European zoos. *Journal of Zoo and Wildlife Medicine*. 50. 3. pp 513-527. [Journal Article.] URL: https://www.zora.uzh.ch/id/eprint/177179/1/177179.pdf Accessed: September 6, 2021.

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Aalborg Zoo	Safari Zoo Mallorca
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Bioparco di Roma	Thoiry ZooSafari
Blackpool Zoo	Tiergarten & Reiterhof Walding
Blairdrummond Safari & Adventure Park	Tiergarten Nürnberg
Borås Djurpark	Tierpark Berlin
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Dudley Zoo and Castle	Yerevan Zoo
Erlebnis Zoo Hannover	ZOO Antwerpen
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Izmir Wildlife Park	Zoo d'Amnéville
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Kharkiv Zoo	Zoo De La Barben Parc
Kiev Zoo	Zoo de la Flèche
Knies Kinderzoo	Zoo de la Palmyre
Knowsley Safari	Zoo de Lyon
Kolmårdens djurpark	Zoo de Pont-Scorff
Kölner Zoo	Zoo Dresden
Krefelder Zoo	Zoo Duisburg gGmbH
Le Monde Sauvage Safari Parc D'Aywaille	Zoo Gdańsk Oliwa
Le Zoo d'Amiens Métropole	Zoo Heidelberg
Lo zoo di Napoli	ZOO Kraków
Longleat Safari Park	Zoo Leipzig
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Pairi Daiza	ZOO Wrocław sp.z o.o.

Parc de la Tête d'Or	Zoo Zagreb
Parco Faunistico Le Cornelle	Zoo Zlín
Parco Natura Viva	Zoo Zürich
Parque de la Naturaleza de Cabárceno	Zoo Bassin d'Arcachon
Parque Marino de la Magdalena	Zoo d'Amneville
Planète Sauvage	Zoologická zahrada hl. m. Prahy
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I hereby confirm that I am familiar with the content of the thesis entitled

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written by Anne Hess …… (student name)

which I deem suitable for submission and defence.

Date: Budapest, 9 day 11 month 2021 year

Dr Gyarmati

……

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I hereby declare that the thesis entitled ……..……..…...

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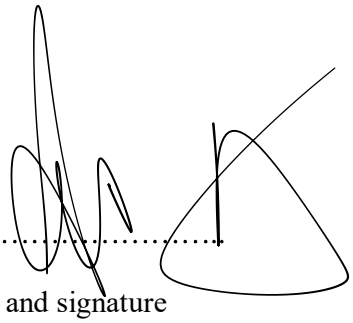

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