General Summary

Our main research projects in 2010 have focused on sudden unexpected infant death due to milk aspiration, diagnosis of drowning by detection of specific DNA fragments of aquatic bacteria from blood samples, analysis of the ubiquitin proteasome system and the autophagy lysosome system in the central nervous system, identification of war-dead remains by DNA analysis, the objective evaluation of the limit of DNA typing based on the intensity of ninhydrin treatment, and quantitative analyses of medicines and poisonous substances in forensic autopsy cases.

Research Activities

Forensic pathology
1. Sudden unexpected infant death due to milk aspiration
To examine the longitudinal change of pathological findings of the lungs and other organs in cases of milk aspiration, an experimental study was performed with a murine model. The results of immunostaining with antibodies against human α-lactalbumin indicated that the lungs, kidneys, and spleen showed positive reactions against the antibody over time. The detection of aspirated milk in organs other than the lungs would be clear evidence of intravital milk aspiration and would suggest previous or recurrent milk aspiration.

2. Diagnosis of drowning by detection of specific DNA fragments of aquatic bacteria
In general, the diagnosis of death by drowning is based on the detection of diatoms in organs other than the lungs. We speculate that bacteria are more useful markers than are plankton for diagnosing death by drowning. From the preserved blood specimens of 30 cases of freshwater drowning, specific DNA fragments of *Aeromonas sobria*, a common aquatic bacteria, were examined with the polymerase chain reaction. With the nested polymerase chain reaction, DNA fragments of the bacterium were detected in most of the cases.

3. Analysis of the ubiquitin proteasome system and the autophagy lysosome system in the central nervous system
Research associated with the ubiquitin proteasome system and the autophagy lysosome system, which play major roles in the degradation of intracellular proteins and organelles, has recently advanced in various areas of medical science. How the ubiquitin proteasome system and the autophagy lysosome system are induced in traumatic intracranial injury was investigated in cases of such injury examined at autopsy at our department. Both degradation pathways were found to be induced in the injured cortex soon after trauma, and the pathway involved in the degradation of unnecessary substances or the cells in which the degradation is activated may be different or be altered over time.
after the traumatic event in the central nervous system.

**DNA analysis**
1. Identification of war-dead remains with DNA analysis
   We performed identification of war-dead remains buried in the former Soviet Union by means of DNA analysis as part of the war-dead remains return project of the Ministry of Health, Labour and Welfare. For genetic markers we used single nucleotide polymorphisms of hypervariable regions of mitochondrial DNA and short tandem repeats of nuclear DNA.
2. The objective evaluation of the limit of DNA typing based on the intensity of ninhydrin treatment
   Shed epithelial cells on a sheet of paper were stained with ninhydrin reagent, and DNA typing was performed. We studied the relationship between the intensity of the purple staining after ninhydrin treatment and the limit of DNA typing as mitochondrial DNA polymorphisms, and we attempted to perform an objective evaluation to determine the target of the staining area for DNA analysis.

**Forensic toxicology**
1. Quantitative analyses of medicines and poisonous substances
   Medicines and poisonous substances (abused drugs, alcohol, carbon monoxide, cyanide, and agricultural chemicals) suspected to have caused deaths were quantitatively analyzed with gas chromatography/mass spectrometry, and spectrum photometry in tissue specimens obtained at autopsy.
2. Qualitative and quantitative analyses of hydrogen sulfide
   We performed autopsies in 17 cases of death due to the inhalation of intentionally generated hydrogen sulfide gas. The concentrations of sulfide and thiosulfate in blood, urine, and cerebrospinal fluid were examined with gas chromatography/mass spectrometry. Although in previous reports of cases of hydrogen sulfide poisoning, the blood concentration of thiosulfate was higher than that of sulfide, in the present study the concentration of sulfide was higher than that of thiosulfate in 9 of 17 cases. Furthermore, the sulfide concentrations were similar to those in previously reported cases of accidental fatal poisoning in 9 of our 17 cases and resembled those in previously reported cases of intentional fatal poisoning in 2 of our 17 cases.
3. Examination of a method for analyzing tandospirone citrate
   We detected tandospirone citrate, a serotonergic anxiolytic, in 2 autopsy cases. Qualitative and quantitative methods of analyzing tandospirone citrate with gas chromatography/mass spectrometry were examined. With quantitative analysis, high concentrations of tandospirone citrate were detected.

**Radiocarbon analysis**
1. Establishment of age estimation
   We studied the estimation of the date of birth from the quantity of radiocarbon isolated from tooth enamel. We have succeeded in specifying the age range from only a single tooth by measuring carbon-14 separately in incisal (occlusal) and cervical regions of the
Publications

