Current State of Forensic Pathology for Animals in Japan: What can we know from the remains of Animals?

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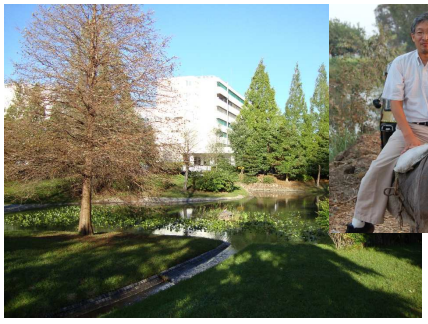
Gifu University
1992-2019 March



Laboratory of Veterinary Pathology

Okayama University of Science
Veterinary College 2019 April

Laboratory of Wildlife / Forensic Pathology

Forensic Science – Why?

Forensic science is the application of science to criminal and civil laws, mainly—on the criminal side—during criminal investigation, as governed by the legal standards of admissible evidence and criminal procedure.

1）Used to determine cause and manner of death, time of death, contributing causes:

2）Determine/exclude other factors – injuries, nutritional condition and diseases

What's responsible to death?

3）Document what is found and what is not and interpret

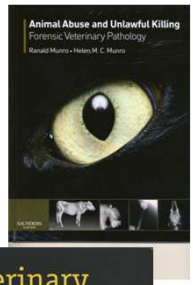
4）Apply to law

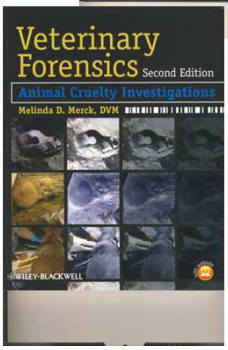
Forensic Science，Forensic Pathology
Forensic Investigation

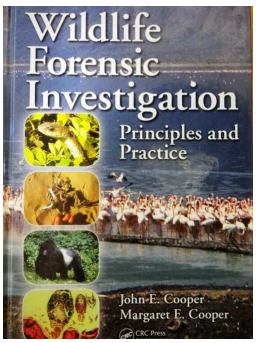
Demands of forensic Science in animal Science: Needs of Veterinary forensics !

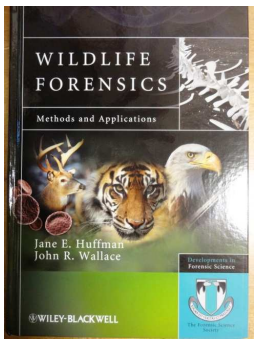
1．Evidences of criminals including animal abuse, unusual killing or neglect for suits

2．Monitoring of wildlife diseases for conservation



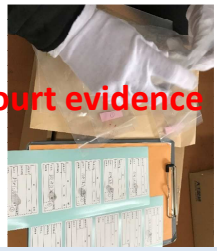






Post-mortem Principle for Forensic Science

1. Follow standard objective protocol
2. Description and measurements
3. Label and photograph everything
4. Complete a full necropsy every time
5. Access contributory factors-pre-existing disease or injures
6. May assist in understanding why an attack occurred
7. Handle with care-introduced errors, contaminations, storage condition
8. Prevention of the second infection to humans; TB, Rabies, Plague etc.



Court evidence

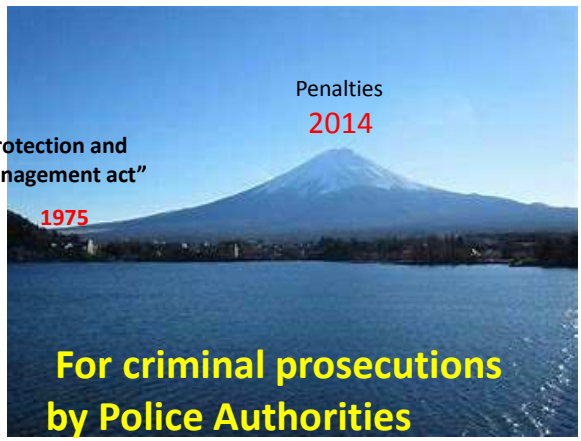
For the forensic diagnosis to set up a crime on animal abuse or unlawful killing, samples are collected as legal evidence in the presence of police. Each sample needs a signature of a forensic pathologist on the label with date, time of collection under the control of police.

What's difference between "Pathology" and "Forensic Pathology" in Medicine

Pathology: Perform autopsy on corpse which died in the hospital due to cancer etc., and study mainly for the research and training of the doctor such as the investigation of the disease condition and the judgment of the treatment effect.

Forensic Pathology: Performed autopsy to maintain the rights and safety of the people by accurately investigating the cause of death. Forensic diagnosis is to determine not only the result of autopsy but also the result of forensic examination such as chemical and other laboratory investigation to identify the cause and time-course of death.
(Standard Forensic Science , Igaku-syoin, 7th edition pp16, 2013)

Epoch-making events for criminal prosecution on animal abuse and unlawful killing in Japan



"Animal protection and proper management act"

1975

"Animal protection and proper management act" in Japan (established in 1975)

(2) Fundamental Principle

In the light of the fact that animals are living beings, no person shall destroy, injure, or inflict cruelty on animals without reason, and every person shall treat animals properly by taking into account their natural habits and giving consideration to the symbiosis between humans and animals. (No penalties!!)

Chapter VI Penal Provisions (By revision in 2014)

Article 44 (1) A person who had destroyed or injured, without reason, a protected animal shall be punished by imprisonment with work for not more than two years or a fine of not more than 2 million yen (18,500 USD).

(2) A person who has, without reason, committed on a protected animal an act of causing debilitation through discontinuation of feeding and/or watering, exploitation of the animal, or restraint of the animal at a place where it is difficult to maintain its health and safety, an act of failing to appropriate protect a protected animal which he/she cares for or keeps and which has a disease or injured, an act of caring for or keeping the animal in a facility which he/she owns where excrement is deposited or where the carcass of another protected animal is left as it is, or any other cruelty shall be punished by a fine of not more than one million yen.

(3) A person who has abandoned a protected animal shall be punished by a fine of not more than 1 million yen.

* What's protective animal?

1. Cattle, horses, pigs, sheep, goats, dogs, cats, rabbits, chickens, domestic pigeons and ducks 2. Other animals that humans have and that belong to mammals, birds or reptiles

Since then, the Police has to make actions for prosecution!

TOTAL CASES FROM POLICE 2010～2019 (MARCH)
GIFU UNIVERSITY/OKAYAMA UNIVERSITY OF SCIENCE

The number of forensic cases from police

A Request letter from police

Problems
1. **The police are not good at animal issue**
2. **Who can conduct forensic necropsy?**
3. **There was no lab for animal forensic science**

So we got a dead animal from the Police or wildlife field researcher…

The choice of who examines, may depend on..

- **Best (Forensic investigational) – expertise in Forensic Science Lab (criminal /wildlife): very few Vet experts, only one lab in Japan (Okayama University of Science : me)**

- **Best (Pathological) + anatomy + tissue changes = Veterinary Pathologists**

An application of Forensic Science to unlawful killing and wildlife conservation

Basic and detailed information on:

*Cause and manner of death

*Pre-existing disease

Body condition-nutritional status

*Any background disease and parasitic exposure

*Biological information –
weight, measurements, age (tooth age), sex and reproductive history, genetic profile

Sample collection – specific purpose, archiving…

Forensic information – evidence on ; abuse, poisoning etc.

Why complete necropsy is needed?

1) Identify (ID) abnormalities including cause of death

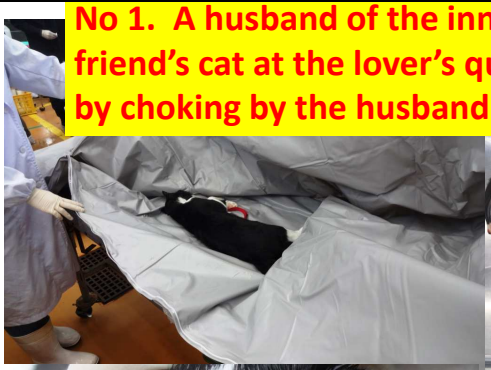
2) ID underling diseases, injuries or conditions contribute to attack

3) Consider additional tests
Microscopic
DNA

All carcass should be examined and reserved for current and future studies

Suspected cases of Animal abuse and unlawful killing

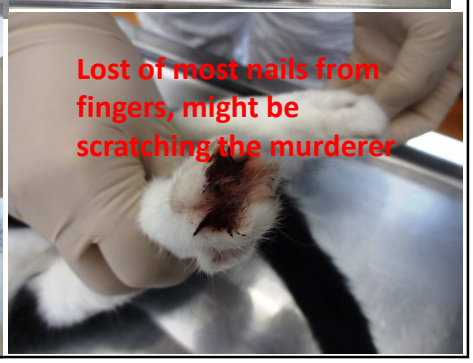
No 1. A husband of the inner edge strangled his girl friend's cat at the lover's quarrels: that female cat died by choking by the husband. The witness was the girl.



A good nutritional condition

The pupil were opened

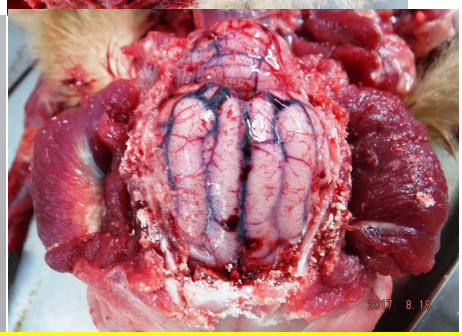
Bloody discharges from the nose



Lost of most nails from fingers, might be scratching the murderer

Forensic Pathological diagnosis: Unusual (Unlawful) killing due to choking and pressing the thoracic area

No.2 Hanging dead Body of a cat

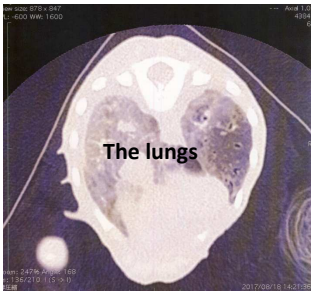


At forensic necropsy, a severe congestion in the neck, cervical spine and lungs as well as brain. When the cat hung? Before death?

Severe Congestions in the lung

Might be asphyxiation？

No other lesions than severe congestion



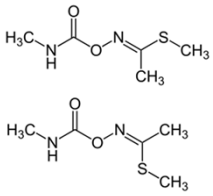
We still have questions
How do we prove abuse?
What's definition of animal Abuse?

May be different from human cases

Need to establish veterinary forensic science!!

No. 3 Poisoning of a cat
A young-adult male cat, age: 1-2 year of age,
Suspected poisoning

Clinical episode: Convulsed the body and died after blowing bubbles from the mouth
Grossly, the cat was in good nutritional condition with rich fat deposition.
The liver, heart, kidneys was dark red in color.
The stomach contained moderate amount of undigested content with slight greenish blue color



methomyl

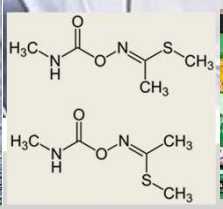
The lungs

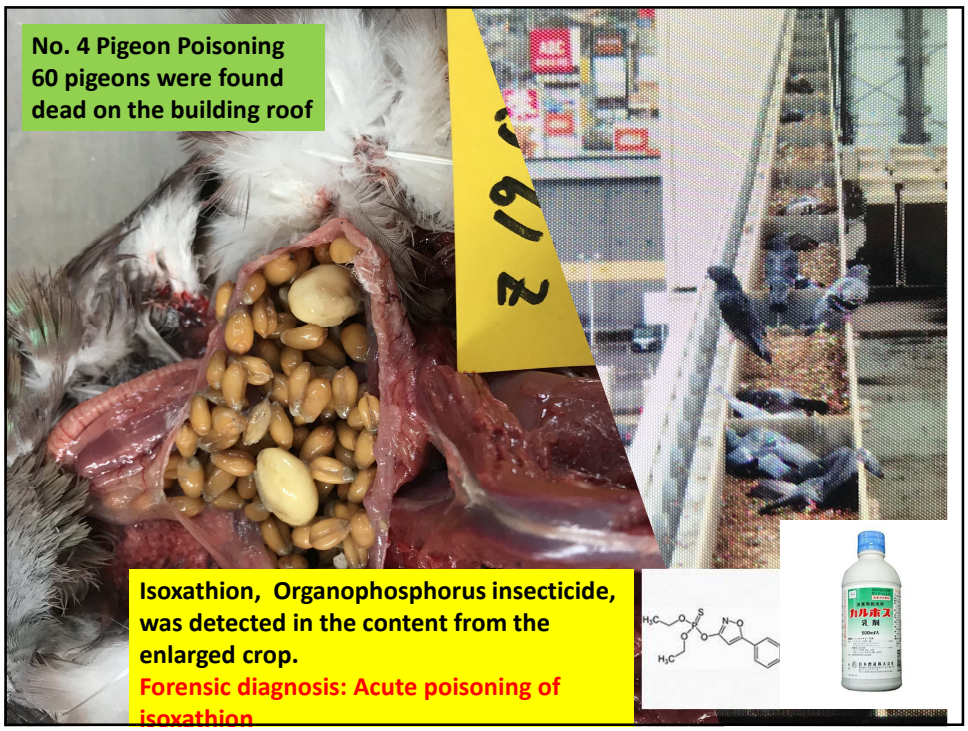
With chemical analysis methomyl, a common carbamate insecticide, was detected from stomach content.
The toxic mechanism is cholinesterase inhibitor.
Lethal dose by oral route: dog 20mg/kg, pigeon 10mg/kg
Forensic diagnosis: Acute toxicity with methomyl poisoning

Severe congestion and pulmonary edema

According to newspapers, an associate professor was arrested with a suspect of violation of Animal Protection Law on April 3rd in 2019 in Tokyo. He was suspected to put on poisoned bait and killed over 100 pigeons.

Methomyl

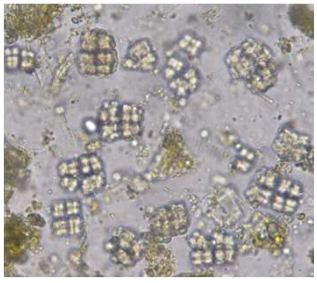




The most Recent Toxic Case in Newspapers www.ashahi.com June 3rd

According to Osaka prefectural police, a 4-year-old male dog died after eating a dog food left in the park of Tsuruhashi and from the dead dog's vomit, a harmful chemical substance "ethylene glycol" used for antifreeze of cars, etc. has detected. The police are investigating on charges of violation of the animal protection law.

On May 3rd the dog plunged its head into the space in the park and ate a dog-food-like substance soaked in the liquid in the container, and then weakens the following day he started vomiting and died of renal failure on the 21st May



Frequent crystals in the stomach contents, which might be calcium oxalate

By the investigation those capybaras died of narcissus intake, which was contaminated in the diet by mistake.

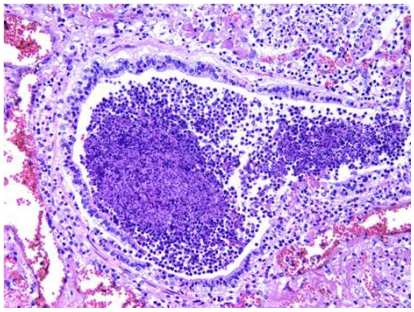
Narcissus contains several toxic materials including Lycorine, galanthamin, tazettine and calcium oxalate

Forensic diagnosis: Acute and fatal poisoning of Narcissus

Case 6. Forensic pathology prevents atonement !!

There were total five dead bodies of stray cats in a residential area. A guy was suspected as a killer by poisoning.

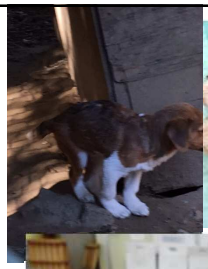
At forensic necropsy, there was severe bronchopneumonia, which might cause frequent fatal bacterial pneumonia in that area. The charges were fine.



Forensic diagnosis: Spontaneous bronchopneumonia

The last case 7: Inappropriate way of keeping. Close to neglect

At a breeding facility for hunting dogs. The owner asked me to investigate a strange disease in puppies which frequently occurred as swelling of the joint and crotch posture from 20 days of age.



Stomach contents: rice and noodle

It was diagnosed with Rickets because it showed marked enlargement of costal cartilage. The most common cause is vitamin D deficiency. This can result from eating a diet without enough vitamin D, or not enough calcium or phosphorus. The stomach content were digesting rice and noodle. The disease disappeared by changing food to commercial dog food. Forensic diagnosis: Rickets due to inadequate diet.

2. The current major role of forensic pathology in wildlife conservation

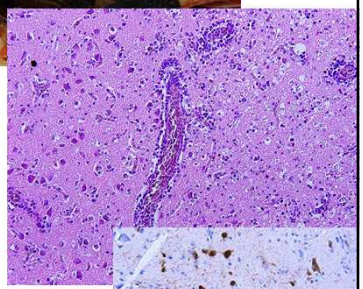
Complete necropsy
Histological exam.
Sampling for PCR

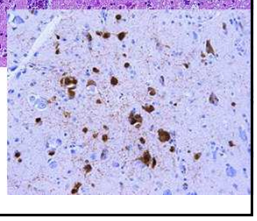
Team Work
Pathologist and field researchers and zoo vets

Front line for wildlife disease surveillance! Major Targets in Priority!

1. Monitoring of infectious diseases, especially zoonosis
2. Monitoring of environmental pollutions, toxicity
3. Monitoring of health condition, including nutritional factors, reproductive activities of endangered wildlife
4. Sampling from the carcasses of wildlife for PCR
5. Evidences for Illegal trades and cruel captivity

Monitoring of infectious diseases, especially zoonosis





In 2011, tufted ducks were found dead And sent for forensic pathology.
Grossly, Marked dark red clotted blood in the pectoral muscles and multifocal hemorrhages on the serosal surface of the pancreas, liver and pericardium and lungs. There was encephalitis with positive IHC for influenza A.
HPAIH5N1 subtype was diagnosed

Infections to be monitored in endangered raptors

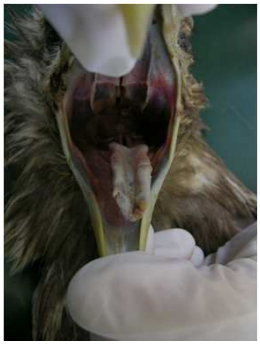
- Virus　Herpesvirus induced hepatitis
  Avian pox
- Bacteria Fowl cholera (*Pasteurella multocida*)
  Mycobacteriosis（*Mycobacterium avium*）
  Mycoplasmosis（*Mycoplasma falconis*
- Fungus　Aspergillosis (*Aspergillus fumigatus*)
- Parasites　*Trichomonas gallinae*
  Intestinal　coccidiosis（*Isospora spp.*）

Steller's sea eagle

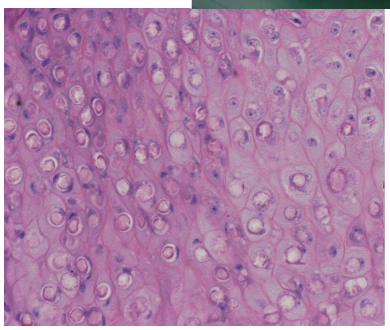
A case of Avian pox infection in white-tailed eagle In 2004

White-tailed eagle in Hokkaido: endangered species

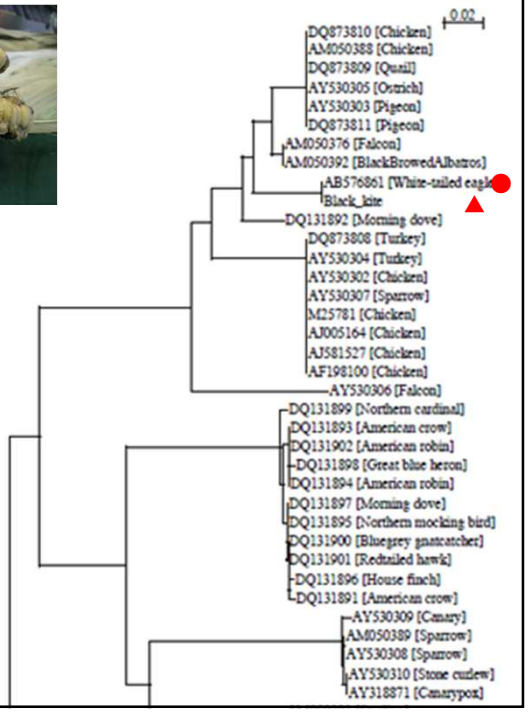
- Poor nutritional condition, the carcass was very thin
- There were multifocal nodular lesions in the beak, eyelid, leg hairless parts and even in oral cavity.



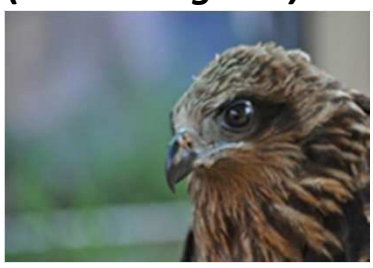
White-tailed eagle



Histologically, there were frequent intracytoplasmic inclusion bodies （Bollinger's body ）in swelling prick cells In the skin

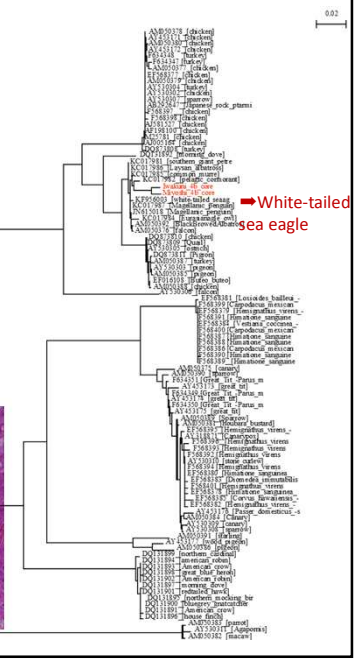


Avian Pox in Kites (*Milvus migrans)*



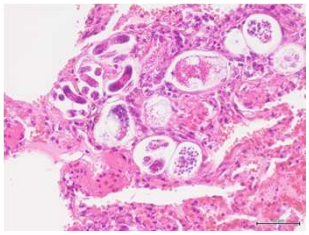
・In 2010 in Tokyo, several cases of avian pox were observed in the flock of kites.
・The affected birds got nodules in the lips and eyelid as well as head. They were very thin, standing denial .
・Histologically, Bollinger's body were observed in swelling prick cells in the skin
Wildlife Forensic; Avian pox, genetically identical to previous one in a white-tailed eagle

An outbreak of avian pox in captured Japanese cormorant in June to July in 2017



Tsushima Leopard Cat *(Prionailurus bengalensis euptilurus)*

Endangered subspecies of a Bengali wildcat: estimated population is around 70-100 (2010). All carcasses found dead were sent to us for forensic necropsies to collect samples for biology, DNA and pathology



Pulmonary nematode infection affect health condition?

Some limitations of Pathological Examination

- Merits of Pathology

Useful for final diagnosis because of clear evidence of histopathology

- Demerits of Pathology

Some difficulties for detection of bone fracture, air embolisms or Pneumothorax
Time consuming

Need of CT Examination

Application of Autopsy Imaging (Ai) to Wildlife Forensic Pathology

- **Autopsy imaging(Ai)＝Image pathological diagnosis at death**
- **New options to compensate for low autopsy rate**
- **Carried out as a screening prior to autopsy**

CT analyzing on traffic accidental cases of Tsushima Leopard Cat

From October in 2017 to August in 2018 13 cases of Tsushima leopard cats were Sent to Gifu University for autopsy.

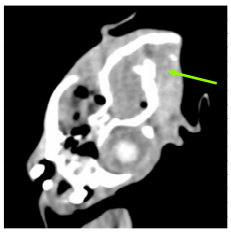
- Method

CT Examination →Analysis by using Analyzing software OsiriX

Gross Examination
Histopathological Examination

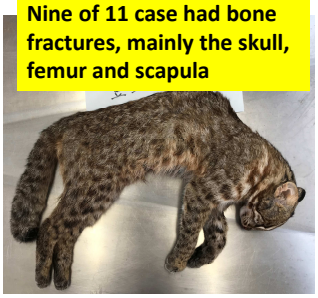
CT is useful to detect Bone fractures in roadkill cases

Skull fracture



頭蓋骨骨折（症例No.4）

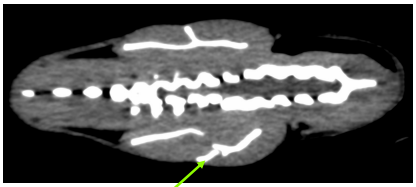
Nine of 11 case had bone fractures, mainly the skull, femur and scapula



右肩甲骨骨折（症例No.8）

左大腿骨骨折（症例No.10）

Left femur fracture

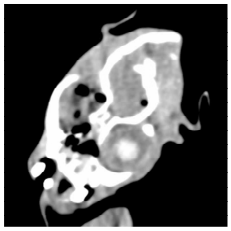


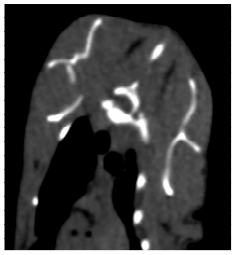
Right scapula fracture

CT can detect bone fractures easily when no obvious change from the appearance

Detailed Analysis by Analysis Software for Bone Damage

（東京慈恵会医科大学 高次元医用画像工学研究所）





Visualizing putting arrow to fracture site

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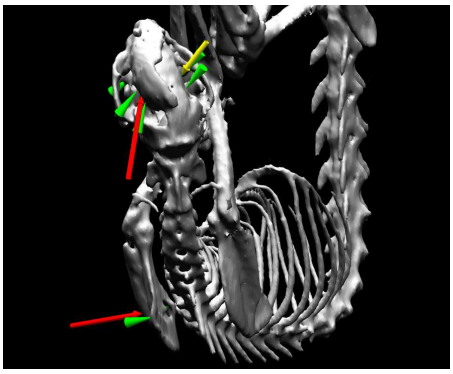
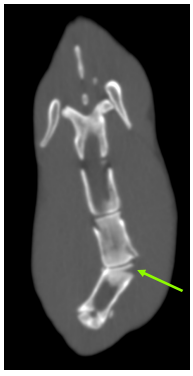
To estimate the direction of the force

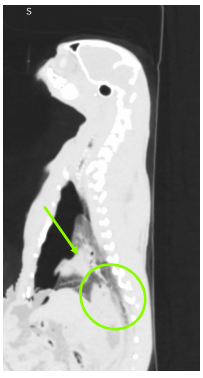


Skull and left scapula fracture 頭蓋骨＆左肩甲骨骨折（症例No.4）

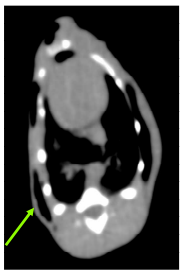
CT is also useful to detect abnormalities of the structure and space in carcass: dislocation, hernia, pneumothorax, emphysema etc.



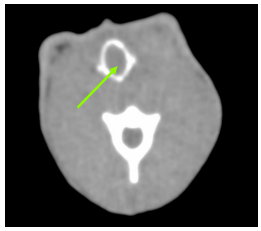
Lumber dislocation
腰椎脱臼



Diaphragmatic hernia
Pneumothorax
気胸
横隔膜ヘルニア



Subcutaneous emphysema皮下気腫



Blood clot in the trachea気管内血液貯留

Conclusion

- Increasing demands of animal forensic science including investigations on animal abuse, unusual killing or neglect as well as wildlife conservation of endangered animals in Japan

- All carcass should be necropsied and samples should be reserved for evidences on forensic studies based on protocol

- The major roles of wildlife forensic pathology; Monitoring of infectious diseases including zoonosis, pollutions, toxicity, health condition and sampling for PCR, and Illegal trade evidence

- CT examination may be useful for forensic investigation

- Forensic necropsy is useful for detection of animal abuse and unlawful killing as well as conservation of endangered wildlife

Asian Networking for Forensic Pathology

2006 Asian Society of Wildlife Pathology
Wildlife pathology group

Reformed
2017 Asian Society of Animal Forensic Pathology
Reformed

Purpose
1. Asian Textbook for Animal Forensic Pathology
2. Archive for Animal forensic cases in Asia
3. Animal Forensic Pathology Workshop
4. Consultant for forensic cases

Thank you!

Photo by Mr. Yamamura