PATHOLOGY RESEARCH DIVISION

Deputy Director & Head	…	Dr. Kyaw Soe MBBS(IM1), PhD(Medical Science)(Nagasaki)
Research Officer	…	Daw Myat Mon Oo BSc(Chemistry)(YU), DA(MSA), Post-Graduate Diploma in English(YU)
	…	Dr. Zayar Chit MBBS(UM2)
	…	Dr. Khin Kant Kaw Oo MBBS(UM2), MMedSc(Pathology)(UM2)
Medical Technician (1)	…	Daw Than Than Swe BSc(Chemistry)(YU)
Research Assistant (2)	…	Daw Khin Myo Set BA(Myanmar)(UDE)
	…	Daw Hnin Nu Htwe BA(History)(UDE), Diploma in Food Technology(YU)
Research Assistant (3)	…	Daw Mya Thandar Win BSc(Botany)(UDE) Post-Graduate Diploma in English(YU)
	…	Daw Kay Thwe Win BSc(Zoology)(UDE)
Research Assistant (4)	…	Daw Khin Zar Chi Aung
Laboratory Attendant	…	Daw Nilar Tun

The Pathology Research Division is actively engaged in research on haemoglobinopathies, anaemia and association of aflatoxin B1with liver diseases. Moreover, research projects on common cancers, particularly carcinogenic, cellular kinetic changes and epigenetic alterations of liver, gastrointestinal and nasopharyngeal cancer were studied by immunohistochemistry (IHC), terminal deoxynucleotidyltransferase mediated deoxyuridine triphosphate biotin nick end labeling(TUNEL), in-situ hybridization (ISH) and polymerase chain reaction (PCR), are being conducted in collaborating with other organizations and departments. The division also performs establishment of in-house phytohaemagglutinin (PHA) reagent for detection of chromosomal disorders.

RESEARCH PROJECTS

1. NON-COMMUNICABLE DISEASES

1.1. Antenatal screening of anemia including iron deficiency anemia, haemoglobinopathies and thalassaemia among the pregnant women living in Bago Region of Myanmar

Anemia in pregnancy is a major health problem in many developing countries (33-75%) and is associated with increased rates of maternal and child mortality, premature delivery and low birth weight. The present study was conducted to screen out anemia including iron deficiency anemia, haemoglobinopathies and thalassaemia among pregnant women attending ante-natal clinic of Maternal and Child Health Center (MCH) in Bago Region during 2014-2015. A cross-sectional descriptive study was carried out in randomly selected 478 pregnant women living in different villages within the age of 18-43 years and 12-40 weeks of gestation. Anemia was determined by using rapid haemoglobin (Hb) analyzer (Hemocue). Past medical, surgical and obstetric history, social and education status were recorded from the study participants. Red blood cell parameters (Hb%, total red blood cells count, mean corpuscular volume, and red cell width), blood film examination, osmotic fragility test (OFT), determination of Hb A2%, Hb F% and serum ferritin, H inclusion test, iso-electric focusing (IEF) were done on pregnant women with Hb less than 11g% in the laboratory at Pathology Research Division, Department of Medical Research. Anemia (mean Hb 10.2 g%) was detected in 268 pregnant mothers (56%). The most common age group was 20-30 years (63.6%). Most of them were dependent (65%) with primary school level of

education (38.1%) and their family income was one hundred to two hundred thousand kyat monthly (71.8%). The commonest gestation of pregnancy was more than 24 weeks (68%) and number of family members was mostly less than 5(70.7%). The commonest hemoglobin A type (normal Hb) was detected in 195 (76.5%) and other abnormal Hb types (Hb EA, EE and AH) were found in 60 subjects respectively (21.6%, 1.5% and 0.4%). Hemoglobinopathies were detected in 61 pregnant women (28%) and iron deficiency anemia (serum ferritin <30 μg/ml) was found in 89 pregnant mothers (52.5%). Alpha thalassaemia minor was detected in one pregnant mother with anemia and silent beta thalassaemia minor was detected in 28 pregnant women with anemia (12.8%). Hb concentration was significantly different in types of hemoglobin variants (p=0.008). There were no significant differences between Hb concentration and gestation of pregnancy (p=0.316), education (p=0.711) and family income (p=0.282). Serum ferritin was significantly decreased in more than 24 weeks of pregnancy (p=0.012). In the present study, iron deficiency anemia is the commonest type of anemia and others are hemoglobinopathies, alpha thalassaemia and silent or minor beta thalassaemia. Anemia is one of the main health problems in pregnant women living in rural areas in Bago Region. Prevention and proper treatment of anemia are important requirements to be included in future health planning for reduction of maternal, child mortality and morbidity. Therefore, public health education or information on reproductive health, monitoring the compliances of women towards ante-natal care services and strengthening of their health care seeking behavior are important measures to be practised in rural communities in Myanmar.

1.2. Determination of serum aflatoxin B1 (Biomarker) adducts in chronic liver disease patients attending in liver unit, 500 Bedded Specialist Hospital, Yangon

Hepatocellular carcinoma (HCC) and cirrhosis of liver (COL) are common in liver unit, 500 Bedded Specialist Hospital, Yangon. Obesity and diabetes are closely associated with liver abnormality called non-alcoholic fatty liver disease (NAFLD) that may increase the risk of liver cancer. Environmental exposure to aflatoxin is one of the risk factor for development of liver cancer in underlying chronic liver disease. A cross sectional hospital and laboratory based study was carried out on total 10 normal healthy persons and 91 chronic liver disease patients (75 males and 16 females with mean aged 50±11 years) including 35 HCC cases and 56 COL cases attending to liver unit, 500 Bedded Specialist Hospital, Yangon. This study was aimed to determine AFB1-albumin adduct level in serum of all subjects by using enzyme link immunosorbent assay (Bioo Scientific ELISA kit). The minimum level (>0.25 ng/mL) of AFB1 was detected by the ELISA kit. AFB1 was detected in 11.4% (4/35 cases) of HCC and 7.1% (4/56 cases) of COL cases. AFB1 concentration was more increased(mean 0.38 ng/ml) in 4 HCC cases (2 cases were hepatitis B positive, 1 case was hepatitis C positive and 1 case was both hepatitis B and C positive) than 4 COL cases (1 case was hepatitis C positive, 3 cases were both hepatitis B and C negative)(mean 0.28 ng/ml). All AFB1 positive HCC cases had markers for hepatitis B and C. However 3 cases of COL with AFB1 positive had no markers for hepatitis B and C. This study provides AFB1 as a potential risk factor for HCC and COL.

1.3. CANCER

1.3.1. Prognostic value of p53 and apoptotic index on gastric cancer tissue

Gastric cancer is one of the most commonly occurring cancer and accounts for 4.87% among all cancers. Tumor suppressor gene p53 is the single most common target for genetic alteration in human cancers and serves as critical gatekeeper against the formation of cancer.

Tumor cells will undergo apoptosis (programmed cell death) if they have normal copies of p53 and they will resist to undergo apoptosis if the p53 gene is mutated. This study was aimed to find out the relationship and prognostic value between histological types, grades, p53 expression and apoptotic index on tissues of 40 gastric cancer cases. Histological types and grades were determined by haematoxylin-eosin stain, detection of p53 expression by immunohistochemistry and apoptotic index by Terminal deoxynucleotidyltransferase mediated deoxyuridine triphosphate biotin nick end labeling (TUNEL) method. The ages of the patients ranges from 35 to 82 years with male preponderance (M:F=3:2). The histological types were classified according to Lauren's classification as intestinal and diffuse, and graded as well, moderate and poorly differentiated. The p53 immunoexpression were scored according to intensity and positivity showing 3+, 2+ and 1+ in 4%, 27% and 9% of total cases, respectively. The apoptotic index (number of TUNEL positive cells per 1000 cells) was determined by counting a thousand tumor cells in randomly chosen areas of neoplastic cells of gastric cancer on a light microscope. The apoptotic index (AI) 40%, 30%, 20% and 10% in 7.5%, 27.5%, 55% and 10% of total cases, respectively. As a result, highest positive p53 expressions (3+) were seen in both intestinal 2/21 (9.5%) and diffuse types 2/19 (10.5%) and moderately 2/15 (13.3%) and poorly differentiated grades 2/25 (8%). The AI were higher in both intestinal 22/40 (55%) and diffuse types 18/40 (45%) and moderately 16/40 (40%) and poorly differentiated types 24/40 (60%). In this study, there were no associations between histological types, grades and p53 expression and also with apoptotic index. There was a significant correlation between the p53 expression and apoptotic index ($p<0.05$). We concluded that these markers might be good indicators of sensitivity to chemotherapy.

SERVICES PROVIDED

ACADEMIC

Sr.	Name	Course	Responsibility
1,	Dr. Kyaw Soe	Research Methodology Workshop(2015) M.Med.Sc (Medical Technology, Medical Jurisprudence)	Facilitator Teaching
2.	Daw Myat Mon Oo	M.Med.Sc (Medical Technology, Medical Jurisprudence)	Teaching
3.	Dr. Khin Kant Kaw Oo	M.Med.Sc (Medical Technology, Medical Jurisprudence)	Teaching

PATHOLOGY RESEARCH DIVISION (POL)

Research Scientist & Head	…	Dr. Khin Moe Aung MBBS, MMedSc(Pathology) (UM 2)
Research Officer	…	Dr. Nyein Nyein Thaung MBBS(UMM)
	…	Dr. Wut Hmon Min MBBS(UMM)
	…	Dr. Thae Thae Moe Han MBBS(UMM)
	…	Dr. Ei PhyoWai MBBS(UM1)
	…	Dr. Su Su Lin MBBS(UMM)
	…	Dr. Nandar Ko MBBS(UMM)
	…	Dr. Yandar Aung Myo Han BVSc(NPW)
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	…	Daw Htay Htay Kywe BSc(Botany)(UDE)
	…	Daw Nan Phyu Phyu Mar BSc(Mathematics)(MU)
	…	Daw Than Than Maw BSc(Chemistry)(UDE)
	…	Daw War War Phwe MSc(Microbiology)(MU)
	…	Daw Thant Sin Win MSc(Microbiology)(MU)
	…	Daw May Thin Kyu BMedTech(Medical Laboratory Technology)(UMTM), Diploma in English
	…	Daw Haung Naw BMedTech(Medical Laboratory Technology)(UMTM)
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	…	U Phone Zin Myint BA(English)(UDE)
	…	Daw Ohn Myint Aye BSc(Botany)(Meikhtila University)
Research Assistant (4)	…	Daw Thandar Win BSc(Chemistry)(UDE)

Pathology research division is primarily involved in research activities of the following diseases such as anaemia, iron deficiency anaemia, haemoglobinopathy, thyroid disorders, intestinal worm infestation and genetic diseases.

RESEARCH PROJECTS

1 COMMUNICABLE DISEASES

1.1 INTESTINAL WORM INFESTATIONS

1.1.1 Study of intestinal worm infestation among adult people at Pin Tee village, Pyin Oo Lwin Township

Globally, more than 3.5 billion people are infected with intestinal worms. People get infected with worms when living in an unclean environment of poor sanitation and unhygienic habits. The purpose of this study was to determine prevalence of worm infestation among adult people in Pin Tee village, Pyin Oo Lwin Township. Cross-sectional descriptive community and laboratory-based study was done to 121 cases, male 49 (40.5%) and female 72 (59.5%) from September to December 2015. Age distribution was 19-83±SD15.54 years. History taking and physical examination was done. Stool sample was collected with wide

mouth sterile plastic bottle. Stool examination for ova detection was performed with saline preparation and iodine preparation at Department of Medical Research (Pyin Oo Lwin Branch), Pathology Research Division. Among them, 77 (63.6%) respondents had attended primary school, 34 (28.1%) secondary and above school level, 7 (5.8%) read and write, 3 (2.5%) illiterate. Income per month between 100,000 and 200,000 kyats were 73 (60.3%) respondents, followed by ≥200,000 kyats 30 (24.8%), ≤100,000 kyats 18 (14.9%). They all used fly-proof sanitary latrine. Type of housing was concrete 74 (61.2%), semi-concrete 24 (19.8%) and non-concrete 29 (19%). Source of water was tap water 60 (49.6%), well 33 (27.3%) and river 28 (23.1%). Among them, 10 (8.3%) cases had history of passing worm. History of taking anthelmintics during last 6 months was 36 (29.8%) cases. Regarding hand washing with soap before eating, 61 (50.4%) washed sometime, every time 52 (43%) and not washing 8 (6.6%). Related to hand washing with soap after toilet, 79 (65.3%) washed every time, 33 (27.3%) sometime and 9 (7.4%) did not wash. The overall prevalence of intestinal worm infestation was 53 (43.8%) cases. The most prevalent intestinal helminthes were *Ascaris lumbricoides* 24 (19.8%), followed by *Taenia* infestation 14 (11.6%) and *Trichuris trichiura* infestation 3 (2.5%) participants. Mixed infestation was observed in 12 (9.8%). Of these, *Ascaris* and *Taenia* mixed infestation were examined in 9 (7.4%) participants. Others were *Taenia* and Hookworm, *Trichuris trichiura* and *Taenia*, *Trichuris trichiura* and *Ascaris* were detected in 1 (0.8%) participant each. A high percentage of cases have intestinal worm infestation and majority of them have *Ascaris lumbricoides* and may need public health intervention and health education for hand washing practice.

2 NON-COMMUNICABLE DISEASES

2.1 ANAEMIA

2.1.1 Prevalence of haemoglobinopathies among pregnant mothers attending 300- bedded Pyin Oo Lwin General Hospital

This hospital and laboratory-based, cross-sectional descriptive study was done at 300-bedded Pyin Oo Lwin General Hospital during September 2014 through February 2015. The purpose of this study was to determine severity of anaemia and types of abnormal haemoglobin among pregnant women who were attending antenatal clinics of study hospital. A total of 500 pregnant women were enrolled and their hematological parameters were assessed by using Pentra-60, haematology auto-analyzer and serum ferritin level by Mini-vidas, fully-automated immunology analyzer, HbH inclusion detection by brilliant cresyl blue dye test and Hb E by NESTROFT (Naked Eye Single Tube Red Cell Osmotic fragility test) were done and Agarose gel electrophoresis by SAS- MX Alkaline Hb-10 kits qualitatively. The mean haemoglobin levels of pregnant women during first trimester was (11.35 ± 1.16 g/dl) and second and third trimester were (10.41 ± 1.17 g/dl) and (10.58 ± 1.22 g/dl) respectively. The overall prevalence of anaemia among these pregnant women was 321 cases (64.2%) and third trimester group was significantly higher than other groups (p value < 0.05). Moreover, 108 cases (21.6%) were found to have iron deficiency anaemia in which serum ferritin level (below 15 µg/l) was used as the cut-off point to diagnose IDA in the present study and 89 cases (17.8%) had haemoglobinopathy in which 43 cases (8.6%) haemoglobin E trait (Hb AE), 6 cases (1.2%) β-thalassaemia trait ($HbAA_2$), 2 cases(0.4%) haemoglobin E β-thalassaemia (HbEF), 36 cases(7.2%) carrier of α-thalassaemia trait (Hb AH) and 2 cases (0.4%) were Hb H disease ($HbAA_2H$). There was positive correlation between haemoglobin level, haematocrit, RBC count, mean corpuscular volume and mean corpuscular haemoglobin with haemoglobinopathy cases (p value= 0.016). The findings of this study indicate high

prevalence rate anaemia among pregnant women and also highlighted that there was anaemia remains a common health problem among pregnant women. Therefore, screening programs of anaemia and clinical management of haemoglobinopathies in pregnant women of this area should be encouraged.

2.1.2 Prevalence of anaemia among elderly persons in Pyin Sar village of Pyin Oo Lwin

Anaemia is one of the most common public health problems of older population especially in developing countries. While decreased hemoglobin levels were previously largely considered a normal consequence of aging, now there is evidence that anemia is associated with an increased risk for morbidity and mortality. The aim of this study was to determine the prevalence of anemia and severity of anemia among elderly persons in Pyin Sa village tract of Pyin Oo Lwin Township. Community and Laboratory-based, cross-sectional descriptive study was done at Department of Medical Research (Pyin Oo Lwin Branch) during September to December 2015. Personal details of 100 elderly people over and age of 60 years old were collected in each case, and taken 2 ml of blood under aseptic condition. Haematological parameters were measured by haematological auto-analyzer and blood film examinations were done. According to hemoglobin (Hb) cut-off levels defined by the World Health Organization (WHO) anemia is defined as Hb <12 g/dL for females and <13 g/dL for males. There are three grades of anemia which are differentiated as severe (Hb <8 g/dL), moderate (Hb 8 to <9.5 g/dL), and mild (Hb≥ 9.5 g/dL) respectively. The overall prevalence of anaemia among the older persons was 30 cases (30%). The majority 28% showed mild anaemia, and moderate anaemia was recorded in 2%. Results showed that prevalence of anemia 30% contains two types of anemia which were hypochromic microcytic anemia 27% and macrocytic anemia 3% in this study. Therefore, the results showed awareness of anemia in this studied population and geriatric persons should be routinely screened for anemia for early diagnosis, effective treatment and prevention are important in rural area of developing country.

2.1.3 Haematological findings in rural community in Pin Tee village, Pyin Oo Lwin

The importance of anaemia as a major public health problem throughout the world is widely recognized. The aim of this study was to determine the prevalence of anemia and severity of anemia among adult rural people in Pin Tee village of Pyin Oo Lwin Township. Community and Laboratory-based, cross-sectional descriptive study was done at Department of Medical Research (Pyin Oo Lwin Branch) during September to December 2015. A total of 263 people in which male 110 cases (41.8%) and female 153 cases (58.2%) were enrolled and age distribution was from 18 to 86 years and the mean age was 44.58 years (SD-15.96 years). After taken 2 ml of blood under aseptic condition, haematological parameters were measured by haematological auto-analyzer and blood film examinations were done. According to hemoglobin (Hb) cut-off levels defined by the World Health Organization (WHO) anemia is defined as Hb <12 g/dL for females and <13 g/dL for males and severity was differentiated as severe (Hb <8 g/dL), moderate (Hb 8 to <9.5 g/dL), and mild (Hb ≥ 9.5 g/dL) respectively. The overall prevalence of anaemia among those participants was 69 cases (26.2%) and severity of anaemia showed 44 cases (16.7%), 24 cases (9.1%) and only one case (0.4%) respectively. There was significant association between sex distribution and severity of anaemia (p value= 0.03). In blood film examination, there were normal blood film 146 cases (54.1%), hypochromic microcytic anaemia 39 cases (14.4%), anaemia with eosinophilia 30 cases (11.4%), only eosinophilia 35 cases (13.3%) , polycythaemia 6 cases (2.2%), thrombocytopenia 4 cases(1.5%) and leukocytosis 3 cases (1.1%) respectively.

Therefore, the results showed awareness of anemia in this studied population and useful for screening programs and clinical management of anaemia in this area.

2.2 METABOLIC SYNDROME

2.2.1 Metabolic risk factors and associated morbidities among adult urban people in Pyin Oo Lwin Township

This Community and laboratory-based cross-sectional descriptive study was conducted at Department of Medical Research (Pyin Oo Lwin Branch) during September to December 2015. The purpose of this study was to identify metabolic risk factors and associated morbidities among adult urban people in Pyin Oo Lwin Township. A total of 355 people in which male 94 cases (26.5%) and female 261 cases (73.5%) were enrolled and systematic sampling procedure were used. Age distribution was from 18 to 85 years and the mean age was 49.98 years (SD-15.22 years). Metabolic risk factors were identified according to NCEP ATP III (National Cholesterol Education Program Adult Treatment Panel III) guideline. Waist circumference male > 90 cm, female > 80 cm, High triglycerides ≥ 150 mg/dl, low HDL-cholesterol male < 40 mg/dl and female < 50 mg/dl, elevated blood pressure ≥ 130/ 85 mmHg, and elevated fasting glucose ≥ 110 mg/dl. Metabolic syndrome was if ≥ 3 of the categorical cut points. In this study, central obesity was the highest component 145 subjects (40.8%) followed by elevated triglycerides 129 (36.3%), elevated blood pressure 104 (29.3%), low HDL-cholesterols 85(23.9%), and elevated fasting glucose only 48 individuals (13.5%).The prevalence of metabolic syndrome was 35.2% with prevalence's in men and women were 26.6% and 38.3% respectively. There were statistical significant associations between all five metabolic risk factors with metabolic syndrome (p value <0.001). Regarding the liver function tests, the range of total bilirubin was from 1.96 to 17.33 µmol/l (mean 5.77± 2.08 µmol/l), AST level from 12.14 to 149.30 IU/L (mean 30.10± 16.87 IU/L), ALT level from 9.09 to 192.40 IU/L (mean 27.31± 17.07 IU/L) and ALP level from 75.46 to 436.90 IU/L (mean 188.87±58.76 IU/L) respectively. Renal function test showed urea level 6.78 to 59.31 mg/dl (mean 29.74± 9.80 mg/dl) and creatinine level from 0.40 to 1.90 mg/dl (mean 0.72± 0.21 mg/dl). Serum calcium level from 4.00 to 16.10 mg/dl (mean 8.01±1.51 mg/dl) and serum uric acid level from 2.40 to 11.00 mg/dl (mean 5.78±1.38 mg/dl) were observed. ECG examinations showed normal 296 cases (83.4%) and abnormal in 59 cases (16.6%). In Bone Mass Density by T- score using quatitative Ultrasound examination, the results showed normal 168 cases (47.3%), osteopenia 174 cases (49.0%) and osteoporosis 13 cases (3.7%) respectively. In conclusion, this study revealed the high prevalence rate of metabolic syndrome and will be depicted about the metabolic risks as the baseline data for the implementation of further activities to reduce the incidence of non-communicable diseases.

3. TRADITIONAL MEDICINE

3.1 ANTI-BACTERIAL ACTIVITY

3.1.1 *In vitro* Antibacterial Activity of *Aloe vera* (L.) Burm.f. (ရှားစောင်းလက်ပတ်) Leaf Extracts

Pathogenic bacteria are major causes of human morbidity and mortality. It is necessary to know antibacterial action of traditional herbal extract. *Aloe vera* (L.) Burm.f. (ရှားစောင်းလက်ပတ်) is one of the most popular medicinal plants in our country, belonging to the family Aloaceae. The present study dealt with *in vitro* antibacterial activity of *Aloe vera* (L.)

Burm.f. (ရှားစောင်းလက်ပတ်) leaf extracts. This was a laboratory based experimental study carried out at the Department of Medical Research (Pyin Oo Lwin Branch) during October 2015 to February 2016. The leaf samples were collected from Mandalay Region in October 2015. The collected plant was identified according to the taxonomic characters based on the Backer & Brick (1968) and Dassanayake (2000) by taxonomist. Extraction of leaves was done by maceration method. The 95 % ethanol, methanol and distilled water were used as solvents. Strains of pathogenic bacteria were tested in this study such as *Pseudomonas aeruginosa, Staphylococcus aureus* and *Escherichia coli.* Agar disc diffusion technique in Modified Kirby and Bauer method was used to determine the zone of inhibition of three extracts of *Aloe vera***.** The minimum inhibitory concentration (MIC) and the minimum bactericidal concentration (MBC) testing were done by broth dilution method. Among different extracts, the 95 % ethanolic extract showed larger zone of inhibition, i.e., 7 mm to 10 mm for *Pseudomonas aeruginosa*. The methanolic extract showed 7.5 mm and the distilled water extract did not show antibacterial activity. Three kinds of extract did not show antibacterial activity on *Staphylococcus aureus* and *Escherichia coli*. The MIC of 95 % ethanolic and methanolic extracts against *Pseudomonas aeruginosa* was 12.5 mg/ml. The MBC of 95 % ethanolic and methanolic extracts against *Pseudomonas aeruginosa* was 25 mg/ml. Therefore, the finding of research was very applicable for further studies of *Aloe vera*.

SERVICES PROVIDED

ACADEMIC

Sr. No.	Name	Course	Responsibility
1.	Dr. Khin Moe Aung	1st yr. M.Med.Sc (Biochemistry)	Demonstration

LABORATORY

Sr. No.	Laboratory Tests	Tested Samples
1.	CP and Blood film report	111 samples
2.	Bone Mass Density	725 samples
3.	ECG	725 samples
4.	Tissue Processing and H & E stain	7 samples
5.	Urea	375 samples
6.	Creatinine	740 samples
7.	Calcium	477 samples
8.	Total Cholesterol	740 samples
9.	Triglycerides	740 samples
10.	HDL	740 samples
11.	Uric Acid	628 samples