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## INSTRUCTIONS TO AUTHORS
MESSAGE FROM THE EDITOR-IN-CHIEF

DEAR READERS

As an Editor-In-Chief of the Mongolian Journal of Health Sciences, it is really honor to introduce this issue to the hundreds of researchers, health professionals, academic staffs, graduate students and others, who are loyal readers and contributors.

In this issue of the Mongolian Journal of Health Sciences we offer our heartfelt thanks to the volunteer peer reviewers who help ensure the quality and integrity of the journal. It is tremendous contribution of all the editors to Mongolian Medical Sciences.

This issue includes broad area of research works in the fields of health sciences including Traditional Medicine, Nursing, Medicine, Public Health and etc. We sincerely believe that scientific articles published in this issue would be useful for all researchers, doctors and health care professionals to enhance their knowledge and obtain new research ideas for their future work.

All the best wishes to the dear readers of Mongolian Journal of Health Sciences!

Editor-in Chief

Academician, Professor Lkhagvasuren Ts (M.D., Ph.D., D.Sc.)
Antithrombotic and Anticoagulation Effects of Caragana Jubata Pall poir

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ABSTRACT
To study the anticoagulation and antithrombotic activities of the medicinal plant Caragana jubata Pall Poir on the experimental model of deep vein thrombosis induced in laboratory animals. Thirty rats were used for the experiment and divided randomly and equally into 3 different groups: control group (saline), test group with C.Jubata and Warfarin treated group as a comparison. The parameters such as Prothrombin time (PT) and the activated partial thromboplastin time (APTT) were measured by means of Quick’s one-stage assay and modified APTT assay respectively in the rats. Additionally, a thrombin activity test was estimated in rats with PT assay using a hemagglutination analyzer. Thrombosis pathologic experimental model was induced.

Thrombi were recorded in all rats after experimentally induced thrombosis. As seen in histopathological assessment in the inferior vena cava of control group had formed thrombosis with the fibrin thread, erythrocytes and leucocytes. However in Caragana jubata pall poir (C.Jubata) group we noticed a little mass of fibrin and few leucocytes, and full of erythrocytes. Hence blood elements were few; formed thrombus was small and vessel cavity was free. Red thrombus formed in vein of Warfarin group animals has fibrin mass, occurring mostly in the wall, where as leucocytes were few, and erythrocytes were full. Fibrin thread was similar to the control group. As a result of research it was revealed that C.jubata prolonged Prothrombin time by 32.5%, thrombin time by 39.8%, where as fibrinogen amount decreased by 43.2%. It shows that C.Jubata has an anticoagulation activity while inhibiting blood coagulation and thinning blood.

C.Jubata medicinal plant has anticoagulation activity and inhibits thrombosis.

Key words: inferior vena cava ligation, thrombosis model, anticoagulation activity, Caragana jubata Pall Poir.

INTRODUCTION
Venous thromboembolism, which includes deep vein thrombosis and pulmonary embolism, is a major cause of morbidity and mortality. It is also a serious health problem in the world, which plays an important role in the pathogenesis and progression of atherosclerosis, cardiovascular diseases and diabetic complications. Nowadays the risk factors for thrombosis include blood stasis, vessel wall injury, and hypercoagulability, as proposed by Virchow over 150 years ago. According to the theory of traditional Mongolian medicine blood essence disturbed by disease development and hence blood disease arises. On the other hand, symptoms of blood thickening and forming thrombosis considered as a disease occurring by means of impure blood. In the Mongolian traditional medicine C.Jubata is used for curing blood thickening or impure blood.

The genus Caragana is a member of the family Fabaceae, subfamily Faboideae and is native to arid and semi-arid areas of the temperate zones of Asia and Eastern Europe. Many species are cultured for dune-fixation, livestock forage and biological resources for fuel energy and fiber production. More than 10 species in this genus have a long history of use in traditional Chinese, Mongolian and Tibetan medicines and are believed to “nourish yin, invigorate the spleen, temper the blood and promote blood flow”. They have been used for the treatment of a wide range of ailments including fevers, inflammation, wounds and infections, dizziness, headache, hypertension, female disorders, arthritis and cancer. Over 100 phytochemicals have been identified with flavonoids and stilbenoids being the major constituents of this genus. Clinical
studies have demonstrated the pharmacological activities of different Caraganum species, e.g. Caragana sinica for the treatment of hypertension, and in vivo and/or in vitro studies have provided some support for other traditional uses, e.g. anti-cancer, anti-inflammatory, phytoestrogenic, immunostimulant and immunosuppressant activities. Thus, we chose to study the anticoagulation and antithrombotic activities of the C.Jubata medicinal plant on the experimental model of deep vein thrombosis induced in laboratory animals.

**MATERIALS AND METHODS**

The study protocol was approved by the Ethics Committee of the Health Sciences of University.

**Materials**

Research was conducted in the laboratory of pharmacology of Traditional Medical Science Technology and Production Corporation. Experiments were carried out at 30 male Wistar albino rats with 180-220 g as an animal model. Rats were kept in cages at room temperature (20 ± 2°C) and humidity (55 ± 5%) with 12 h cycle of light and dark. They were given free access to commercial animal plated diet and water.

**Decoction preparation**

C.Jubata decoction was prepared in the traditional medical factory of Traditional Medical Science Technology and Production Corporation of Mongolia. The plants were collected and authenticated by the expert at the Department of Botany, Institute of Biology and Mongolian Academy of Sciences. 10 g crushed dried plant material was suspended in 250ml water and boiled till water evaporated to 100 ml. Experimental group

Thirty rats randomly divided into three groups of ten rats of each (25 thrombotic rats, 5 normal rats) used for the study. Group 1: normal control rats received distilled water daily, orally by gavages, for a week, Group:2 received Zomoshin-6 decoction 200 mg/kg and Group:3 Warfarin 3.23 mg/kg daily for a week.

**Determination of PT, APTT and thrombin activity in rats**

PT, APTT and thrombin activity were estimated by reported standard methods as indication for blood coagulation. PT was measured by means of Quick’s one-stage prothrombin test and APTT by means of modified APTT assay using an EA-containing APTT reagent. Thrombin activity was determined by PT assay with a PK-B hemagglutination analyzer (Zhongshan Peikang Limited Company for Medical Electronic Instruments, Zhongshan, China).

**Induction of thrombosis**

Stasis-induced thrombosis was tested after injection of tissue thromboplastin. Thrombus formation was induced by a combination of stasis and hypercoagulability as described by Herbert et al. Rats were anesthetized with ketamin hydrochloride (90 mg/kg, intraperitoneally).

**Blood collection and dissection**

At the end of the experiment, blood samples were collected from each rat by cardiac puncture into heparinized tubes and the plasma was separated by centrifugation at 1500 g for 10 min. The rats were sacrificed and dissected. The thrombosis was collected, fixed in 10% formalin and processed routinely for histopathological evaluations.

**Statistical analysis**

The data are expressed as the mean±standard error of mean (S.E.M.). The data were elevated by a one-way ANOVA SPSS program and the means were assessed using Tukey’s test. Statistical significance was considered at p<0.05.

**RESULTS**

Effects of C.Jubata on anticoagulation activity in rats

As a result of the research it was revealed that C.Jubata prolonged prothrombin time by 32.5%, thrombin time and activated partial thromboplastin time by 39.8% where as fibrinogen amount decreased by 43.2%. It shows that C.Jubata has an anticoagulation activity, while inhibiting blood coagulation and thinning blood (table 1).

<table>
<thead>
<tr>
<th></th>
<th>PT (sec)</th>
<th>APTT (sec)</th>
<th>TT (sec)</th>
<th>Fibrinogen (g/l)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>10.7±0.56</td>
<td>19.6±0.5</td>
<td>21.6±4.1</td>
<td>3.0±0.1</td>
</tr>
<tr>
<td>C.Jubata</td>
<td>15.86±6.4*</td>
<td>42.3±6.4**</td>
<td>30.2±2.6*</td>
<td>1.7±0.3*</td>
</tr>
<tr>
<td>Warfarin</td>
<td>9.97±0.4</td>
<td>45.0±0.3**</td>
<td>49.2±8.6**</td>
<td>2.6±0.02</td>
</tr>
</tbody>
</table>

*-p<0.05, **p<0.01 Values are mean ± SE. Values within a column having different superscripts are significantly different at p<0.05 and p<0.01.

**Histological studies**

The effect of the C.Jubata was further confirmed by histopathological examination. Thrombi were detected in all rats after experimentally-induced thrombosis. Histopathological analysis demonstrated the presence of thrombi in the inferior vena cava (IVC) of the control group, composed of fibrin thread, erythrocytes, and leucocytes and obstructed lumen. During the experiment red thrombus formed in the vein was blocked in the capillaries, and in the large vessel it has a shape of walling (Figure 1). Red thrombus blocked in the vein of control group animal had a lot of threads, which stacked with each other and contain few leucocytes, but many erythrocytes. Only a small amount of fibrin clot, containing a few leucocytes and large numbers of erythrocytes, was observed in the C.Jubata
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treated group. Few blood elements; minimal thrombus and free vessel cavity were observed in this group (Figure 2). In comparison with control group fibrin thread was less and thrombin forming process was not so quick. Red thrombus features formed in the veins of the Warfarin treated animals consisted of fibrin clot, which was mostly attached to the wall, with few leucocytes but abundant erythrocytes, and it was similar to the control group. Fibrin thread features was variable (Figure 3).

**DISCUSSION**

Blood coagulation has long been considered an important factor in the pathogenesis of venous thrombosis, a pathological condition particularly occurring in post traumatic and postoperative periods⁵. The inferior vena cava is ligated to cause complete or partial stasis measured the extent of thrombus formation 15 and 60 minutes after IVC ligation in rats. Even after 15 minutes, thrombi were detected. These thrombi contained fibrin, platelets, and red blood cells, along with the occasional leukocyte⁶. Thrombosis is associated with blood coagulation and endothelial lesions⁷. Thus, we propose that C.Jubata can inhibit thrombosis. To confirm the rat model of inferior vena cava ligation was used. The results suggested that C.Jubata had an inhibition effect on thrombosis and markedly reduced thrombus formation. Blood coagulation is not only the result of a complex process initiated by the intrinsic system or the extrinsic system and/or a common pathway, but also a highly regulated process involving interactions between platelets, plasma coagulation factors, and the vessel wall ⁸. In the clinical test of blood coagulation, APTT is used to evaluate the intrinsic clotting index. A prolonged APTT usually represents deficiency in factors VIII, IX, XI, XII and V on Willebrand’s factor. PT is used to evaluate the extrinsic clotting pathway. A prolonged PT indicates a deficiency in coagulation factors V, VII and X ⁹. TT is a test of fibrin formation, which is directly induced by the addition of thrombin. The test therefore only detects disturbances in the final stages of coagulation, especially dysfibrinogenaemia or the presence of thrombin inhibitors¹⁰. These tests have proved useful in identifying bleeding risks so they can be utilized to evaluate thrombotic risks. Thus, the APTT, PT and TT were tested. The results demonstrated that C.Jubata prolonged APTT, PT and TT but showed more a powerful effect on APPT and TT than PT, suggesting that C.Jubata mainly inhibited extrinsic and instrinsic clotting pathway of coagulation and fibrin formation. In this study, was tested the effectiveness of C.Jubata on inferior vena cava ligation thrombosis model of anticoagulation assay was tested on rats. The results presented above indicated that C.Jubata had significant anti-thrombotic and anticoagulation effects in vitro. In conclusion, C.Jubata has anticoagulation activity and inhibits thrombosis.

**Acknowledgements**

This work was supported by a project from the Science Technology Foundation of Mongolia.

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Anti-Liver-Cancer Effect of Scutellaria Baicalensis Georgi.


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5 Traditional Medical Science Technology and Production Corporation of Mongolia

ABSTRACT
The number of incidences of liver cancer has been increasing and in the usage of phytogenic drug for medical treatment has been dominated due to its less side effects. Thus, the number of studies concerning on these issues have intensively increased. The goal of this study was to study effectiveness of Scutellaria baicalensis Georgi. extract on the liver cancer cells via in vitro. There is an expectation that the etanol extract prepared from Scutellaria baicalensis Georgi. might have an inhibition effect on primary cell of liver cancer. In this study, we used primary cancer cell-PCC of liver cancer, MDCK cell line derived from dog kidney and etanol extract prepared from the upper side of Scutellaria baicalensis Georgi. In addition, Skullcap-a medicinal plant extracted from the root part of Scutellaria baicalensis Georgi, the plant extract of Scutellaria baicalensis Georgi. Skullcap preparation did not show any destructive and decomposition effect on DNA of the cell genom. However, DNA concentration extracted from a single cell was relatively different from each other. This might be due to the number of the live cells remained after influencing by plant extraction. The plant extraction of Scutellaria baicalensis Georgi and Skullcap preparation do not have direct effect on the cell DNA but kill them by other ways. According to our study, 100µg/ml extract of Scutellaria baicalensis Georgi. can activate Bcl-2 gene expression of the cell with 6%. Skullcap preparation (100 µg/ml) and Scutellaria baicalensis Georgi. inhibited ZFYVE1 gene expression by 4.8% and 13.2% respectively. GAPDH gene expression was inhibited by 100 µg/ml each of preparation and extract by 13.4% and 18.9% respectively. The plant extract of Scutellaria baicalensis Georgi. and Skullcap preparation did not show any destructive and decomposition effects on DNA of the cell genom. The etanol extraction of Scutellaria baicalensis Georgi. derived from upper part of the plant does not induce apoptosis, but transmembrane proteins can block cancer cell division by inhibiting their gene expression.

Key words: Scutellaria baicalensis Georgi. nuclear DNA of cancer cell, gene expression.

INTRODUCTION
The number of incidences of liver cancer has been increasing and in the usage of phytogenic drug for medical treatment has been dominated due to its less side effects. Thus, the number of studies concerning on these issues have intensively increased. Nowadays, there are many studies carrying out on medicinal plants, such as, whether they can be used as original herb or mixed as medicinal compound, and purification of substances from plants and producing new medicines from them, and explanation of function and their mechanisms. In Mongolian Traditional Medicine, Scutellaria baicalensis Georgi. is used for drying blood, lowering blood pressure, reducing dithering fever and internal organs’ fever.

Liver cancer and its complications occupy a special place among morbidity and mortality in our population, and they tend to increase. However, diagnostics and treatment of liver cancer have not been fully solved. Therefore, we have selected Scutellaria baicalensis Georgi. For our study, in order to study the effectiveness of Scutellaria baicalensis Georgi. extract on the liver cancer cells via in vitro. Scutellaria baicalensis Georgi. belongs to the Lamiaceae family and seven species of this family had been identified in Mongolia. This is one of the important ingredients of 242 medicines of the East Medicine and occupies 17th place by its frequency (B.Boldsaikhan).

MATERIALS AND METHODS
We carried out this study in Molecular Biological Laboratory of Institute of Biological Sciences, the Mongolian Academy of Sciences, and Reference Laboratory for Medicines of the Public Health Institute of by using experimental model. We have collected plant specimen of Scutellaria baicalensis Georgi. from the Ulz river valley of Norovlin soum, Khentii aimag. We dried the plant of Scutellaria baicalensis Georgi.
and grinded in Reference Laboratory for Medicines and then made an extract from them by using 70% ethyl alcohol as described in percolation method. We had thicken this extraction with vacuum thickener. H2O was added to the extraction to make a watery extract and then 70% ethyl acetate was used as a solvent. Extract of *Scutellaria baicalensis* Georgi. and preparation have been solved in 25 µg/ml, 50 µg/ml, 100µg/ml, 200-250 µg/ml, 500 µg/ml media and incubated in cancer cell. In this study, we used primary cancer cell-PCC of liver cancer, MDCK cell line derived from dog kidney and etanol extract prepared from the upper side of *Scutellaria baicalensis* Georgi. and Skullcap-a medicinal plant extracted from the root part of *Scutellaria baicalensis* Georgi. PCC cell have been cultured in RPMI-1640 and MDCK cells in DMEM medium. From PCC, MDCK cell lines 6 x 10^12-12 x 10^12 cell/ml isolated and cultured in 24 hole-cell culture with 5% CO₂ media for 24-48-72 hours in 37˚C. Extract of *Scutellaria baicalensis* Georgi. and preparation have been solved in 25 µg/ml, 50 µg/ml, 100 µg/ml, 200-250 µg/ml, and 500 µg/ml media and incubated in cancer cell 12-14.

Method for DNA ladder assay: We used 6 hole cell culture panel and 3 of them have been cultured with same number of cells. By using I-genomic DNA extraction kit, we extracted genomic DNA from experimental and control cells, and we analyzed them by 0,8% agarose gel electrophoresis method and took photos. **

Method for gene expression: Bcl-2, ZFYVE1, GAPDH genes are involved in cancer cell division and apoptosis and their gene expression carried out by using RT-PCR in RNA isolated from the experimental and control cells with extract and without extract. RT-PCR products tested with agarose gel and cell genomic products with extract compared with control cells and they were calculated by ImageJ program.

RESULTS

**Result of the DNA ladder assay:** The plant extract of *Scutellaria baicalensis* Georgi. and Skullcap preparation did not show any destructive and decomposition effect on DNA of the cell genom. However, DNA concentration extracted from a single cell was relatively different from each other. This might be due to the number of the live cells remained after influencing by plant extract. The plant extract of *Scutellaria baicalensis* Georgi. and Skullcap preparation do not have direct effect on cell DNA, but killing them by other ways (Figure 1).

**Figure 1.** Cell DNAs affected by etanol extract and preparation, prepared from *Scutellaria baicalensis* Georgi. and control without them.

C – Control, S.B – *Scutellaria baicalensis* extract, S.C – Skullcap preparation

**Result of the gene expression:**

**Bcl-2 gene expression:** Bcl-2 gene located on chromosome 18q21 and 197bp long. This gene encodes Bcl-2 protein that regulates apoptosis. This protein is called Apoptosis regulator Bcl2 and consist of 65 amino acids. 2.4кDa=2400Da and located in outer mitochondrial membrane and nuclear membrane. If function of Bcl-2 gene becomes abnormal, then cell division will be uncontrollable. Over-expression of anti-apoptotic Bcl-2 gene can result in the lack of cell death and uncontrollable cell division that is characteristic of cancer. According to our study, 100 µg/ml extract of *Scutellaria baicalensis* Georgi. can activate Bcl-2 gene expression of the cell with 6% (Figure 2).

**Figure 2.** Bcl-2 gene of the cell affected by etanol extract and preparation, prepared from *Scutellaria baicalensis* Georgi. and control without them.

A. 1%-agarose gel М-100bp DNA marker, (Х)- control, (S)- with Skullcap preparation (100 µg/ml), S.B – *Scutellaria baicalensis* extract, 

B. Changes occurring in Bcl-2 gene expression with 100 µg/ml extract (%)

**Result of ZFYVE1, GAPDH gene expression:** ZFYVE1 gene is located on chromosome 14q24 and 285bp long. Protein of this gene plays a role for membrane transferring and cell receptor. Thus, this gene regulates apoptosis
induced by death receptor or outer receptor. According to our study, 100 µg/ml extract of Scutellaria baikalensis Georgi, and Skullcap preparation inhibited ZFYVE1 gene expression by 4.8% and 13.2% respectively (Figure 3-B1). GAPDH gene is located on chromosome 12p13 and 426bp long. Protein participates in the processes of transcription, RNA expression, DNA replication, and apoptosis. According to our study, GAPDH gene expression was inhibited by 100 µg/ml each of preparation and extract by 13.4% and 18.9% respectively (Figure 3-B2).

Figure 3. Result of ZFYVE1, GAPDH gene expression of the cell affected by ethanol extract and preparation, prepared from Scutellaria baikalensis Georgi, and control without them. B.1, B.2 - Changes occurring in ZFYVE1 and GAPDH gene expression with 100 µg/ml extract (%)

DISCUSSION
The clinical management of cancer invariably involves diverse conventional modalities, including surgery, chemotherapy and radiation. The complex characteristics of human cancer may also require alternative management to improve the therapeutic efficacy of conventional treatment and /or the quality of life for cancer patients. Complementary and alternative medicine (CAM) has recently gained attention for cancer management, since CAM covers a wide spectrum of ancient to modern approaches that expand options for preventing and treating diseases. Botanicals have been a valuable source of therapeutic candidates for new compounds since tremendous chemical diversity is found across the millions of species of plants. Thus, botanicals have contributed significantly to cancer therapy for the past 30 years, and it is likely that this class of medication will continue to be important in cancer therapeutics. Over the years, the anticancer activities of Scutellaria baikalensis extract and its constituents have been evaluated. Published studies have focused on prostate and liver cancer.15-17

CONCLUSION
The plant extraction of Scutellaria baikalensis Georgi, and Skullcap preparation did not show any destructive and decomposition effect on DNA of the cell genom. The ethanol extract of Scutellaria baikalensis Georgi, derived from upper part of the plant does not induce apoptosis, but transmembrane proteins can block cancer cell division by inhibiting their gene expression.

ACKNOWLEDGEMENT
This study is carried out in the framework of the Mongolian foundation for Science & Technology project called “Pharmacological studies on anti-bacterial and anti-viral actions of some medicines and preparations of Mongolian Traditional Medicine”. Thus, we would like to thank the support and interest of Pr. Oldokh S and its colleagues of TMS, HSUM, Academician. Oyunsyren Ts and its colleagues of Mongolian Academy of Sciences and colleagues of Reference Laboratory for Medicines of the Public Health Institute and research worker Munkhjargal B of the Botanical Institute, Mongolian Academy of Sciences.

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Atherosclerotic Findings by C-Reactive Protein and Cardio-Ankle Vascular Index of Mongolian and Japanese People

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ABSTRACT

Mongolian people suffer from atherosclerotic diseases more than Japanese people, while both people are thought to share similar genetic background under different lifestyles and environments. Comparative studies using novel atherosclerotic parameters of Mongolian and Japanese people may demonstrate the atherosclerotic features of both populations. We have recently reported the findings regarding atherosclerosis of both populations using circulating high sensitivity C-reactive protein (CRP: as low-grade inflammatory parameter) and cardio-ankle vascular index (CAVI: as arterial stiffness parameter), herein summarizing the data. Our studies revealed that in comparison to the Japanese subjects, the Mongolian subjects had higher levels of CRP and CAVI, in addition to a higher percentage of current smoking and higher levels of body mass index, heart rate and blood pressure and insulin, even though there were not so higher levels of serum total cholesterol and glucose. These results were confirmed in healthy young subjects and patients with hypertension and diabetes mellitus. These comparative studies used recent parameters suggest that Mongolian people may be at higher risk for cardiovascular disease than Japanese people. The management strategies of atherosclerotic diseases are also expected in the near future.

Key words: atherosclerosis, altitude, ethnic, cardio-ankle vascular index, C-reactive protein

INTRODUCTION

Atherosclerosis is leading pathophysiologic basic pathway of cardiovascular disease (CVD) including cerebrovascular disease, coronary heart disease (CHD) and hypertension (HT) is nowadays a possible worldwide cause of premature death1. Recently, in a clinical practice arterial stiffness has been established blood pressure independent a novel atherosclerotic parameter cardio-ankle vascular index (CAVI). The increased level of CAVI is associated to risk of CVD and predictive to HT, CHD, chronic kidney disease and diabetes mellitus (DM)2,3. Vascular inflammation is one of the pathophysiologic processes of atherosclerotic lesion, investigated in serum high sensitive C-reactive protein (hsCRP) which induces adhesion molecule expression in human endothelial cells4. Small elevation in serum levels of hsCRP are associated to higher risk of atherosclerosis and ischemic heart disease in apparently healthy population5,6. Higher level of CAVI and CRP are established in Mongolian people than Japanese people are showed there have higher risk of CVD7,8. Morbidity and mortality rates of CVD, levels of CAVI and serum CRP are different in all of ethnical countries associated to their food intake, unhealthy lifestyle, genetic background and environmental risk factors (exp: altitude difference, oxidative stress…). Ethnical comparative studies significant for each country to detect a crucial features of lifestyle related diseases and important to select preventive suitable methods. There have been several ethnical comparative studies of Mongolian and Japanese population which are including in Asian country7,8,10,11. Atherosclerosis related early preventive method is important to decline premature deaths of cardiovascular events.

EPIEMIOLOGIC FEATURES OF ATHEROSCLEROTIC DISEASE IN JAPAN AND MONGOLIA

Japanese people are the world highest life expectancy population and have been maintained over 20 years12. Global feature of CVD in Japan is high stroke and low CHD mortality rate among industrialized country however serum total cholesterol (TC) and smoking consumption rate was high13. In 1965, age-adjusted all-stroke mortality rate in Japan was highest in the world after then the mortality rate was gradually reduced until 199014. Mortality rate of CHD
also reduced from 1970 recorded that diagnosis of heart failure has a higher proportion in Japan than in the United States. According to the World Health Organization (WHO) data 2009 reported age-standardized death rate per 100 000 people stroke was 42.20, CHD was 32.14, DM was 4.53 and HT was 1.74 in Japan, respectively. HT and smoking are the main reason to reduce stroke and CHD in Japan. The prevalence of HT was decreased by 17% in 1990, prevalence of smoking is decreased by 43.2% in men and 17.5% in women compared to 1965, respectively. Current health global situation in Japan is a high level of serum TC however mortality rate of CVD is decreased related to CHD.

The WHO statistical data in 2008 reports Mongolian people to have a 15-years shorter life expectancy than Japanese people however their genetic backgrounds are similar. Mongolian statistical data in 2008 investigated, non-communicable diseases (NCD) such as diseases of the respiratory system, digestive system, genitor-urinary system, circularity system and injury, poisoning is leading five causes of morbidity and increased 25-40% compared to 2001. Since 1991, mortality rate of circularity system has been increasing and become global first health issue in Mongolia. The statistical data showed morbidity and mortality rates of NCD such as CVD had been rapidly increasing in Mongolia. The data WHO 2009 reported age-standardized death rate per 100 000 people stroke was 185.60, CHD was 92.82, DM was 2.05 and HT was 50.98 in Mongolia, respectively. This data was showed mortality rate of stroke, CHD and HT is 3-5 folds higher in Mongolia than Japan. Ischemic heart disease morbidity rate of inpatients accounted for 19.2% diseases of the circularity system in 2000, 25.7% in 2004, 30.1% in 2008. Mortality rate caused by NCD such as stroke, CHD and HT had been Mongolian health global issue however the concentration of TC is lower than Korean people and Japanese people. Therefore comparative study is helpful to clarify required ethnical health feature issue and predict premature deaths.

Therefore, there are required country special new preventive methods to decrease premature deaths, mortality and morbidity rates of CVD in both countries.

**NEW MARKERS OF ATHEROSCLEROTIC DISEASES: CARDIO-ANKLE VASCULAR INDEX (CAVI) AND C-REACTIVE PROTEIN (CRP)**

CAVI is arterial stiffness measurement blood pressure independent new marker. This index is measured VaSera VS-1000 made by Fukuda Denshi Co., Ltd in Japan. Formulation of CAVI is basic of the stiffness parameter-β theory and the values are calculated by: a \{(2ρ/ΔP)xIn(Ps/Pd) xPWV^2\} + b (ρ: blood density, Ps: systolic blood pressure, Pd: diastolic blood pressure, ΔP: Ps-Pd, a and b: constant). Measurement method of CAVI is non-invasive and superior of brachial-ankle pulse wave velocity which had been currently using in a clinical practice. CAVI is able to investigate whole body vascular stiffness including from aorta to ankle. There have several clinical studies regarding CAVI and their relation to CVD. In fact, CAVI is associated with atherosclerotic risk factors and predict the future of CVD. In patients with HT or CHD, CAVI is positively correlated to carotid intima-media thickness (IMT) and helpful to manage anti-hypertensive medication. Furthermore, increased CAVI level is associated to diabetic micro-vascular complication such as neuropathy and nephropathy patients with type II DM. The clinical significance of the CAVI for CVD has been established recently.

Recent studies investigated that inflammation plays a role in the pathogenesis of CVD associated to the initiation and progression of atherosclerosis. CRP is an acute-phase inflammatory protein primarily synthesized and released from hepatocytes. Several studies found CRP may be occurring other sites such as macrophages and smooth cells which play pathogenesis of vascular atherosclerosis. For example, Morimoto J et al also noted CRP found early atherosclerotic lesion of human coronary arteries such around foam cells. High concentration of serum CRP may be related to accumulation of CRP in early atherosclerotic vascular wall lesion and thought to be predictive of CVD.

**COMPARATIVE STUDIES FINDINGS OF JAPANESE AND MONGOLIAN PEOPLE**

Regarding to the comparative study, Mongolian healthy young (age 18-25) subjects had significantly higher percentage of meat intake and current smoking (47.0% vs 19.0%) compared with that Japanese subjects (13% vs 2.6%), respectively. Other lifestyle factors including vegetable intake, salt intake and physical activity were not significant differences. It may be related the study participants were young and the data questionnaire was a general. The study, who participated residents of Murun (prefecture) in Mongolia showed a daily calorie intake is 1.3 fold higher in Mongolian people and their have association with lifestyle related diseases than Japanese people. Salt intake is one of the lifestyle related risk factor to increase blood pressure associated to cardiovascular function. Nutritional data 2000 reported a total daily salt intake is 16.7g in Mongolian population and 12.5g in Japanese population, respectively. Additionally, Mongolian people have higher level of oxidative stress markers including malondialdegyde-modified low density lipoprotein cholesterol (LDLC), urinary 8-hydroxy-2'-deoxyguanosine and serum reactive oxygen metabolism compared to Japanese people. Several researchers found lower level of plasma n-3 polyunsaturated fatty acid (n-3 PUFA) in Mongolian people which related to higher
risk of CVD [21,25]. N-3 PUFA includes in a fish which have high consumption Japanese people. The lower level of n-3 PUFA negatively related to triglyceride (TG) in Mongolian people, higher n-3 PUFA associated to HDL-c and TG in Japanese people while it was associated systolic blood pressure in Korean people31. Recently, collision between traditional and westernized lifestyles has been seen in the urban areas of Mongolia. The previous studies showed Mongolian people may be having higher risk of cardiovascular events related to their lifestyles.

One of the health crucial issues to prevent CVD is the detection of atherosclerotic alterations among asymptomatic subjects. There have been a few comparative studies which including healthy young aged 18-25 years subjects. This study was first time used a new atherosclerotic parameters, CAVI and ABI in Mongolian subjects. Modern high quality techniques may help to find new important scientific detailed information which did not detected last years. The data investigated that body mass index (BMI), heart rate (HR), diastolic blood pressure (DBP), CAVI and ABI levels were significantly higher and TC and glucose levels were significantly lower in Mongolian subjects than those in Japanese subjects. Similar results, in Mongolian over age (age 30-60) subjects than that of Japanese subjects and Korean subjects was found in early survey 10. Furthermore, interestingly Mongolian healthy young subjects have significantly higher level of CAVI and ABI than that of Japanese subjects. Researchers explained the higher level of CAVI and ABI in Mongolian subjects may be related to their hematocrit 32, oxidative stress and peripheral resistance which are including hypoxia-related factors at higher altitude level 20. Mongolia is landlocked country located 1580 m above sea level which is higher than Japan (Tochigi prefecture; 50 m). The highest altitude in Mongolia is 2180 m above sea level (Gobi-Altai) and lowest is 625 m above sea level (Selenge). CVD such as blood pressure and hematocrit are increased with altitude level which was reported in previous study 32.

Although, rising level of CAVI is showing vascular elasticity changing but the Mongolian subjects were young. Therefore future study needs to clarify higher level of CAVI in Mongolian young subjects which is including normal level. Additionally, this study determined correlation patterns of CAVI and ABI with other atherosclerotic parameters between the Mongolian and Japanese subjects was different 7. In 1965, serum lower level of TC in Mongolian healthy young subjects was first time found compared to Czechoslovak people (age 19-23)10, 33. This result is confirmed other further studies including Japan and Korean population 10. Some of the data showed low density lipoprotein cholesterol (LDL-c) level is lower in Mongolian people than Asian ethnic countries however physiologic pathway is not yet explained clearly. Carotid atherosclerosis is assessed by carotid IMT with non-invasive high frequency ultrasound which is widely useful in clinical practice. Carotid IMT is surrogate marker for CVD and preventive cardiological fields. Carotid IMT in Mongolian subjects was (median [interquartile range]; 0.63 [0.49-0.86]) greater and strongly associated to age when indirectly compared other ethnic studies results 11. Furthermore, carotid IMT related parameters were different in Mongolian people compared other Asian country 34,35.

Ethnical diseased group study was attempted to compare atherosclerotic parameters between hospital-based of Mongolian and Japanese population with hypertensive and/or DM 8. There have a few comparative studies on atherosclerotic parameters between Japanese and Mongolian patients. Significantly higher BMI, HR, DBP and CAVI, significantly lower TC and glucose in Mongolian patients than that of Japanese patients was consistent with the previous study results which were indicated in healthy and young Mongolian subjects. Additionally, systolic blood pressure, CRP, carotid IMT and insulin were significantly higher in Mongolian patients than that of Japanese patients. Higher serum CRP levels were seen with increased metabolic syndrome risk factors in both Japanese people and Mongolian people9. These higher atherosclerotic parameters in Mongolian patients may be showing the patients have more vascular stiffness. CVD risk factors such as higher percentage of current smoking and higher BMI in Mongolian population (apparently healthy subjects, young subjects and patients) are detected in all of comparative studies. Smoking such as nicotine is one of the accelerator factors of atherosclerosis which have pathophysiologic affect to endothelial cell injury, inhibitor of prostacyclin and to increase LDLc level. Furthermore, interestingly Mongolian patients have similar level of insulin resistance while higher insulin and lower glucose than Japanese patients. Schindler et al reported that body weight is increasing with insulin resistance, chronic inflammation and is related to impairment of endothelial dependent coronary vasomotion in overweight individuals 36. One possible conjecture explanation is affect existence of differences in environmental factors. The lowered effect of high altitude on plasma glucose has also been reported in previous study 17.

Researchers confirmed atherosclerotic parameters such as higher CAVI and CRP in Mongolian patients not associated to blood pressure8. There have several studies in Mongolia which blood pressure is increasing with altitude level between different regions in Mongolia32. Additionally, different and unknown some genetic polymorphism may be played roles on atherosclerosis however genetic background in Japanese and Mongolian people is generally similar. These factors are thought to merit further investigation to elucidate such differences between Japanese and Mongolian people, while more unmeasured factors might remain hidden.
PERSPECTIVE STRATEGY OF ATHEROSCLEROSIS IN MONGOLIAN PEOPLE
The previous studies showed the level of CAVI is different in ethnical countries. The higher level of CAVI is not suspected Mongolian young subject’s artery is stiffer than Japanese subjects. Adequate reference range of CAVI is required to Mongolian people to get down to back-rock of artery stiffness. In the previous study was suggested the level of CAVI is may be dependent on environment factors such altitude difference. The highest altitude is 2180 m above sea level (Gobi-Altai) and lowest is 625m above sea level (Selenge) in Mongolia. The CAVI level is may be different among Mongolian residents who are living at different altitude. Additionally, atherosclerotic parameters including hemoglobin, oxidative stress and elasticity of peripheral artery and their associations are needed in future study. Peripheral vascular (radial and brachial artery) elasticity will measure transcutaneous ultrasonography by phased tracing method. Further study need specific comparative atherosclerotic study between Japanese and Mongolian people including lifestyle differences, meat intake, fruit intake, vegetable intake, salt intake and physical activity. Comparative studies such as arterial stiffness related study is required to understand pathophysiologic essence and feature of higher level of CAVI and CRP in ethnic countries.

SUMMARY
The higher level of CAVI and atherosclerotic risk factors were detected in Mongolian young healthy population and diseased population. In the previous comparative study such as young healthy subjects data showed CAVI is may be different in ethnical country. This suggests future study need to indicate normal range of CAVI. Higher level of atherosclerotic parameters in Mongolian young healthy people and patients with HT and DM showed Mongolian people have high risk to increase morbidity and mortality rate of CVD in future. Therefore Mongolian people need to clarify the essence of atherosclerotic parameters and arterial wall stiffness, and their association of environment risk factors including lifespan, altitude level and weather features in the further study. These results may be a science basic key of the why CAVI is higher in Mongolian people than Japanese people.

REFERENCES


Breast Cancer Incidence and Mortality Trend in Mongolia


1School of Public Health, HSUM 2National Cancer Center

ABSTRACT
Breast cancer morbidity and mortality rates remain relatively high despite implementation of programs such as “National Cancer Control program”, “Sub-program on cancer prevention and control”, “National program on the Prevention and Control of Non-communicable diseases”, and having greater possibilities for the cancer to be prevented, detected earlier and treated.

A retrospective descriptive study was conducted for forecasting incidence and mortality trends. Time series analysis data on morbidity and mortality from breast cancer dated 1998-2012 were collected. Incidence per 100000 female populations increased from 5.8 to 10.4 in 2002 and 2012 respectively. Mortality from breast cancer per 100000 female populations was 2.0 and this increased to 3.2 per 100000 female populations in 2012.

Time series analysis data for the last 15 years predicts gradual increase in incidence trends of breast cancer in 2013-2022. The incidence of breast cancer will be 10.27, 10.48 and 12.58 in 2013, 2014 and 2022 respectively.

Keywords: Breast cancer, incidence, mortality, trend

INTRODUCTION
Epidemiology transition has been occurring since 1990s in Mongolia.1 Breast cancer morbidity and mortality rates remain relatively high despite implementation of programs such as “National Cancer Control program”, “Sub-program on cancer prevention and control”, “National program on the Prevention and Control of Non-communicable diseases”, and having greater possibilities for the cancer to be prevented, detected earlier and treated.1 Cancer has become the second leading cause of death in Mongolia since 1990s.4 In other words there were 152 diagnosed cases of the cancer in 2012 compared to 58 cases in 1998. Some 74.2% of women diagnosed with breast cancer for the last 12 years were in the third and forth stages of the disease.5 According to the 2012 health statistics 75% of diagnosed cases in 2012 were in the late stages of the disease. In our country breast cancer is the sixth most common cancer after cancers of liver, stomach, esophagus, cervix uteri and ovaries.6 The incidence rate of breast cancer was 10.4 cases per 100000 female population and it is 2.62 fold increase of breast cancer in 2012 compared to 1998.9 Therefore, the aim of the study is to study incidence and mortality trends for breast cancer in women.

MATERIALS AND METHODS
This is a retrospective descriptive study. Time series data on morbidity and mortality from breast cancer dated 1998-2012 were collected for forecasting incidence and mortality trends by:
- Time, duration
- 15 years trend

Statistical analysis was performed using STATA 11, incident, mortality and prevalence data on breast cancer were analyzed applying ARIMA modeling.

RESULTS
In 2002, 73 new cases of breast cancer were registered among women whereas in 2012 there was 2.08% increase in new diagnosed cases reaching 152. Incidence per 100000 females increased from 5.8 to 10.4 in 2002 and 2012 respectively.
Table 1. Incidence and mortality from breast cancer, 2002-2012 /per 100000 population/

<table>
<thead>
<tr>
<th>Year</th>
<th>All</th>
<th>Females</th>
<th>All</th>
<th>Females</th>
<th>per 100.000 pop</th>
<th>per 100.000 females</th>
<th>per 100.000 pop</th>
<th>per 100.000 females</th>
</tr>
</thead>
<tbody>
<tr>
<td>2002</td>
<td>74</td>
<td>73</td>
<td>26</td>
<td>26</td>
<td>3</td>
<td>5.8</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>2003</td>
<td>71</td>
<td>71</td>
<td>24</td>
<td>24</td>
<td>2.8</td>
<td>5.6</td>
<td>0.9</td>
<td>1.8</td>
</tr>
<tr>
<td>2004</td>
<td>81</td>
<td>78</td>
<td>34</td>
<td>34</td>
<td>3.2</td>
<td>6</td>
<td>1.3</td>
<td>2.6</td>
</tr>
<tr>
<td>2005</td>
<td>95</td>
<td>95</td>
<td>24</td>
<td>24</td>
<td>3.7</td>
<td>7.3</td>
<td>0.9</td>
<td>1.8</td>
</tr>
<tr>
<td>2006</td>
<td>89</td>
<td>88</td>
<td>29</td>
<td>29</td>
<td>3.4</td>
<td>6.6</td>
<td>1.1</td>
<td>2.1</td>
</tr>
<tr>
<td>2007</td>
<td>76</td>
<td>76</td>
<td>36</td>
<td>35</td>
<td>2.9</td>
<td>5.6</td>
<td>1.3</td>
<td>2.6</td>
</tr>
<tr>
<td>2008</td>
<td>97</td>
<td>97</td>
<td>35</td>
<td>35</td>
<td>3.6</td>
<td>7</td>
<td>1.3</td>
<td>2.5</td>
</tr>
<tr>
<td>2009</td>
<td>82</td>
<td>80</td>
<td>37</td>
<td>36</td>
<td>3</td>
<td>6</td>
<td>1</td>
<td>2.6</td>
</tr>
<tr>
<td>2010</td>
<td>126</td>
<td>123</td>
<td>48</td>
<td>47</td>
<td>5</td>
<td>8.7</td>
<td>2</td>
<td>3.3</td>
</tr>
<tr>
<td>2011</td>
<td>140</td>
<td>139</td>
<td>44</td>
<td>43</td>
<td>5</td>
<td>9.7</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>2012</td>
<td>152</td>
<td>152</td>
<td>49</td>
<td>47</td>
<td>5</td>
<td>10.4</td>
<td>2</td>
<td>3.2</td>
</tr>
</tbody>
</table>

Moreover, mortality from breast cancer increased from 26 up to 47 in 2002 and 2012 respectively. Mortality from breast cancer per 100000 females was 2.0 and this increased to 3.2 per 100000 females in 2012.

Figure 1. Breast cancer Incidence trend /per 100000 population/

It was estimated that there will be 2.4 times increase in trend of the total incidence of breast cancer in 2022 compared to 1998 reaching 12.58 (CI 95% 8.66, 16.49) per 100000 population (Figure 1.)

Table 2. Breast cancer incidence trend in 2013-2022 /per 100000 population/

<table>
<thead>
<tr>
<th>Year</th>
<th>Breast cancer</th>
<th>Lower</th>
<th>Upper</th>
</tr>
</thead>
<tbody>
<tr>
<td>2013</td>
<td>10.27</td>
<td>8.20</td>
<td>12.35</td>
</tr>
<tr>
<td>2014</td>
<td>10.48</td>
<td>7.87</td>
<td>13.08</td>
</tr>
<tr>
<td>2015</td>
<td>9.48</td>
<td>6.20</td>
<td>12.77</td>
</tr>
<tr>
<td>2016</td>
<td>10.68</td>
<td>7.03</td>
<td>14.32</td>
</tr>
<tr>
<td>2017</td>
<td>10.19</td>
<td>6.46</td>
<td>13.93</td>
</tr>
<tr>
<td>2018</td>
<td>11.31</td>
<td>7.57</td>
<td>15.05</td>
</tr>
<tr>
<td>2019</td>
<td>10.85</td>
<td>7.03</td>
<td>14.68</td>
</tr>
<tr>
<td>2020</td>
<td>11.95</td>
<td>8.11</td>
<td>15.78</td>
</tr>
<tr>
<td>2021</td>
<td>11.51</td>
<td>7.60</td>
<td>15.43</td>
</tr>
<tr>
<td>2022</td>
<td>12.58</td>
<td>8.66</td>
<td>16.50</td>
</tr>
</tbody>
</table>
Figure 2. Breast cancer mortality trend /per 100000 population/

Mortality trends of breast cancer for 2013-2022 were estimated applying time series analysis using data from 1998-2012 and it stays at stable level each year. It was estimates as 3.44, 3.17 and 3.21 per 100000 population in 2013, 2014 and 2022 respectively (Table 3).

DISCUSSION

Our study results showed 2.4 times increase in trend of the total incidence of breast cancer in 2022 compared to 1998. Amarsanaa et al. studied risk factors of breast cancer and noted that morbidity from breast cancer increases by 1-2% each year in many countries. Moreover, in some international studies there has been an increase of incidence of breast cancer since 1998 and incidence goes up each year among women in Japan, China and Philippines.

CONCLUSION

Incidence trends of breast cancer steadily increase over years whereas mortality trends stay at stable level.

REFERENCES

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Changes of Hemeostasis in Nicotine-Dependent Subjects

Ulziisaikhan S et al

ABSTRACT

This case control study aimed to clear possible pathogenetic mechanisms of homocysteine influence on hemeostasis in nicotine-dependent subjects using comparative analysis of coagulation and vascular-platelet system features. Twenty 20 male volunteers (mean age 20.0±1.0 year) were randomly selected into 2 groups (n=10 in each group). Case groups included tobacco smokers (mean index of smoke 4.9 pack/year, mean history of smoking 2.2±0.5 year) and non smokers were included in the control group.

Peripheral blood samples were collected from all a participants. Concentration of homocysteine was determined using high performance liquid chromatography. Level of anti-thrombin antibodies were determined using ELISA. Aggregation activity of platelets was evaluated in agrometer (Biola, Russia) using adenosine diphosphate (ADP) (5 mg) and ristomycin monosulfate as an inductors.

Evaluation of changes in hemeostasis was performed using standard methods and determined the following features: prothrombin time (PTT, sec.), prothrombin time-international normalized ratio (PTT-INR), thrombin time (TT, sec.), activated partial thromboplastine time (APTT, sec.) and level of fibrinogen (g/L). Absolute count of platelets was measured by an automatic hematology analyzer (ABX PENTRA 120, Block Scientific, Inc.). Titer of total homocysteine did not exceed was reference range, but a found significant difference between case and control groups. Mean titer of total homocysteine in control group was significantly low comparing to case group (p=0.0031).

Features of blood clotting did not exceed a reference range, but was found shortened tromboplastine time (p=0.0003) and activated partial thromboplastine time (p=0.001) in case group.

In the control group titer of anti-thrombin antibodies was siginificantly lower than in case group (p=0.022). There were elevated homocysteine levels and changes in system of hemeostasis such as dysfunction of platelet aggregation, changes in blood clotting features, indicating indirectly hyperthrombinemia in subjects with relatively short tobacco smoking history. Elevated titer of anti-thrombin antibodies in nicotine-dependent subjects indicates the mobilization of mechanisms directed to regulate hemeostasis.

Key words: Homocysteine, hemeostasis, anti-thrombin antibody.

INTRODUCTION

Thrombin is a multifunctional enzyme and a major acting component in forming fibrin clots. In addition thrombin activates blood platelets through stimulation of their adhesion, aggregation and releasing of biologically active substances, acts as a chemoattractant for neutrophils, macrophages and monocytes, induces mitosis of various cells and supports the retraction of neurons. A new principal function of thrombin discovered in the last decade is triggering neurotropic effects in central and peripheral nervous system by changing neuronal cell condition in order to activate surface receptors to thrombin.

The role of thrombin in reactions of inflammation and tissue reparation, which always leads to blood clotting and generation and concentration of thrombin in site of vessel alteration and neighboring tissue is much more crucial, but less investigated. Thrombin was defined as...
an anti-inflammatory factor and its modulating effect on cells still has not been considered.5, 6 According to these findings the detailed study of the role of thrombin in reactions directed to limitation of inflammatory processes, blockade of cell adhesion to altered sites of blood vessels and inhibition of releasing of inflammatory mediators by mast cells. Thrombin generated in site of blood vessel alteration activates various cells attending to the process of inflammation and repairation, including monocytes, T lymphocytes of peripheral blood and mast cells, localized along vessels.7, 8

However, normally thrombin is absent in the bloodstream. Its generation requires the appearance of tissue factor, which is expressed by altered endothelial cells, macrophages, fibroblasts and smooth muscle cells, or released due to the destruction of monocytes. Elevated level of homocysteine has shown as one factor, stimulating release of tissue.9, 10 The role of tobacco smoke and nicotine in dysfunction of endothelial cells was demonstrated; nicotine in particular can cause the decreased concentration of vitamin B<sub>6</sub> required for transformation of homocysteine to cystathionine which leads to an elevated level of homocysteine.11 We aimed to determine possible pathogenetic mechanisms of homocysteine influence on hemostasis in nicotine-dependent subjects using comparative analysis of coagulation and vascular-platelet system features. Features of hemostasis in tobacco smokers significantly increased in spontaneous and as well as induced aggregation of platelets.

MATERIALS AND METHODS

Case-control study design was used. Twenty male volunteers (mean age 20.0±1.0 year) were randomly selected into 2 groups (n=10 in each group). The case group included tobacco smokers (mean index of smoking 4.9 pack/year, mean history of smoking 2.2±0.5 year) and non smokers were included to control group.

Peripheral blood samples were collected from all participants. Concentration of homocysteine was determined using high performance liquid chromatography with ultraviolet detection at 330 nm of wave length and separation in Chromolith 100X 4.6 mm column. 0.05 M citric acid (10:90, v/v) was used as eluent acetonitrile.12

The level of anti-thrombin antibodies was determined using following method: Beforehand wells of 96 well microtiter plate were treated with human thrombin (Sigma) (200 mg per well). Plate was incubated for 24 hour in +4°C, then washed and solution of glycine was added to each well and incubated for another 24 hour at +4°C. Wells were washed and 100 mL serum sample diluted in 1:100 was added. Conjugates with secondary antibodies to human IgG labeled with horseradish peroxidase (Vector best Co. Ltd., Russia) were added to each well. The following procedures were made according to standard procedures of ELISA and optic density (OD) was measured in regimen of wave length 450 nm.

Aggregation activity of platelets was evaluated in agrometer (Biola, Russia) using adenosine diphosphate (ADP) (5 mg) and ristomycin monosulfate as an inductors. The following parameters were considered in interpretation of received data.

1. In the curve of mean size of aggregates (MSA): degree of aggregation – maximum prevalence of mean size of aggregates (unit); aggregation speed: maximum inclination of curve of mean size of aggregates (unit/minute).

Evaluation of changes in hemostasis was performed using standard methods and determined the following features: prothrombin time (PTT, sec.), prothrombin time-international normalized ratio (PTT-INR), thrombin time (TT, sec.), activated partial thromboplastine time (APTT, sec.) and level of fibrinogen (g/L).

Absolute count of platelets was measured by an automatic hematology analyzer (ABX PENTRA 120, Block Scientific, Inc.).

Statistical analysis of study data was provided using software StatistikaÔ 6.1 (StatSoft). Descriptive statistics were presented by medians and interquartile intervals (25th and 75th percentiles); comparison of independent variables was performed using Mann-Whitney’s U test, significance of statistical hypothesis were considered in p<0.05.

RESULTS

Titer of total homocysteine did not exceed reference range, but significant difference between case and control group was found. Mean titer of total homocysteine in control group was 7.83 (6.08-10.55) mmol/L and in case group 212.0 (179.0-215.0) ´10<sup>9</sup>.

Low concentration of thrombin in blood leads to activation of all platelets, but it doesn’t cause their apoptosis. If the amount of thrombin is able to trigger clotting in circulation, up to 30-40% of platelets will exposed to apoptosis.1 Significant difference between case and control groups in count of platelets was not found. Count of platelets were determined in control group 238.0 (198.0-249.0) ´10<sup>9</sup> and in case group 212.0 (179.0-215.0) ´10<sup>9</sup>. In tobacco smokers a dysfunction of aggregation activity of platelets was found (Table 1).
Table 1

Changes in vascular-thrombocyte hemostasis in tobacco smoking and non-smoking young men

<table>
<thead>
<tr>
<th>Features</th>
<th>Non-smokers (n=10)</th>
<th>Smokers (n=10)</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Spontaneous aggregation</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Degree of aggregation (MSA; unit)</td>
<td>1.05 (0.99-1.05)</td>
<td>1.48 (1.42-1.76)</td>
<td>p=0.004</td>
</tr>
<tr>
<td>Aggregation speed (MSA; U/min)</td>
<td>1.12 (1.10-1.14)</td>
<td>2.26 (2.08-2.49)</td>
<td>p=0.002</td>
</tr>
<tr>
<td>Degree of aggregation (LR; %)</td>
<td>0.25 (0.18-0.31)</td>
<td>2.11 (1.69-2.23)</td>
<td>p=0.002</td>
</tr>
<tr>
<td>Aggregation speed (LR; %/min)</td>
<td>0.93 (0.56-1.27)</td>
<td>4.32 (3.79-5.57)</td>
<td>p=0.002</td>
</tr>
<tr>
<td>ADP induced aggregation</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Degree of aggregation (MSA; unit)</td>
<td>4.02 (3.70-4.51)</td>
<td>8.38 (8.33-10.12)</td>
<td>p=0.003</td>
</tr>
<tr>
<td>Aggregation speed (MSA; U/min)</td>
<td>6.24 (5.64-8.84)</td>
<td>67.41 (57.68-76.18)</td>
<td>p=0.002</td>
</tr>
<tr>
<td>Degree of aggregation (LR; %)</td>
<td>6.92 (4.44-13.70)</td>
<td>24.65 (18.61-29.88)</td>
<td>p=0.004</td>
</tr>
<tr>
<td>Aggregation speed (LR; %/min)</td>
<td>7.70 (6.08-10.30)</td>
<td>83.91 (54.68-94.61)</td>
<td>p=0.002</td>
</tr>
<tr>
<td>Ristomycin sulfate induced aggregation</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Degree of aggregation (MSA; unit)</td>
<td>4.50 (3.670-5.12)</td>
<td>9.15 (8.15-10.15)</td>
<td>p=0.002</td>
</tr>
<tr>
<td>Aggregation speed (MSA; U/min)</td>
<td>28.5 (23.30-35.60)</td>
<td>127.5 (74.88-253.25)</td>
<td>p=0.002</td>
</tr>
<tr>
<td>Degree of aggregation (LR; %)</td>
<td>5.00 (4.76-5.65)</td>
<td>11.15 (9.33-13.53)</td>
<td>p=0.002</td>
</tr>
<tr>
<td>Aggregation speed (LR; %/min)</td>
<td>27.70 (25.5-30.6)</td>
<td>134.5 (103.71-259.5)</td>
<td>p=0.002</td>
</tr>
</tbody>
</table>

As seen in Table 1 the features of hemostasis in tobacco smokers significantly increased in spontaneous and as well as in induced aggregation of platelets.

Table 2

Changes in blood clotting in tobacco smoking and non-smoking young men

<table>
<thead>
<tr>
<th>Features</th>
<th>Non-smokers (n=10)</th>
<th>Smokers (n=10)</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Concentration of fibrinogen (g/L)</td>
<td>1.06 (1.01-1.08)</td>
<td>1.05 (1.01-1.07)</td>
<td></td>
</tr>
<tr>
<td>PTT</td>
<td>14.0 (13.0-14.1)</td>
<td>16.6 (15.7-17.9)</td>
<td>p=0.000293</td>
</tr>
<tr>
<td>TT</td>
<td>29.35 (28.9-29.75)</td>
<td>32.15 (30.55-34.3)</td>
<td>p=0.0013</td>
</tr>
<tr>
<td>APTT</td>
<td>248 (212-274)</td>
<td>224 (213-254)</td>
<td></td>
</tr>
</tbody>
</table>

Prolonged thrombin time and APTPT in smokers in comparison to non-smokers indicates the disorder of polymerization of fibrin monomers in these subjects.

Previously predominant secretion of nitric oxide, prostacyclin was established, and tissue activator of plasminogen and activation of protein C was observed by the appearance of low concentration of thrombin in circulation. The thrombin acts on the cells through protease activated receptors (PARs). Different mechanisms are observed when endothelial cells are altered. Low amounts of thrombin, secreted due to blood vessel alteration leads to local forming of clots. Tobacco smoking may alter blood vessels in various ways: direct alteration of endothelial cells; change of blood neutrophil structure and function; transformation of monocytes into secretory macrophages and secretion of cytokines; fixation of tobacco glycoprotein (hapten) on surface of endothelial cells with following production of autoantibodies and antibody-dependent cellular cytotoxicity of endothelial cells, release of tissue damaging mediators; and elevated concentration of homocysteine. Hypercoagulation was compensated for via increased production of autoantibodies to thrombin. We have demonstrated the confirmation for this issue in current study. In control group titer of anti-thrombin antibodies was 0.61 (0.03-0.1) ng/mL and in case group 0.276 (0.165-0.310) ng/mL (p=0.022).

In conclusion, we have found elevated homocysteine levels and changes in the system of hemostasis such as dysfunction of platelet aggregation, changes in blood clotting features, indicating indirectly hyperthrombinemia in subjects with...
relatively short tobacco smoking history. Elevated titer of anti-thrombin antibodies in nicotine-dependent subjects indicates the mobilization of mechanisms directed to regulate hemostasis.

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Clauses of Cesarean Section in Multiparae

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¹ Maternity hospital of Mongolia
² Health Sciences University of Mongolia

ABSTRACT

Our objective was to determine clauses of Cesarean section in multiparae. Analysis materials are the laboratory samples and birth histories of mothers having cesarean sections. Research Methodology Sampling Frame: The study will involve mothers (approximately 70 mothers) who are hospitalized in Maternal Departments 1 and 2 of the Clinical Maternity Hospital № 1 and who will be delivered of their children by Cesarean sections. In order to determine frequency and cause of Cesarean section, frequency of organ system and obstetrics disease among the women who delivered many times and were performed cesarean section we did retrospective study and considered the result in the delivery of Cesarean section of 5 years. If we compare 5797 cases who were delivered by Cesarean section with number of delivery; in 2001-21%, in 2002-21%, in 2003-26.2%, in 2004-27%, in 2004-27%, in 2005-28.1%

When we determined many years tendency of percentage of women delivered many times by the method of quadratic and did statistic probability by 2010 it has tendency to become 12.8%. It was 3.6% in 2001 but in 2002 it increased steeply and became 7.6%, since that it is in steady level, it proves that it doesn’t have tendency of decreasing. This is the result. mean age of mothers who were performed Cesarean section for the first time is 26.6±0.09, for the second time 31.7±0.1 and mothers delivered many times 36.9±0.19. It shows that number of delivery and mothers age has direct dependence. When number of delivery increases number of age increases. (p<0.01)

Key words: Multiparae, complication, cesarean section, mortality rate

INTRODUCTION

According to a 2005 social survey, the Mongolia’s average population growth reached 1.5 percent and 50.4 percent of the population are females including those of reproductive age making up 28.3 percent. Health and sociological surveys conducted in recent years have shown that the number of multiparae and multigravidae is unlikely to fall in the coming years, probably in the next 10 years. CS increases 3 times in the late 10 years in the countries of the world.¹,²,³,⁴

It was proved that in nowadays branch of obstetrics CS occupies specific place, frequency of CS delivery increases, direction of performing this surgery often changes as the case of performing by comparative indication increases. The surgery affects to prenatal disease of mother and mortality positively and negatively.⁵,⁶ Because of hormone change on woman’s body and increase of pressure to vessel of lower limbs. In the process of vein vessel expands therefore speed of systemic circulation decreases and vessel wall hurts. Due to vein vasodilatation complication of blood coagulation and bleeding occur after delivery in 30-50% blockade is formed.⁵,⁶,⁷

MATERIAL AND METHODS

Analysis materials are the laboratory samples and birth histories of mothers having cesarean sections. Research Methodology Sampling Frame: The study will involve mothers (approximately 70 mothers) who are hospitalized in Maternal Departments 1 and 2 of the Clinical Maternity Hospital № 1 and who will be delivered of their children by Cesarean sections.

Sampling Frame: Our study involved 40 mothers who have given birth to 4 or more fetuses and are required to have Cesarean section comprising the main group and 30 mothers who have been pregnant for the first time and required to have Cesarean sections comprising the control group. We conducted our survey using questionnaires with 81 questions which are later keyed into SPSS-16.0 for the statistical process of emphasizing and grouping the important indicators. Survey results are keyed into the software, whereas images are processed by the software. The questions and answers in the questionnaires are coded in order to key in the computer.
RESULTS

Seeing from the table (Table 1) mean age of deliverer for the first time is 28±0.94 deliverer of many times is 37±0.59% (p<0.01).

Table 1

<table>
<thead>
<tr>
<th>№</th>
<th>Indices</th>
<th>M±m</th>
<th>Str.D</th>
<th>Max</th>
<th>Min</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Prime gravidea</td>
<td>28±0.94</td>
<td>5.2</td>
<td>19</td>
<td>38</td>
</tr>
<tr>
<td>2.</td>
<td>Multiparae</td>
<td>37.1±0.59</td>
<td>3.7</td>
<td>28</td>
<td>44</td>
</tr>
</tbody>
</table>

M- mean age, m – standard correlation, Str.D- standard curve

Table 2

<table>
<thead>
<tr>
<th>Number of delivery</th>
<th>N</th>
<th>M±m</th>
<th>Median</th>
<th>Mode</th>
<th>Std.D</th>
<th>Min</th>
<th>Max</th>
</tr>
</thead>
<tbody>
<tr>
<td>For the first time deliverer</td>
<td>30</td>
<td>476.7±22.6</td>
<td>425</td>
<td>400</td>
<td>123.7</td>
<td>400</td>
<td>850</td>
</tr>
<tr>
<td>Many times deliverer</td>
<td>40</td>
<td>836.3±65.7</td>
<td>750</td>
<td>600</td>
<td>416</td>
<td>400</td>
<td>2500</td>
</tr>
</tbody>
</table>

Seeing from Table 2 average amount of bleeding (during operation) of women with many times delivery is 836±416ml, for the first time deliverer 476±123 ml. When we analyzed this by student criteria it is different in t=5.22 (P<0.001) level. Average amount of bleeding during CS is 652±50 ml. When we measured bleeding amount during operation by gravimeter method for the first time deliverer it was up to 500 ml 81.8% for women with many times delivery up to 501-1000ml (90.3%) more. In comparison with deliverer for the first time for women with many times delivery bleeding during operation occurs 2.1 times more. Among the directions of CS performed on the mothers involved in control group and basic group 80% of them have obstetrics and organ system disease, 75% combined form of late gestation of pregnancy and deliverers for the first time have breech presentation -83.3%, no result for delivery impetus treatment 66.7%. These indices have high percentage (Table3).

Table 3

<table>
<thead>
<tr>
<th>Direction</th>
<th>Prime gravidea</th>
<th>Multiparae</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Combined form of obstetrics and organ system disease</td>
<td>3</td>
<td>12</td>
<td>15</td>
</tr>
<tr>
<td>Combined form of late toxication</td>
<td>%</td>
<td>10±5.4</td>
<td>30±7.2</td>
</tr>
<tr>
<td>Lat deliverer for the first time was not prepared for delivery</td>
<td>%</td>
<td>23.3±7.7</td>
<td>55±7.9</td>
</tr>
<tr>
<td>Late deliverer</td>
<td>%</td>
<td>23.3±7.7</td>
<td>0</td>
</tr>
<tr>
<td>No result for delivery impetus treatment</td>
<td>%</td>
<td>6.6±4.5</td>
<td>2.5±2.5</td>
</tr>
<tr>
<td>Scar fragile of caesarean section</td>
<td>%</td>
<td>0</td>
<td>5±3.4</td>
</tr>
<tr>
<td>Breech delivery</td>
<td>%</td>
<td>16.6±6.8</td>
<td>2.5±2.5</td>
</tr>
<tr>
<td>Out flow of liquid near fetus which is not prepared for delivery</td>
<td>%</td>
<td>3.3±3.3</td>
<td>0</td>
</tr>
<tr>
<td>Big fetus with abbreviated deposition of obstetrics</td>
<td>%</td>
<td>3.3±3.3</td>
<td>0</td>
</tr>
<tr>
<td>Was not prepared for delivery tract, with obstetrics abbreviated deposition</td>
<td>%</td>
<td>0</td>
<td>2.5±2.5</td>
</tr>
<tr>
<td>Abbreviated deposition of obstetrics</td>
<td>%</td>
<td>0</td>
<td>2.5±2.5</td>
</tr>
<tr>
<td>Serious form of late toxication of pregnancy</td>
<td>%</td>
<td>6.6±4.5</td>
<td>0</td>
</tr>
<tr>
<td>Organ system disease of stage and which serious process</td>
<td>%</td>
<td>3.3±3.3</td>
<td>0</td>
</tr>
<tr>
<td>With more period of pregnancy and obstetrics deposition</td>
<td>%</td>
<td>3.3±3.3</td>
<td>0</td>
</tr>
<tr>
<td>Total</td>
<td>%</td>
<td>100</td>
<td>100</td>
</tr>
</tbody>
</table>
For women with many deliverers (Table 4) bleeding occurs 72.2, for the first time deliverer (27.39) 2.5 times more. It is seen that for women with many times delivery cardiovascular, liver, vascular and hypertension disease occur more. (P<0.001) Organ system disease occurs in 57.1 for women with many delivery, in 42.9% for the first time deliverer.

Table 4

<table>
<thead>
<tr>
<th>Frequency of organ structural disorder</th>
</tr>
</thead>
<tbody>
<tr>
<td>Organ system disease</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>Bleeding</td>
</tr>
<tr>
<td>%</td>
</tr>
<tr>
<td>Renal disorders</td>
</tr>
<tr>
<td>%</td>
</tr>
<tr>
<td>Cardiovascular disorders</td>
</tr>
<tr>
<td>%</td>
</tr>
<tr>
<td>Vascular disorders</td>
</tr>
<tr>
<td>%</td>
</tr>
<tr>
<td>Hypertension</td>
</tr>
<tr>
<td>%</td>
</tr>
<tr>
<td>Hepatic disorders</td>
</tr>
<tr>
<td>%</td>
</tr>
<tr>
<td>Ophthalmologic disorders</td>
</tr>
<tr>
<td>%</td>
</tr>
<tr>
<td>Others</td>
</tr>
<tr>
<td>%</td>
</tr>
<tr>
<td>Total</td>
</tr>
</tbody>
</table>

Seeing from Table 5 complications after CS occurs more on women with many times delivery. From this bleeding – 82.6%, endometritis 75.0%, wound opening and septic occur up to - 100% it shows that complex measure of preventing from complication before, during and after surgery is not sufficient (P<0.001).

Table 5

<table>
<thead>
<tr>
<th>Complication after Caesarean section</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of delivery</td>
</tr>
<tr>
<td>For the first time deliverer</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>Many times deliverer</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>Total</td>
</tr>
<tr>
<td></td>
</tr>
</tbody>
</table>

* P<0.001

**DISCUSSION**

Seeing from result of research material frequency of CS increases year by year and further it has tendency of increasing suits to the result of research work by Bayasgalan. G (2007), Jav.B (1998). CS increases 3 times in the late 10 years in the countries of the world.1,8 It was proved that in nowadays branch of obstetrics CS occupies specific place, frequency of CS delivery increases, direction of performing this surgery often changes as the case of performing by comparative indication increases. The surgery affects to prenatal disease of mother and mortality positively and negatively.4 For women with many delivery we chose mothers with 4 and more delivery not considering age and did the study. It suited to index by H.M.Khamidov (1986) and A.A.Oradmuradov (1994).7,9 In B.Oraevs study it was determined that for most percentage of women with many delivery pregnancy and organ system disease are conducted at the same time. It was revealed that these affect to process of pregnancy and delivery. For factors of pregnancy and delivery organ system disease play main role but other factors affect too.7,10 R.J.Stepanyan (1989) considers that period between pregnancy of women with many delivery is one of
the affecting factors. In many books and works it was mentioned that majority percentage of various kinds of organ system disease occupy anemia of iron deficiency during being pregnant. Frequency of this disease case occupies 1.8-72% of all the delivery. By scholars study of Dagestan university ‘among diseases of organ system anemia occurs more” suits to the result of our study. But on the second and the 3rd place of disease cardiovascular and renal disease are included, this doesn’t suit to the result of our study.

By our study on the second renal disease (17.1%) on the 3rd cardiovascular disease (8.7%) occur more.

By G.Lhagvasuren and B.Jav’s study among the indications of CS renal disease (65.1%), cardiovascular disease (17.1%). The result of our study suits to this result. Anemia of iron deficiency shows bad consequence in the process of delivery in particular premature delivery is 3 times more, diastole of delivery force 4 times, chronic bleeding 2.4 times more occur. Anemia affects to fetus and infant negatively. It was noted definitely. Anemia of iron deficiency one of the dangerous serious complications and it shows a lot of bad consequence. The main index of diagnose is reduction of erythrocyte number 3.5*10^9/l, hemoglobin up to 100 gr/l.7,10,15

This suits to criterian index which is shown women has anemia of iron deficiency. This conclusion suits to the result of our study. By SH.D. Muratov’s conclusion above mentioned anemia is formed in connection with organ system disease before being pregnant and pregnancy. By M.A. Omarov A.SH.Khasaev’s (1989) study anemia of iron deficiency occurs plentifully on women with many delivery or repeated delivery it suits to our study result that.7,10

Being pregnant many times delivery, baby suckling decrease accumulation of iron in womens body, slow down blood structure forming and blood sanguification. This suits to the result of our study B.Oraev’s (1991) study on women with late gestation of pregnancy and anemia occurs 66.6% and for healthy women 4-12% pregnancy complicates due to hypotension increase of amniotic fluid, immature delivery, placental a rupture, malposition of fetus. Also anemia affects to fetus and infant condition negatively.7,10,15

It was noted definitely on all the women involved in the study gestation of later period (55.5%), light (41.4%), serious (0,5%), eclampsia (1.3.) and in the form of combined disease occur.7,13 It suits to the result of study by Bayasgalan.G (2000) organ system diseases affect much to form risk of gestation of later period of pregnancy therefore renal disease and on the base of it hypertension and vascular disease are involved in the practice.11 Our study suits to this proof. From the material by B.Jav when age of deliverer for the first time is elder it increases frequency of forming complication and become the indication which completes delivery with CS. On women with many delivery, vasodilatation disease occurs not few. If on the base of vasodilatation disease pregnancy gestation combined it increases coagulation process, hurts and changes wall structure of vessel. Gestation deepens vasodilatation of pregnant women more. Vasodilatation of vein mostly occurs on lower limbs, process of disease complicates at the second part of pregnancy.

Because of hormone change on woman’s body and increase of pressure to vessel of lower limbs. In the process of vein vessel expands therefore speed of systemic circulation decreases and vessel wall hurts.

Due to vein vasodilatation complication of blood coagulation and bleeding occur after delivery in 30-50% blockade is formed.5,7,11

For forming blood coagulation system loss affects much. Study 1997 of scholars M.A.Repin (1988) of Dagestan university proves this.5,7 Material of study by Jav still becomes indication of complicating the delivery by CS because when the age of deliverer for the first time is elder it increases frequency of forming complication of obstetric disease in combination of organ system. For mothers with many delivery premature delivery in particular obstetric disease in combination with and organ system delivery was observed4.

Our study result suits to the study result by SH.SH. Rudjabova, M.A.Omarov (1997). If we see mothers involved in study age of woman with many delivery is more (36.9±0.19) than the woman for the first time delivery (26.6±0.09).By M.KH. Khamidov (1994) mean age of women with many delivery fluctuates between (34.8±37.9) and also study of scholars of Dagestan university proves this (1997)7.

Number of age and delivery has direct dependence. It was proved by our study.(p<0.01)5,7. Among the complications of CS. metritis and peritonitis, bleeding of uterus diastole, wound opening , septic , wound suppuration occur more. It shows that complex measure of preventing from complication before after and during surgery is not sufficient. Our conclusion suits to the study result conclusion by researchers Radjabova.SH.SH. Omarov M.A (1997), J.Lhagvasuren, B.Jav (2004)5,7. When organ system disease is combined with gestation of later period of pregnancy, it constitutes condition of complication process of pregnancy becomes cause and factor of forming disease. It suits to M.M.SHekhtman’s(2000). In forming later period
gestation of pregnancy organ system disease affects much therefore renal disease on the base of it hypertension and vascular disease depend on it. It was proved in practice. Obstetric disease organ system, age and delivery number of women become factor of forming later period gestation of pregnancy. Later period gestation of pregnancy becomes factor of increasing frequency of CS and when clinical process complicates frequency of performing surgery. It suits to the study result of researcher J.Lhagvasuren. Amount of blood of women with many delivery during CS is more. It is connected with organ system and obstetric combined disease of serious form. It has probability (p<0.01). This has risk of increasing frequency of surgery following risk of bleeding during and after delivery and reveals by blood coagulation disease. It was proved by our study and suits to study of researchers; V.N Serov (1987), A.D.Makatsariya (2000), V.I.Kulakov (2001), Ch.Shagdar (1977), B.Shijirbaatar (1996)6,7. If we see bleeding during surgery – 81.9% of all the women up to 500ml (17.1%) , 500-1000ml (0.6), 1000-1500ml (0.4%), over 1500ml.

By our study it was proved that for women with many delivery bleeding during surgery is more. Our study result suits to the result of study of researchers; M.erland (1986), V.J.Krainopoliski, V.E.Razdinskii (1993), Sh.Sh. Radjabova (1997) that the most repeatedly occurring case after and before surgery bleeding. Bleeding occurs 3-5 times more occurs than the mothers delivery of delivery tract. It suits to the conclusion that bleeding during surgery is approximately 650±50ml. It also suits to study conclusion of researcher J.Lhagvasuren (2006). Giving birth many times constitutes unpleasant condition of period of being pregnant increases risky factors frequency of forming disease, fetus asphyxia, nutrition insufficiency, abnormal development increases and it reaches prenatal mortality. Hemoglobin amount has direct dependence with bleeding amount during and after surgery. It was proved by statistic of dependence (p<0.01). Some researchers of Mongolia studied and determined blood coagulation system disease, they concluded that at the end of pregnancy all the indices of coagulation reaches the highest point and by adaptation mechanism over coagulating condition was formed. By our study about adaptation we tried to clear up difference of women for the first time delivery and many times delivery by test change of blood coagulation and on the base of structure. We also cleared up hemostase and structure change when inadaptation is formed, pregnant organism is an important mechanism which adapts itself. If adaptation mechanism is too much compensation is lost and coagulation disease is formed. We proved this. From the tests which defines blood coagulation system number of platelet, period of activated recalcification, period of activated thromboplastin, amount of serum fibrinogen is very important, by S.A.Markin (1997), V.N.Serov (1987) the considered that period of blood coagulation is a test of having less clinical information. It doesn’t suit to our study.

In fact these tests it expresses coagulating factor of inner mechanism of coagulating deficiency of prekallekrein and kallekrein or dominance. Seeing from the study for women for the first time deliverer they doesn’t have particular change but for deliverer of many times it is shortened during all the stages of surgery. It proves risk of forming blockade with thrombus. During pregnancy with normal process amount of procoagulant substances of fibrinogen protein increases amount of fibrinogen reaches the highest amount that’s why protein amount is high during normal pregnancy. In other words change of blood coagulating system has direct dependence (p<0.01).

For deliverers for the first time amount of fibrinogen has tendency of increasing as usual for deliverer with many times it increases during and after surgery, for these women pregnancy occurs in combination with obstetrics and organ system a lot.

That’s why our study shows that disease. During pregnancy VII, V, II, X factors loss of foreign tract factors which involves in coagulating but in connection with inner mechanism amount of foreign tract factor increases in blood a little and process of overcoagulating may be conducted. During normal pregnancy fibrinogen protein as procoagulant or priority substances amount increases fibrinogen protein amount increases and reaches the highest amount. The result of our study suits to study material by G.M.Savelieva, G.D.Djiveleuova (1986). Therefore tests of women with many delivery are shortened expresses overcoagulating, defines risk of forming blockade with thrombus. It suits to the study results of scholars tests expresses overcoagulating in particular activation of hemostase inner tract, result of our study proved risk of forming blockade with thrombus. It suits to the study of scholars A.Ts.Makatsaria (1997), V.N.Kulakova (2001), Sh.Sh.Radjabova (1997) and researchers of Dagestan university7,3,15.

As the researchers determined the first stage of coagulating starts from vessel – platelet blood stopping, normal condition is lost pregnancy process is complicated in combination with obstetric and organ system disease prevalent trauma of vessel inside was hurt, activation of platelet activates, hemostase of plasma activates. This proof is important result of our study. By the study …and period of coagulating has direct dependence and informative quality which expresses loss of coagulating system.
Many scholars determined this and it suits to the result of our research work. Besides diagnosing and treating, in order to prevent from it, it is necessary to do test of coagulation system in order to reveal coagulating disease related with pregnancy for pregnant woman.

The result of our study proves that it is required to prevent from forming blockade with thrombus in early and late period, to do antiagregant and antiagulant treatment with proper dose, to determine degree of light and serious form, to start proper treatment not wasting time, to prevent from late gestation of pregnancy. In order to prevent from bleeding it is important to analyse activity of blood coagulating system its loss.

This was proved by our study. If we see period of being pregnant during CS delivery problem 2.2-7.2% of women with many delivery on the 28-36 weeks, for women of this group pregnancy is conducted in combination with obstetric and organ system disease. It shows that it has risk of forming complication from the side of mother and fetus. For women with many delivery with up to 4 score occupies 1.9%, 5-6 score 10.3% this constitutes condition of forming chronic deficiency of oxygen to fetus and increase of prenatal morbidity because of obstetric and organ system disease.

93% of infants of deliverers for the first time was born with Apgar 7-8 score. Result of our study suits to the result of study of researchers J. Lhagvasuren (2006), Sh. Sh. Radjabova (1997) and scholars of Dagestan university. In the period after CS the following complications are formed in particular; metritis (7%), wound opening (2.7), septic (6%), wound suppuration (5.2%), hysterectomy (27.9%), total hysterectomy (5.2%). This is an important result which shows that complex measure of preventing from complication before, during and after surgery.

CONCLUSION
1. Among women who was performed CS organ system disease occurs (21.3%). Due to frequency of obstetric disease occupies 48.4% among women with many delivery.
2. In other word in comparison with deliverer for the first time above mentioned complication occurs 1.5 times more.
3. Women with many delivery are involved in high risky group. By involving them under control of pregnancy as early as possible, revealing body and organ system disease early, treating and curing and changes which appear hemodynamic and hemostase must become normal.

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Comparison Study Luteinizing Hormone, Testosterone Levels of Polycystic Ovary Syndrome


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2 School of Bio-Medicine, Health Sciences University of Mongolia

ABSTRACT
Polycystic ovary syndrome (PCOS) is one of the most common female endocrine disorders affecting women of reproductive age, with a prevalence of about 5%–10% in the general population. PCOS is characterized by menstrual disturbances due to hirsutism, chronic anovulation or oligoovulation, and acne due to excessive androgen production (hyperandrogenemia). This aim of the study was to compare testosterone (T) and luteinizing hormone (LH) levels in serum of reproduction age for comparative healthy women and polycystic ovary syndrome (PCOS). An analysis cross-sectional study was performed comprising 425 comparative healthy women of reproductive age group 18-35, 62 with PCOS patients. Control and patients were divided 3 groups. To collect blood sample 4 times (early follicular phase, late follicular phase, ovulation and mid-luteal phase) of healthy, normal menstrual cycle and body mass index (BMI) of women. To determine LH and T hormone in serum by ELISA. The mean age was group A 26.33±5.28, group B 24.9±4.93, and group C 29.75±4.12, BMI a; 22.35±2.95, b; 28.88±3.85, c; 28.0±3.19. The mean level was LH a; 6.42±3.3 mIU/ml, b; 39.43±15.89 mIU/ml, c; 10.89±2.53 mIU/ml of PCOS.

In conclusion, women with PCOS have elevated levels of LH and T strongly correlates with the clinical degree of amenorrhea and hyperandrogenism. It seems that LH, T could be a crucial diagnostic and predictive factor among women with menstrual disorders or presence of polycystic ovaries.

Key words: PCOS, LH, T, reproductive hormone

INTRODUCTION
Polycystic ovary syndrome (PCOS) is the most common endocrine disorder in women with a prevalence between 6 and 10% based on the National Institute of Health criteria and as high as 15% when the broader Rotterdam criteria are applied. PCOS is typically first identified during the early reproductive years. The clinical expression varies but commonly includes oligo or anovulation (Oligo), hyperandrogenism (HA) (either clinical or biochemical) and the presence of polycystic ovaries (PCO) (the “billiard ball” sign on ultrasound examination).1 Dewailly, Welt and Pehlivanov divided the patients with PCOS into 4 phenotype groups: A, B, C, and D.2,3,4,5 Regarding the Rotterdam criteria for PCOS both endocrinal and clinical, we distinguish 4 different phenotypes of the syndrome. (A) Oligo + HA + PCO, (B) Oligo + HA, (C) HA + PCO, (D) Oligo + PCO. 7 The most common irregularities of PCOS include elevated serum levels of free testosterone (T), androstenedione, dehydroepiandrosterone sulfate (DHEAS), excessive amount of luteinizing hormone (LH), increase in LH peak pulse frequency and its response to GnRH (Gonadotropin-releasing hormone), and change in LH pulse frequency. Insulin resistance, obesity, dyslipidemia, elevated laboratory findings associated with inflammation, high blood pressure, and increased risk of cardiovascular diseases are the common symptoms of PCOS. At the conference in Hamburg, the additional diagnostic criteria for PCOS were added: acne, hirsutism, elevated blood levels of androgens, and increased insulin resistance. Today’s definition of PCOS was defined by a consensus workshop sponsored by ESHRE/ASRM in Rotterdam in May 2003.13 As well as, 20-30 % of PCOS is diagnosed by ultrasound scan.6

It is beneficial to the results controlled when during the PCOS, hormone level and endocrine disorder are diagnosed. Therefore, it is vital to determine the level of LH and T healthy women or with PCOS.

MATERIALS AND METHODS
Subjects. The study was approved of the Ethical Committee of Minister of Health Mongolia, followed by principles outlined in the Helsinki Declaration.
PCOS patients
In the present study, sixth-two women with PCOS 18-35 years of age were diagnosed on the basis of the ESHRE/ASRM criteria.6 The physical examination included measurements of circumferences of their waist and hips and calculation of the body mass index (BMI). We also evaluated the extent of hyperandrogenism (hirsutism-using the Ferriman Gallwey hirsutism evaluation system, acne using the subjective 0–10 scale). The presented results were prepared by classifying the women into two groups (Rotterdam Study) according to their phenotype ((B=32 (HA+PCO) C=30 (Oligo+HA)). In each patient, we determined the serum levels of LH and testosterone. An ultrasound examination of the reproductive system, in particular the ovaries, was performed on each patient. The diagnosis of the hirsutism was established by gathering at least six points using the Ferriman Gallwey hirsutism evaluation system. Acne was diagnosed in these patients who received four or more points using the subjective 0–10 scale, in which we assessed face, back, and other body parts. Group B was divided menstrual phase four phase.

Healthy and control groups
We enrolled 425 healthy women, age 18-35 who had regular menstrual cycles. All women were healthy and never used any type of hormonal contraception or any other medical treatments for at type the last 3 months before entering the study.

Laboratory methods
Fasting morning (0800-1100) blood samples were collected and centrifuged and stored at -20°C until measurements were performed. Blood sample were collected 4 times from healthy women and type of hyperandrogenism and PCO. Between early follicular phase (day 1-3), late follicular phase (day 7-12), ovulation phase (day 13-14), mid luteal phase (day 21-24). Specially, A single blood sample were collected of amenorrhea and hyperandrogenism. We measured serum LH and testosterone concentrations with Enzyme-linked immuno sorbent assay (ELISA) in School of Bio medical, microbiological immunology.

Statistical analysis
All calculations were performed using SPSS V15.0 for Windows software. The geometrical mean and SD were used for serum LH and testosterone. The mean values of different parameters from different groups were compared with each other for significant differences and assessed using one-way ANOVA, Post host, Tukey.

RESULTS
The research involved 62 women who met the Rotterdam criteria for PCOS and 425 healthy women they are from in Ulan-Bator. We split the women who got involved in the research into three groups by their clinical sign as Group A (healthy women), Group B (Oligo+HA) and Group C (HA+PCO), then determined the level of LH and T. The characteristics of the participants are summarized in Table 1.

The women with PCOS among amenorrhea, their average age was 24.9±4.93, but when androgen is increased 29.75±4.12. In addition, during the research the participants’ body weight relevantly noticed as it is included in international classification (p<0.05).

<table>
<thead>
<tr>
<th>Variables</th>
<th>Group A</th>
<th>Group B</th>
<th>Group C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>26.33±5.2</td>
<td>24.9±4.93</td>
<td>29.75±4.12</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>22.35±2.95</td>
<td>28.88±3.85</td>
<td>28.0±3.19</td>
</tr>
<tr>
<td>Age of Menarche</td>
<td>13.59±1.94</td>
<td>16.2±1.03</td>
<td>15.75±1.37</td>
</tr>
<tr>
<td>Pregnancy</td>
<td>2.12±1.16</td>
<td>0</td>
<td>0.06±0.25</td>
</tr>
<tr>
<td>First pregnancy age</td>
<td>21.3±4.7</td>
<td>0</td>
<td>19±2.83</td>
</tr>
<tr>
<td>Birth</td>
<td>0.9±1.4</td>
<td>0</td>
<td>0.03±0.18</td>
</tr>
<tr>
<td>Abortion</td>
<td>1.55±0.8</td>
<td>0</td>
<td>1.03±0.18</td>
</tr>
</tbody>
</table>

Table 1.
Table 2 shows the group-related mean values of subjects serum LH and T.

### LH and T levels of healthy women and patients with PCOS

<table>
<thead>
<tr>
<th>Groups</th>
<th>n</th>
<th>m±SD (mIU/ml)</th>
<th>p value</th>
<th>m±SD (mIU/ml)</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group A</td>
<td>425</td>
<td>6.42±3.3</td>
<td>0.001</td>
<td>0.61±0.2</td>
<td>0.001</td>
</tr>
<tr>
<td>Group B</td>
<td>30</td>
<td>39.43±15.89</td>
<td>0.001</td>
<td>1.24±0.52</td>
<td>0.001</td>
</tr>
<tr>
<td>Group C</td>
<td>32</td>
<td>10.89±2.53</td>
<td>0.05</td>
<td>0.74±0.61</td>
<td>0.01</td>
</tr>
</tbody>
</table>

From the above table we can see that group A LH level 6.42±3.30 mIU/ml, amenorrhea group B 39.43±15.89 IU/ml, group C while androgen increased 10.89±2.53 mIU/ml (p<0.05). T level was determined group A 0.61±0.2 ng/ml, group B 1.24±0.52 ng/ml whereas group C 0.74±0.61 ng/ml (p<0.001). Although Group C is androgen increased and with PCOS, it is regular menstruation cycle. LH and testosterone is defined by its measured into the 4 menstruation cycles and demonstrated in the group A and C. Table 3

### Comparisons between serum LH and T levels with menstrual phase

<table>
<thead>
<tr>
<th>Menstrual cycle</th>
<th>T</th>
<th>m±SD</th>
<th>n=32</th>
<th>n=425</th>
<th>p value</th>
<th>LH</th>
<th>m±SD</th>
<th>n=32</th>
<th>n=425</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Early follicular phase</td>
<td>0.7±0.65</td>
<td>0.7±0.27</td>
<td>&lt;0.05</td>
<td>10.89±2.53</td>
<td>6.42±3.3</td>
<td>&lt;0.001</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Late follicular phase</td>
<td>0.88±0.72</td>
<td>0.56±0.21</td>
<td>&lt;0.05</td>
<td>16.37±14.78</td>
<td>10.46±4.95</td>
<td>&lt;0.001</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ovulation phase</td>
<td>0.62±0.61</td>
<td>0.58±0.17</td>
<td>&lt;0.05</td>
<td>33.55±26.72</td>
<td>26.8±9.56</td>
<td>&lt;0.001</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mid luteal phase</td>
<td>0.77±0.6</td>
<td>0.59±0.18</td>
<td>&lt;0.05</td>
<td>10.52±7.23</td>
<td>4.93±1.35</td>
<td>&lt;0.001</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>0.74±0.61</td>
<td>0.61±0.22</td>
<td>&lt;0.05</td>
<td>16.74±2.95</td>
<td>14.48±10.05</td>
<td>&lt;0.001</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Testosterone level was increased in follicular late phase by 0.88±0.56 ng/ml. In conclusion, when amenorrhea and androgen increased, testosterone level is the highest as 1.24 ng/ml. (p<0.001). From the result, although LH mid level is greater during ovulation phase, there is statistical standard deviation. (p<0.05).

**DISCUSSION**

By our study, the mean age of PCOS was detected comparative early diagnosed by 24.9±4.93 years old. Such as in Iran, generally type of amenorrhea was early diagnosed by 28.98±8.9 age of PCOS (Behnaz Khani et al 2011).11 As in other studies, mean serum testosterone level was 0.81 ng/ml for amenorrhea and hyperandrogenism, 0.41 ng/ml for hyperandrogenism and PCO.15 In Poland, Olgierd Głuszak et al. reported testosterone level had below 0.76±0.3 ng/ml.16 Similar results have been reported elsewhere. The mean of testosterone level was 0.74±0.61 ng/ml of PCOS, 0.61±0.22 ng/ml of healthy women. In China, mean serum testosterone was 0.97±0.11 ng/ml of PCOS, 0.51±0.09 ng/ml of control (Shou-Kui Xiang 2012 et al).17 But in Bahrain women testosterone level was 0.95±0.07 ng/ml, 1.42±0.32 ng/ml of PCOS (Jamal Golbahar et al 2008).18 Studies in Western countries have shown that the prevalence of LH level increased 95%. In this study, the prevalence of LH higher were all 62 cases of PCOS.16 The LH level was 8.3±7.2 ng/ml of PCOS by Bertha Pangaribuan et al.9 Our study, significant differences between PCOS and healthy women were observed for LH level and group B.

**CONCLUSIONS**

In conclusion, women with PCOS have elevated levels of LH and T strongly correlates with the clinical degree of amenorrhea and hyperandrogenism. It seems that LH, T could be a crucial diagnostic and predictive factor among women with menstrual disorders or presence of polycystic ovaries.

**ACKNOWLEDGEMENTS**

We thank the study participants, field staffs, of the School of Biomedicine of Health Sciences University of Mongolia.
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Comparative Study on Medical Ethics

Shagdarsuren D, Damdinjav Kh

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2 School of Public Health, Health Sciences University of Mongolia

ABSTRACT

Humanistic principles are commonly mentioned in both western and eastern medical ethics. However, there is a lack of detailed comparative analysis on strengths and features of these two approaches. Briefly, humanism can be defined as following the principle of compassion and prioritizing others’ interests and welfare before your own ones. In modern medicine, humanistic approach can serve to find solutions for highly debatable issues such as euthanasia or testing on animals. In traditional eastern medicine, all actions of a medical professional such as treatment and diagnosis are conducted based on clearly defined ethics. According to famous script “Four Tantras of Traditional Medicine,” ethics are based on compassionate conscience “love for all beings”. This paper analyzes humanism in medical ethics stated in “Four Tantras of Traditional Medicine” with comparative perspective to western medical science.

INTRODUCTION

Humanistic principles are commonly mentioned in both western and eastern medical ethics. However, there is a lack of detailed comparative analysis on strengths and features of these two approaches. Briefly, humanism can be defined as following the principle of compassion and prioritizing others’ interests and welfare before your own ones. In modern medicine, humanistic approach can serve to find solutions for highly debatable issues such as euthanasia or testing on animals. In traditional eastern medicine, all actions of a medical professional such as treatment and diagnosis are conducted based on clearly defined ethics. According to famous script “Four Tantras of Traditional Medicine,” ethics are based on compassionate conscience “love for all beings”. This paper analyzes humanism in medical ethics stated in “Four Tantras of Traditional Medicine” with comparative perspective to western medical science.

PURPOSE AND GOALS

The study was conducted to compare ethical precepts of physicians and other health care professionals between western and eastern medicine and analyze the ethical principles methodically.

1) Finding the fundamental universal principles of Western and Eastern medical ethics
2) Determining effectiveness of the fundamental principles of medical ethics
3) Aiming to executing ethical standards by health care professionals and examine the ways to resolve ethical dilemmas correctly based on historical facts and the primary texts

MATERIALS AND METHODS

Indian, Tibetan, Mongolian, Chinese, Greek and Arabian medical texts, its explications, modern biomedical ethical theories and facts were used.

Document analysis, logical argumentation, analytic-synthetic distinction, hermeneutics, grouping and content analysis were utilized in the abstract.

RESULTS

1) The principle of compassion is more brought up in eastern medical ethics compared to western medical ethical principles.
2) Being highly ethical medical professional is essential to diagnose and treat diseases, prevent, cure the untreatable patients, enhance the strength of drug, purify the poison of drug raw material (i.e., mercury) and generate psychotropic skills of physicians.
3) The beginning of ethical education starts with loving own parents. It is not useful when physicians swear “I will give all my love for others” and nurses swear “I will possess genuine compassion” if they do not love their own mother properly.

According to Mongolian and Tibetan traditional ethics, nothing is created by power of God, the existence of everything is explained by interrelatedness and positive impacts of others. Moreover, it is not worth to have sworn statement that “We prefer public priorities than our own interest” in ethical principle of clinical professionals when they do not realize the law of causality.

DISCUSSION

“Ashtanga”, “Charaka”, “Sushruta” are the primary texts of Indian ayurvedic medicine and its ethical theories are
based on the philosophies of Brahma, Shiva and Vishnu of Hindu religion, ethical theories of ancient Chinese medical text, “Neijing” are from the philosophies of Confucius and Dao, and ethical theories in Arabian medical book, “Avicenna” are based on ethical principles of the Koran of Muslim religion. “Dumta” is the primary philosophical basics of Mongolian medicine. According to the Dumta, philosophy of “I” or “creator” is faulty that is mentioned in Christianity and Shamanism. Love and compassion are taught in these teachings. In Buddhism life is cherished, but at the same time the end of it seen as step to the next life. Table 1 reveals some differences between Western and Eastern views on euthanasia. The euthanasia debate, which started in Western world just in last century, was answered already in detail in ancient documents of East. Furthermore, the love and compassion in Western practice is subject to discrimination and shortsighted principles, while in Eastern side love is universal for any living beings. Tibetan and Mongolian traditional medicine teach to love every living being as your own mother and it is based on compassion, integration, causes, the law of causality of Mahayana philosophy in “Four Tantra”, “Four Amrita”. For instance animals are treated same as humans, and animal testing is forbidden as it is seen as testing on own mother (Table 2). Table 2 and 3 show that karma plays significant role in defying ethics in Eastern medicine. Physician who made suffer animals through the testing can have same pain and sufferings later in life or in the next life. In abortion case, parents who aborted their child could have death from abortion in their next life. Therefore, there were two traditional ways such as “seven masteries of causality” and “being in one’s position” to train physicians and medical professionals to have compassion for others.

Table 1.

<table>
<thead>
<tr>
<th>Euthanasia in western and eastern medicine</th>
</tr>
</thead>
<tbody>
<tr>
<td>In Western medicine</td>
</tr>
<tr>
<td>Preparation</td>
</tr>
<tr>
<td>Preparation started by somebody himself or by family members</td>
</tr>
<tr>
<td>Time of administration</td>
</tr>
<tr>
<td>Time of administration</td>
</tr>
<tr>
<td>Methods</td>
</tr>
<tr>
<td>Methods</td>
</tr>
<tr>
<td>Medicine used</td>
</tr>
<tr>
<td>Medicine used</td>
</tr>
<tr>
<td>Who administers?</td>
</tr>
<tr>
<td>Who administers?</td>
</tr>
<tr>
<td>When first applied?</td>
</tr>
<tr>
<td>When first applied?</td>
</tr>
<tr>
<td>Non-drug methods</td>
</tr>
<tr>
<td>Non-drug methods</td>
</tr>
<tr>
<td>Ultimate goal</td>
</tr>
<tr>
<td>Ultimate goal</td>
</tr>
</tbody>
</table>
Passive euthanasia is administered when terminally ill patient requests to end treatment or life support when it is possible to cure for persons who: - pose threat to public - harm environment - disrespectful to service of physician - offend physician - breaks the Buddhist religion when it is impossible to cure - 9 kinds of terminal illnesses - with signs and symptoms of near death

Active euthanasia is administered in countries where it is legalized for patient with obvious signs of near death when the body is dying and the breath is disturbed when external breath stops but spirit is still inseparable.

<table>
<thead>
<tr>
<th>Table 2. Testing on animals in western and eastern medicine</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>In Western medicine</strong></td>
</tr>
<tr>
<td><strong>Animal</strong></td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td><strong>Testing on animals</strong></td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td><strong>Theory</strong></td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td><strong>After the testing</strong></td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td><strong>Animal killing</strong></td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td><strong>Effects on researcher who conducts the testing</strong></td>
</tr>
<tr>
<td></td>
</tr>
</tbody>
</table>
Table 3.

<table>
<thead>
<tr>
<th></th>
<th>In Western medicine</th>
<th>In Eastern medicine</th>
</tr>
</thead>
<tbody>
<tr>
<td>Allowed</td>
<td>If legalized in that country, less than 12 weeks of pregnancy</td>
<td>If there is danger for mother’s life</td>
</tr>
<tr>
<td>Prevention</td>
<td>Pregnancy prevention methods</td>
<td>Avoid improper intercourse, maintain sexual ethics</td>
</tr>
<tr>
<td>Abortion hazards</td>
<td>Illegal abortion is punished</td>
<td>Considered same as murder, and pay it all through karma</td>
</tr>
<tr>
<td>Start of life</td>
<td>With first heartbeat of the fetus</td>
<td>At the moment of egg fertilization</td>
</tr>
<tr>
<td>Parent consciousness</td>
<td>The consciousness of being parent not developed toward the zigot</td>
<td>Abortion seen as child killing</td>
</tr>
<tr>
<td>When abortion is performed</td>
<td>Abortion performed even if the normal birth is possible</td>
<td>When it is inevitable and no other means are left</td>
</tr>
<tr>
<td>Abortion effects on parents</td>
<td>Divorce, secondary infertility, death</td>
<td>Divorce, secondary infertility, death, shortening of life, terminal uterus disease, death through dismemberment from abortion in next life</td>
</tr>
<tr>
<td>Abortion effects of physician</td>
<td>Legal punishment if performed wrongfully</td>
<td>Shortening of life, loosing treatment skills</td>
</tr>
<tr>
<td>Reason for abortion</td>
<td>Unwanted pregnancy, fetal deformity, genetic illness, population control policy</td>
<td>Save mother’s life</td>
</tr>
</tbody>
</table>

**CONCLUSION**

In order to find ways to execute medical ethical standards in Mongolia and resolve ethical dilemmas we also have to obtain Mongolian traditional medical ethical training that is one of the traditional culture and wisdom. Mongolians need to learn some ethical resolution related to modern medical precise activities such as genetic engineering and human cloning. However, Mongolian traditional medicine has a richer ethical traditional culture with ethical issues of physician-patient and physician-active or inactive nature.

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Endometrial Epithelial Cell Changes Associated with Uterine Leiomyoma

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ABSTRACT

The uterus comprises the myometrium and the endometrium. The endometrium undergoes dynamic reorganization through each menstrual cycle in response to the steroid hormones. Uterine leiomyomas account for more than 75% of the benign tumours in women of reproductive age group. They are dependent upon the steroid hormones for their growth and maintenance. Hence, it is not surprising that the endometrium would respond to the pathobiologic state of the leiomyomatous uteri. We aimed to study the histopathological changes in endometrium in association with uterine leiomyomas and to identify endometrial changes which help to suggest a diagnosis of uterine leiomyomas on endometrial curettings.

100 cases wherein the uteri revealed leiomyoma were studied. Following receipt of the specimen, a detailed gross examination was done after formalin fixation. Further, microscopic examination was performed after processing, obtaining sections of 5 microns thickness and staining with routine H & E.

Leiomyomas occurred mostly in women aged between 41-50 years and in multiparous women. Menorrhagia was the commonest presentation. Endometrial pattern commonly seen was proliferative phase or hyperplasia. Other epithelial cell changes seen were dilated, elongated or distorted glands, glands parallel to muscle fibres and glands separated by muscle fibres.

Leiomyomas are steroid dependent tumours wherein the endometrium manifests mostly as proliferative phase or hyperplasia suggesting estrogentic prevalence. Association with multiparity explains the need for progesterone in maintenance of leiomyomas. The presence of mixed findings such as glandular atrophy, endometrial hyperplasia or polyposis, together with distorted, dilated or elongated glands and muscle fibres between glands in endometrial curettings, could suggest a possibility of uterine leiomyoma.

Key words: Leiomyoma; Steroid hormones; Endometrial changes

INTRODUCTION

The endometrium is a dynamic tissue which shows structural changes during the menstrual cycle. Structural reorganisation occurs with each menstrual cycle in preparation for implantation. If no implantation occurs, the superficial layer is partially / completely shed and remodelled in preparation for the next cycle. 1

Studies have shown the presence of estrogen and progesterone receptors in the endometrium and myometrial tissue. They play an important role in regulating their growth.3

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Uterine leiomyomas account for more than 75% of the benign tumours in women of reproductive age group.2 The content of the hormone receptors has been shown to be higher in the fibroid tissue.2

In our hospital, hysterectomies are mostly performed for the management of leiomyomas and dysfunctional uterine bleeding. Hence, this study will help to study the histopathologic changes in endometrium in cases of uterine leiomyomas.

The cause of uterine leiomyoma is undetermined. Many factors have been considered on theoretical grounds but practical support is lacking. Heredity is considered to be a factor “fibroids run in families”. From the clinical and epidemiological surveys in the USA, it is known that fibroids are 3-9 times more common in Negroes than in Caucasians. It has been suggested that, this is due to the higher prevalence of pelvic infections in black women,
with abnormal uterine growth occurring secondarily to the myometrial irritation caused by these infections. An alternative view may be the presence of a gene encoding for fibroid development, as there is often a positive family history of fibroids in patients who develop these tumours. They are true neoplasms rather than hyperplastic proliferations. This has been convincingly demonstrated by analysis of iso-enzymes of glucose-6-phosphate-dehydrogenase.

The factors involved in the initiation and growth of leiomyomas remain poorly understood. They probably involve complex interactions of sex steroid hormone and local growth factors with somatic mutations in the normal myometrium. Uterine leiomyomas appear during the reproductive years and regress after the menopause, indicating ovarian steroid-dependent growth potential. Further evidence for the role of female sex hormones in the growth of leiomyomas is their occasional rapid growth and haemorrhagic degeneration associated with pregnancy, clomiphene and progesterone treatment. Oestrogen and progesterone promote the development of myofilaments and dense bodies ultrastructural features of smooth muscle differentiation.

**MATERIALS AND METHODS**

The study conducted at the First Maternity Hospital, Gynecology department, Ulaanbaatar city, Mongolia from June 2010 to December 2012. The hysterectomy specimens received at the Pathology Department, Medical College, Health Science University of Mongolia.

A total of 100 cases were studied in which leiomyoma was present. The study material was obtained from patients admitted at Department of Gynecology, 1st Maternity Hospital, Ulaanbaatar, Mongolia who underwent hysterectomy.

Brief clinical data was obtained from patients or patient records with respect to age, clinical presentation, parity and menstrual phase. Following the receipt of surgical specimens, they were fixed in 10% formalin for 24-48 hours. A detailed gross examination was performed with respect to size and weight of uterus, location and size of fibroids, secondary changes like cystic change, red degeneration, calcification, mucoid degeneration or fatty degeneration and status of endometrium and endometrial polyp if any was noted.

Tissue bits from representative areas of the fibroids and endometrium were taken for histopathological examination, processed and paraffin blocks were made. Sections were cut at 5 micron thickness and stained with hematoxylin and eosin. Microscopic sections were studied and following histologic features were recorded:

1. Endometrial parameters- thickness of endometrium, phase, number and appearance of glands within the given area and stromal changes.

The endometrial area was calculated using a standard 2-mm length multiplied by the measured width. Further, the obtained parameters were evaluated using descriptive statistical analysis. Significance was assessed at 5% level of significance. Chi-square/ Fisher Exact test have been used to find the significance of study parameters on categorical scale between two or more groups. 95% Confidence Interval has been computed to find the significant features. Confidence Interval with lower limit more than 50% is associated with statistical significance.

**RESULTS:**

Endometrial phase on microscopy: Of the 100 cases, proliferative endometrium was noted in 38%, followed by secretory endometrium in 24%, endometrial hyperplasia in 20% and atrophic endometrium in 18% of the cases. (Table 1)

<table>
<thead>
<tr>
<th>Endometrial Phase</th>
<th>Number (n=100)</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Proliferative phase</td>
<td>38</td>
<td>38.0</td>
</tr>
<tr>
<td>Secretory phase</td>
<td>24</td>
<td>24.0</td>
</tr>
<tr>
<td>Simple hyperplasia</td>
<td>20</td>
<td>20.0</td>
</tr>
<tr>
<td>Senile cystic atrophy</td>
<td>18</td>
<td>18.0</td>
</tr>
<tr>
<td>Secretory hyperplasia</td>
<td>8</td>
<td>8.0</td>
</tr>
</tbody>
</table>

Endometrial epithelial cell changes: Among the various epithelial cell changes encountered in the endometrium of leiomyomatous uteri, dilated or distorted glands and arrangement of glands parallel to the long axis of myometrium were seen in 43% of the cases, followed by 31% of the cases showing endometrial glands separated by muscle fibres. Other changes seen are as listed in the table 2.
Table 2.

<table>
<thead>
<tr>
<th>Epithelial Cell Changes</th>
<th>Number (n=100)</th>
<th>%</th>
<th>95%CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dilated/ distorted glands</td>
<td>43</td>
<td>43.0</td>
<td>43.29-62.49</td>
</tr>
<tr>
<td>Endometrial glands parallel to myometrium</td>
<td>53</td>
<td>53.0</td>
<td>43.29-62.49</td>
</tr>
<tr>
<td>Endometrial glands separated by muscle fibres</td>
<td>31</td>
<td>31.0</td>
<td>22.78-40.63</td>
</tr>
<tr>
<td>Total glandular atrophy</td>
<td>13</td>
<td>13.0</td>
<td>7.76-20.98</td>
</tr>
<tr>
<td>Subtotal glandular atrophy</td>
<td>4</td>
<td>4.0</td>
<td>1.78-9.83</td>
</tr>
</tbody>
</table>

Further, total and subtotal endometrial glandular atrophy showed significant association (p value<0.001 and <0.002 respectively) with submucosal leiomyoma.

The above findings suggest that if endometrial curettings obtained show a mixed picture of glandular atrophy, endometrial hyperplasia or polyposis, together with many distorted, elongated or dilated glands and muscle fibres between glands, one can suggest the presence of uterine leiomyoma.

Endometrial Stromal Changes: of the various stromal changes, haemorrhage was seen in 33% of the cases and stromal edema in 3% (Table 3)

Table 3.

<table>
<thead>
<tr>
<th>Endometrial stromal changes</th>
<th>Number (n=100)</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Haemorrhage</td>
<td>28</td>
<td>28.0</td>
</tr>
<tr>
<td>Edema</td>
<td>15</td>
<td>15.0</td>
</tr>
<tr>
<td>Chronic Endometritis</td>
<td>1</td>
<td>1.0</td>
</tr>
<tr>
<td>Absent</td>
<td>56</td>
<td>56.0</td>
</tr>
</tbody>
</table>

Endometrial hyperplasia, polyposis, edema and haemorrhage can result from hormonal disturbances, mainly hyperestrogenism.

Correlation of presenting illness with endometrial area

Majority of the patients who had endometrial area of more than 1 sq.mm presented with symptoms of excessive bleeding. (Table 4)

Table 4

<table>
<thead>
<tr>
<th>Presentillness</th>
<th>Number of patients (n=100)</th>
<th>Endometrial area (sq mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>&lt;1.0</td>
</tr>
<tr>
<td>Excessive bleeding</td>
<td>58</td>
<td>1.72%</td>
</tr>
<tr>
<td>Mass per abdomen</td>
<td>21</td>
<td>4.76%</td>
</tr>
<tr>
<td>Pain abdomen</td>
<td>26</td>
<td>0%</td>
</tr>
<tr>
<td>Mass per vagina</td>
<td>22</td>
<td>22.73%</td>
</tr>
</tbody>
</table>
DISCUSSION

The present study included topographical investigation of the pathological changes of the endometrium with special reference to the site of leiomyoma within the uterus. We suggest that the contradictory descriptions and interpretations that have been reported may be partly explained by the varied changes.

Proliferative endometrium and endometrial hyperplasia, both of which represent estrogenic phase accounted for 57% of the endometrial phase, suggesting a prevalence of estrogenic activity in the leiomyomatous uteri. The incidence of complex hyperplasia was only 2%, which substantiates for the higher content of estrogen and progesterone receptors in the myometrium than in the endometrium. Thereby, the brunt of excess hormones was borne by the myometrium in the form of leiomyoma, while endometrium mostly showed either proliferative phase or simple hyperplasia.

These different pathological patterns may be the result of a mechanical factor and a hormonal factor. Atrophy of the endometrium, elongation, and distortion of the glands may result from mechanical pressure exerted by the nodular mass of the leiomyoma on the overlying or nearby endometrium.

CONCLUSION

Alterations of the endometrial cycle in these 100 women were due to hormonal variations which may represent a common cause for both the leiomyoma and some of the endometrial changes. The remaining endometrial abnormalities appeared to have been due to mechanical factors that mainly affected the endometrium lying over, opposite, or close to the leiomyoma.

REFERENCES

Emergency and Disaster Preparedness of City Health Facilities

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ABSTRACT

Due to current climate change, new and emerging infectious disease, negative impacts affected by the nature and humans and increasing number and frequency of natural disaster such as earthquake, it is becoming more important to accurately predict the possibility of risks and disasters in order to prevent death, and especially, to ensure the safe operation of health organizations.

From recent earthquake examples such as earthquakes in China, Haiti and Chile, researches show us it is very important to ensure emergency preparedness which needs to be started by any individual, family, company and nationwide.

Eleven thousand health departments were collapsed and damaged during the earthquake in Sichuan, China in 2008. Also, an earthquake in March, 2011 in Japan gave the world an understanding of the dangers of natural disaster. Mongolia is located in active earthquake zone of Central Asia. Earthquakes were 50.9% of the overall natural disasters that occurred in XX century. Based on the statistics by the International Earthquake Association in the USA, there were 170 earthquakes with magnitude of 8 or greater, and four of them occurred in Mongolia. Earthquake UB 2005. page 6/. During these years in Mongolia the construction development was poor, and low population density was a main reason for our country being able to pass through these disasters without much economic loss or damages.

INTRODUCTION

Based on research from the last 6 years, it is been observed that there has been a high rise in the number of earthquakes around the western zone of Ulaanbaatar city, which means focus is more intensified near the city area. /appendix of ordinance 98 of Mar, Sep 2010/

By soil research, Ulaanbaatar is considered to be a highly effective earthquake zone with the possibility of a 7.5 magnitude earthquake, based on detailed space photos and field research. Twenty five percent of construction areas of Ulaanbaatar city belong to earthquake zone of 6 magnitude, 52% in 7 magnitude, and 23% in 8 magnitude. Sixty point nine percent of the population are in city areas/ pages 8 and 9 of information about disasters and injuries happened in Mongolia/

It is registered that approximately 1000 – 1500 earthquakes occur per year in our country and 3 to 5 of these earthquakes are quite strong and visible (4-5 magnitude). /page 27, Book on information about disasters and injuries happened in Mongolia/

Soil is comparatively solid, and because it belongs to 6-8 magnitude zone, it is estimated that 556 residential apartments buildings and 65.3-66.1 thousand people who reside in these buildings may be at risks in earthquake of 7 magnitude. /NUBHH ‘Earthquake management system of Ulaanbaatar city’ MoH/99/301 project/

Therefore it becomes mandatory to make an assessment in health buildings and construction. Our objective is to assess current buildings of second and third level hospitals in Ulaanbaatar city and to determine emergency preparedness of hospitals involved in the survey.

MATERIALS AND METHODS

Assessment Sheets were developed and based on the Assessment Model of Safe Hospitals issued by WHO and using Instant Methodology of Analytical Research. A research methodology was approved by the Board Meeting of Scientists’ from the Public Health Institute at the Health Sciences University of Mongolia (HSUM). Once it was approved, the meeting of Medical Ethics Sub-Committee was held on 13 June 2011, which issued ethical permission to start research. Twenty second level hospitals and 10 third level hospitals were involved in the research, and SPSS-17.0 software was used for survey information processing.
RESULTS

More than 83% of buildings of 2nd and 3rd level hospitals involved in the research were built from brick; 33.3% (10) were connected by bridge with the city and 36.7% (11) had not built for the purpose of a hospital. 100% had not demolished loadbearing walls and 98.3% (28) had not changed plan drawings. 43.3% had been used for 21-57 years and (13) had been used for more than 40 years. 100% had not completed a risk assessment.

‘A’ type buildings which were built without estimation and are sorted by districts, with 22.1% in Bayangol district, 34.3% in Chingeltei, 9.9% in Khan-Uul, 5.4% in Bayanzurkh and 13.8% in Songino Hairhan. Thirteen (43%) of hospitals buildings are located in these districts.

![Figure 1. Buildings of health organizations in Ulaanbaatar city](image1)

![Figure 2. Years of building constructions of health organizations](image2)

Ninety percent of 2nd and 3rd level hospitals have no fire alarm system; 36.7% (11) have no surgery operation room; 56.7% (17) have no access doors and stairs for carrying beds and disabled patients; 73.3% (22) have no water source; 26.7% (8) have no additional power source; 30% (9) have no gas system and 56.7% (17) were using mobile gas.

More than 96% (29) of hospitals included in the research have signs and labels, 73.3% (22) have colour level system; 96.7% (29) have working groups during emergencies and 90% (27) have emergency assessment systems.

The emergency preparedness plan of 60% (18) had no relation with other hospitals; 83.3% (25) run emergency preparedness training, although 53.3% are not issuing funding sources; 33.3% has no plan to leave building during emergency; 43.3% has no security worker to give instruction during emergency; 70% (21) can estimate target area for healthcare service; 53.3% (16) have no blood resource; and (13) have no resources to present mobile hospital.

DISCUSSION

Around 83% of 2nd and 3rd level hospital buildings that were involved in the research are built from brick and stone; 43.3% have been used for 21-57 years and (13) have been used for more than 40 years, and although emergency preparedness training is planned, 53.3% have no funding allocated, and 60% (18) have no connection with other hospitals on emergency planning. All have no emergency assessment and are increasing risk to people and health workers during earthquake.

There is research on earthquakes in Mongolia, however there is much less research on the strength of hospital buildings. From government agency research it is estimated that 300-350 buildings would collapse in an earthquake of 7 or more magnitude, but in our research this number increased to be 50% (10) of the 20 second level hospitals.

From our research it becomes important to deliver the results to decision makers and issue instruction on necessary actions.

CONCLUSIONS

1. Buildings of 2nd and 3rd level hospitals that are operating at this moment in Ulaanbaatar city are not meeting with safe hospital requirements, which is increasing possible risks.

2. The emergency preparedness plan prepared is without proper reference to other organisations and does not satisfy the expected result due to lack of funding, no plan to leave organisation and no blood source during emergency, meaning that operation and capacity of health departments during emergency is in poor condition.

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Effect of Calcite Compound on Bone Mineral Contents and Bone Markers in Rat Model of Osteoporosis Induced by Retinoic Acid

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2 Clinical and Pharmacological Institute of Traditional Mongolian Medicine, National University of Inner Mongolia
3 Centre of Research and Training, School of Health Technology, HSUM

ABSTRACT

The aim of this study was to examine the effects of calcite compound on bone mineral contents and bone markers of rat model of osteoporosis induced by retinoic acid. Sprague-Dawley rats were randomly divided into 4 groups. Retinoic acid was given at concentration of 70 mg/kg for 14 days. Calcite compound and CalciD-Denk were administered at concentrations of 0.5 g/kg and 1.16 g/kg respectively.

Ash weight of vertebral bones and concentrations of Ca and P in the ashes were determined by using Inductively Coupled Plasma-Atomic Emission Spectrometry (ICP-AES). Blood plasma levels bone resorption marker- Cross-linked C-telopeptide of collagen type 1 (CTx) and Inflammatory-Associated Marker - receptor activator of nuclear factor kappa-B ligand (RANKL) were measured by ELISA.

Ash weight of vertebral bones and Ca concentration in the bone ashes were significantly reduced after administration of retinoic acid (р<0.001) and they were significantly increased by calcite compound (р<0.001). Levels of CTx and RANKL in the blood plasma significantly increased in rats received retinoic acid (р=0.0001). Calcite compound significantly reduced increases in CTx and RANKL induced by retinoic acid (р=0.0001).

Calcite compound can be beneficial for treatment of osteoporosis through enhancing osteoblast differentiation and bone formation.

Key words: osteoporosis, retinoic acid, calcite, bone ash, CTx, RANKL

INTRODUCTION

Osteoporosis is called the silent epidemic. Change of lifestyle is regarded the potential risk factor of osteoporosis. According World Health Organization, osteoporosis ranks fourth place after cardiovascular disorders, cancer, and diabetes mellitus. Over 8.9 incidences of osteoporosis have been reported worldwide and 4.5 of them have occurred in America and Europe. While 2.8 million of them have disabled due to fracture. According to a study carried out in Mongolia, 57.5 % of women who were diagnosed with osteoporosis had sedentary lifestyle and 42.5 % had physical activities suggesting that sedentary lifestyle can be one of risk factors of osteoporosis. Another study carried out in “Medsco sunny” diagnostic center showed that 88 % of women who were going through menopausal stage had osteoporosis.

Diagnosis, treatment and prevention issues of osteoporosis have been encountering problem of health care system worldwide.

In this study, we examined the effect of calcite, a bone essence as written in traditional sourcebooks of medicine, on rat osteoporosis induced by retinoic acid.

MATERIALS AND METHODS

Sprague-Dawley rats weighing 170-200 g were purchased from Experimental Animal Centre of Jilin University, China. The study was carried out in Clinical and Pharmacological Institute of Traditional Mongolian Medicine, National University of Inner Mongolia. Retinoic acid was purchased from Shaanxi Sciphar Biotechnology Company, China. Ca content of calcite was 44 % as determined in Geological Laboratory, Mongolia. CalciD-Denk was manufactured in Denk Pharma, Germany. ELISA kits for rat collagen type 1 cross-linked C-telopeptide (CTx) and receptor activator of nuclear factor kappa-B ligand (RANKL) were purchased from Khu-Gao Company of Biological Test, China.
Animal model of osteoporosis
Sprague-Dawley rats were randomly divided into 4 groups with 12 animals in each group: 1. control group received distilled water, 2. retinoic acid group received retinoic acid, 3. calcite group received calcite and retinoic acid, and 4. CalciD-Denk group received CalciD-Denk and retinoic acid. Osteoporosis was induced by retinoic acid administration to rats at concentration of 70 mg/kg for 14 days.\textsuperscript{4,5,6} Calcite compound and CalciD-Denk were given orally at concentrations of 0.5 g/kg and 1.16 g/kg respectively for 30 days.\textsuperscript{7}
Determination of mineral contents in bone ashes
Ash weight of rat vertebral bones and Ca and P contents in the ashes were determined by using Inductively Coupled Plasma-Atomic Emission Spectrometry.

**RESULTS**
Ash weight of vertebral bones and Ca, P concentration in the ashes were reduced by 500 mg and 27.66 mg, 6.54 mg after administration of retinoic acid respectively (р<0.001). Calcite compound increased (р<0.001) the ash weight and Ca, P contents in the bone ashes of rats given retinoic acid by 500 mg and 25.18 mg, 6.19 mg respectively (Table 1).

<table>
<thead>
<tr>
<th>Groups</th>
<th>Ash weight (g)</th>
<th>Ca (mg)</th>
<th>P (mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (n=10, distilled water)</td>
<td>0.18±0.006</td>
<td>53.49±1.4</td>
<td>18.66±0.4</td>
</tr>
<tr>
<td>Retinoic acid (n=12, Retinoic acid, 70 mg/kg)</td>
<td>0.13±0.007*</td>
<td>25.83±5.9</td>
<td>12.12±2.6</td>
</tr>
<tr>
<td>Calcite (n=12, Calcite 0.5 g/kg+retinoic acid 70 mg/kg)</td>
<td>0.18±0.026</td>
<td>51.01±7.8**</td>
<td>18.31±3.7</td>
</tr>
<tr>
<td>CalciD-Denk (n=12, CalciD-Denk 1.16 g/kg +retinoic acid 70 mg/kg)</td>
<td>0.15±0.013</td>
<td>41.29±6.8</td>
<td>16.82±0.4</td>
</tr>
</tbody>
</table>

* p<0.05, ** p<0.001

CTx was increased by 8.85 ng/ml in retinoic acid administered rats compared to control (p=0.0001). Calcite compound decreased CTx by 12.45 ng/ml (p=0.0001). RANKL levels were increased by 1.94 pmol/L compare to control (p=0.0001). It was decreased by calcite compound by 2.6 pmol/L (p<0.001, Table 2).

<table>
<thead>
<tr>
<th>Groups</th>
<th>CTx (ng/ml)</th>
<th>RANKL (pmol/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (n=10, distilled water)</td>
<td>74.88±0.85*</td>
<td>51.11±0.77</td>
</tr>
<tr>
<td>Retinoic acid (n=12, Retinoic acid, 70 mg/kg)</td>
<td>83.73±3.90*</td>
<td>53.05±0.91*</td>
</tr>
<tr>
<td>Calcite (n=12, Calcite 0.5 g/kg+retinoic acid 70 mg/kg)</td>
<td>71.28±1.18**</td>
<td>50.45±0.96**</td>
</tr>
<tr>
<td>CalciD-Denk (n=12, CalciD-Denk 1.16 g/kg +retinoic acid 70 mg/kg)</td>
<td>75.08±2.28</td>
<td>51.70±0.56</td>
</tr>
</tbody>
</table>

Table 1

Table 2
**DISCUSSION**

While studying causes and pathogenesis of osteoporosis, researchers have developed various models of osteoporosis. Animal models of osteoporosis developed by feeding animals with Ca deficient food materials or ovariectomy are most commonly used. Rodents have been regarded the most suitable animals for osteoporosis models.

Short-term osteoporosis is caused by retinoic acid in rats. Retinoic acid is a derivative of vitamin A, which mainly used in the treatment of Leukemia at present that has a few adverse/side effects such as osteoporosis.

Ash weight and Ca and P contents in the ashes or bone resorption markers have commonly been used for examinations of effects of medicines with high Ca contents (shell, oyster, coral) on osteoporosis. In this study, ash weight of vertebral bones and Ca and P contents in the ashes were significantly decreased in animals given retinoic acid compare to control. These decreases in ash weight and mineral contents were significantly increased in rats treated with calcite. Also bone resorption markers were increased in animals with osteoporosis induced by retinoic acid. Calcite and Calcid-Denhk which have high contents of Ca significantly decreased the plasma levels of bone resorption markers.

**CONCLUSION**

Calcite compound improves bone strength of osteoporosis model of rats induced by retinoic acid through increasing bone minerals, osteoblast differentiation, and tissue proliferation.

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Effect of Yuna-4 on Alloxan-Induced Diabetes in Mice

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3Doctor and Professor of Health Sciences of the Health Sciences University of Mongolia

ABSTRACT

Yuna-4 was chosen in our research. The Influenced function of Yuna-4 was researched having irritated Pathogenetic diabetes in the testing animal by Rajagopal.K and Sasikala.K of 2008 method. Yuna-4 inhibited increases in serum aspartate aminotransferase (31.3 %), alanine aminotransferase (50.8 %), alkaline phosphatase (33.2 %), total cholesterol (48.3 %), triglyceride (21.7 %) and low density lipoprotein (21.2 %) in mice administered alloxan monohydrate (100 mg/kg). Serum levels of high density lipoprotein (43.9 %) was significantly increased by Yuna-4 (p<0.05).

Yuna-4 also decreased alloxan-induced increases in blood levels of glucose and uric acid. Body weight and water consumption of mice given alloxan monohydrate were also reduced by Yuna-4 treatment.

Key words: traditional medicine, Yuna-4, alloxan, diabetes

INTRODUCTION

Diabetes is one of the most pernicious diseases which were become epidemic and called as a silence plague throughout the world. Diabetes becomes the reason of death one person in every second and 3 million people in every year pursuant to the sickness mices and complication of diabetes. In developed countries, which are having higher development of industries, Diabetes accounted as the 4th reason of death. Venus complication of diabetes becomes main reason to loss work ability and death in early time. Also, the blood pressure is 2-3 time high, the reason of blind is 10 times more , kidney disease incident is 12-15 times more, and food depravation is 20 times more for the people who diseased by diabetes.

In accordance the research of the World Medical Organization, the number of people whom diseased by diabetes counted about 100 million already, because this number is increased by 5-7% every year. Those testimony shows that diabetes causes much danger, hazard, after affects and consequence to our human being.1 Therefore, we, researchers decided to study Yuna-4 between prescription for diabetes and retention, which is sued to use in Traditional Medicine. Our goal was to study effect of Yuna-4 on alloxan-induced 2 kinds of diabetes in testing animals.

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MATERIALS AND METHODS

The research was conducted based on the research laboratory of the Science and technology corporation of Traditional Mongolian Medicine of Ulaanbaatar / UASHUTUK/. There was involved 40 mices of Vistar breed who was grown up under same standard condition of vivart care and the scientific experiment was conducted in accordance with medical esthetics. Rajagopal.K and Sasikala K of 2008 method researched the influenced function of Yuna-4 having irritated Pathogenetic diabetes in the testing animal 2. 40 mices were chosen in scientific experiment was divided into 4 category; healthy, controlling, comparison and experimental. Alloxan monohydrate by dose of 100mg/kg vaccinated into the abdominal cavity of mices of controlling and experiment categories. Therefore blood glucose of the chosen animals increased to 200mg/dl after 72 hours and took under inspection. The inspection was conducted for 21 days and during this period, there was taken physiological solution to animals of inspection category, glibenclamide to comparison category, and 100mg/kg Yuna-4 to the animals of experimental category once a day for 21 days. Before and after experiment, there was checked weight of those animals, as well as tested blood glucose by “one touch glucometer” before experiment and after 72 hours as well as after 21 days. At the 22nd days, we tested uric acid, ASAT, ALAT SHF, total cholesterol, triglyceride, LDL and HDL of the 4 categories animals.
RESULTS

Table 1

Weight changes of animals, alloxan-induced diabetes and effect of Yuna-4

<table>
<thead>
<tr>
<th>Group</th>
<th>O day</th>
<th>21 day</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Healthy group</td>
<td>194.6±15.9</td>
<td>201.4±17.8</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>Controlling</td>
<td>188.4±12.9</td>
<td>174.5±10.4</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>Glibenclamide</td>
<td>190.1±10.5</td>
<td>195.9±12.7</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>Yuna-4</td>
<td>192.6±15.8</td>
<td>200.5±9.7</td>
<td>&gt;0.05</td>
</tr>
</tbody>
</table>

According to the table, weight of animals of group of Yuna-4 (3.84%) and health category (3.4%) increased little bet and accessed to the normal data. (P>0.05), but weight of animals of inspection category and decreased as 7.97% (P<0.05).

And we tested blood glucose of the animals before starting experiment and after 3 and 21 taking Alloxan.

Table 2

Blood glucose test of animals, alloxan-induced diabetes and effect of Yuna-4

<table>
<thead>
<tr>
<th>Group</th>
<th>O day</th>
<th>4 day</th>
<th>21 day</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Healthy group</td>
<td>47.6±2.9</td>
<td>51.9±3.1</td>
<td>50.7±2.4</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>Controlling</td>
<td>51.6±3.9</td>
<td>254±16.1</td>
<td>238.5±11.4</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>Glibenclamide</td>
<td>49.3±2.8</td>
<td>250.5±15.9</td>
<td>100.2±10.9</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>Yuna-4</td>
<td>48.4±2.4</td>
<td>286.5±23.7</td>
<td>176.3±17.6</td>
<td>&gt;0.05</td>
</tr>
</tbody>
</table>

Based on table record blood glucose of Yuna-4 group mices reduced by 38.5%, compare with blood glucose test after 4 day and after 21 days of experiment. (P<0.05) as well as blood glucose of animals of Yuna-4 category reduced by 26% or 1.4 times comparing to animals of controlling category.

Table 3

Changes of uric acid (mg/dl) and dose of drinking water (mg/day) of animals, alloxan-induced diabetes and effect of Yuna-4

<table>
<thead>
<tr>
<th>Group</th>
<th>Uric acid (mg/Dl)</th>
<th>Dose of drinking water (mg/day)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Healthy group</td>
<td>1.37±0.26</td>
<td>210.65±18.20</td>
</tr>
<tr>
<td>Controlling</td>
<td>3.7±0.42</td>
<td>352.42±20.25</td>
</tr>
<tr>
<td>Glibenclamide</td>
<td>1.81±0.11</td>
<td>250.1±19.7</td>
</tr>
<tr>
<td>Yuna-4</td>
<td>2.15±0.34</td>
<td>284.45±26.72</td>
</tr>
</tbody>
</table>

Apparantly table 16, mice’s uric acid and dose of drinking water of the animals of Yuna-4 category is less than animals of inspection category. (P<0.05)

Table 4

Biochemical test of animals, alloxan-induced diabetes and effect of Yuna-4

<table>
<thead>
<tr>
<th>Group</th>
<th>ASAT</th>
<th>ALAT</th>
<th>SHF</th>
<th>Cholesterol</th>
<th>Triglyceride</th>
<th>HDL</th>
<th>LDL</th>
</tr>
</thead>
<tbody>
<tr>
<td>Healthy</td>
<td>44.3±3.51</td>
<td>63.33±5.22</td>
<td>168.6±8.94</td>
<td>52.3±2.35</td>
<td>98.7±7.85</td>
<td>51.63±4.76</td>
<td>51.62±1.99</td>
</tr>
<tr>
<td>Controlling</td>
<td>85.1±6.35</td>
<td>122.67±10.98</td>
<td>287.2±15.64</td>
<td>173.9±9.84</td>
<td>168.3±10.65</td>
<td>41.12±2.58</td>
<td>80.56±7.91</td>
</tr>
<tr>
<td>Glibenclamide</td>
<td>65.7±7.78</td>
<td>88.9±9.65</td>
<td>234.8±12.45</td>
<td>65.6±6.34</td>
<td>137.1±11.45</td>
<td>63.6±5.67</td>
<td>67.5±6.23</td>
</tr>
<tr>
<td>Yuna-4</td>
<td>64.8±5.67</td>
<td>81.33±6.11</td>
<td>215.6±18.23</td>
<td>64.56±5.55</td>
<td>131.76±10.23</td>
<td>59.2±4.77</td>
<td>63.53±4.51</td>
</tr>
</tbody>
</table>

50
There was less data ASAT (31.3%), ALAT (50.8%) and SHF (33.2%) and total cholesterol (48.3%) and Triglyceride (21.7%), and LDL (21.2%) in blood serum of animals of Yuna-4 category that animals of controlling category but HDL was less by 43.9%. (p<0.05)

**DISCUSSION**

In accordance with the research of the effect of Yuna-4 to mice by alloxan-induced diabetes, there was shown that glucose in blood serum of mices of Yuna-4 category was decreased by 38.5% comparing 4th and 21st day tests. (P<0.05)

According to the research, Blood glucose of animals of Yuna-4 category reduced by 38.5% according to the comparison of blood glucose test after 4 day of experiment and after 21 days experiment. (P<0.05) as well as blood glucose of animals of Yuna-4 category reduced by 26% or 1.4 times comparing to animals of inspection category. But, there were no glucose changes in blood serum of animals of inspection category. (P<0.05)

The research of our result is being confirmed to the result of scientist of other countries. For example: the research result, which was conducted by of Choi J, Tantawy W.H, and Chaulan.A in 1991-2010. This research is shown that the total alkaloid and flavonoid including rutinozid, steroid and saponin of zelen zanguu improves insulin in beta cell of pancreas and improves insulin concentrate of blood serum. As well as, our research result is accepted to the research in 2011 of the scientist Raghavendra H.G, that the water extract of zelen zanguu reduces the glucose in blood serum of the mices with alloxan-induced diabetes.

**CONCLUSION**

The result our research shows that effect of Yuna-4 reduces glucose of blood serum by 38.5% during the diabetes. As well as, it effects to reduce uric acid, body weight and to normalize dose of drinking water and metabolism.

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Effect of Corticosteroids on Exacerbations of Chronic Obstructive Pulmonary Disease

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1State Third Central Hospital
2Health Sciences University of Mongolia School of Medicine
3State First Central Hospital

ABSTRACT
Chronic obstructive pulmonary disease (COPD) continues to be a major medical problem and is now the 6th leading cause of death in the world.16 Exacerbations are a common cause of mortality in COPD patients. The Global Initiative for COPD has recommended strategies for managing acute exacerbations of COPD.14 These strategies include beta-2 agonists, anticholinergics, the intravenous or oral administration of corticosteroids, methylxanthines,15 antibiotic therapy when indicated. There are limited data on the use of inhaled corticosteroids in the treatment of exacerbations of COPD, and its role in this setting must be more clearly defined. We studied the effect of high dose of inhaled corticosteroids compared to systemic corticosteroids in acute exacerbation of COPD.

Key words: Chronic obstructive pulmonary disease, exacerbation, inhaled corticosteroids, quality of life, St. George’s Respiratory Questionnaire.

INTRODUCTION
COPD is an increasingly prevalent disease and is the 6th leading cause of death in the world today.1 The depending Characteristics of COPD are inflammations and damage to the lung tissue, with irreversible airflow obstruction.2 Patients with COPD often present with acute exacerbations of increased symptoms that frequently require a change in their usual medications. Exacerbations are a common cause of mortality in COPD patients. Of COPD exacerbations approximately 50-70% are caused by bacterial infection. Other causes include viral infections and exposure to air pollutants. Approximately one third of exacerbations have no known cause3. Systemic and local inflammation is central to the pathophysiology of COPD. Several effective medications are used to treat COPD exacerbations. All patients should be placed on oxygen. The treatment of COPD exacerbations include anticholinergic agents such as ipratropium bromide, tiotropium bromide and short-acting β2-agonists such as albuterol, ventoline, methylxanthines, antibiotics are indicated if a bacterial infective cause is suspected. The Global Initiative for COPD recommend corticosteroid treatment for COPD exacerbations with methylprednisolone 40mg daily for 10 days.5 Short courses of systemic corticosteroids increase the time to subsequent exacerbation, decrease the rate of treatment failure, shorten hospital stays, and improve hypoxemia and forced expiratory volume in one second (FEV1).5,6,12, 21,22 Some patients have serious contraindications for systemic corticosteroids. In that case the role of inhaled corticosteroids must be more clearly defined. There are limited data on the use of inhaled corticosteroids in the treatment of acute exacerbations of COPD. We studied the effect of high dose of inhaled corticosteroids (fluticasone and budesonide) compared to systemic (parenteral) in acute exacerbations of COPD.

Our goal was to study effect of corticosteroids on exacerbation of COPD. Objectives were:
1. To evaluate the effect of systemic and inhaled corticosteroid therapy in acute exacerbation of COPD using clinical and functional criteries
2. To evaluate the quality of life in patients with acute exacerbation of COPD using international Sr. George’s Respiratory Questionnaire

MATERIALS AND METHODS
We compared the efficacy of inhaled corticosteroids (fluticasone and budesonide) with systemic corticosteroids (intravenous) in the treatment of acute exacerbation of COPD. In this study a total of 45 patients with acute exacerbation of COPD according to Global Initiative for COPD criterion were selected. Patients randomly divided into two groups. Patients in first group were used inhaled corticosteroids (Flixotide 1000 mcg/daily or Frenolyn 800-1200 mcg/daily). Patients in second group were administered parenteral corticosteroid (prednisolone 30-60mg/daily). Effect of corticosteroid therapy we evaluate
using clinical and functional criteries (symptoms, FEV₁, FVC). The quality of life we evaluated using international St. George’s Respiratory Questionnaire. The permission was obtained to carry out clinical study at the Medical Science Ethic Committee of the Health Science University of Mongolia.

RESULTS
The mean age of 45 participants were 59.6±7.9 years old. (21) 46.7% of them lived in Mongolian ger, (9) 20% - in heated houses, and (15) 33.3% in apartments. (28) 62.2% of patients were smokers. Cramer’s V-criteria shows that COPD depends on the age, living environment and smoking behavior. Average value and the student T-criteria show that the average smoking history was 30.3±10.0 years, smoked pack year was 14.1±12.7. After 10 days corticosteroid therapy the mean forced expiratory volume in one second (FEV₁) increased from 65.7% to 69.4% by 3.7 points (figure 1).

![Figure 1. The mean of forced expiratory volume in 1 second (FEV1) (per cent)](image1)

The mean forced vital capacity (FVC) raised from 80.5% to 88.3% by 7.8 points (figure2).

![Figure 2. The mean of forced vital capacity (FVC) (per cent)](image2)

FEV₁/FVC ratio increased with corticosteroid treatment from 65.1% to 69.5% by 4.4 points (figure 3).

![Figure 3. The mean of forced expiratory volume in 1 second and forced vital capacity ratio (FEV1/FVC)](image3)

The quality of life evaluated by St. George’s Respiratory Questionnaire that assess 1. Symptoms; 2. Activity; 3. Impact of COPD. The mean of symptoms with corticosteroids decreased from 50.8 to 27.2 (figure 4), the mean of activity- from 63.9 to 40.9 (figure 5), the mean of impact- from 45.2 to 22.7 (figure 6).

![Figure 4. The mean of symptoms of quality of life](image4)

![Figure 5. The mean of activity of quality of life](image5)

![Figure 6. The mean of impact of quality of life](image6)
DISCUSSION
Acute exacerbations in COPD are common and systemic corticosteroids play an important role in the management of these cases. Inhaled corticosteroids is being used in the acute attacks of asthma. But the role of inhaled steroids in acute exacerbation of COPD is not much studied in the literature. In this clinical study we evaluate the role of inhaled corticosteroids in the management of acute exacerbation of COPD. We found a few studies which had evaluated the role of nebulized corticosteroids and combined with long acting β₂ agonists (LAB) inhaled corticosteroids in exacerbations of COPD. 16,18-20,23 All these studies had used nebulized budesonide or combined with LAB inhaled corticosteroids compared with both either parental or oral steroids and standard bronchodilator therapy. All the studies had found the clinical efficacy of nebulized or inhaled corticosteroids to be of similar extend to that of either parental or oral steroids in acute exacerbations of COPD. Side effects of nebulized or inhaled corticosteroids was minimal and acceptable as compared to systemic steroids.

CONCLUSION
1. Both systemic and inhaled corticosteroid therapy in the management of acute exacerbations of COPD are effective
2. High dose of inhaled corticosteroids may be an alternative to parental prednisolone in the treatment of acute exacerbation of COPD
3. Systemic and inhaled corticosteroids increased the quality of life in patients with acute exacerbations of COPD

REFERENCES
Hepatitis B Virus Infection Among Population Over 40 years of Age in Ulaanbaatar City

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3Division of Virology, Department of Infection and Immunity, Jichi Medical University, Japan

ABSTRACT

In Mongolia the incidence of hepatitis was 14672 in 2011 which determine 34.3 percent of total number of communicable diseases. Of total viral infections 5.1 and 1.0 percent account for hepatitis B and C respectively. Mongolia has high prevalence of viral infections which end up to severe conditions such as liver cancer, liver cirrhosis the leading causes of population morbidity and mortality. This study aimed to evaluate the prevalence of hepatitis B virus (HBV) infection and its risk factors among Ulaanbaatar city population aged over 40 years. Population-based cross-sectional study is conducted and statistical analysis performed utilizing SPSS version 17. Study results indicated prevalence of hepatitis B viral infection estimated as 9.1 %. HBV infection occurred in 9.0% of population who had dental procedure, 10.5% and 9.5% who had injections at home and blood or blood products infusions respectively, 10.0% those who had tattoo. Study participants comprised of 37.6% male, 63.4% female. 21.8% aged 45-49 years, 21.0% aged 50-54 years. Prevalence of HBV infection was 9.1%. There is no difference of HBV infection by age category but there is statistically significant difference by sex. 12.2% and 7.2% of male and female were HBV positive. (p=0.0027)

Key words: HBV, prevalence, risk factors

INTRODUCTION

Worldwide, 2 billion people contacted hepatitis B virus globally there are about 400 million people became careers that lead to chronic liver pathologies particularly raising risks for hepatocellular carcinoma. It is estimated that more than 5% of those contacted hepatitis D virus are careers of HbsAg. If the number of HBsAg carriers is 300 million globally the number of persons with hepatitis D would reach 15 million. Hepatitis C virus disseminate as endemic in most of parts of the world. 3% of World population is infected with HCV which demonstrate that about 170 million people are at high risk of developing hepatocellular carcinoma. WHO classifies the prevalence of HCV among population according to presence of antibody to HCV as high (over 10%), intermediate (2.5-10%) and low (1-2.5%). In Mongolia the incidence of hepatitis was 14672 in 2011 which determine 34.3% of total number of communicable diseases. Of total viral infections 5.1% and 1.0% account for hepatitis B and C respectively. A number of scholars have investigated prevalence of hepatitis B and C in Mongolia and ways of its decreasing.

MATERIALS AND METHODS

This is a population-based cross sectional study. Multistage random sampling method was employed to select 1289 observations above 40 years old with equivalent sex and locality from Chingeltei, Khan-Uul, Songino-Khairkhan, Bayanzurkh districts. Approval obtained from Ethical Review Board. HBsAg was determined by ELISA (enzyme-linked immunosorbent assay) method in virology laboratory of Jichi Medical University, Japan. Descriptive and advanced statistical analysis was executed using SPSS statistical package version 17.0. After ensuring normal distribution of variables Pearson’s chi-square test performed to compare proportions, t-test to compare means, univariate and multivariate regression analysis. 2-tailed p-value of less than 0.05 was considered as statistically significant.
Table 1.

<table>
<thead>
<tr>
<th>HBV infection by sex, age</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>HBV (+)</td>
<td>HBV (-)</td>
<td>P value</td>
</tr>
<tr>
<td>n</td>
<td>%</td>
<td>n</td>
</tr>
<tr>
<td>Male</td>
<td>59</td>
<td>12.2</td>
</tr>
<tr>
<td>Female</td>
<td>58</td>
<td>7.2</td>
</tr>
<tr>
<td>40-44</td>
<td>23</td>
<td>10.7</td>
</tr>
<tr>
<td>45-49</td>
<td>30</td>
<td>10.7</td>
</tr>
<tr>
<td>50-54</td>
<td>26</td>
<td>9.6</td>
</tr>
<tr>
<td>55-59</td>
<td>20</td>
<td>10.0</td>
</tr>
<tr>
<td>above 60</td>
<td>16</td>
<td>5.3</td>
</tr>
</tbody>
</table>

RESULTS

Study participants comprised of 37.6% male, 63.4% female. 21.8% aged 45-49 years, 21.0% aged 50-54 years. Prevalence of HBV infection was 9.1%. There is no difference of HBV infection by age category but there is statistically significant difference by sex. 12.2% and 7.2% of male and female were HBV positive. (p=0.0027) The percentage of HBV infection is 10.7% in age group 40-44, 9.6% in age group 50-54, 10% in 55-59 and 5.3% in age above 60 but no statistically significant differences observed.

Table 2.

<table>
<thead>
<tr>
<th>Some risk factors of HBV infection</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Risk factors</td>
<td>Total</td>
<td>HBV (+)</td>
<td>HBV (-)</td>
</tr>
<tr>
<td></td>
<td>n x</td>
<td>n</td>
<td>%</td>
</tr>
<tr>
<td>Dental procedure</td>
<td>0.908</td>
<td>143</td>
<td>14</td>
</tr>
<tr>
<td>Yes</td>
<td>1143</td>
<td>103</td>
<td>9.0</td>
</tr>
<tr>
<td>No</td>
<td>268</td>
<td>26</td>
<td>9.7</td>
</tr>
<tr>
<td>Injection in hospital</td>
<td>0.746</td>
<td>1020</td>
<td>91</td>
</tr>
<tr>
<td>Yes</td>
<td>586</td>
<td>43</td>
<td>7.3</td>
</tr>
<tr>
<td>No</td>
<td>729</td>
<td>70</td>
<td>9.6</td>
</tr>
<tr>
<td>Injection at home</td>
<td>0.138</td>
<td>558</td>
<td>47</td>
</tr>
<tr>
<td>Yes</td>
<td>813</td>
<td>73</td>
<td>9.0</td>
</tr>
<tr>
<td>No</td>
<td>475</td>
<td>44</td>
<td>9.3</td>
</tr>
<tr>
<td>Surgery</td>
<td>0.528</td>
<td>729</td>
<td>70</td>
</tr>
<tr>
<td>Yes</td>
<td>558</td>
<td>47</td>
<td>8.4</td>
</tr>
<tr>
<td>No</td>
<td>813</td>
<td>73</td>
<td>9.0</td>
</tr>
<tr>
<td>Acupuncture</td>
<td>0.927</td>
<td>475</td>
<td>44</td>
</tr>
<tr>
<td>Yes</td>
<td>1118</td>
<td>101</td>
<td>9.0</td>
</tr>
<tr>
<td>No</td>
<td>169</td>
<td>16</td>
<td>9.5</td>
</tr>
<tr>
<td>Infusion of blood/blood products</td>
<td>0.906</td>
<td>1274</td>
<td>116</td>
</tr>
<tr>
<td>Yes</td>
<td>13</td>
<td>1</td>
<td>7.7</td>
</tr>
<tr>
<td>Dialysis</td>
<td>0.890</td>
<td>1047</td>
<td>93</td>
</tr>
<tr>
<td>Yes</td>
<td>240</td>
<td>24</td>
<td>10.0</td>
</tr>
</tbody>
</table>

Tattoo | 0.780 | 1274 | 116 | 9.1 | 1158 | 90.9 | 0.890 |

| Yes | 13 | 1 | 7.7 | 12 | 92.3 |
| No | 1047 | 93 | 8.9 | 954 | 91.1 |
| No | 240 | 24 | 10.0 | 216 | 90.0 |
Majority of participants (approximately 90%) who had risk factors such as dental procedure, injections, surgery, tattoo, acupuncture, dialysis, blood/ blood products infusion were HBV negative whereas only 10% had HBV positive results. Comparison of each risk factor among HBV positive and negative groups we didn’t observe any statistically significant differences. For instance HBV infection occurred in 9.0% of population who had dental procedure, 10.5% and 9.5% who had injections at home and blood or blood products infusions respectively, 10.0% those who had tattoo.

**DISCUSSION**

According to the study by Oyunbileg J. et al. in 1992, overall prevalence of HBsAg was 6.8% and it increased with age group. 7.1% in age 0-9 years, 9.3%, in 21-30 years, 15.7% in 31-40 years, 15% in age group above 50 years. The latter goes in line with our study. (15% in age group above 50). Khurelbaatar N. Et al. found (2001) 29.7-31.6% HBsAg positive individuals among 167 cohorts aged 19-53 years. In study conducted by Oyunsuren Ts. et al. prevalence of HBsAg was 23.1%, 15.7% among healthy population. Prevalence of HBsAg among male was 16.7% found in study by Nyamdavaa P. et al. which is similar to our finding. In our study prevalence of HasAg among male was 12.2% which has statistically significant difference comparing to female (p=0.0027). In cross sectional study by Dagvadorj Ya et al. in 2001-2003 among population aged 0-80 years prevalence of HasAg found to be 11.8%. Our data found the prevalence of positive HasAg being 9.1% which display decreasing tendency of HasAg prevalence.

**CONCLUSION**

HBV infection has statistically significant difference in gender which does not apply to age groups. Although some risk factors such as tattooing, infusion of blood/ blood products, having injections at home were not statistically significant but could be potential risk factors.

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Human Brucellosis Among Rural Population of Mongolia

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ABSTRACT

Brucellosis is a public health problem in Mongolia. The aim of our study was to estimate a representative seroprevalence of Brucella spp and to determine risk factors for brucellosis seropositivity among rural people of selected aimags of Khangai region. A cross sectional study conducted in 3 aimags (provinces) of Mongolia. A total of 1191 randomly selected rural people aged 4-84 years were enrolled in the study. All study participants were interviewed using a questionnaire to obtain their brucellosis history and most likely risk factors. Blood samples were drawn to determine the brucellosis seroprevalence. The overall apparent seroprevalence of Brucella spp among rural people in selected 3 aimags was estimated at 17.0 percent [95 percent CI 15.7-18.2] and true seroprevalence 16.4 percent [95 percent CI 15.0-17.7]. Assistance in livestock obstetric work without personal protective equipments (OR=1.9, p=0.001), contact with aborted fetus and retained placenta of animals without personal protective equipments (OR=1.5, p=0.02) were significantly associated with seropositivity and risks for brucellosis. Our study confirms that human brucellosis seroprevalence among rural people of Khangai region is high. Many risk factors have had among people who participated in the study. Education and regular training for rural people are required. Also important issues are livestock mass vaccination and cooperation between the medical and veterinary sectors to fight with brucellosis.

Key words: seroprevalence, human brucellosis, three aimags, Mongolia

INTRODUCTION

Brucelloses are either acute or chronic infections of animals and humans caused by gram-negative bacteria of the genus Brucella, which may infect one or several organs. The disease have also been called Bang’s disease and Malta fever (Hartmut Krauss et al. 2003). Brucellosis is an important human disease in many part of the world, especially in the Mediterranean countries of Europe, north and east Africa, the Middle East, south and central Asia and Central and South America (Corbel MJ 2006).

Seven republics of the former Soviet Union are included in the 25 countries with the highest incidence of the disease worldwide, while another country of this region, Mongolia ranks second (Georgios Pappas et al. 2006). After the transition from socialism to the market economy in 1990, human brucellosis incidence rapidly increased and has re-emerged in Mongolia. The evolution from a socialist state to free market economy resulted in the loss of strict livestock control. Furthermore, the transition of the health system during this period precluded early recognition of the disease, or interventions for the emerging trends in human beings and animals (Georgios Pappas et al. 2006).

Annual reporting human brucellosis incidence per 10 000 population sharply increased over the previous 20 years (Report of the National Center for Communicable Diseases, Mongolia, 2012). The aim of our study was to estimate the seroprevalence of Brucella spp and to determine risk factors for brucellosis seropositivity among rural people of selected aimags of Khangai region.

MATERIALS AND METHODS

Study design and population
A cross sectional study was conducted between November and December, 2011 in 3 aimags such as Arkhangai, Khuvsgul and Selenge. Aimag (province), soum (village), bagh (smallest administrative unit), families and people were selected by simple randomization. We were included in the study 4 soums per aimag and 2 baghs per soum, 10 families per bagh and 4-5 people per selected family for blood sampling and filling an individual questionnaire. The sample size was selected based on the feasibility and the available budget.

We were enrolled in the study a total of 1191 randomly selected rural people aged 4-84 years (median age 37±6.8) including 843(70.8 percent) herders, 32(2.7 percent) office workers, 73(6.1 percent) school pupils, 43(3.6 percent) workers, 25(2.1 percent) retired, 18(1.5 percent) entrepreneur, 13(1.1 percent) unemployed, 7(0.6 percent) preschool children under 6 years, 6(0.5 percent)
veterinarians, 2(0.2 percent) students and 129(10.8 percent) other residents.

Sample size calculation
The sample size calculation assumes a human brucellosis seroprevalence among rural people of 27 percent according to the seroprevalence study conducted in Zavkhan and Sukhbaatar aimags, 2010.

The sample size calculated the relevant formula for a 95 percent confidence interval is:

\[ n = \frac{Z^2_{\alpha/2} \times (1 - p_{exp})}{d^2} \times \frac{0.27(1-0.27)}{0.05^2} = 302.7 \]

Where: \( n \) - required sample size; \( Z_{\alpha/2} \) – confidence interval; \( p \) - expected sero-prevalence (by percent); \( d^2 \) - desired absolute precision

The required sample size multiplied by design effect which is appropriate for a human brucellosis is:

\[ n \times D = 302.7 \times 1.2 = 363.2 \sim 363 \]

Where: \( n \) - sample size; \( D \)-design effect

We predicted that 5 percent of all selected people do not participate in the study due to refuse to participate, migration and natural disaster because the study conducted in winter time. Therefore the required sample size was increased by 5 percent is:

\[ n + 5\% = 363 + 18.15 = 381.15 \sim 381 \]

\( n = 381 \) or required number of study participants per aimag

The total number of study participants is:

\[ n \times 3 \text{ aimags} = 381 \times 3 = 1143 \]

Data collection
Study questionnaire
All study participants were interviewed using a questionnaire to obtain brucellosis history and risk factors information.

Blood sample collection and handling
The study was conducted by the trained teams of soum physicians, nurses and laboratory staffs for blood sampling and data collection. Venous blood was taken with 5 ml Vacutainer® tubes. The blood sample was centrifuged for 5 minutes, separated 1.5 ml of serum, kept at cool box a day and collected in soum hospital until finish the study.

Then the blood samples were transported to the provincial laboratories to store them cooled before cooled shipment to the serological laboratory of the National Center for Communicable Diseases of Ulaanbaatar, where they were tested for brucellosis.

Serological tests
Sera was tested with the Rose Bengal Test from the Tulip Diagnostic Ltd, India. Positive sera were re-tested with modified Rose Bengal test diluted from \( \frac{1}{2} \) to \( \frac{1}{32} \) and indirect enzyme linked immunoassay “Nova Lisa”, Germany for determination of IgG class antibodies against Brucella spp.

Statistical analysis
The study data was double entered in the Access-2007 and compared in Epi-info 3.5 and analyzed using STATA 10. Univariate analyses were first done using Pearson chi-square or Fisher’s exact tests for categorical explanatory variables. We have done univariate logistic regression on the binary serological outcome with risk factors. Adjusted odd ratios and confidence interval were calculated for explanatory variables. A p value of \( \leq 0.05 \) was defined as statistically significant.

Apparent seroprevalence of Brucella spp estimated with positive result of Rose Bengal test and the true prevalence by the Rogan-Gladen estimator is:

\[ (P-RB) = (AP + Sp - 1)/(Se + Sp - 1) \]

where: \( AP \) is the apparent prevalence, \( Se \) is the sensitivity and \( Sp \) is the specificity of Rose Bengal test

True prevalence is:

\[ TP = AP - (P-RB) \]

We considered that \( Se = 0.99 \) and \( Sp \) of >0.90 of the Rose Bengal test because the result certified from the European Brucellosis Reference Center.

RESULTS
Estimation of apparent and true seroprevalences:
Overall apparent seroprevalence of Brucella spp was estimated at 17.0 percent [95 percent CI 15.7-18.2] and ranged between the 3 aimags from 15.8 percent to 18.9 percent. Apparent seroprevalences of Brucella spp in selected aimags are shown in Table 1. The overall true seroprevalences in 3 aimags estimated 16.4 percent [95 percent CI 15.0-17.7].
### Table 1.

**Apparent sero-prevalence of Brucella spp in selected 3 aimags**

<table>
<thead>
<tr>
<th>Name of aimag</th>
<th>Number of soum</th>
<th>Number of people</th>
<th>Number of positive</th>
<th>% of positivity</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arkhangai</td>
<td>4</td>
<td>400</td>
<td>63</td>
<td>15.8</td>
<td>13.9 - 17.2</td>
</tr>
<tr>
<td>Khuvsugul</td>
<td>4</td>
<td>400</td>
<td>66</td>
<td>16.5</td>
<td>12.9 - 20.1</td>
</tr>
<tr>
<td>Selenge</td>
<td>4</td>
<td>391</td>
<td>74</td>
<td>18.9</td>
<td>15.0 - 22.7</td>
</tr>
<tr>
<td>Total</td>
<td>12</td>
<td>1191</td>
<td>203</td>
<td>17.0</td>
<td>15.7-18.2</td>
</tr>
</tbody>
</table>

In Table 2, the seroprevalences of Brucella spp are presented for every soum included in the study. The highest seroprevalence of Brucella spp is found in Tsagaannuur soum of Selenge aimag (31.0 percent) and the lowest in Jargalant soum of Arkhangai aimag (6.0 percent).

### Table 2.

**Apparent sero-prevalence of Brucella spp in selected soums of the aimags**

<table>
<thead>
<tr>
<th>Aimag name</th>
<th>Soum name</th>
<th>n</th>
<th>Positive</th>
<th>% of seropositivity</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arkhangai</td>
<td>Erdenemandal</td>
<td>100</td>
<td>23</td>
<td>23.0</td>
<td>19.4-26.6</td>
</tr>
<tr>
<td></td>
<td>Jargalant</td>
<td>100</td>
<td>6</td>
<td>6.0</td>
<td>1.8-13.7</td>
</tr>
<tr>
<td></td>
<td>Khangai</td>
<td>100</td>
<td>17</td>
<td>17.0</td>
<td>12.7-21.3</td>
</tr>
<tr>
<td></td>
<td>Undur-Ulaan</td>
<td>100</td>
<td>17</td>
<td>17.0</td>
<td>12.8-21.5</td>
</tr>
<tr>
<td></td>
<td>Ihk-Uul</td>
<td>100</td>
<td>16</td>
<td>16.0</td>
<td>11.5-20.5</td>
</tr>
<tr>
<td>Khuvsugul</td>
<td>Khankh</td>
<td>100</td>
<td>18</td>
<td>18.0</td>
<td>13.8-22.1</td>
</tr>
<tr>
<td></td>
<td>Ulaan-Uul</td>
<td>100</td>
<td>19</td>
<td>19.0</td>
<td>14.9-23.0</td>
</tr>
<tr>
<td></td>
<td>Bayanzurkh</td>
<td>100</td>
<td>13</td>
<td>13.0</td>
<td>7.9-18.1</td>
</tr>
<tr>
<td></td>
<td>Sant</td>
<td>100</td>
<td>19</td>
<td>19.0</td>
<td>14.9-23.0</td>
</tr>
<tr>
<td>Selenge</td>
<td>Mandal</td>
<td>91</td>
<td>12</td>
<td>13.2</td>
<td>7.8-18.5</td>
</tr>
<tr>
<td></td>
<td>Tsagaannuur</td>
<td>100</td>
<td>31</td>
<td>31.0</td>
<td>28.0-33.9</td>
</tr>
<tr>
<td></td>
<td>Bayangol</td>
<td>100</td>
<td>12</td>
<td>12.0</td>
<td>6.7-17.3</td>
</tr>
</tbody>
</table>

Seroprevalences of Brucella spp in selected age groups ranged from 4 percent to 20.6 percent. Seroprevalence in women was more than 2 percent compared with men. The highest seroprevalence among veterinarians observed 66.7 percent [95 percent CI 44.8-88.5] and the lowest among school pupils 6.8 percent [95 percent CI 1.5-15.1] (Table 3).

### Table 3.

**Seroprevalences of Brucella spp by age group, gender and occupation**

<table>
<thead>
<tr>
<th>Age group</th>
<th>n</th>
<th>positive</th>
<th>% of positivity</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>0-9 yrs</td>
<td>25</td>
<td>1</td>
<td>4.0</td>
<td>2.7-31.7</td>
</tr>
<tr>
<td>10-14 yrs</td>
<td>32</td>
<td>4</td>
<td>12.5</td>
<td>3.1-21.8</td>
</tr>
<tr>
<td>15-19 yrs</td>
<td>46</td>
<td>3</td>
<td>6.5</td>
<td>4.9-17.9</td>
</tr>
<tr>
<td>20-44 yrs</td>
<td>724</td>
<td>120</td>
<td>16.6</td>
<td>14.9-18.3</td>
</tr>
<tr>
<td>45 and above</td>
<td>364</td>
<td>75</td>
<td>20.6</td>
<td>18.5-22.6</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Gender</th>
<th>n</th>
<th>positive</th>
<th>% of positivity</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Women</td>
<td>448</td>
<td>83</td>
<td>18.5</td>
<td>16.5-20.4</td>
</tr>
<tr>
<td>Men</td>
<td>743</td>
<td>120</td>
<td>16.5</td>
<td>14.8-18.1</td>
</tr>
</tbody>
</table>
Analysis of risk factors for brucellosis:

Univariate analysis of some risk factors for brucellosis are shown in Table 4. Assistance in livestock obstetric work without personal protective equipments (OR=1.9, p=0.001), contact with aborted fetus and retained placenta of animals without personal protective equipments (OR=1.5, p=0.02) were significantly associated with seropositivity and risks for brucellosis.

Table 4. Univariate analysis of some risk factors for brucellosis

<table>
<thead>
<tr>
<th>Risk Factor</th>
<th>n</th>
<th>Positive</th>
<th>% of positivity</th>
<th>p value</th>
<th>Adjusted OR</th>
<th>95%CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Assistance in obstetric work of animals without PPE*</td>
<td>395</td>
<td>94</td>
<td>23.8</td>
<td>0.001</td>
<td>1.9</td>
<td>1.4-2.6</td>
</tr>
<tr>
<td>Contact with aborted fetus and retained placenta of animals without PPE*</td>
<td>227</td>
<td>50</td>
<td>22.0</td>
<td>0.02</td>
<td>1.5</td>
<td>1.0-2.1</td>
</tr>
<tr>
<td>Consumption of raw milk</td>
<td>194</td>
<td>29</td>
<td>14.9</td>
<td>0.397</td>
<td>0.8</td>
<td>0.5-1.2</td>
</tr>
<tr>
<td>Consumption of raw liver</td>
<td>3</td>
<td>1</td>
<td>33.3</td>
<td>0.467</td>
<td>0.24</td>
<td>0.22-0.27</td>
</tr>
<tr>
<td>Consumption of half done liver</td>
<td>405</td>
<td>74</td>
<td>18.3</td>
<td>0.419</td>
<td>1.1</td>
<td>0.83-1.5</td>
</tr>
<tr>
<td>Consumption of animal fresh blood</td>
<td>77</td>
<td>14</td>
<td>18.2</td>
<td>0.784</td>
<td>1.0</td>
<td>0.6-1.9</td>
</tr>
</tbody>
</table>

* Personal protective equipment, OR=odds ratio, CI=confidence interval

Consumptions of half done liver and raw milk were very common in all age groups. Risk factors such as assistance in livestock obstetric work and contact with aborted fetus and retained placenta of animals without personal protective equipments were the highest in 20-44 years of age (Figure 1).

In Figure 2, all studied risk factors have had either men or women. But among men the risk factors were predominantly compared with women. Assistance in obstetric work of livestock and direct contact with aborted fetus and retained placenta of livestock without personal protective equipments were 39.7 percent [95 percent CI 37.8-41.5] and 23.2 percent [95 percent CI 19.6-26.7] in men, respectively.

**Figure 1. Risk factors for brucellosis by selected age group and percent**

**Figure 2. Risk factors for human brucellosis by gender and percentage**
DISCUSSION
This study was designed as a cross sectional assessment to estimate both the seroprevalence of Brucella spp and determine risk factors for brucellosis among rural people. The study is a part of the baseline to estimate seroprevalence of Brucella spp in humans before the livestock mass vaccination campaign and will be used for brucellosis surveillance to monitor efficacy of the vaccination. As the result of our study overall apparent seroprevalence of Brucella spp among rural people who participated from 3 aimags of Khangai region was estimated at 17.0%. Since 2010 the National Center for Communicable Diseases has been conducting seroprevalence study of Brucella spp among rural people in several aimags. Compared with other aimags of Western and Govi regions in 3 aimags of Khangai region the seroprevalence was similar but lower than about 11% from Eastern Sukhbaatar aimag. This likely reflect the human brucellosis prevalence is lower in forest and sub forest areas. Some author noted that in steppe area of Mongolia brucellosis spreads widely and forest area is lower (Tserendash Ch, 1960). On the other hand the result of the study shows brucellosis is an endemic and widespread in our country. In Mongolia have conducted several studies to estimate the seroprevalence of Brucella spp but the studies were not comparable for time and study design (Dashdavaa J 1969, Baldandorj Ts 1972, Gombosuren T 1982, Dagvadorj Ya 2001).

In selected all age groups and occupations with exception of students were prevalent for brucellosis. It is may show that rural people of all age groups and occupations including children have risks for brucellosis related to participation in lambing/calving works and consumption of animal raw and undercooked products.

For some risk factors for brucellosis such as assistance in obstetric work of livestock (p=0.001) as well as directly contact with aborted fetus and retained placenta of livestock without personal protective equipments (p=0.02) were important risk factors for brucellosis and the odds were 1.5-1.9 times higher among the study participants. The results did concord with the case control and cross sectional studies conducted by Kozukeev et al., and Omer et al., in Kyrgyzstan and Eritrea, respectively. The risk factors can be an indicator of big and small ruminant brucellosis infection in our country. Because from 23.7 percent to 24.1 percent of people assisting in obstetric work of goat, sheep and cattle were seropositive.

In age groups of 20-44 years, assistance in animals obstetric work and direct contact with aborted fetus and retained placenta without personal protective clothes (p=0.0001) and seroprevalences of Brucella spp were the highest. It is may indicate that people of the age group predominantly participate in animal herding and lambing/calving works.

We found no the association between brucellosis seropositivity and risk factors such as consumption of raw milk, animal fresh blood, raw and half done liver among rural people of the aimags.

CONCLUSION
Our study confirms that human brucellosis seroprevalence among rural people of Khangai region is high. Many risk factors related to assistance in livestock obstetric work and contact with aborted fetus and retained placenta of animals without personal protective equipments and consumption of raw and undercooked animal products have had among people who participated in the study. Education and regular training for rural people are required. Also important issue is mass vaccination of cattle and small ruminants to stop the brucellosis transmission between livestock and from livestock to human. Cooperation between the medical and veterinary sectors in view of a One Health approach is needed to fight with brucellosis.

ACKNOWLEDGEMENTS
We would like to thank Health departments of the Arkhangai, Khuvsgul, Selenge aimags’ and soums’ physicians and laboratory personnel for helping data collection. We thank the Ministry of Health for funding of the study and the Institute of Veterinary Medicine for assisting, especially Professor Erdenebaatar Janchivdorj.

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Impact of Accreditation
On Institutional Development

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1 HSUM, School of Health Technology
2 HSUM, School of Medicine
3 HSUM, Division of educational policy and management

ABSTRACT
Higher education accreditation is a type of quality assurance process which identifies institutional development policies and implementation of procedures are evaluated by an external body. Inspection of external evaluation assesses differences between current level of development and future perspectives.

Also to update forward planning of organization and to determine implementation for institutional positive changes.

The School of Health Technology was evaluated by the Asia-Pacific Accreditation and Certification Commission (APACC) which has been accredited and awarded our school.

The aim of this study was to evaluate the impact of appreciative inquiry based accreditation on quality assurance and institutional positive changes.

This study was conducted in a descriptive (retrospective) and a cross-sectional study and involved total of 72 respondents (included faculty staff and officers employed since 2007) who were divided into 4 groups by the duty of involvement for accreditation process at the School of Health technology. Statistical analysis was performed by using SPSS 17 version.

We have concluded “The Accreditation” is a voluntary process that positively impacted for the institutional change. Individual development was changed by the participating on the accreditation review process. The level of evaluation and perceiving of individual participant was poor associated with enrollment of accreditation process.

Keyword: Accreditation process, institutional change, Impact of accreditation

INTRODUCTION
Accreditation of higher educational institution is important process which to identify the development of institutional policies and implementation procedures to developing.1,2 Accreditation has important responsibility for the global educational development throughout the world.3 The accreditation requirements, standards and criteria would be more similar in all of country that individual occupation, education could be chance increased convertibility.4 The accreditation process has a lot of benefits such as faculty and staff evaluate current level of development and contribute to planning of institution, more training and learning.5 Inspection of external body evaluation assesses differences between current level of development and planned future perspectives. And also to update strategic planning of organization and determine implementation of institutional change.6 School of Health Technology was evaluated by the Asia-Pacific Accreditation and Certification Commission (APACC) has been accredited and awarded our school. The aim of this study was evaluating the impact of accreditation programs on the quality assurance and institutional change. Our goal was to evaluate the impact of appreciative inquiry based accreditation on the institutional change. Objectives were to study of impact appreciative inquiry –based accreditation on institutional change and to identify the parties which have been changed mostly during the accreditation review process.

MATERIALS AND METHODS
This study was conducted in a descriptive (retrospective) and a cross-sectional study and involved total of 72 respondents (included faculty staff and officers employed since 2007) who were divided into 4 groups by the duty of involvement for accreditation process at the School of Health technology.

1. This group did not participate in the accreditation review process /n=4/
2. This group participate in the some action of accreditation review process /n=37/
3. This group participate during of accreditation review process \( n=28 \)
4. This group participate to leading responsibility in the accreditation review process \( n=3 \)

We used the total of 25 items. This form was filled 3 questions on general information of respondents, there are 10 questions focused about the impact of appreciative inquiry – based accreditation process on the institutional change, 10 questions were focused on impact of 10 questions accreditation process on the individual change and remained 2 were optional questions.

Statistical analysis was performed by using SPSS 17 version.

**RESULTS**

The mean years of work experiences were 7.8\( \pm \)6.3 in this study. Team leader constituted 3 (4.2%), team member 27 (37.5%), teachers 36 (50.0) remaining 6 (8.3%) were officers to duty of participants in the accreditation process. The position was 26 (40.62%) of them were junior lecturer and lecturer 24 (37.5%), senior lecturer 10 (15.62%), associate professor 4 (6.25%).

5.6% of respondents were given answer “I could not participate in the accreditation” 51.4% of them “I had been participated in the some activities of accreditation”, 38.9% of them “I had been participated in the all activities of accreditation” 4.2% of them had answered “I had been participated to leading duty in the accreditation process” for question “Can you identify your performance of accreditation enrollment”.

In this question “Who was initiated to accredit for your institute” 38 /53.5% of respondents answered “Team of administration planned this accreditation” and 23 (32.4%) of them answered “Dean of school initiated and appealed for attend this accreditation”.

This study didn’t find significantly differences between groups of personal role and enrollment in the accreditation review process. Most of respondents answered that team of administration had planned and appealed to attending for this accreditation process not regarding other faculty staff’s involvement and performances in the activities.

10 group questions were related to impact of appreciative inquiry based accreditation on institutional change in this study. The most of answers were good and very good changed development institution (Figure 1).

58.6 – 68.4% of respondents answered “good changed” for question “Our faculty and staff would be innovating and seeking for new idea related to accreditation process” “Good changed” answer dominantly was “average changed” and “very good changed” approximately were answered to other questions. 0-8.6% of respondents answered “not changed and poor changed” above questions. We studied the impact of accreditation on institutional change how to depends on the individual change and involvement on the accreditation process. This study showed first group (not participated) answered “average changed” for 5 questions and “poor changed” for 3 questions. This group was compared with total respondents in this study which the impact of accreditation on the organization was concluded “poor changed”. But we are suspecting this result may related to few respondents.
We have not found statistical true value that total respondents were compared with second group (participating in the some activities of accreditation review process /n=37/) 66,7% of third group evaluated “good changed” for 2 questions and “very good changed” for 5 questions 33,3% of them answered “poor”, “average” for other answers. 13 /1,9%/ of respondents evaluated that accreditation reviews process was not effect of change to the organization development. According to this study, If individual duty increase, they would be more positively evaluate the impact of appreciative inquiry accreditation on institutional change (rxy=0.16).

**DISCUSSION**

This study /the scientific at Lincoln University in Nebraska/ explored the perceptions of change resulting from Appreciative Inquiry applied to accreditation and related institutional effectiveness activities. The result of their study described perceptions of positive changes same as our survey. At the result of survey “The power of accreditation” which is from Harvey described that the accreditation is not just evaluation process of external body who determines the levels it is an appreciative inquiry accreditation activities related to perception of change for individuals and institutions. Also our study shows as a same positive result.

**CONCLUSIONS**

1. “The Accreditation” is a voluntary process that positively impact for the institutional changes.
2. Individual change was changed by the participating on the accreditation review process. The level of evaluation and perceiving of individual participant was poor associated with enrollment of accreditation process.

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4. Development documentation - School of Health technology, UB 2012 year.
5. Education accreditation in the USA, UB 2011 year.
Microsurgical Treatment of Anterior Circulation Aneurysms

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2Neurological department, State Central Hospital

ABSTRACT

A cerebral aneurysm has been surgically treated since the early twentieth century. Since then, numerous new surgical methods and technologies have been developed in neurosurgical practice to improve treatment outcomes in neurosurgery. The pterional or the fronto-temporo-sphenoidal approach, developed by Yasargil, has been widely used in practice and became the standard treatment for anterior circulation aneurysms. In Mongolia, aneurysms have been surgically treated since early 1980s. Particularly, the bitemporal approach has been used since 2000, the pterional approach has been used since 2007, and the simplified pterional approach has been adopted since 2010 as the main treatment options for anterior circulation aneurysms in Mongolian neurosurgical practice. The keyhole approach, cultivated by German neurosurgeon A. Perneczky, was introduced to Mongolian neurosurgical practice in 2011 as another treatment option for the anterior circulation aneurysms. We would like to note that the pterional approach was successfully used for the first time in removing brain tumor in Mongolia.

In this study, we introduce a new keyhole surgical technique with four small burr holes that is based on the method of German neurosurgeon A. Perneczky for treating anterior circulation aneurysms. We also report the impact of this new keyhole surgical technique on the rate of surgically related complications based on the data collected during this study.

Keywords: Cerebral aneurysms; Keyhole approach; Minimally invasive; Supraorbital; Microneurosurgery; Microscope-assisted; dissection.

INTRODUCTION

Microsurgical methods and technologies have greatly improved effectiveness of neurosurgery and surgery in general by inflicting far less damage to the surrounding areas compared to the traditional surgical techniques. The new approach that we are introducing is one example of this kind of advanced microsurgery.

According to statistics on brain surgeries conducted in our country, anterior circulation aneurysms have consistently been on the rise in the past decade. For instance, between 2000 and 2002, only 59 patients with anterior circulation aneurysms were treated in the neurosurgical department of Third State Central Hospital, while 412 cases were diagnosed and treated between 2009 and 2012. On the other hand, when arterial aneurysms rupture, the mortality rate is extremely high (60%-87%) and the traditional surgical techniques are relatively less effective due to their long durations and more intrusive nature and result in high death rate in two weeks following the surgery (16%). Therefore, it was absolutely critical to improve the existing neurological methods and develop more effective and advanced surgical techniques for treating anterior circulation aneurysms in Mongolia. The main goal of this research work is to develop a new and more effective open surgical method for anterior circulation aneurysms and apply this new technique to current neurosurgical practice to decrease the mortality rate and the related complications that occur both post and during brain surgery. Goals are:

1. Using our new microsurgical technique conduct brain surgeries on patients with anterior circulation aneurysms and compare its effectiveness with the traditional surgical techniques.
2. Develop a complete step-by-step methodology for carrying out this new surgical technique and determine the effectiveness of the new method based on the statistical significance of the collected data.

MATERIALS AND METHODS

1. This study is based on the results of 235 neurosurgeries performed by Dr. Enkhbold Dorjgotov (the researcher of this study) on the patients with anterior circulation aneurysms between 2011 and 2012 in the neurosurgical department of Third State Central Hospital. In order to compare the effectiveness of the
new technique with the traditional techniques, the researcher selected 103 surgeries that utilized the new neurosurgical technique as an experimental group and another 103 surgeries that utilized the traditional methods as a control group for this comparative study. This comparative study does not include 54 brain surgeries that utilized other surgical methods.

2. In this research study, the researcher developed a new surgical method for anterior circulation aneurysms that employs pterional approaches and key-hole approaches and determined the effectiveness of the new method over the traditional methods with statistical significance.

We conducted 103 surgeries on the patients with anterior circulation aneurysms that utilized the key-hole approaches that utilized 3 main approaches: paranasal approach, subfrontal approach, and subfrontal approach. This new technique is improved and refined key-hole approach that was originally designed by German neurosurgeon A. Perneczky. In this technique, four burr holes of 0.5cm size are drilled into the skull through key-hole approaches and then quadrangle shaped bone flap is cut with 45° angle. Even though it prolongs the duration of the surgery by about 20 minutes, putting the removed bone flap back becomes easier, produces more stable fitting without the need for microplates that cost 500-600 thousand tugrugs, and results in cosmetically better appearance. To increase the size of the opening for the surgery, it was enough to drill off the inner edge of pterion. This allows the bone to sit firmly into the surrounding 3 walls and heal in that position. There was not a single case where either the patient had an infection in the area of surgery or the bone flap was displaced after the surgical operations. Both pre and post-surgical conditions of the patients were evaluated using the SAH grading scale of the World Federation of Neurosurgical Societies (WFNS) and the Hunt and Hess scale (see Table 1).

<table>
<thead>
<tr>
<th>World Federation of Neurosurgical Societies (WFNS) SAH grading scale and Hunt-Hess scale</th>
</tr>
</thead>
<tbody>
<tr>
<td>World Federation of Neurosurgical Societies, SAH grading scale</td>
</tr>
<tr>
<td>GCS score</td>
</tr>
<tr>
<td>----------</td>
</tr>
<tr>
<td>I</td>
</tr>
<tr>
<td>II</td>
</tr>
<tr>
<td>III</td>
</tr>
<tr>
<td>IY</td>
</tr>
<tr>
<td>Y</td>
</tr>
</tbody>
</table>

From the total of 235 patients, who were diagnosed with anterior circulation aneurysms, 198 (84.2%) of them were classified as having the SAH grade level-I of the WFNS of which 126 (91.9%) were patients who were treated with the new surgical method. There were only 2 patients who had the SAH grade level-IV and no patient with the SAH grade level-V was present in the selected period. We believe that this is attributed to the patients’ choice whether to take a significant risk at that SAH grade level. Since the WFNS SAH grading is based on the Glasgow Coma Scale, we did not apply both scales separately in the study. To evaluate the patients’ mental state, we employed the mini-mental state examination test (see Table 2).

<table>
<thead>
<tr>
<th>The mini-mental state examination test</th>
</tr>
</thead>
<tbody>
<tr>
<td>Test</td>
</tr>
<tr>
<td>------</td>
</tr>
<tr>
<td>score</td>
</tr>
</tbody>
</table>

From the total of 235 patients, who were diagnosed with anterior circulation aneurysms, 198 (84.2%) of them were classified as having the SAH grade level-I of the WFNS of which 126 (91.9%) were patients who were treated with the new surgical method. There were only 2 patients who had the SAH grade level-IV and no patient with the SAH grade level-V was present in the selected period. We believe that this is attributed to the patients’ choice whether to take a significant risk at that SAH grade level. Since the WFNS SAH grading is based on the Glasgow Coma Scale, we did not apply both scales separately in the study. To evaluate the patients’ mental state, we employed the mini-mental state examination test (see Table 2).
Hemiparesis after surgery was tested by checking muscle strength, pathological reflexes, and hyper reflexes, which are considered as internationally accepted method. Temporal atrophy was evaluated by comparing the size and strength of the temporal muscles of the healthy side with the effected side.

**The new refined key-hole approach**

In the preparation phase of the new refined key-hole approach, lumbar drainage is set under general anesthesia. The patient is positioned supine while the head is elevated above the thorax to facilitate venous drainage and retroflexed from $5^\circ$ to $15^\circ$ degrees depending on the aneurysm locations. During the last positioning step, the head is rotated to the contralateral side about 10 to 15 degrees until the eyebrow of the targeted surgical side becomes the highest point. The initial skin incision of 5-6cm is made within the eyebrow. To achieve better cosmetic results, the incision must follow the orbital rim. Skin flap is temporarily retracted with two stitches very gently, and then frontal and temporal muscles are cut and carefully dissected in a frontal direction. After local hemostasis, 4 small burr holes are drilled using a hand drill. The first burr hole is made posterior to the temporal line, while the second burr hole is drilled 1 cm above the first. The third burr hole is drilled just above the orbital rim and the fourth burr hole is made 1cm in a frontal direction. Quadrangle shaped bone flap of size 2.5x2cm is sawn through these holes and removed. Then only the inner edge of the bone above the orbital rim is drilled off while the remaining three edges are left as is. These three edges that are not drilled allow the bone flap to sit firmly in place touching the inner walls of the bone when the wound is closed in the end of the operation. The dura should be opened in a curved fashion along the bone edge toward to the supraorbital rim and fixed with two sutures. In the following step, a sufficient amount of cerebrospinal fluid (CSF) is drained by opening the chiasmatic and carotid cisterns, after which the frontal lobe deflects by itself. By dissecting the arachnoid membranes and opening the ipsilateral Sylvian fissure we expose A1 segment, ACA, ipsilateral ICA, and M1 segment (see Picture 1). We put temporary clips to control blood flow about 5-6 minutes before using the permanent clips.

**Picture 1. Refined subfrontal key hole approach**

**Statistical Analysis Tools**

Microsoft Excel and statistical analysis program SPSS were used for conducting statistical analysis on the collected data for this study.

**Results**

The outcome from total of 157 patients who were diagnosed with anterior circulation aneurysms and treated with surgery was analyzed in this study. This includes 6 patients aged between 20-29 (2 males, 4 females), 23 patients aged between 30-39 (10 males, 13 females), 57 patients aged between 40-49 (25 males, 32 females), and 71 patients aged between 50-59 (42 males, 29 females). The average age of the group was 46.3±2.3 while 78 patients were females and 79 were males. Hence, the ratio of males to females is 1:1 in the population. The data indicates that the rupturing of anterior circulation aneurysms happened at the rate of 36.2% in the 40-50 age group while the rate increased to 45.1% in the 50-60 age group. Out of all anterior circulation aneurysms considered in this study, 23 (18%) cases were multiple aneurysms. The data also shows that aneurysms of the anterior communicating artery tend to occur to male patients significantly more than females while aneurysms of the middle cerebral artery tend to occur to female patients significantly higher than males. The same trends were observed in male patients with multiple aneurysms 5 times more while in female patients grading indicates that 198 patients were in SAH grade level-I, 27 patients were in SAH grade level-II, 8 patients were in SAH grade level-III, and 2 patients were in SAH grade level-IV (see Table 4).
with internal carotid artery 3 times more (see Table 3).

Table 3.

<table>
<thead>
<tr>
<th>Arterial aneurysms</th>
<th>20-29</th>
<th>30-39</th>
<th>40-49</th>
<th>50-59</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>ACoA*, ACA*</td>
<td>6 (3.8)</td>
<td>3 (1.9)</td>
<td>15 (9.5)</td>
<td>9 (5.7)</td>
<td>18 (11.4)</td>
</tr>
<tr>
<td></td>
<td>7 (4.4)</td>
<td>4 (2.5)</td>
<td>12 (7.6)</td>
<td>12 (7.6)</td>
<td>15 (9.5)</td>
</tr>
<tr>
<td>MCA*</td>
<td>2 (1.3)</td>
<td>4 (2.5)</td>
<td>3 (1.9)</td>
<td>6 (3.8)</td>
<td>12 (7.6)</td>
</tr>
<tr>
<td></td>
<td>12 (7.6)</td>
<td>4 (2.5)</td>
<td>15 (9.5)</td>
<td>15 (9.5)</td>
<td>35 (22.2)</td>
</tr>
<tr>
<td>PCoA*</td>
<td>1 (0.6)</td>
<td>1 (0.6)</td>
<td>2 (1.3)</td>
<td>2 (1.3)</td>
<td>5 (3.1)</td>
</tr>
<tr>
<td>ICA*</td>
<td>1 (0.6)</td>
<td>3 (1.9)</td>
<td>2 (1.3)</td>
<td>6 (3.8)</td>
<td>18 (11.4)</td>
</tr>
<tr>
<td>Mutiple</td>
<td>1 (0.6)</td>
<td>3 (1.9)</td>
<td>18 (11.4)</td>
<td>1 (0.6)</td>
<td>19 (12.1)</td>
</tr>
<tr>
<td>BA*</td>
<td>1 (0.6)</td>
<td>1 (0.6)</td>
<td>1 (0.6)</td>
<td>1 (0.6)</td>
<td>4 (2.5)</td>
</tr>
<tr>
<td>Total</td>
<td>2 (1.3)</td>
<td>4 (2.5)</td>
<td>10 (6.3)</td>
<td>13 (8.2)</td>
<td>25 (15.9)</td>
</tr>
</tbody>
</table>

*ACoA- anterior communicating artery,*ACA – Anterior cerebral artery* MCA-middle cerebral artery,*PCoA-Posterior communicating artery, *ICA-Internal carotid artery,*BA-Basilar artery

Table 4.

<table>
<thead>
<tr>
<th>Grade</th>
<th>GCS</th>
<th>New approach</th>
<th>Traditional approach</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>15</td>
<td>126 (91.9%)</td>
<td>72 (73.5%)</td>
<td>198 (84.2%)</td>
</tr>
<tr>
<td>II</td>
<td>13-14</td>
<td>11 (8%)</td>
<td>16 (16.3%)</td>
<td>27 (11.5%)</td>
</tr>
<tr>
<td>III</td>
<td>13-14</td>
<td>0 (0%)</td>
<td>8 (8.2%)</td>
<td>8 (3.4%)</td>
</tr>
<tr>
<td>IV</td>
<td>7-12</td>
<td>0 (0%)</td>
<td>2 (2%)</td>
<td>2 (0.8%)</td>
</tr>
<tr>
<td>V</td>
<td>3-6</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>137</td>
<td>100%</td>
<td>100%</td>
<td>237 (100%)</td>
</tr>
</tbody>
</table>

We compared the outcome of the new keyhole surgical approach with the outcome of traditional approaches based on the severity of after surgery complications, focal neurological deficits, postoperative mental changes and functional impairments, and duration of hospital stay (recovery period). We selected 103 patients, who were treated with traditional approaches, as a control group and selected other 103 patients, who were treated with our new approaches, as a treatment group to conduct our
comparative research on the effectiveness of the new neurosurgical technique.

### Table 5.

<table>
<thead>
<tr>
<th></th>
<th>New approach</th>
<th>Traditional approach</th>
<th>Total</th>
<th>P mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hemiparesis</td>
<td>n</td>
<td>%</td>
<td>n</td>
<td>%</td>
</tr>
<tr>
<td>Yes</td>
<td>5</td>
<td>4.85</td>
<td>43</td>
<td>41.7</td>
</tr>
<tr>
<td>No</td>
<td>98</td>
<td>96.0</td>
<td>60</td>
<td>58.2</td>
</tr>
<tr>
<td>Total</td>
<td>103</td>
<td>100</td>
<td>103</td>
<td>100</td>
</tr>
</tbody>
</table>

According to Table 5, 43 out of 103 patients treated with the traditional approaches demonstrated some degree of hemiparesis symptoms on either side of their body while only 5 out of 103 patients treated with the new approach demonstrated the same. This 4.85% rate of postoperative hemiparesis in the treatment group is highly statistically significant compared to 41.7% rate of postoperative hemiparesis in the control group (P<0.0005).

### Table 6.

<table>
<thead>
<tr>
<th></th>
<th>New approach</th>
<th>Traditional approach</th>
<th>Total</th>
<th>P mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>Atrophy</td>
<td>n</td>
<td>%</td>
<td>n</td>
<td>%</td>
</tr>
<tr>
<td>Yes</td>
<td>7</td>
<td>6.7</td>
<td>79</td>
<td>76.6</td>
</tr>
<tr>
<td>No</td>
<td>96</td>
<td>93.2</td>
<td>24</td>
<td>23.3</td>
</tr>
<tr>
<td>Total</td>
<td>103</td>
<td>100</td>
<td>103</td>
<td>100</td>
</tr>
</tbody>
</table>

Postoperative temporalis muscle atrophy is mostly caused by the injury to the facial nerve branches and superficial temporal artery and gradually affects the patient’s chewing process and eventually causes significant cosmetic disfigurement in the face. When we compare the results of new and traditional approaches based on this criterion, only 6.70% of the patients treated with the new approach showed postoperative temporalis muscle atrophy while 76.6% of the patients treated with the traditional approaches demonstrated the same (see Table 6). This result is again highly statistically significant at level P<0.0005.

### Table 7.

<table>
<thead>
<tr>
<th></th>
<th>New approach</th>
<th>Traditional approach</th>
<th>Total</th>
<th>P mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mental change</td>
<td>n</td>
<td>%</td>
<td>n</td>
<td>%</td>
</tr>
<tr>
<td>Yes</td>
<td>4</td>
<td>3.8</td>
<td>46</td>
<td>44.6</td>
</tr>
<tr>
<td>No</td>
<td>99</td>
<td>96.1</td>
<td>57</td>
<td>55.3</td>
</tr>
<tr>
<td>Total</td>
<td>103</td>
<td>100</td>
<td>103</td>
<td>100</td>
</tr>
</tbody>
</table>

We also compared the results of the new approach with the traditional approaches based on the patients’ postoperative mental changes during the short period (24 weeks) immediately following the operation. Using this criterion, 44.60% of the patients treated with the traditional approaches demonstrated some degree of postoperative mental changes while only 3.80% of the patients treated with the new approach demonstrated the same (see Table 7). This result is again highly statistically significant at level P<0.0005.

DISCUSSIONS

Based on our study, the male to female ratio of the incidents, where anterior circulation aneurysms rupture and cause subarachnoid hemorrhage, looks like approximately one (1:1).

This result is somewhat different from the results of research conducted in Japan, Austria, and Canada where the rupture of anterior circulation aneurysms occurs two times more frequently to women than men. From the treatment group, two patients who were admitted to our hospital with the SAH grade level III-IV died after the operation due to cerebral vasospasm and five patients result in hemiparesis due to cerebral vasospasm. After 3 months of rehabilitations, 4 of those 5 patients recovered from the hemiparesis completely.
male, 2:1). According to our data, the majority of aneurysms (68.70%) are either anterior cerebral artery aneurysms or middle cerebral artery aneurysms. This result is consistent with the results from a number of research works conducted in other countries. More importantly, we achieved significantly better results in postoperative hemiparesis, temporal muscle atrophy, and mental changes with our new neurosurgical technique in contrast to the traditional approaches. These superior results are attributed to the new technique’s ability to address major problems in the traditional approaches where brain and brain tissues get easily damaged due to continuous retraction and large and long exposure to external environment (non-physiological) during the operation. In terms of the average duration of hospital stay after the operation, the new approach resulted in 8.2 days while the traditional approaches resulted in 12.7 days. This indicates that our new approach significantly shortened the healing time of the wounds compared to the traditional approaches.

CONCLUSIONS

1. By employing the new keyhole approach, we were able to decrease the exposed brain area during the surgery up to 3-4 times and greatly reduced brain and brain tissue damage (4.80%) during the operation since the new approach does not put much retraction on the brain.

2. The new approach requires only small incision of 5-6 cm. This small incision reduces blood loss and keeps surgically induced trauma to a minimum, which in turn allow a rapid healing of the wound and quicker return to normal live and decrease the expenses of surgery up to 2-3 times.

3. For anterior communicating artery aneurysms that are located 8 cm higher from the planum and measured larger than 10 mm in size and intracerebral hematoma of more than 25 ml, we recommend pterional approaches instead of keyhole approaches since the keyhole approaches have a limited access to aneurysms in these specific conditions.

REFERENCES:

Optic Nerve Compression by an Enlarged Carotid Artery: A Case Report

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Department of neurosurgery, State Third Hospital, Mongolia

ABSTRACT

Compression of the optic nerve by an internal carotid artery aneurysm is a rare and correctable cause of visual loss. A young woman presented with right eye visual loss without obvious cause and was found to have compression of the right optic nerve by the ipsilateral internal carotid artery, as revealed by magnetic resonance imaging. Pterional craniotomy and decompression of the optic nerve by unroofing the optic canal resulted in improvement of vision in the affected eye. Compression of the optic nerve by an enlarged internal carotid artery may produce visual loss. The disorder is well demonstrated by magnetic resonance imaging, timely intervention can improve vision. The optic nerve is susceptible to compression by space occupying lesions, such as tumors or aneurysms, as it enters the intracranial cavity. We report an unusual case of visual loss in a young woman that was caused by compression of the optic nerve by an enlarged internal carotid artery (ICA), which was demonstrated by magnetic resonance imaging (MRI) and successfully treated by microneurosurgery.

Key words: Internal carotid artery, Optic nerve diseases, Vision disorders

CASE REPORT

A previously healthy 34 year-old woman noted 2 weeks of progressive right eye visual loss, with no ocular pain or other neurological symptoms. There was no history of migraine, demyelinating disease, connective tissue disorder. The results of the general physical and neurological examinations were normal. The ophthalmological examination documented best corrected visual acuity of 20/40 right eye and 20/20 left eye. The ocular motility and the funduscopic examination were normal. Fullthreshold Humphrey automated static perimetry showed marked visual field constriction for the right eye, whereas the left eye visual field was normal (fig. 1a ). Repeat visual field testing 2 weeks later documented progressive right eye visual field loss. MRI of the brain revealed elevation and compression of the right optic nerve and optic chiasm by the right ICA (fig 2). Cerebral angiography showed unfolding of the siphon of the right ICA relative to the left ICA, and no aneurysm or vascular malformation (fig 3). Lumbar puncture yielded normal cerebrospinal fluid analysis without oligoclonal bands. The patient underwent right pterional craniotomy and, at surgery, the right internal carotid artery was observed to elevate and compress the right optic nerve. The bony optic canal was unroofed and the dural sheath of the nerve opened (fig 4). A focal area of pallor was noted in the optic nerve, where it was compressed by the overlying dural fold. Three months postoperatively, visual acuity was 20/30 right eye and 20/20 left eye. Automated perimetry showed almost complete improvement of the right eye visual field, with a mild residual arcuate scotoma (fig 1B).

DISCUSSION

As the optic nerve enters the intracranial cavity, it shares a narrow anatomic space with the internal carotid artery and the ophthalmic artery, bounded by bone and fold of dura mater. An enlarged or elongated ICA van act as a space-occupying lesion, forcing the optic nerve against the dural fold, producing a compressive optic neuropathy. A prefixed optic chiasm, which decreases the mobility of the optic nerve, or an excessively pneumatized sphenoid sinus, which compromises the lumen of the optic canal, may predispose to this condition.

Figure 1. A, preoperative Humphrey automated perimetry showing marked constriction of the right eye visual field. B, 3 months postoperatively, showing almost complete resolution of the right eye visual field defect.
Compression of the optic nerve by the ICA was demonstrated both by MRI and at surgery for our patient. Cerebral angiography showed some unfolding of the right carotid siphon compared to the left, but otherwise, the study revealed nothing abnormal. However, angiography cannot show the relationship of the vessels to structures such as optic nerve or chiasm. Moreover, three of the four cases of optic nerve compression by the ICA reported by Mitts and Mc-Queen (5) showed normal angiography, as did the cases reported by Begaust (2) and Matsuo et al. (4). Our case suggests that MRI demonstrates the disorder well and is diagnostic technique of choice, although angiography is also necessary to ensure there is no aneurism.

Our patient’s vision was restored by microneurosurgical decompression of the optic nerve. The operation unroofed the bony and dural canal of the optic nerve, thereby relieving the knife-like pressure on the nerve by the edge of the dural fold as the nerve enters the intracranial cavity and allowing the nerve to escape from the pulsations of the underlying ICA. Identical decompressive procedures with resulting improvement in vision have been described elsewhere (2,4,6).

Although modern imaging studies may suggest compression of the optic nerve by the ICA (3), caution must be exercised in attributing visual loss to vascular compression. Optic neuritis may produce similar visual loss. However, >90% of patients with optic neuritis present with pain, particularly with eye movement, which was not present in our patient (1). Additionally, MRI did not demonstrate signal abnormalities or demyelinating plaques, as seen in optic neuritis associated with multiple sclerosis. Also, the results of the cerebrospinal fluid analysis were normal. Other causes of visual loss, such as ischemic optic neuropathy, infiltrative processes of the meninges and optic nerve (such as sarcoidosis), and low-tension glaucoma, must be carefully considered. If no other causes of progressive visual loss can be demonstrated and optic nerve compression is unequivocally shown by MRI, then microneurosurgical decompression of the optic nerve is an effective treatment when performed before optic nerve damage is permanent, as suggested by optic atrophy. An awareness of this cause of reversible visual loss will hopefully result in its timely recognition and lead to salvaging vision in appropriate patients.

REFERENCES

Dolichoectasia of the ICA causing optic nerve compression has been typically described in older patients, particularly in those with hypertension or diabetes mellitus (4-6). There is no clear explanation of why this disorder occurred in such a young woman, although the disorder has been reported in a 38-year-old woman (5). The visual loss in this disorder may be sudden or gradual. The optic nerve head may be normal or may exhibit optic disc edema with subsequent optic atrophy (6).


Oxidant-Antioxidant Status in Chronic Obstructive Pulmonary Disease: Relationship with Disease Severity

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ABSTRACT

An oxidant-antioxidant imbalance is thought to play an important role in the pathogenesis of chronic obstructive pulmonary disease (COPD). We hypothesized that antioxidant capacity reflected by cytochrome c oxidase (COX), free radical scavenging substances (FRSS), and levels of the lipid peroxidation product malondialdehyde (MDA) in erythrocyte, plasma and urine may be related with disease severity in COPD patients. We measured several parameters of oxidant-antioxidant status in erythrocyte membrane and cytosol, plasma and urine in 188 patients with COPD and 48 healthy controls (HC). Lung function was measured by spirometry. Partial oxygen pressure in blood (PaO2) was determined in correlation with an oxy-hemoglobin saturation (SaO2) estimated measured by finger pulse oximeter. Free radical scavenging substances in erythrocyte cytosol (FRSSc) and membrane (FRSSm) were significantly lower, but greater in urine (FRSSu) in patients with COPD as compared to HC (FRSSc: 0.331±0.025 vs. 0.311±0.015 mcg/ml, p<0.05, FRSSm: 0.526±0.024 vs. 0.481±0.011, mcg/ml, p<0.05, FRSSu: 0.045±0.008 vs. 0.052±0.008 mcg/ml, p<0.05). In contrast, no differences were seen between the two groups in the plasma FRSS (0.320±0.013 vs. 0.32±0.011 mcg/ml). COX in plasma (COXp), urine (COXu) and erythrocyte membrane (COXm) were significantly lower, but greater in erythrocyte cytosol (COXc) in patients with COPD as compared to those in HC (COXp: 3.40±0.21 vs. 2.92±0.11, COXu: 24.39±1.34 vs. 21.92±1.27, COXm: 37.01±1.21 vs. 33.67±1.11, Oc: 14.19±0.71 vs. 19±1.2, p<0.05). Plasma, urinary and membrane MDA levels were significantly higher in study group as compared to HC (MDAp: 0.080±0.007 vs. 0.057±0.004, MDAu: 0.101±0.02 vs. 0.054±0.005, MDAm: 0.145±0.01 vs. 0.109±0.007, p<0.05). Linear regression analysis revealed a significant direct relationship between FVC, FEV1 and COXm (r=0.299, p<0.01, r=0.260, p<0.01), and a significant inverse relationship between FVC, FEV1 and MDAm levels (r=-0.277, p=0.01, r=-0.235, p<0.05). Findings of the present study suggest that antioxidant capacity reflected by COX and the lipid peroxidation products MDA in erythrocyte membrane are linked to the severity of COPD.

Key words: Chronic obstructive pulmonary disease, Free radical scavenging substances, Cytochrome c oxidase, Lipid peroxides products, Malondialdehyde

INTRODUCTION

COPD represents a major health problem, and its prevalence and mortality rates are increasing worldwide. Oxidative stress, defined as an imbalance between increased exposure to oxidant and/or decreased antioxidative capacities, represents one of the key pathogenetic mechanisms in the development of COPD. A number of antioxidant disturbances have been observed in patients with COPD. Lipid peroxidation products, one of the key indicators of oxidative stress, are elevated in sputum and exhaled breath condensate of patients with COPD. At the same time, the antioxidant mechanisms are attenuated in these patients, as indicated by reduced glutathione levels in the lungs, reduced glutathione peroxidase activity in erythrocytes and lower antioxidant capacity in plasma during exacerbations of COPD. Nevertheless, studies on the relationships between the oxidant-antioxidant imbalance and pulmonary functions showed inconsistent results. On the one hand, airway obstruction, reflected by reductions in forced expiratory volume in one second (FEV1), was shown to correlate with antioxidant substances such glutathione and myeloperoxidase levels. Furthermore, lipid peroxidation products measured as malondialdehyde (MDA) content correlated inversely with the degree of small airway obstruction. On the other hand, more recent studies showed that there was no significant relationship between plasma antioxidant capacity and pulmonary function in patients with COPD. The aim of the present study was to assess the relationships between the COPD stages and antioxidant activity reflected by free...
radical scavenging capacity, cytochrome c oxidase and MDA levels in erythrocyte membrane and cytosol, plasma and urine.

**MATERIALS AND METHODS**

Subjects: Patients with COPD were consecutively recruited to the study in 2008, 2010 and 2012, at the Pulmonology Department of First National Central Hospital (Ulaanbaatar, Mongolia). All patients had COPD according to the American Thoracic Society /European Respiratory Society guidelines\(^6\). Patients with other respiratory disorders than COPD, malignancy, overt cardiac failure, recent surgery, severe endocrine, hepatic or renal diseases were not included. The control group included 48 healthy persons with similar ages, having normal pulmonary function tests. Ethical considerations: The study was conducted in accordance with national policies on ethics for surveys involving human subjects. The study protocol was approved by Ethical Review Committee of Health Science University of Mongolia. In accordance with their approval, all participants signed an informed consent form before participating in the study.

Pulmonary function testing: Pulmonary functional tests were evaluated with using of spirometer ST-320 (Mitsubishi, Japan). All pulmonary function tests were performed at the 10-15 minute after inhaling short-term β2-agonist salbutamol in dosage 200 mcg. Forced inspiratory volume in one second (FEV1) and forced vital capacity (FVC) were expressed as a percentage of the predicted values for age, sex, and height. Three technically acceptable measurements were performed in each patient, and the best value was included in the analyses. Partial oxygen pressure in blood (PaO2) was determined in correlation with an oxyhemoglobin saturation (SaO2) estimate measured by finger pulse oxy-meter (504-US; Criticare Systems, Waukesha, WI) and expressed in a percentage.

Parameters related to oxidant-antioxidant status: Fasting venous blood samples were collected in EDTA vial and in plain vials (without anticoagulant). Samples were used for the estimations of cytochrome c oxidase, free radical scavenging substances, lipid peroxidation products in plasma, erythrocyte cytosol and membrane suspension. Assessment of similar parameters were performed in urine, taken under standardized condition.

Free radical scavenging substances, measured as protonated products in plasma, urine, erythrocyte cytosol and membrane suspension were determined with method, using of the stable free radical 2,2-diphenyl-pircyl-hydrazyl (DPPH), described by Brand-Williams (1995) and expressed as microgramm per milliliter.

Cytochrome c oxidase (COX) in plasma, urine, erythrocyte’s cytosol and membrane was estimated using of HIMEDIA oxidase disks, based on the method described by Kovaec, developed by Gaby and Hadley (1957) and expressed as a minute.

Lipid peroxidation in erythrocyte membrane, plasma and urine were assessed by measuring concentration of thiobarbituric acid reactive substances (MDA-TBA) in spectrophotometry at 535 nm. MDA levels are expressed as nanomoles of thiobarbituric acid reactive substances formed per liter of erythrocyte membrane suspension, plasma and urine.

Statistical analyses: Statistical analysis was carried out using SPSS 20. Continuous variables are shown as means ± S.E.M. To assess the relationship between selected variables, linear regression analyses was used. P-value less than 0.05 (P<0.05) was considered as significant.

**RESULTS**

One hundred and eighty eight patients, 127 men and 61 women, were included in this study. They were generally late middle-aged (mean age 59.3±5.5 years), with the average smoking history of 31.3±7.3 pack-years. COPD severity in patients was classified as Stage I, II, III, or IV, depending on the FEV1% predicted, as described in the GOLD guidelines\(^8\). Stages III and IV were combined into one group designed “Stage III+IV”, because the number of subjects in Stage IV was too small for separate examination. The control group included 48 healthy persons with similar ages, smoking history of 4.5±1.2 pack-years and having normal pulmonary function tests. No differences were found in the demographic data between the groups (Table 1). FVC, FEV1, and the ratio of FEV1/FVC were all significantly lower in patients with COPD compared to HC (p<0.001 for all spirometric variables). Examination of SaO2 and PaO2 revealed significantly lower values in COPD group compared to HC (p<0.001, p=0.08, respectively) (Table 1). Erythrocyte cytosol FRSS were significantly lower in Stage II and Stages III+IV (p<0.05), but greater in Stage I. Erythrocyte membrane FRSS were lower in COPD group as compared to HC (p<0.05), although not all differences were statistically significant. Urinary FRSS were lower in Stage I and greater in the other Stages as compared to HC. In contrast, no differences of plasma FRSS were seen between the control and COPD groups. COX activity in plasma, urine and erythrocyte’s membrane are significantly lower, but having greater in erythrocyte cytosol in patients with COPD compared to HC (p<0.05). Plasma and urine COX in Stage III+IV were significantly lower than that in both Stage II and Stage I. Erythrocyte membrane COX also tended to decrease with COPD progression, although not all differences were statistically significant. Plasma, urinary and membrane lipid peroxides measured as MDA-TBA products were
greater in COPD group significantly, compared to HC (p<0.05) (Table 2). Erythrocyte membrane MDA in Stages III+IV was significantly greater than that in both Stage II and Stage I. This suggests that MDAm increased with the progression of COPD. Linear regression analysis revealed a significant direct relationship of FVC and FEV1 with COX activity of erythrocyte membrane (r=0.260, p<0.01), and a significant inverse relationship of FVC and FEV1 with membrane MDA levels (r= - 0.235, p<0.05). Findings of the present study suggest that oxidant-antioxidant capacity reflected by erythrocyte’s membrane cytochrome c oxidase and membrane levels of the lipid peroxidation product MDA are linked to the severity of COPD.

**DISCUSSION**

By studying patients with different stages of COPD we have demonstrated that the erythrocyte’s membrane COX activity, plasma COX activity and membrane MDA levels correlate with disease severity as assessed by FVC and FEV1. Our present study suggests a significant negative relationship between COX activity of erythrocyte’s membrane and the stage of COPD. These findings are extend investigations of imbalance between oxidant and antioxidant capacities, represents one of the key pathogenetic mechanisms in the development of COPD3,5,11. The present study yields several interesting and novel findings. First, the antioxidant activity measured as FRSS in erythrocyte’s cytosol were increased in Stage I and decreased in progressing COPD. This finding may be related to the activation of compensate mechanism in the early stage of disease. The previous investigation by Halliwell B. also suggested the increase of antioxidant activity in smokers12. Numerous studies have shown depletion of antioxidant activity in plasma and erythrocyte3,5,6,12. Indeed, several13 but not all14 studies documented that certain markers of oxidative stress may be related to the severity of obstructive lung impairment in patients with COPD. Relationships between anti-oxidative enzymatic systems and lung function impairment were also reported in previous studies, where the anti-oxidative enzymes were measured in erythrocytes15 but not in plasma3. Similarly, we didn’t observe any relationship between plasma antioxidant capacity and spirometric variables in present study. One reason for failing to find a significant relationship between FRS activity and pulmonary function parameters may be related to the earlier described phenomenon that various enzymatic systems differ substantially in their responses to smoking-induced increases in oxidative stress3.

Second, COX activity in erythrocyte cytosol was increased in all stages of COPD, in contrast to erythrocyte’s membrane, plasma and urine COX, where they decreased respectively. This finding may be related to migration of Fe++ from hemoglobin to erythrocyte’s cytosol due to damage of their membrane. A previous investigation by Sauleda et al.16 also suggested that activity of COX is increased in circulating lymphocytes, although numerous studies have shown depletion of COX in lungs, plasma and erythrocytes of patient with COPD17,18.

Finally, a significant inverse relationship between erythrocyte membrane MDA levels and the degree of obstructive lung impairment reflected by FEV1 and FVC was observed in the present study. Previously, lipid peroxidation products measured as MDA content in plasma correlated inversely with the degree of small airway obstruction reflected by low maximal expiratory flow rates in smokers9. Our findings support these original reports by suggesting that high level of MDA in erythrocyte membrane as well as in plasma might be associated with lung function in patients with COPD. The numerous of studies showed elevated levels of other markers of lipid peroxidation such as urinary and plasma concentrations of 8-isoprostane19 and exhaled ethane20 in patients with COPD. Lipid peroxidation products were elevated in sputum, exhaled breath condensate4 and plasma21 of patients with stable COPD. Moreover, exacerbations of COPD lead to even further elevations in various markers of oxidative stress22. In contrast, we didn’t found any significant correlation of MDA levels, measured in plasma and urine with COPD stages in present study.

In conclusion, our results indicate that the COX activity is increased in erythrocyte’s cytosol, but decreased in membrane. Moreover, FRSS are reduced and lipid peroxidation activity is increased with progressing of COPD. Further studies are needed to analyze the pathophysiological mechanisms involved in lung injury related to oxidant-antioxidant imbalance.
### Table 1.
Demographic data and pulmonary functional tests in control and study groups.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Control group n=48</th>
<th>COPD group Total n=188</th>
<th>COPD group Stage I n=45</th>
<th>COPD group Stage II n=90</th>
<th>COPD group Stage III+IV n=53</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age (years)</strong></td>
<td>59.9±5.6</td>
<td>59.32±5.5</td>
<td>58.18±11.23</td>
<td>59.66±12.61</td>
<td>60.8±10.74</td>
</tr>
<tr>
<td><strong>Men/women</strong></td>
<td>32/16</td>
<td>127/61</td>
<td>25/20</td>
<td>57/33</td>
<td>45/8</td>
</tr>
<tr>
<td><strong>Pack-years</strong></td>
<td>4.5±1.2</td>
<td>31.3±7.3*</td>
<td>27.9±14.8</td>
<td>30.6±15.5</td>
<td>35.4±12.8</td>
</tr>
<tr>
<td><strong>Smoke index</strong></td>
<td>11.3±2.2</td>
<td>21.7±16.5*</td>
<td>18.6±10.8</td>
<td>20.99±16.9</td>
<td>27.6±17.4</td>
</tr>
<tr>
<td><strong>Body mass index (kg/m²)</strong></td>
<td>27.2±4.4</td>
<td>26.9±6.1*</td>
<td>28.2±6.10</td>
<td>26.92±5.71</td>
<td>25.7±6.8**</td>
</tr>
<tr>
<td><strong>FVC (%)</strong></td>
<td>96.04±3.40</td>
<td>90.54±21.6**</td>
<td>114.6±12.3</td>
<td>92.9±12.4</td>
<td>66.1±12.7**</td>
</tr>
<tr>
<td><strong>FEV1 (%)</strong></td>
<td>83.98±11.77</td>
<td>57.82±17.07**</td>
<td>78.1±6.2</td>
<td>59.97±9.1</td>
<td>36.9±8.02**</td>
</tr>
<tr>
<td><strong>FEV1/FVC (%)</strong></td>
<td>0.83±0.11</td>
<td>0.62±0.08**</td>
<td>0.68±0.03</td>
<td>0.64±0.05</td>
<td>0.55±0.09**</td>
</tr>
<tr>
<td><strong>SaO₂ (%)</strong></td>
<td>97.6±0.77</td>
<td>92.6±4.63**</td>
<td>95.6±1.8</td>
<td>92.97±3.5</td>
<td>89.55±5.9**</td>
</tr>
<tr>
<td><strong>PaO₂ (%)</strong></td>
<td>89.9±3.4</td>
<td>69.4±14.2**</td>
<td>79.02±10</td>
<td>69.3±12</td>
<td>61.2±14**</td>
</tr>
</tbody>
</table>

* p<0.05; ** p<0.01 Data are means ± S.E.M.

### Table 2.
Parameters of oxidant-antioxidant status in control and study groups.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Control group n=48</th>
<th>COPD group Total n=188</th>
<th>COPD group Stage I n=45</th>
<th>COPD group Stage II n=90</th>
<th>COPD group Stage III+IV n=53</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plasma FRSS (mcg/ml)</td>
<td>0.321±0.01</td>
<td>0.320±0.013</td>
<td>0.310±0.013</td>
<td>0.311±0.015</td>
<td>0.350±0.014</td>
</tr>
<tr>
<td>Urinary FRSS (mcg/ml)</td>
<td>0.052±0.01</td>
<td>0.045±0.008</td>
<td>0.061±0.012</td>
<td>0.039±0.005</td>
<td>0.042±0.07</td>
</tr>
<tr>
<td>Cytosol FRSS (mcg/ml)</td>
<td>0.311±0.02</td>
<td>0.331±0.025*</td>
<td>0.28±0.02</td>
<td>0.37±0.03</td>
<td>0.38±0.03*</td>
</tr>
<tr>
<td>Membrane FRSS (mcg/ml)</td>
<td>0.481±0.01</td>
<td>0.526±0.024*</td>
<td>0.57±0.03*</td>
<td>0.50±0.02</td>
<td>0.54±0.02*</td>
</tr>
<tr>
<td>Plasma COX (min)</td>
<td>2.92±0.11</td>
<td>3.40±0.21*</td>
<td>3.18±0.23</td>
<td>3.39±1.79</td>
<td>3.58±0.24</td>
</tr>
<tr>
<td>Urinary COX (min)</td>
<td>21.92±1.27</td>
<td>24.39±1.34*</td>
<td>24.3±1.42</td>
<td>24.2±1.34</td>
<td>24.9±1.31</td>
</tr>
<tr>
<td>Cytosol COX (min)</td>
<td>19±1.2</td>
<td>14.19±0.71*</td>
<td>13.7±0.5</td>
<td>14.8±0.7</td>
<td>14.1±0.7</td>
</tr>
<tr>
<td>Membrane COX (min)</td>
<td>33.67±1.11</td>
<td>37.01±1.21*</td>
<td>33.7±1.2</td>
<td>36.6±1.3</td>
<td>42.5±1.3*</td>
</tr>
<tr>
<td>Plasma MDA (mmol/L)</td>
<td>0.057±0.01</td>
<td>0.080±0.007*</td>
<td>0.075±0.006</td>
<td>0.089±0.009*</td>
<td>0.071±0.005</td>
</tr>
<tr>
<td>Urinary MDA (mmol/L)</td>
<td>0.054±0.01</td>
<td>0.101±0.02*</td>
<td>0.054±0.004</td>
<td>0.122±0.002*</td>
<td>0.106±0.012*</td>
</tr>
<tr>
<td>Membrane MDA (mmol/L)</td>
<td>0.109±0.01</td>
<td>0.145±0.01*</td>
<td>0.096±0.01</td>
<td>0.143±0.03</td>
<td>0.170±0.01*</td>
</tr>
</tbody>
</table>

* p<0.05 Data are means ± S.E.M.
REFERENCES


Oxygen Saturation in Healthy Infants During the First 10 minutes of Life

Tungalag.L1*, Gerelmaa.Z2
1 Third Maternity hospital
2 School of Medicine, Health Sciences University of Mongolia

ABSTRACT
To study the normal levels of oxygen saturation (saturation by pulse oximetry, SpO2) in healthy newborns during the first 10 minutes of life and the possible relationship to birth weight or mode of delivery. In this observational study, asymptomatic newborns with gestational age ≥37 weeks at birth that did not require supplemental oxygen had continuous recordings taken of the postductal oxygen saturation over the first 10 minutes of life. A total 132 deliveries were monitored. Oxygen saturation readings were obtained 1, 5, 10 minutes of life. The median oxygen saturation at 1 minute was 64%, at 5 minutes 83%, at 10 minutes 97%. Oxygen saturation was significantly related to mode of delivery. Infants delivered by cesarean delivery had 1.5% lower oxygen saturation than in those delivered vaginally. (95% confidence interval, P<0.001). No relationship exists between oxygen saturation and birth weight. In healthy newborns, levels oxygen saturation measured during the first 10 minutes of life are negatively related to birth weight and related to mode of delivery. The process of transitioning to normal postnatal oxygen saturation requires more than 5 minutes in healthy newborns breathing room air.

Keywords: Newborns; oxygen saturation; pulse oximeter.

INTRODUCTION
All newborns are “cyanotic” at birth. During the few minutes of life, oxygen saturation (saturation by pulse oximetry, SpO2) increases from intrapartum levels of 30-40%. Traditionally, oxygenation levels of newly born infants have been assessed clinically. In algorithms for neonatal resuscitation published by the International Liaison Committee for Resuscitation,1 European Resuscitation Council2 and Australian Resuscitation color and heart rate during neonatal transition is Counsil,3 clinical assessment of an infant’s color and heart rate used as major action points. However, studies have shown that clinical assessment of unreliable.4,5 Long before the discovery of pulse oximetry, arterial blood gases were the prime diagnostic tests to measure the saturation of oxygen in patients. Pulse oximetry was invented by Takuo Aoyagi, a biomedical engineer working for the Shimadzu Corporation in Kyoto, Japan, in the early 1970s.6 Pulse oximetry is a very simple, non-invasive procedure of monitoring the oxygen saturation of the hemoglobin component of the red blood cell. Basically, it works by measuring the absorption of red and infrared by pulsatile blood. In most literature, the accepted normal value of a pulse oximeter reading is between 95-100%.6 that in the process of postnatal adaptation, a normal newborn undergoes a period of transitional physiological cyanosis. Administering 100% oxygen to a spontaneously breathing neonate based only on visual assessment of cyanosis may be unnecessarily invasive and lead to potentially harmful hyperoxia.7-10 Several perinatal factors such as birth weight, gestational age (GA), gender or mode of delivery could possible influence the levels of SpO2 in newborns.

The objective of our study was to document the arterial oxygen saturation in healthy newborns ≥37 week gestation during unassisted transition in the delivery room and to examine the association between the delivery method (vaginal or cesarean) and oxygen saturation at birth.

MATERIALS AND METHODS
We conducted a prospective observational study of oxygen saturation newly born infants between February 2012 and March 2012. The study was endorsed by the Human Research and Ethics Committees of Health Sciences University of Mongolia. Newborns born at ≥37 weeks gestational age and weighing ≥2500 g admitted to the nursery from the maternity ward were included in the final study. Children with symptoms of disease or receiving supplementary oxygen were admitted to the neonatal intensive care unit and therefore excluded. Infants also were excluded if they had a congenital malformations, that might interfere with the normal transition to extra uterine life. All infants were assessed at birth, were dried and wrapped with warmed towels. Immediately after birth, the Apgar timer was started, and a pulse oximeter sensor was placed on the infants as soon as possible and then was connected to an oximeter. The sensor was then connected
to the oximeter, because this leads to the fastest acquisition of data. \( \text{SpO}_2 \) was measured post ductal (foot) by the pulse oximeter “EDAN” measuring functional \( \text{SpO}_2 \) over the first 10 minutes of life. \( \text{SpO}_2 \) was measured post ductal (foot) by the pulse oximeter “EDAN” measuring functional. The value of \( \text{SpO}_2 \) as well as birth weight, mode of delivery, GA and the time of measurement (hours after delivery), were registered. Data were analyzed by the SPSS 17.0 statistical package.

RESULTS

A total 132 children fulfilled the criteria for inclusion in the final analyses. Of these, gender was registered 62 (46.9%) boys and 70 (53.1%) girls. The median birth weight of the children was 3448 g and median gestational age was 38.5 weeks (Table 1).

Figure 1 shows the graph of median \( \text{SpO}_2 \) values over the first 10 minutes of age for infants born by cesarean and vaginal delivery. Median \( \text{SpO}_2 \) values (IQR) at 5 minutes were 86%, for newborns delivered vaginally and 79% for those delivered by cesarean section. By 10 minutes of age, the median \( \text{SpO}_2 \) (IQR) rose to 97%, in the vaginal delivery group and 95% in the cesarean delivery group. The median \( \text{SpO}_2 \) values at 1, 5, 10 minute were 64%, 83%, 97% respectively. (Figure 2)

Table 1. Infant characteristics

<table>
<thead>
<tr>
<th>Included infants</th>
<th>N=132</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gestational age, mean (range)</td>
<td>38.5 (37- 41.5)</td>
</tr>
<tr>
<td>Birth weight, median (range)</td>
<td>3448 (2500 - 4850)</td>
</tr>
<tr>
<td>Gender:</td>
<td></td>
</tr>
<tr>
<td>Boys</td>
<td>62 (46.9%)</td>
</tr>
<tr>
<td>Girls</td>
<td>70 (53.1%)</td>
</tr>
<tr>
<td>Delivery route:</td>
<td></td>
</tr>
<tr>
<td>Vaginal</td>
<td>88 (66.6%)</td>
</tr>
<tr>
<td>Cesarean</td>
<td>44 (33.3%)</td>
</tr>
<tr>
<td>Apgar score:</td>
<td></td>
</tr>
<tr>
<td>1 minute, median (IQR)</td>
<td>7 (7-8)</td>
</tr>
<tr>
<td>5 minute, median (IQR)</td>
<td>9 (8-10)</td>
</tr>
</tbody>
</table>

Table 2. Comparison of \( \text{SpO}_2 \) values in the first 10 minutes after birth for vaginal and cesarean births

<table>
<thead>
<tr>
<th>Time after birth</th>
<th>( \text{SpO}_2 ) Median (IQR)</th>
<th>Vaginal Birth</th>
<th>Cesarean Birth</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 min</td>
<td>65 (41-73)</td>
<td>59 (40-72)</td>
<td>0.002</td>
</tr>
<tr>
<td>5 mins</td>
<td>86 (58-96)</td>
<td>79 (56-92)</td>
<td>0.001</td>
</tr>
<tr>
<td>10 mins</td>
<td>95(68-100)</td>
<td>97 (69-100)</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

DISCUSSION

We demonstrated in this study that levels of \( \text{SpO}_2 \) during the first 10 minutes of life were related birth weight and mode of delivery in healthy newborns. \( \text{SpO}_2 \) was higher in children born by delivered vaginally than in those cesarean section. It has been demonstrated that children born by cesarean section have lower levels of \( \text{SpO}_2 \) during the first few minutes of life, probably due to increased amount of lung fluid.\(^{11,12}\) \( \text{SpO}_2 \) is <60% in the fetus just before birth\(^1\) and can decrease to 30% during labor.\(^2\) This study reports how \( \text{SpO}_2 \) values changed of infants in the first 10 minutes after birth. We found that it took up to 9 minutes to reach a median \( \text{SpO}_2 \) of 90%; other studies have reported times of between 8 and 15 minutes.\(^{15,16,17}\) Our results support the assertion that during normal neonatal transition, it often takes 9 minutes or longer to achieve an oxygen saturation ≥90%. In the first 5 minutes after birth, infants born through cesarean section had significantly lower \( \text{SpO}_2 \) measurements than those delivered vaginally. This is consistent with the findings of Rabi et al\(^{12}\) and Harris et al.\(^1\) In contrast, other researchers found no significant differences between infants delivered vaginally or cesarean section.\(^{5,18,19}\) The median \( \text{SpO}_2 \) values at 5 minutes were 83%, 10 minutes 97% which is similar to other studies. We recommend clinicians could use pulse...
oximeter during stabilization and resuscitation, when monitoring SpO2 values. This is especially important when treating at risk of hyperoxia. Deckardt et al20 and Kenotic and Lindner21 suggested that using SpO2 measurements in the delivery room was valuable in managing resuscitation. Recently, Finer and Leone22 advocated use of a targeted SpO2 protocol in the DR. Kattwinkel23 suggested that “we should be aiming to restore normoxia quickly and to achieve normal levels of blood oxygen throughout and beyond the resuscitation process. More aggressive use of the pulse oximeter in the delivery setting may facilitate achieving this goal”. The best definition of “normoxia” is that which leads to the best short- and long-term outcomes after resuscitation.

CONCLUSIONS

We recommend that clinicians consider using one of the charts when monitoring SpO2 values during transition. Before oximetry is advocated for routine use in the delivery room, more research is needed to define normoxia, and more importantly, how to interpret and apply SpO2 readings to clinical practice to improve short-term and long-term outcomes.

REFERENCES

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Prevalence and Risk Factors of Latent Tuberculosis Infection Among Health Care Workers in Ulaanbaatar City, Mongolia

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2Mongolian Anti Tuberculosis Coalition
3Department of Epidemiology and Biostatistics, Health Sciences University of Mongolia

ABSTRACT
Tuberculosis (TB) has been recognized as an occupational hazard for health care workers in many countries. In the last few years, number of MDR TB cases and even deaths registered among health care workers. Therefore, the screening of people employed in the health care sector for active and LTBI is fundamental to infection control programmes in health care facilities. A cross sectional study was conducted including 844 health care workers who working in randomly selected health care facilities in the capital Ulaanbaatar city from January to June 2012. The overall prevalence rate of latent tuberculosis infection among health care workers was 52.1% (95% CI 48.8-55.5). The prevalence rate of latent TB infection among men is 55.7% (95% CI: 45.3%; 65.4%) while among women this rate is 51.7% (95% CI: 48.2%; 55.2%) (p=0.481). Presence active tuberculosis was 6 (0.7%) cases and inactive tuberculosis was 53 (6.3%) cases among health care workers, the overall prevalence rate of TB disease is 699/10000 and prevalence of TB disease for HCWs working in TB-related health care facilities 19.4% was significantly high (p<0.001).

On multivariate analysis, the risk factors found to be significantly associated with latent tuberculosis infection were working at tuberculosis facilities aOR=6.8 and working at the secondary level health care service aOR=2.2, working night shift aOR=1.5 (p<0.001).

The prevalence of latent tuberculosis infection among health care workers in Mongolia was relatively same level of low and middle income countries. The result of our study demonstrates that TB is a significant occupational problem among health care workers in Mongolia.

Key words: tuberculosis; health care workers; occupational risk; tuberculin skin test

INTRODUCTION
The fundamental ethic of health care is sick persons must receive care. This premise carries an unstated consequence: an occupational risk to health care workers (HCWs) who respond to the needs of contagious patients. This predicament was shown yet again during the severe acute respiratory syndrome (SARS) epidemic. During the past 2 decades, occupationally acquired hepatitis B, HIV infection, multidrug resistant tuberculosis, and viral hemorrhagic fever, among others, have killed health care workers [1]

Tuberculosis (TB) has been recognized as an occupational hazard for health care workers in many countries [2,3]. The risk of latent tuberculosis infection (LTBI) and active TB as an occupational disease is well established and HCWs are still recognized as a high-risk group for LTBI [4-10]. Health care workers at risk of Mycobacterium tuberculosis infection during participation in the diagnosis and treatment of patients with active tuberculosis [11,12].

More than half of all health care workers in the high tuberculosis incidence, low and middle income countries are estimated to be latently infected with tuberculosis and this high prevalence is attributable to increased occupational exposure to the M. tuberculosis, in addition to possible exposure in the community [13,14].

According to WHO records, the average rate of tuberculosis reported in the general population in Mongolia is 162/100000; it still has the highest incidence rate in the Western Pacific Region [15].

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In the 2007 Drug Resistance Surveillance (DRS) in Mongolia, 850 patients had *M. tuberculosis* isolated. Of these, 650 were new, and 200 were previously treated. Any drug-resistance was found in 7.5% (95% CI 5.9-9.5). While the MDR-TB rate among new cases was low at 1.4% (95% CI 0.7-1.6), the MDR rate among retreatment cases was quite high, 27.5% (95% CI 21.8-34.1) [16,17].

Therefore, the screening of people employed in the health care sector for active and LTBI is fundamental to infection control programmers’ in Mongolian hospital. It was the aim of the study to determine the prevalence of active and latent TB, and risk factors associated with latent tuberculosis infection among health care workers.

**MATERIALS AND METHODS**

**Study design and sampling**

A cross sectional study was conducted from January to June 2012 among HCWs working in health care facilities in the capital city of Mongolia. Therefore, 42 health care facilities were selected by simple random sampling. The HCWs involved in this study were nurses and doctors. Sample size calculation was based on a prevalence rate of 43% among nurses and 54% among doctors based on another study in low and intermediate tuberculosis burden country. The minimum sample size required was 840 in two groups. The respondents were selected randomly using probability proportionate to size approach.

**Tuberculin skin test**

The dose of two tuberculin units of protein derivative was Purified protein derivative (tubersol) from Ukraine was used. All skin testing was done by one-step protocol. The Mantoux technique was used and 0.1ml of protein derivative was applied by an intradermal injection in the middle third of the inner forearm of participants.

Skin tests were read 72 hours after; the transverse diameter of the indurations was measured using calipers and recorded in millimeters. Tuberculin skin test (TST) was considered positive if induration was ≥10 mm in diameter (after the one-step protocol), as recommended for TB endemic populations, such as these.18 Five trained personnel (nurses for district TB dispensary) administered and read the tuberculin skin tests. The Bacilli Calmette-Guerin (BCG) vaccination status was confirmed by the presence of vaccination scars.

**Interview and Questionnaire items**

HCWs enrolled in the study signed an informed consent and filled in a standardized self-administered questionnaire containing question about socio-demographic features, BCG vaccination history, duration of employment, possible exposure to tuberculosis in their households and workmates, job title, workplace, co-morbid conditions, substance abuse, history of previous tuberculosis.

**Interpretation of chest radiographs**

One of the several objectives of this study was to screened for the presence of active TB by clinical examination and standard postero-anterior view chest radiographs.

In present study one radiologist, with more than 15 years of post-residency clinical experience, interpreted all radiographs of the participating. All discordant radiographs were evaluated by a senior radiologist with 30 years of clinical experience whose interpretation was considered as final. We used standard criteria for reading x-rays, based on the radiological abnormalities present; each radiograph was classified as either: 1) normal, 2) suggestive of inactive TB, 3) suggestive of active TB, 4) non tubercular pulmonary abnormalities.

**Bacteriology examinations**

All symptomatic HCWs, and those had radiological lesions suggestive of active TB disease were further investigated by sputum smear and cultures and repeat chest radiographs as indicated. All bacteriological examinations were done in TB Reference Laboratory of National Center for Communicable Disease in Mongolia.

Any health care workers with active TB were treated with standard first line chemotherapy as by national guidelines, using a DOTS strategy.

The study protocol was approved by Ethical Review Committee Health Science University of Mongolia. In accordance with their approval, all participants signed an informed consent form before participating in the study.

**Statistical analyses**

Data entry and all statistical analyses were performed using EPI-INFO Version 7.1 and SPSS Version 20 software. Associations between the tuberculin skin test status and potential risk factors were assessed using x² significance testing and the calculation of odds ratios (OR) with 95% confidence intervals (CI).

Logistic regression methods were used to determine the risk factors associated with tuberculin reactivity, and to adjust for confounding factors. A multivariate model was used for all variables with a P value less than 0.20 in univariate analysis. Interactions between the variables were examined. The goodness-off–fit of this final regression model was checked using the Hosmer-Lemeshow test.

**RESULTS**

**Study population**

Of 844 eligible HCWs, 100% answered the questionnaire and agreed to be tested for LTBI with one-step TST and chest X-ray. The mean age of the participants was 40.3±18.9 years, majority of the respondents were females.
89.6% (P<0.001). 55.6% of the total study population was nurses, 44.4% physicians. Almost all of the respondents (92.3%) had with BCG vaccination scar. 774 (91.7%) working non-TB health care facilities and 70 (8.3%) them working at TB-related health care facilities (P<0.001).

Prevalence of LTBI
The overall prevalence of LTBI among HCWs was 52.1% (95% CI 48.8-55.5). Prevalence of LTBI among physicians was 53.1% (95% CI 48.0-58.0), nurses was 51.4% (95%CI 46.9- 55.9) (P=0.62). The prevalence of LTBI for HCWs working in secondary health care facilities was 66% (95% CI 59.6-71.7), which was higher than any others health care facilities (P<0.001).

The prevalence of LTBI for HCWs working in TB-related health care facilities was 84.3% (95% CI 74.0-91.0) and Non-TB health care facilities 49.2% (95% CI 45.7-52.7).

The prevalence of LTBI for HCWs working in TB-related health care facilities was higher than Non-TB health care facilities (Table 1).

### Table 1

**Prevalence of latent tuberculosis infection by health care facilities**

<table>
<thead>
<tr>
<th>Variable</th>
<th>Non-TB health care facilities</th>
<th>TB-related health care facilities</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Total</td>
<td>TST positive</td>
</tr>
<tr>
<td>Age group</td>
<td></td>
<td></td>
</tr>
<tr>
<td>20-29</td>
<td>154</td>
<td>72</td>
</tr>
<tr>
<td>30-39</td>
<td>170</td>
<td>88</td>
</tr>
<tr>
<td>40-49</td>
<td>302</td>
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<tr>
<td>50+</td>
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<td>79</td>
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<tr>
<td>Middle</td>
<td>292</td>
<td>146</td>
</tr>
<tr>
<td>High</td>
<td>482</td>
<td>235</td>
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<tr>
<td>Duration of employment (years)</td>
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<td></td>
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<tr>
<td>&lt;1</td>
<td>32</td>
<td>15</td>
</tr>
<tr>
<td>1-5</td>
<td>145</td>
<td>78</td>
</tr>
<tr>
<td>6-10</td>
<td>110</td>
<td>53</td>
</tr>
<tr>
<td>&gt;11</td>
<td>487</td>
<td>235</td>
</tr>
<tr>
<td>Type of health care service</td>
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<td></td>
</tr>
<tr>
<td>Outpatient</td>
<td>399</td>
<td>196</td>
</tr>
<tr>
<td>Inpatient</td>
<td>251</td>
<td>124</td>
</tr>
<tr>
<td>Administrative</td>
<td>24</td>
<td>12</td>
</tr>
<tr>
<td>Intensive care unit Surveillance and monitoring</td>
<td>65</td>
<td>32</td>
</tr>
<tr>
<td>Surveillance and monitoring</td>
<td>22</td>
<td>9</td>
</tr>
<tr>
<td>Other</td>
<td>13</td>
<td>8</td>
</tr>
<tr>
<td>Total</td>
<td>774</td>
<td>381</td>
</tr>
</tbody>
</table>

**Risk factors associated with LTBI**

Some socio-demographic factors including age, sex, marital status, average household monthly income were not significantly associated with LTBI.

The occupational factors found to significantly associated with higher prevalence of LTBI were every day health care service to TB patients (OR=1.9 [95% CI: 1.2 - 2.9]), and had history of active TB among workmates (OR=1.9 [95% CI: 1.3 - 2.9]), working at secondary level of health care service (OR=2.7 [95% CI: 1.7 - 4.3]), working in tuberculosis facility (OR=5.5 [95% CI: 2.9 - 10.7]), working at night shift (OR=1.4 [95% CI: 1.0 - 1.8]).
Univariate analysis showed significant risk factors among living in the same house with close contact family members who have active tuberculosis (OR=4.0 [95% CI: 1.7 - 9.2]) (P<0.001), BCG vaccinated (OR=1.7 [95% CI: 1.0 - 2.8]), smoker (OR=1.6 [95% CI: 1.0 - 1.7]), drinker (OR=1.6 [95% CI: 1.3 - 2.0]).

**Table 2.** Final model for logistic regression analysis of tuberculin reactivity status (n=844)

<table>
<thead>
<tr>
<th>Covariates</th>
<th>OR</th>
<th>95% CI</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Night shift</td>
<td>1.4</td>
<td>1.0 - 1.8</td>
<td>0.04</td>
</tr>
<tr>
<td>Working secondary level</td>
<td>2.9</td>
<td>2.0 - 4.2</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Working in TB sector</td>
<td>6.0</td>
<td>2.8 - 12.6</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Logistic regression analysis of tuberculin skin test status demonstrated that working at tuberculosis facilities, working at secondary level health care service, working in night shift for tuberculin reactivity (Table 2).

**TB disease prevalence**
Chest radiography was performed for 844 (100%) HCWs. After the discordant was resolved by the radiologist, 255 (30.2%) of the 844 chest radiographs were classified as normal, 6 (0.7%) as suggestive of possible active TB and 53 (6.3%) as suggestive of inactive TB and remaining 530 (62.8%) non tubercular lesion.

The overall TB disease prevalence among HCWs was therefore 71/10000. The characteristics of TB occurrence among the HCWs are shown in the Table 3. The prevalence rate of TB was significant higher 714.3 (95% CI 308.9-1565.5) for those working in TB-related health care facilities compared with other health care facilities (P<0.001).

**Table 3.** Characteristics of health care workers with tuberculosis

<table>
<thead>
<tr>
<th>Variable</th>
<th>Total</th>
<th>No of incidence</th>
<th>Prevalence rate*</th>
<th>95% CI</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Health care facilities</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tuberculosis</td>
<td>70</td>
<td>5</td>
<td>714.3</td>
<td>308.9-1565.5</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Non tuberculosis</td>
<td>774</td>
<td>1</td>
<td>12.9</td>
<td>2.3-72.8</td>
<td></td>
</tr>
<tr>
<td>Professional</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.21</td>
</tr>
<tr>
<td>Physician</td>
<td>375</td>
<td>1</td>
<td>26.7</td>
<td>4.7-149.5</td>
<td></td>
</tr>
<tr>
<td>Nurses</td>
<td>469</td>
<td>5</td>
<td>106.6</td>
<td>45.6-247.1</td>
<td></td>
</tr>
</tbody>
</table>

*prevalence rate; per 10000 HCWs

**Clinical characteristics of HCWs who had TB**
Of the 6 HCWs who developed TB, 4 (66.7%) had pulmonary TB, 2 (33.3%) had extra-pulmonary TB (TB pleurisy, born). Sputum acid-bacilli (AFB) smear and bacteriological culture were performed for 4 of the 4 HCWs with pulmonary TB; positive sputum smear were obtained 2, negative sputum smear 2 cases. All patients were treated with standard first line drugs in TB dispensary.

The prevalence of inactive TB among HCWs was therefore 627.9/10000 and prevalence rate of LTBI was 73.6% (95% CI 60.4-83.6). All 53 HCWs with inactive TB were asymptomatic, 27 (50.9%) of them had fibrotic scar, 16 (30.2%) of them calcified nodules as the only abnormality.

**DISCUSSION**
TST described in the 19th century, remains a good tool for the diagnosis of *M. tuberculosis* infection. It is indicated for people who are at risk of infection and progression to active disease, people who would benefit from prophylactic treatment with isoniazid [19]. There is growing awareness about the problem of nosocomial TB and the need to protect healthcare workers in the era of multidrug-resistant and extensively drug-resistant tuberculosis [18].

The prevalence of LTBI and TB disease among HCWs was higher than observed in the national TB prevalence survey that conducted in 1959-1961 in Mongolia [20]. Studies elsewhere showed varying LTBI rates amongst HCWs. Our results from Ulaanbaatar city showed high LTBI rates amongst HCW similar to other studies in low and middle income countries and compared well with an average LTBI prevalence of 54% among HCWs. Our study results are also show an increasing prevalence of LTBI with work location with higher exposure to TB patient’s health care facility.
In our study, working at TB-related health care facilities was associated with a higher LTBI prevalence among HCWs. This study suggests that nosocomial transmission of TB is an important occupational problem among HCWs. The reduction of this risk should be a priority. Occupational contracted TB can lead to the loss of skilled workers, and this can adversely impact on health care services in the long term. Health care workers may also avoid working with TB centers due to the high risk of TB transmission to HCWs. This problem is particularly serious with MDR-TB strains. Implementation of effective TB infection control measures can promote awareness of the disease among HCWs, and help in the adoption of good practices for diagnosis and treatment of TB [21].

In this study, 52.1% of TST positivity was observed among HCWs from hospitals in Ulaanbaatar city. Similar studies involving HCWs described high TST positivity rates, ranging from 26.7%-69.5%. Muzy de Souza (2000) reported 51% of TST positivity in HCWs in a general hospital in Rio de Janeiro, similar to the data of our study [19]. The high proportion of LTBI in HCWs evaluated could also be related to the effect of BCG vaccination. The influence of BCG vaccination on TST was not investigated in the present study because none vaccinated HCWs were not available. In the further studies, another alternative method to TST, such as interferon-gamma release assays (IGRA), could be carried out exclude possible false-positive due to BCG vaccination.

Many risk factors for TB bacillus infection have been reported. The most common are overcrowding, and age, gender, and corticosteroid treatment. In the present study, some of these characteristics were evaluated and not significant association to TST was observed.

This is the first study to examine tuberculin reactivity among Mongolian health care workers. In our study, we observed a higher frequency of TST positivity among HCWs who had been at the job for less than two years (54.7%). In a study conducted by Karen Gisele Person Severo et al. (2010), a higher frequency of TST positivity among HCWs who had been at the job for less than one year (63.6%) [19]. This could represent a previously acquired infection while working as a health care professional or a recently acquired infection during their work in the hospital. However, we cannot be certain of that, as TST is not routinely applied as a pre-employment screening test.

HCWs are essential in the fight against TB and their health needs to be protected as well. The results of our study provide data for hospital infection control committees, safety engineering specialized services and occupational medicine to develop preventive measures to reduce the LTBI rates among their employees.

In a person with a newly acquired LTBI, the lifetime risk to develop reactivation is about 5-10 percent. Such risk estimates vary depending on the immune status of the individual, and gradually decline as the time elapsed since infection increases. It is know that the individuals with radiological lesions suggesting inactive TB have a higher risk of developing TB disease as compared to those with normal chest radiographs [19].

**Limitation**

Our results showed that people with BCG vaccination had higher prevalence of LTBI. BCG vaccination can also lead to positive results of the TST, thus we may have overestimated the LTBI rate in BCG vaccination group. However, a couple of international reviews have reported that the influence of BCG is relatively small in the adult population, especially ≥ 10 years after BCG vaccination.

**CONCLUSION**

The prevalence of latent tuberculosis infection among health care workers in Mongolia was relatively same level of low and middle income counters. TB infection control practices in TB related health care facilities and secondary care should be strengthened in Ulaanbaatar, including administrative measures, renovation of buildings, and use of respirators and masks. Moreover, regular screening of HCW for TB disease and infection needs to be considered. 

**ACKNOWLEDGEMENTS**

The authors would like to thanks the Project Coordination Unit of the HIV and TB project supported by Global Fund in Mongolia for providing funding and support for this study. We would like to express doctors and researchers of the Department of Tuberculosis Surveillance and Research, NCCD who successfully conducted this study and fruitful collaboration with the staff Mongolian Anti Tuberculosis Coalition and lecturers and researchers of the Department of Epidemiology and Biostatistics, SPH, HSUM. We especially would like to thank all the health care workers who participated in the screening for their cooperation.

**REFERENCES**

Plasmid Analysis of ESBL Producing Gram Negative Bacilli in Mongolia


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ABSTRACT

It has been using the many classes of antibiotics and antibacterial activity chemotherapeutic preparation in clinical practice and antibiotic resistance has become a very serious problem for these antibiotics in worldwide. Purpose: To determine molecular mechanism of resistance among ESBL producing Gram-negative bacteria. A total of 32 ESBL producing Gram-negative bacteria were enrolled in this study and antibiotic susceptibility of the clinical isolates was determined by VITEK-1 system. The presence of ESBL was determined using the Double Disc Synergy test (DDST). Strains were screened for 3 types of ESBL gene groups (blaCTX-M-uni (blaCTX-M3), blaTEM, blaSHV) by PCR. Conjugative plasmid analysis was performed by agar mating technique with Rnal recipient E.coli and the number of conjugative plasmid was determined. Among ESBL producing Gram-negative bacilli, 78.1%, 18.8% and 3.1% were K.pneumoniae, E.coli and E.cloaceae, respectively. The production of the ESBL of all strains was confirmed by the Double Disc Synergy test (DDST). Strains were positive for blaCTX-M-uni, blaCTX-M3, blaTEM, blaSHV genes by PCR. The PCR product (blaCTX-M-uni)s were subjected to direct sequencing. All ESBL producing gram negative bacilli were contained 3-4 plasmids (<8 mDa). A total of 78.1% (25/32) of all strains were contained one conjugative plasmid. Aminoglycoside (gentamicin, tobramycin) resistance was transferred through conjugation together with ESBL gene. A total of 32 (100%) ESBL producing gram negative bacilli contained a blaTEM, blaSHV and blaCTX-M3 gene. However, ESBL producing gram negative bacilli were carried 3 to 4 plasmid (<8 mDa)s, only one plasmid was transferable by the conjugation. Aminoglycoside (gentamicin, tobramycin) resistance gene was transferred by conjugation together with ESBL genes.

Key word: ESBL producing GNB, HCWs, colonization, CTX-M, blaTEM, blaSHV

INTRODUCTION

The Family of Enterobacteriaceae is the most important family of bacteria in human medicine, which includes several genera and species that cause disease in community setting and as well as nosocomial infections. Most Enterobacteriaceae are normal component of the gastrointestinal flora of humans and animals, and many of them widespread in the environment. These bacteria can cause different infections, from mild to severe, potentially life-threatening infections such as septicemia, urinary tract infection (UTI)s, pneumonia, cholecystitis, cholangitis, peritonitis, wound infections, meningitis, gastroenteritis and etc. Furthermore, infections caused by these organisms can give rise not only sporadic infections, but also outbreaks.

β-lactam antibiotics are the most common treatment for many bacterial infections including enteric gram negative bacterial infections 1. Production of β-lactamases is the main mechanism of resistance to these classes of antibiotics. Many Gram-negative bacteria possess natural chromosomally mediated β-lactamases and natural β-lactamases have physiologic role in peptidoglycan assembly or may evolved to defend bacteria against β-lactams produced by environmental bacteria and fungi 2. The first acquired or plasmid-mediated β-lactamase was reported in 1965 from an Escherichia coli isolate, and named as a TEM-1, due to the patient’s name Temoniera 3. Since that time plasmid-mediated β-lactamases became common resistance mechanism in staphylococci, enterobacteria, Haemophilus influenzae, and Neisseria gonorrhoeae. These enzymes capable to hydrolyze narrow to broad spectrum beta-lactam antibiotics.

Since 1980’s, the extended-spectrum β-lactams such as third generation cephalosporins is widely used in the treatment of serious infections caused by β-lactamase producing Gram-negative bacteria. But resistance to these
antibiotics emerged quickly in clinical setting 4-8. The first report of plasmid-encoded extended-spectrum ß-lactamase (ESBL) which is capable to hydrolyze the extended-spectrum cephalosporins was published in 1983 9. Now, total number of ESBLs already exceeded 200 (http:www.lahey.org/studies/webt.htm) and these ESBLs are differed from each other by single nucleotide and subsequent single amino acid substitutions 10, 11. ESBLs able to hydrolyze antibiotics containing an ß-lactam ring such as penicillins, first-, second-, and third generation cephalosporins (ceftazidime, ceftriaxone, cefotaxime and etc) as well as oxyimino-monobactam (aztreonam) 4, 7. They are not active against cephemycins and carbapenems and inhibited by ß-lactamase-inhibitors such as clavulanate and tazobactam 4, 7.

ESBLs are very common in many Gram-negative rods, especially in family Enterobacteriaceae and Klebsiella pneumonia and Escherichia coli are the most common ESBL producer worldwide 4. In most instances, ESBL producing GNB infections have clinical manifestations that are similar to infections caused by susceptible bacterial strains. However, treatment options caused by ESBL producing organisms are extremely limited, has increased lengths of stay, costs, morbidity and mortality are associated with ESBLs 12-24.

The ß-lactamases are usually classified according to the Bush-Jacoby-Medeiros functional classification system (ESBL = 2be) or the Ambler structural classification (ESBL = class A) 25, 26. ESBLs can be divided into groups, which are designated the TEM, SHV, CTX-M, Toho, OXA, PER, VEB and other enzymes (http://www.lahey.org/studies/). Among them, TEM (approx. 200 variants), SHV (over 140 variants), CTX-M (approx. 130 variants) types are the most prevalent types of ESBL.

Therefore, plasmid profiling one of the important tool for epidemiological typing. Multiple plasmids carrying ESBLs were seen in bacteria and ESBL carrying plasmid could be endemic in certain area 27-30.

We aimed to study conjugative plasmid of ESBL-producing isolates of Enterobacteriaceae, confirming the transfer of resistance to recipients by antimicrobial susceptibility testing and PCR amplification of the encoding resistance genes. In addition the role of plasmid in co-transfer resistance was investigated.

**MATERIALS AND METHODS**

A total of 32 ESBL producing Gram-negative bacteria were enrolled in this study and antibiotic susceptibility of the clinical isolates was determined by API GN strip or VITEK-1 system (Biomerieux, Marcy L’Etoile, France). 

**BACTERIAL STRAINS**

Thirty two isolates of ESBL producing E. coli (n =6), K. pneumoniae (n =25) and other bacteria (n =1) were enrolled in this study. The strains were chosen from the strain collection of the Central Research Laboratory, Health Sciences University of Mongolia and identified to the species level using the API 20E and/or Vitek (Biomerieux, Marcy L’Etoile, France). All clinical isolates were used as donor or parent strains in conjugation experiments.

Conjugation experiments

Conjugative plasmid analysis was performed by agar mating technique with R<sub>col</sub> recipient E.coli and transconjugants were selected using MacConkey agar plates containing nalidixic acid plus 2 mg/l cefotaxime (Figure 1) 31.

Plasmid extraction

Alkaline lysis method by Sambrook, Russell and plasmid DNA extraction kit (Wizard Plus SV Minipreps. Promega, USA) was used to extract plasmids from the wild strains and their transconjugants 32. Their sizes were estimated by comparison with those of 6 – 8 MD plasmids after electrophoresis.

**Figure 1. Selective agar of the transconjugants (nalidixic acid plus 2 mg/l cefotaxime).**

Antimicrobial susceptibility testing

Antimicrobial susceptibility was tested by the disk diffusion method and broth microdilution test as recommended by Clinical Laboratory Standards Institution (CLSI, USA) (Figure 2) 33.

MICs of the antibiotics for the both donor strains and the transconjugants were determined using the VITEK GNI card (Biomerieux, Marcy L’Etoile, France).

**Figure 2. Antimicrobial susceptibility testing by agar diffusion method according to the CLSI guideline.**
ESBL production was detected with the double-disk synergy test with cefotaxime, ceftazidime, aztreonam, cefepime, and amoxicillin/clavulanic acid disks. The phenotypic confirmatory tests as recommended by the CLSI also were performed.

Polymerase Chain Reaction (PCR)
Types of the ESBL were determined by PCR using the following universal primers (Table 1.). Primers of all genes were used for PCR amplification on the both genomic DNA and extracted plasmids from the wild strain and the transconjugants (Table 2.). PCR products were run on 1.5% agarose gel, stained with ethidium visualized under U.V. light and photographed.

### Table 1. The primer sequences used in this study

<table>
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<th>№</th>
<th>Primer sequences</th>
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<tbody>
<tr>
<td>CTX-M-uni-F</td>
<td>5'-CGA TGT GCA GTA CCA GTA A-3'</td>
<td>19</td>
<td>46.1°C</td>
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<td>CTX-M-uni-R</td>
<td>5'-ATA TCG TTG GTG GTG CC-3'</td>
<td>12</td>
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<td>BLAT-F</td>
<td>5'-ATA AAA TTC TTG AAG ACG AAA-3'</td>
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<td>49.8°C</td>
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<td>BLAT-R</td>
<td>5'-GAC AGT TAC CAA TGC TTA ATC A-3'</td>
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<tr>
<td>CEPH-F</td>
<td>5'-TCA GCG AAA AAC ACC TTG-3'</td>
<td>18</td>
<td>49°C</td>
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<tr>
<td>CEPH-R</td>
<td>5'-TCC CGC AGA TAA ATC ACC A-3'</td>
<td>19</td>
<td>51°C</td>
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<tr>
<td>CTX-M-3a</td>
<td>5'-CCC ATG GTT AAA AAA TCA CTG-3'</td>
<td>21</td>
<td>51.9°C</td>
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<tr>
<td>CTX-M-3b</td>
<td>5'-CCG TTT CCG CTA TTA CAA AC-3'</td>
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### Table 2. PCR condition

<table>
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<th>Denaturation</th>
<th>Annealing</th>
<th>Extension</th>
<th>Final extension</th>
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<tr>
<td>bla SHV</td>
<td>40</td>
<td>94°C-5 min</td>
<td>94°C-1min</td>
<td>43°C-30sec</td>
<td>72°C- 1.15min</td>
</tr>
<tr>
<td>bla TEM</td>
<td>40</td>
<td>94°C-5 min</td>
<td>94°C-1min</td>
<td>46°C-30sec</td>
<td>72°C- 1.15min</td>
</tr>
<tr>
<td>blaCTX-M</td>
<td>30</td>
<td>94°C-5 min</td>
<td>94°C-30sec</td>
<td>54°C-30sec</td>
<td>72°C- 1min</td>
</tr>
<tr>
<td>blaCTX-M-3</td>
<td>30</td>
<td>94°C-5 min</td>
<td>94°C-30sec</td>
<td>55°C-1min</td>
<td>72°C- 1min</td>
</tr>
</tbody>
</table>

### RESULT
Among ESBL producing Gram-negative bacilli, 78.1% (25/32), 18.8% (6/32), and 3.1% (1/32) were K. pneumonias, E.coli and E.cloaceae, respectively. The strains were collected from 1 maternity home (n=20), Maternal and Child Health Research Center (n=11) and First General Hospital (n=1).

The isolates were determined resistant to ampicillin, piperacillin, 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup> generation cephalosporins except cephemycins (cefoxitine). All isolates were susceptible to carbapenems including imipenem and meropenem. From 53% to 87.5% of the isolates were resistant to aminoglycosides including gentamycin and tobramycin. The resistance rate of tigecycline was determined low 3.1% (1/32). Moreover, 28% of the isolates were determined as a resistant to the ciprofloxacin (Table 4).
<table>
<thead>
<tr>
<th>№</th>
<th>Antibiotics</th>
<th>Breakpoint (ug/ml)</th>
<th>Interpretation</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Ampicillin</td>
<td>&gt;32</td>
<td>8&lt;</td>
</tr>
<tr>
<td>2</td>
<td>Ampicillin-sulbactam</td>
<td>&gt;32/16</td>
<td>8/4&lt;</td>
</tr>
<tr>
<td>3</td>
<td>Piperacillin</td>
<td>&gt;128</td>
<td>16&lt;</td>
</tr>
<tr>
<td>4</td>
<td>Piperacillin tazobactam</td>
<td>&gt;128/4</td>
<td>16/4&lt;</td>
</tr>
<tr>
<td>5</td>
<td>Cefazolin</td>
<td>&gt;32</td>
<td>8&lt;</td>
</tr>
<tr>
<td>6</td>
<td>Cefuroxime</td>
<td>&gt;32</td>
<td>8&lt;</td>
</tr>
<tr>
<td>7</td>
<td>Cefoxitin</td>
<td>&gt;32</td>
<td>8&lt;</td>
</tr>
<tr>
<td>8</td>
<td>Cefpodoxime</td>
<td>&gt;8</td>
<td>2&lt;</td>
</tr>
<tr>
<td>9</td>
<td>Cefotaxime</td>
<td>&gt;64</td>
<td>8&lt;</td>
</tr>
<tr>
<td>10</td>
<td>Ceftazidime</td>
<td>&gt;32</td>
<td>8</td>
</tr>
<tr>
<td>11</td>
<td>Cefepime</td>
<td>&gt;32</td>
<td>8&lt;</td>
</tr>
<tr>
<td>12</td>
<td>Imipenem</td>
<td>&gt;16</td>
<td>4&lt;</td>
</tr>
<tr>
<td>13</td>
<td>Meropenem</td>
<td>&gt;16</td>
<td>4&lt;</td>
</tr>
<tr>
<td>14</td>
<td>Gentamicin</td>
<td>&gt;8</td>
<td>4&lt;</td>
</tr>
<tr>
<td>15</td>
<td>Tobramycin</td>
<td>&gt;8</td>
<td>4&lt;</td>
</tr>
<tr>
<td>16</td>
<td>Ciprofloxacin</td>
<td>&gt;4</td>
<td>1&lt;</td>
</tr>
<tr>
<td>17</td>
<td>Levofloxacin</td>
<td>&gt;8</td>
<td>2&lt;</td>
</tr>
<tr>
<td>18</td>
<td>Tigecycline</td>
<td>1</td>
<td>-</td>
</tr>
<tr>
<td>19</td>
<td>SXT****</td>
<td>&gt;8/152</td>
<td>2/38&lt;</td>
</tr>
</tbody>
</table>

Note: R*- resistant, S**- susceptible, I***-intermediate, SXT****- Trimethoprim/sulfamethoxazole

The production of the ESBL of all strains was confirmed by the Double Disc Synergy test (DDST) (Figure 4). All 32 strains were positive for blaCTX-M-uni, blaCTX-M3, blaTEM, blaSHV genes by PCR, respectively (Figure 5, 6, 7, 8).

**Figure 3.** Double disk synergy test of the isolates

**Figure 4.** Gel electrophoresis result of blaCTX-M specific PCR.
Figure 5. Gel electrophoresis result of bla SHV specific PCR.

Figure 6. Gel electrophoresis result of bla TEM specific PCR.

Figure 7. 78.1% (25/32) of all strains were contained one conjugative plasmid (Figure 8) (Table 5).

Figure 8. Gel electrophoresis result of conjugative plasmid

The PCR product (blaCTX-M-uni)s were subjected to direct sequencing and the CTX-M type of the ESBL were confirmed by sequencing analysis.

All ESBL producing gram negative bacilli were contained 2-4 plasmids and conjugative plasmids were determined by agar mating technique. The approximate size of the plasmid extracted from the wild strain were determined <8 mDa (Figure 7).

All transconjugant were checked for cefotaxime and ciprofloxacin resistance and PCR with CTX-M gene specific primer. CTX-M gene was transferred to the transconjugants through conjugation (Figure 9).
<table>
<thead>
<tr>
<th>№</th>
<th>Isolate ID</th>
<th>Hospital</th>
<th>Sample</th>
<th>Species</th>
<th>PCR TEM</th>
<th>SHV</th>
<th>CTX-M-uni</th>
<th>CTX-M3</th>
<th>Conjugation</th>
<th>PCR (CTX-M-uni) from transconjugant</th>
</tr>
</thead>
<tbody>
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<td>1</td>
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<td>-</td>
</tr>
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<td>-</td>
<td><em>K. pneumoniae</em></td>
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<td>-</td>
</tr>
<tr>
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<td><em>K. pneumoniae</em></td>
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<td>Positive</td>
<td>Positive</td>
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<td>-</td>
</tr>
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<td>-</td>
</tr>
<tr>
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<td><em>E.coli</em></td>
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<td>-</td>
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<td>Positive</td>
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<td>-</td>
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<td>Positive</td>
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<td>Positive</td>
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<td>Positive</td>
<td>conjugative</td>
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Table 5: Conjugation test results
### Table 5. Conjugation test results (cont.)

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<th>Hospital</th>
<th>Sample</th>
<th>Species</th>
<th>PCR TEM</th>
<th>PCR SHV</th>
<th>PCR CTX-M-UNI</th>
<th>PCR CTX-M3</th>
<th>Conjugation</th>
<th>PCR (CTX-M-uni) from transconjugant</th>
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<td>Positive</td>
<td>Positive</td>
<td>conjugative</td>
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<tr>
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<tr>
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<td>conjugative</td>
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<td>27</td>
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<td>Positive</td>
<td>Positive</td>
<td>Positive</td>
<td>conjugative</td>
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<tr>
<td>28</td>
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<td>Positive</td>
<td>Positive</td>
<td>Positive</td>
<td>conjugative</td>
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<tr>
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<td>K.pneumoniae</td>
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<td>Positive</td>
<td>Positive</td>
<td>Positive</td>
<td>conjugative</td>
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<td>Ach-10</td>
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<td>environment</td>
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<td>Positive</td>
<td>Positive</td>
<td>Positive</td>
<td>conjugative</td>
<td>Positive</td>
</tr>
</tbody>
</table>
Antibiotic susceptibility were determined for all transconjugants by broth microdilution test and aminoglycoside (gentamicin, tobramycin) resistance was transferred through conjugation together with ESBL gene.

**Table 6.**

**Antibiotic susceptibility test result of transconjugants (MIC)**

<table>
<thead>
<tr>
<th>№</th>
<th>Antibiotics</th>
<th>Breakpoint (µg/ml)</th>
<th>Interpretation</th>
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<tbody>
<tr>
<td></td>
<td></td>
<td>R*</td>
<td>S**</td>
</tr>
<tr>
<td>1</td>
<td>Ampicillin</td>
<td>&gt;32</td>
<td>8&lt;</td>
</tr>
<tr>
<td>2</td>
<td>Ampicillin-sulbactam</td>
<td>&gt;32/16</td>
<td>8/4&lt;</td>
</tr>
<tr>
<td>3</td>
<td>Piperacillin</td>
<td>&gt;128</td>
<td>16&lt;</td>
</tr>
<tr>
<td>4</td>
<td>Piperacillin tazobactam</td>
<td>&gt;128/4</td>
<td>16/4&lt;</td>
</tr>
<tr>
<td>5</td>
<td>Cefazolin</td>
<td>&gt;32</td>
<td>8&lt;</td>
</tr>
<tr>
<td>6</td>
<td>Cefuroxime</td>
<td>&gt;32</td>
<td>8&lt;</td>
</tr>
<tr>
<td>7</td>
<td>Cefoxitin</td>
<td>&gt;32</td>
<td>8&lt;</td>
</tr>
<tr>
<td>8</td>
<td>Cefpodoxime</td>
<td>&gt;8</td>
<td>2&lt;</td>
</tr>
<tr>
<td>9</td>
<td>Cefotaxime</td>
<td>&gt;64</td>
<td>8&lt;</td>
</tr>
<tr>
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<td>Ceftazidime</td>
<td>&gt;32</td>
<td>8&lt;</td>
</tr>
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<td>Cefepime</td>
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<td>Imipenem</td>
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<td>Tobramycin</td>
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<td>4&lt;</td>
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<td>Ciprofloxacin</td>
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<td>1&lt;</td>
</tr>
<tr>
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<td>Levofloxacin</td>
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<td>2&lt;</td>
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<td>-</td>
</tr>
<tr>
<td>19</td>
<td>SXT****</td>
<td>&gt;8/152</td>
<td>2/38&lt;</td>
</tr>
</tbody>
</table>

Note: R*- resistant, S**- susceptible, I***-intermidate, SXT****-Trimethoprim/sulfamethoxazole
DISCUSSION

The most common type of plasmid mediated ESBL in worldwide is CTX-M group. Natural CTX-M enzymes producer is Kluyvera spp and gene coding CTX-M type of b-lactamase was found in the chromosomes of those bacteria, transferred to a plasmid, distributed to other species bacteria. The first isolates with plasmid mediated resistance (acquired) were determined in Japan and Germany in 1986 and 1989, respectively. Our study showed that CTX-M 3 group of ESBLs is also common in Mongolia.

Moreover, our findings suggest that aminoglycoside together with antimicrobials like carbapenem including imipenem, meropenem (susceptible, n=32), flouroquinolones including ciprofloxacin and levofloxacin (susceptible, n=22) and cefoxitin (susceptible, n=29). These groups of antimicrobials are useful antibiotics to treat ESBL producing organisms. However, other study showed high level of resistance to above mentioned group of antimicrobials. Conclusion. The present report is first report which describes the plasmid profiles of ESBL producing strains in Ulaanbaatar, Mongolia. Further results will be available and our study shows that large scale molecular epidemiological study is needed to determine ESBL encoding plasmids circulating in Mongolia.

REFERENCE

Some Cytocine Levels and Microstructure of Brown Adipose Tissue in Marmot and Mouse

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¹School of Bio-Medicine, HSUM
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ABSTRACT

Function of brown adipose tissue (BAT) is maintenance to thermogenesis in hibernators and immunity depression in deep hypothermia are suggested there may be have relation between BAT and immunity. However, according to some researchers’ demonstration, BAT is assist to function of immune activity, but immune cells in BAT is not clear so far. Methods of general and selective histology were used in preparation of microstructure slides of BAT in marmot. BAT CD4+ and CD8+ were counted by Fluorescence-activated cell sorting (FACS) method. Microstructure of BAT in the marmot is showed rich network of blood vessels in lobules of connective tissue, lymphoid tissue accumulation of follicle and blood cells detected in vessels. In this study selected BALB/c model 80-90 day-old healthy male mouse. In the BAT detected total of 31095/μL lymphocytes, as total of lymphocytes CD4+ T cells were 3742/μL (5.20%) and CD8+ T cells were 1868/μL (4.99%), respectively. There have different value of T cell in the peripheral vessel blood and in the BAT is suggested BAT is assist to developing blood cell.

Keywords: brown adipose tissue, immune cell, CD4+, CD8+

INTRODUCTION

Study of brown adipose tissue (BAT) structure and function is started from 1551.¹ Morphologically, BAT cells are composed of typical adipocytes with multilocular distribution of fat and smaller than white adipose tissue. Brown fat cell (BFC) has ball shape and nucleus is located in center of cell.² Location and function of white fat cell is different from BFC. Functionally, the results of last year study are showed BAT is active endocrine tissue which enrolled thermogenesis and lipid metabolism.³⁴ In BFC mitochondria included 50-100 times more than other cells. Uncoupling protein1 (UCP1) and thermogenin’s are special proteins of BAT which related to thermogenesis and located inside of cell membrane.⁶ Stimulation of sympathetic nerve is related to BAT. Norepinephrine is stimulating adenylatecyclase to release lipoproteinlipase which active to lipolysis.⁷

The importance of BAT for the maintenance of body temperature in hibernators and suppression of immune reactions by deep hypothermia suggested a relationship between BAT and immunity.⁴ A few studies in this area have indicated that extracts obtained from BAT of hibernators are capable of suppressing the production of antibody in-vitro. However the studies have demonstrated both in vivo and in non-hibernators the immunosuppressive activity of BAT; it was found that injections of extracts which prepared from rat BAT produced a considerable reduction of immune response. Furthermore, removal of BAT from newborn rats enhanced to a great extent the reactions of cell-mediated immunity.⁸⁻¹⁰

Metabolism of hibernator is exclusive and study of immunosuppressive function is important and necessary to hibernators than non-hibernators. However researchers demonstrated immune function of BAT, there did not have any studies to detect immune cells in BAT so far.

MATERIALS AND METHODS

Our study was performed at Laboratory of Research Training Center, School of Health Technology, HSUM. Disengaged brown fat tissue of marmot was inserted to 96% of alcohol and in 10% formalin for fixation and histology slides were prepared by using general and selective methods. In this study was used BALB/c model, 80-90 days-old healthy male mouse. Detection of CD4 and CD8 was performed by Fluorescence-activated cell sorting (FACS) method.
RESULTS

Our study termed to demonstrate other researchers suggestion related to BAT and immunity. Microstructure of BAT in marmot is showed rich network of blood vessels in lobules of connective tissue, accumulation of follicle with lymphoid tissues and vessels with blood cells surrounded by BAT. Furthermore, in the BAT of mouse was detected CD4+ and CD8+ T cells using standard reagents for FACS method. According to histogram results, in the first sample of brown fat tissue were detected total of 6313/μL lymphocytes in the 1-1 tube and 1347/μL lymphocytes in the 1-2 tube, respectively. The ratio of CD4+ and CD8+ T cells were 369/μL (5.68%) vs 113/μL (5.88%), respectively (Figure 2, Table 1).

![Figure 1. BAT microstructure of marmot. (40x10). A. BAT B. Lymphoid tissue C. Connective septum between lobules](image)

**Table 1.**

<table>
<thead>
<tr>
<th>Samples</th>
<th>BAT Cells connected to CD4 antibody</th>
<th>Cells connected to CD8 antibody</th>
</tr>
</thead>
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<tr>
<td></td>
<td>Lymphocytes (1/μL)</td>
<td>Lymphocytes (1/μL)</td>
</tr>
<tr>
<td>Sample 1-1</td>
<td>6313</td>
<td>1839</td>
</tr>
<tr>
<td>Sample 1-2</td>
<td>1347</td>
<td></td>
</tr>
<tr>
<td>Sample 2-1</td>
<td>60551</td>
<td>18545</td>
</tr>
<tr>
<td>Sample 2-2</td>
<td>31095</td>
<td></td>
</tr>
</tbody>
</table>

![Figure 2. CD4+T, CD8+T cells in the BAT of healthy mouse](image)

![Figure 3. CD4+T and CD8+T cells in BAT of healthy mouse](image)

In the second sample of brown fat tissue were detected total of 60551/μL in the 2-1 tube and 31095/μL lymphocytes, respectively. The ratio of CD4+ and CD8+ T cells were 3746/μL (5.20%) vs 1868/μL (4.99%), respectively (Figure 3, Table 1).

**DISCUSSION**

In this study, relation between immunity and BAT, spleen of adipectomized rat showing normal density of lymphocytes in periarteriolar white pulp, but thymo-adipectomized rat showing lack of periarteriolar lymphocytes. Histology of axillary lymphoid nodes showed adipectomized rat dense lymphocytes in paracortical area and thymo-adipectomized rat with scattered lymphocytes in paracortical zone. This function is may be related to activation of site reaction in the deep hypothermia. Suppression of antibody associated to immunoglobulin synthesis related to low regulation of metabolism. Furthermore, neonatal and 21 days-old rats that repeated injections of extracts from BAT cold-adapted rats, the formation of antibody was not greatly affected. This findings are in complete agreement with the results presented here which show that neonatal adipectomy enhances the reactions of cell-mediated immunity while leaving intact the production of antibody against BSA an SRBC. Metabolism of hibernator is exclusive and study of immunosuppressive function is important and necessary to hibernators more than non-hibernators.

The discrepancy between the humoral and cell-mediated immune response in adipectomized rats,
the neonatal excision of the interscapular brown adipose tissue represents, in fact, an incomplete adipectomy, which leaves intact other deposits of BAT, so that the amount of the tissue in adipectomized rats is still sufficient to allow a normal production of antibody. Reasoning of this kind would imply that in non-hibernators the mechanisms which underlie cell-mediated immunity is more sensitive to the lack of BAT than the mechanisms primarily involved in humoral immunity. 

Accumulation of lymphocytes in the center of BAT in marmot, similar to young of a marmot of hamster BAT is demonstrated by study of Dagdanbazar B et al (1990). Interestingly, the study results were showed that BAT in young of a marmot of hamster smooth muscle layer of vessels in BAT was more developed than marmot. However, accumulation of lymphocytes in the BAT of marmot is reach to connective tissue and lymphocytes, we could not detect T cell and B cell.

We had been agree to other researchers suggestion which suggested T lymphocytes (cell-mediated immunity) developed in the thymus and B lymphocytes (humoral) may be developed in the BAT. Khongorzul B. et al study showed percentage of CD4+ T and CD8+ T cell in the peripheral vascular blood of mouse. This study and our study results showed CD4+ and CD8+ T cells percentage is different in blood and tissue. There do not have enough study to compare generation of lymphocytes in the BAT.

Conclusions

In conclusion, BAT detected rich network of blood vessels in lobules of connective tissue, accumulation of follicle with lymphoid tissues with vessels which included blood cells. Detection of CD4+ T cell and CD8+ T cell in the BAT of mouse suggests that brown fat is involved to immune system.

References

Some Issues of Antihypertensive Medication Nonadherence in Mongolia

Burmaa.B1, Otgonbayar.R1, Chimedsuren.O2, Ser-od.Kh2
1 School of Medicine, Health Sciences University of Mongolia
2 School of Public health, Health Sciences University of Mongolia

ABSTRACT

In Mongolia, 1 out of 3 deaths are caused by CVD which is leading cause of mortality and estimated 6000-6500 deaths caused by CVD annually. Failure to adhere to antihypertensive medication regimes will increase the risk of complications. According to the WHO survey, 50-70% of hypertensive patients don’t follow the medication regimes as doctor suggested. In Mongolia, hypertension prevalence is increasing therefore data on the antihypertensive medication adherence is needed. We conducted cross sectional study and selected 1676 people who were randomly chosen by registrations of family practitioner. As international standard, uncontrolled hypertension includes those who are unaware of their hypertension, those who are aware but not treated with medication, and those who are aware and treated with medication but still have uncontrolled hypertension.

The prevalence of uncontrolled hypertension was 68.2%. In the uncontrolled hypertensive group, 6.2% were unaware that they were hypertensive, 27.5% were aware that they were hypertensive but were untreated and 56% were aware of their hypertension and treated but were not controlled with their current medical regimen.

Also the result shows that 60.9% of doctors counseled about significance of BP home control, 65.4% of doctors counseled about reducing salt intake, 67.6% of doctors counseled benefits of reducing animal derived or saturated fat, 53.6% of doctors counseled of physical activity benefits, 21.2% of doctors counseled of smoking cessation and 24% of doctors counseled of reducing alcohol consumption.

Poor medication adherence considered as possible underlying cause of uncontrolled hypertension which is further negatively influencing hypertension health care.

Keywords: Arterial hypertension, uncontrolled hypertension, antihypertensive medication nonadherence

INTRODUCTION

Hypertension, heart attack and stroke are the main causes of cardiovascular mortality. Untreated hypertension predisposes coronary heart disease, congestive heart failure and stroke. According to the WHO studies, 13% of total mortality in worldwide is caused by the arterial hypertension [1]. Since 1980, the urbanization, high prevalence of risk factors, lifestyle changes and epidemiological transition are contributing to the increase of non communicable diseases (NCDs) in developing countries and the associated rate is steadily increasing [2-12].

In Mongolia, 1 out of 3 deaths are caused by CVD which is leading cause of mortality and estimated 6000-6500 deaths caused by CVD annually [12].

Failure to adhere to antihypertensive medication regimes will increase the risk of complications. According to the WHO survey, 50-70% of hypertensive patients don’t follow the medication regimes as doctor suggested [13]. In Mongolia, hypertension prevalence is increasing therefore data on the antihypertensive medication adherence is needed.

This study surveyed the prevalence of uncontrolled hypertension in four Mongolian Aimags (provinces: Orkhon, Dornod, Khovd and Dornogovi) in order to compare regions and determine the prevalence of hypertension control and assess reasons for non-compliance with medical treatment.

MATERIALS AND METHODS

We conducted cross sectional study and selected 1676 people who were randomly chosen by registrations of family practitioner. Age was restricted persons 18 – 69 years old. We chose four aimags from each of 4 geographical regions.
in Mongolia. The survey methodology was approved by the Medical ethics committee at the HSUM and the permission to carry out the survey was granted by the order #14-1/1B of July 22nd, 2011. As international standard, uncontrolled hypertension includes those who are unaware of their hypertension, those who are aware but not treated with medication, and those who are aware and treated with medication but still have uncontrolled hypertension.

The Mongolian health care system is largely government run especially in the outside of the city of Ulaanbaatar which is the only major urban area. Public facilities are classified by three major levels: Level I representing primary care hospitals and clinics staffed by primary care physician. These facilities are called soum and inter-soum hospitals in the aimags. Level II or secondary hospitals are called aimag general hospitals in the aimags. Level III facilities are considered to be specialized professional centers in the regions and have some level of tertiary care. As a part of our survey, we compared the completeness of counseling by medical professionals at each hospital/clinic level.

Medical treatment adherence was assessed by the Morisky Medication Adherence Score. Blood pressure was measured on both arms by the National Hypertension Guideline as blood pressure measured on both side and highest reading was taken. Statistical analysis was performed by SPSS 20 statistical software.

**RESULTS**

Out of the total 1676 participants, 550 people’s blood pressure was within normal range and 1126 people were hypertensive.

The prevalence of uncontrolled hypertension was 68.2%. In the uncontrolled hypertensive group, 6.2% were unaware that they were hypertensive, 27.5% were aware that they were hypertensive but were untreated and 56% were aware of their hypertension and treated but were not controlled with their current medical regimen.

We have investigated some influencing factors to the uncontrolled hypertension, the result shows that 60.9% of doctors counseled about significance of BP home control, 65.4% of doctors counseled about reducing salt intake, 67.6% of doctors counseled benefits of reducing animal derived or saturated fat, 53.6% of doctors counseled of physical activity benefits, 21.2% of doctors counseled of smoking cessation and 24% of doctors counseled of reducing alcohol consumption.

<table>
<thead>
<tr>
<th>Doctor counseling by healthcare level. (by percentage)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Answer</strong></td>
</tr>
<tr>
<td>--------------------------------------------------------</td>
</tr>
<tr>
<td>BP home control</td>
</tr>
<tr>
<td>yes</td>
</tr>
<tr>
<td>no</td>
</tr>
<tr>
<td>Reducing salt intake</td>
</tr>
<tr>
<td>yes</td>
</tr>
<tr>
<td>no</td>
</tr>
<tr>
<td>Reducing saturated fat intake</td>
</tr>
<tr>
<td>yes</td>
</tr>
<tr>
<td>no</td>
</tr>
<tr>
<td>Physical activity</td>
</tr>
<tr>
<td>yes</td>
</tr>
<tr>
<td>no</td>
</tr>
<tr>
<td>Smoking cessation</td>
</tr>
<tr>
<td>yes</td>
</tr>
<tr>
<td>no</td>
</tr>
<tr>
<td>Alcohol reduction</td>
</tr>
<tr>
<td>yes</td>
</tr>
<tr>
<td>no</td>
</tr>
</tbody>
</table>

Table 1.
Among the uncontrolled hypertensive patients (reasons for stopping are not exclusive) 71.7% had stopped medications when their symptoms resolved, 61.2% admitted to forgetting to obtain their medication, 66.4% do not obtain their medication regularly and 60.5% stopped their medication when other complaints appear.

Figure 1. Adherence to medical treatment regimen

Other reasons for nonadherence to the medical treatment regimen were: a) the doctor did not suggest to use medication regularly 38.6%, b) doctor suggested multiple drugs 20.2%, c) the patient was unable to purchase the drug 14.0 and d) 12.1% were felt discomfort or other side-effect while using medication.

**DISCUSSION**

According to the “2009 Mongolian STEPS Survey on the prevalence of non communicable disease and injury risk factors” result shows that aware, untreated hypertension prevalence were 61.1% and treated, uncontrolled hypertension prevalence were 25.6% of patients using medication but which is not effective. Our result of aware, untreated hypertension level were lower than STEPS survey result but treated, uncontrolled hypertension prevalence were lower than previous study[14].

Tsolmon U, Naranchimeg S et all carried out survey among Ulaanbaatar city population, according to their result nonadherence of medical treatment regimen was 68.3 %[15], which was close to our result. This result shows antihypertensive medication nonadherence is quite high throughout Mongolia, therefore we need to improve community health education.

By the comparison of international researchers, medical regimen adherence was 52-74%, but nonadherences were 12-57%, these results were close to our results. Marta Pereira et all made meta analysis on 44 studies performed in 35 countries, as a result aware, untreated hypertension level were between 34-89%[16]. According to our result aware, untreated hypertension level were 56% which was close to previous study.

**CONCLUSION:**

Uncontrolled hypertension prevalence was 68.2%, which shows we need to improve healthcare quality of arterial hypertension. The physician did not suggest using medication regularly for 38.6% of uncontrolled hypertension patients. These factors might be influencing to hypertension healthcare service negatively.

**REFERENCE**

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Some Issues on Arterial Hypertension Health Care Service in Mongolia

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²Health Sciences University of Mongolia, School of Public health

ABSTRACT
Since arterial hypertension has high prevalence in country, we need to emphasize some issues of health care service of the arterial hypertension. Our goal was to study current situation of arterial hypertension health care service in Mongolia. Using a cross sectional study design, we have recruited 87 doctors from health care centers and hospitals in 5 aimags (Orkhon, Dornogovi, Uvurkhangai, Dornod and Khuvd) and Ulaanbaatar. We used a questionnaire composed of 7 chapters with 80 questions.

26.5-50% of family, soum and 31.6% of internist and cardiologist answered as they are not able to communicate with their patients because of the working condition is inconvenient. Non depending with the health care service level, almost half of the doctors (39-47.0%) couldn’t check up blood pressure by the standard. Counselling on changeable risk factors such as smoking cessation, 66.7% of soum health care center doctors, 50% of cardiologist and 65.1% of internist were regularly performing, there were statistically significant differences between these three groups (p>0.05). Of the respondents, 39.7 of physicians counselled that regularly lifetime use of antihypertensive medications for the patient with the 160/100 mm.Hg blood pressure. Due to the nomadic lifestyle, 51.3% of physician mentioned that transportation petrol insufficiency is the main difficulty on arterial hypertension control. Health care centers human resource, financing and working load difficulties negatively influencing on hypertension healthcare service. Non depending from the health care service level physicians are inadequately providing hypertension control and medical treatment, therefore we need to organize re-training among doctors.

Key word: arterial hypertension, health care service, Mongolia

INTRODUCTION
CVDs are the number one cause of death globally: more people die annually from CVDs than from any other cause. Low- and middle-income countries are disproportionally affected: over 80% of CVD deaths take place in low- and middle-income countries and occur almost equally in men and women. 7.5 million deaths each year, or 13% of all deaths can be attributed to raised blood pressure. This includes 51% of deaths due to strokes and 45% of deaths due to coronary heart disease. According to the Mongolian Health Statistics, the cardiovascular disease mortality was 7.1% in 1951, 23.4% in 1985, 30.8% in 1995 and in 2000s. It has increased into the 38%. Recent surveys show that hypertension, heart attack and stroke are dominating in the cardiovascular mortality. Since 1980, the urbanization, high prevalence of risk factors, lifestyle changes and epidemiological transition are leading the way to increasing of non communicable diseases in developing countries. Mortality rate is steadily increasing. Since arterial hypertension has high prevalence in country, we need to emphasize some issues of health care service of the arterial hypertension.

Goal:
To study current situation of arterial hypertension health care service in Mongolia

Objectives:
1. To study some impacting factors for the hypertension health care service
2. To emphasize problems and difficulties facing on arterial hypertension health care service

MATERIALS AND METHODS:
Using a cross sectional study design, we have recruited 87 physicians from family and soum health care centers and provincial and central hospital in 5 aimags (Bulgan, Dundgovi, Uvurkhangai, Dornod and Khuvd). Out of the 87 doctors, 58.6% were involved from the II, III health care service level and 41.4% were from the family and soum health care centers. We used a questionnaire composed of 7 chapters with 80 questions. The survey methodology was discussed and approved by the Academic Council of School of Public Health. The survey methodology was approved by the

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Medical Ethical Branch Committee at the Health Sciences University of Mongolia.

**RESULT:**
Of the doctors involved in this survey, 69.2% responded that they have a convenient physical location or room supply communicating with the patients and time load is acceptable. Even though 26.5-50% of soum and family doctors and 31.6% of provincial physicians answered as they are not able to communicate with their patients because of the working condition is inconvenient. In addition, 22.22% of family doctors, 42.86% cardiologist and 37.5% of internist answered as they do not ask about the CVD risk factors because of the time limitation per patient. Same on each health care service level, a physician examine 40-60 patients per day.

<table>
<thead>
<tr>
<th>Availability to ask CVD risk factors</th>
<th>I level</th>
<th>II level</th>
<th>III level</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n</td>
<td>%</td>
<td>n</td>
<td>%</td>
</tr>
<tr>
<td>Yes</td>
<td>28</td>
<td>77.78</td>
<td>25</td>
<td>62.50</td>
</tr>
<tr>
<td>No</td>
<td>8</td>
<td>22.22</td>
<td>15</td>
<td>37.50</td>
</tr>
<tr>
<td>Total</td>
<td>36</td>
<td>100.0</td>
<td>40</td>
<td>100.0</td>
</tr>
</tbody>
</table>

Counselling on changeable risk factors such as smoking cessation, 66.7% of family and soum healthcare center doctors, 50% of cardiologist and 65.1% of internists were regularly performing, there were statistically significant differences between these three groups (p>0.05). 58.5-64.7% of doctors have provided by the sphygmomanometer which can meet with the standard.

Even doctors communication skill is satisfied (82.4-100%) 75.6% of family and soum health care center doctors and 94.1% of internists have talked about the CVD risk factors. 36.6 of soum health care centers didn’t check up blood pressure in both side. Non depending with the health care service level, almost half of the doctors (39-47.0%) couldn’t check up blood pressure by the standard. 29.3-41.2% were checked BP twice. 91.7 of doctors answered correctly regarding the right combination of antihypertensive medications. Regarding the counselling skill aimag doctors skill were lower than soum doctors, especially insufficient (64.7 percent) counselling on alcohol, smoking and physical activity. Counselling on changeable risk factors such as smoking cessation, 66.7% of family and soum health care center doctors, 50% of cardiologists and 65.1% of internists were regularly performing, there were statistically significant differences between these three groups (p>0.05).

Of the respondents, 39.7 of doctors counselled that regularly lifetime use of antihypertensive medications for the patient with the 160/100 mm.Hg blood pressure. Which shows that uncontrolled hypertension might be caused by the doctors inappropriate counselling of antihypertensive medications.

Regarding the arterial hypertension control, 55.6-54.3% were performed by planned control (dispansery), 8.6-15.6% were check up on call, 17.8-22.9% were outpatient care and 11.1-14.3% were performed by active check up.
Figure 2 shows that there were no significant differences in hypertension control types of health care service levels (p>0.05). Most of the doctors use a planned control method for hypertension control. There has any hypertension registering and controlling system in hospitals. Table 2 shows frequency of hypertensive patient check up.

### Table 2.

<table>
<thead>
<tr>
<th>Frequency</th>
<th>II level n</th>
<th>II level %</th>
<th>I level n</th>
<th>I level %</th>
<th>Total n</th>
<th>Total %</th>
</tr>
</thead>
<tbody>
<tr>
<td>7-14 days</td>
<td>33</td>
<td>68.75</td>
<td>18</td>
<td>54.55</td>
<td>51</td>
<td>62.96</td>
</tr>
<tr>
<td>14-30 days</td>
<td>9</td>
<td>18.75</td>
<td>3</td>
<td>9.09</td>
<td>12</td>
<td>14.81</td>
</tr>
<tr>
<td>Once per month</td>
<td>3</td>
<td>6.25</td>
<td>11</td>
<td>33.33</td>
<td>14</td>
<td>17.28</td>
</tr>
<tr>
<td>Twice per month</td>
<td>3</td>
<td>6.25</td>
<td>1</td>
<td>3.03</td>
<td>4</td>
<td>4.94</td>
</tr>
</tbody>
</table>

Total 48 100.0 33 100.0 81 100.0

Regarding the question “when will you follow up if patient had high blood pressure 140/90-159/99 mm.Hg”, of the respondents 62.96% were answered as within 7-14 days, 14.8% responded as within 14-30 days and 17.28% stated that should be checked in once per month. Out of the doctors, 72.6% answered as blood pressure should be checked in every month if patients blood pressure is higher than 160/100 mm.Hg, there has any statistically significant differences between the hospital level (70.9%-74.3%).

According to the Adult Hypertension Guideline of Mongolia in 2010, when the BP is 140/90-159/99 mm.Hg, the patient should be checked once in 2 months and when the BP is 160/100 – 179/109 mm.Hg, the patients should be checked once a month. These findings shows that, there has not enough adherence to the guidelines.

In addition, 37.4-68% of the total respondents were defined as the main reason of the intact high prevalence of changeable risk factors are caused by the health education is low in general population,14-40.7% were lack of community support for changing behavior and 12-14.8% were considered as there has insufficient training for the community.

Due to the nomadic lifestyle, 51.3% of physician mentioned that transportation petrol insufficiency is the main difficulty on arterial hypertension control. Also 71.4% of physician mentioned equipment insufficiency, 42.9-66.7% of respondents mentioned that there has human resource deficiencies in countryside hospital.

### Table 3.

| Arterial hypertension health care service problems /percentage/ |
|--------|----------------|----------------|----------------|--------|
|        | SHC / FHC n | %*            | Central  hospital n | %*       | Provincial / Aimag hospital n | %*       | Total n | %*       |
| Criteria                                    |
| Human resource insufficiency                | 20 | 57.1          | 3 | 42.9        | 24 | 66.7       | 47 | 60.3     |
| Medical equipments insufficiency           | 25 | 71.4          | 5 | 71.4        | 17 | 47.2       | 47 | 60.3     |
| Transportation budget insufficiency       | 19 | 54.3          | 5 | 71.4        | 16 | 44.4       | 40 | 51.3     |
| Early detection equipments budget insufficiency | 23 | 65.7          | 5 | 71.4        | 23 | 63.9       | 51 | 65.4     |
| People live far away from the hospital   | 14 | 40            | 4 | 57.1        | 8 | 22.2       | 26 | 33.3     |
| People move often                        | 10 | 28.6          | 2 | 28.6        | 9 | 25         | 21 | 26.9     |
| Not limited                                | 0  | 0             | 0 | 0           | 2 | 5.6        | 2  | 2.6      |
DISCUSSION

The survey result carried out in Germany showed that 200 patients were seen by physicians per week. Due to these hyperload, physicians mentioned that there has time limitation for giving prompt counselling on lifestyle changes. These result was same as our result. Sarah L. Cutrona et al proved that hypertension control is not only doctors job, for achieving adequate control need to work as team. Especially, nurse role was significant on hypertension control. In Mongolia, we should have to increase nurse and social workers involvement on hypertension control.

In 2004, the survey carried out in Australia shoed that hypertension guideline adherense were 51.8%, even 75% of physicians mentioned that lifestyle change has significant role besides antihypertensive medication, there has only 12% of patients were regularly providing healthy lifestyle counselling, which was lower than our result. In USA, the survey carried out among 5400 patients whether they have counselled of healthy lifestyle or not, the result show only one fourth of the patients get the healthy lifestyle counselling. Our result shows 60.6 percent of lifestyle counselling, which is higher than previous survey. Our result shows that there has 40% of guideline non-adherense. The survey carried out in South Africa shows physicians don’t check all patients blood pressure, even measured sometimes perform on only side or measure only time. Also there has unsusage of home measurements on antihypertensive medication selection or effectiveness.

The survey carried out in South Africa shows that doctors have insufficient knowledge of antihypertensive medication indication, combination and incorrect medication selections on different diseases such as systolic blood pressure, myocardial infarction and chronic kidney disease. The knowledge of physicians medical treatment indication, combination was relatively higher than these survey, which may justify that there has more clinical practice usage.

Some countries such as Germany, physicians re-training level was 42.4% or average, regarding nurses and other medical specialists (nutritionist, social worker, psychologist) re-training level was 21.2%. In our country there medical specialists -re-training level is inadequate.

CONCLUSION

Healthcare centers human resource, financing, physical condition and working load difficulties negatively influencing on hypertension health care service. Non depending from the hospital level physicians are inadequately providing hypertension control and medical treatment, therefore we need to organize re-training among doctors.

REFERENCE

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Study of Pharmacological Activity of Traditional Mongolian Drug Garidi-5

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1Traditional Medical Science, Technology and Production Corporation of Mongolia
2Health Sciences University of Mongolia
3National University of Mongolia, School of Chemistry, Chemical Engineering

ABSTRACT
Garidi-5, a traditional medicine composed of 5 herbs including Terminalia chebula Retz., Aconitum Kusnezoffii Reichb., Acorus calamus L., Saussurea lappa L., and musk of Moschus moschiferus, has been used in traditional Mongolian medicine as an analgesic and antibacterial medicine.

Bioactive compounds of Garidi-5 were determined by the Folin–Ciocalteu method. An antioxidant potential of the methanol extract of Garidi-5 was evaluated by the 1,1-diphenyl-2-picrylhydrazyl (DPPH) scavenging assay. Writhing was induced in mice by an intraperitoneal injection of 0.6 % acetic acid solution (10 ml/kg). Water extract of Garidi-5 was given orally at concentrations of 20 mg/kg, 80 mg/kg and 200 mg/kg. Aspirin was used as a standard medicine (100 mg/kg).

Gallic acid, α-azarone, costunolide, alkaloids and testosterone were detected in Garidi-5. Methanol extract of Garidi-5 showed the highest (95.11%) DPPH radical scavenging activity at concentration of 0.05 µg/ml. Moreover, Garidi-5 significantly reduced the number of writhes induced by acetic acid in mice by 40.4-47.9% suggesting that it has peripheral antinociceptive effect.

Garidi-5 (P < 0.05) inhibited carrageenan induced hind rat paw edema.

Conclusion: Biological activity substances were detected in the Traditional drug Garidi-5. Traditional drug Garidi-5 has low toxicity, anti-inflammatory and analgesic effects.

Key words: Garidi-5, antioxidant, antinociceptive effect, Anti-inflammatory; Carrageenan;

INTRODUCTION
Garidi-5 has been used in traditional Mongolian medicine as an antibacterial and analgesic agent for treatments of various diseases including typhus, diphtheria, joint conditions, neurological and skin disorders. Garidi-5 is composed of 5 herbs including Terminalia chebula Retz., Aconitum Kusnezoffii Reichb., Acorus calamus L., Saussurea lappa L., and musk of Moschus moschiferus. As written in traditional medical sourcebooks the compounds of Garidi-5 mainly balance humors, detoxify the body, kill bacteria and relieve pain.

A variety of bioactive substances have been detected in the components of Garidi-5. Terfalvin B and gallic acid have been found in Terminalia chebula Retz. Diterpene alkaloids and polysaccharides have been isolated from the root of Terminalia chebula. Saussurea lappa L. is rich in sesquiterpenoid lactones and terpenoids.

In the present study, antioxidative and antinociceptive activities of Garidi-5 were examined. Phytochemical screening was also performed to determine the presence of some bioactive components including gallic acid, total alkaloids, α-azarone, costunolide, and testosterone.

MATERIALS AND METHODS
Animals. Specific pathogen-free white mice (20-30 g) and wistar rats (180-220 g) were used for the study and all were housed in a quiet room with 12 h light/dark cycle. The study protocol was approved by the Ethical Committee of the Health Sciences University of Mongolia and the care and handling of animals were in accordance with the principles of the Helsinki Declaration.
Material. Garidi-5 produced in the Traditional Drug Manufacture of Traditional Medical Science, Technology and Production Corporation of Mongolia, was used in this study.

Detection of bioactive compounds. Garidi-5 and Terminalia chebula were extracted in 70 % ethanol for 24 h. These extracts and gallic acid standard were analyzed by a thin layer chromatography (TLC). Gallic acid was separated using a benzene-ethylacetate-formic acid-acetone (5:5:2:0.5) solvent system and detected by spraying with 3 % FeCl3, with following dark blue spot [9]. Garidi-5 and Aconitum kusnezoffii were extracted with diethyl-ether plus ammonia solution. Total alkaloids were determined in a TAMA - toluene-acetone-methanol-ammonia (49.5:41.5: 8:5) solvent system by spraying with Dragendorff reagent with following orange spots.

Garidi-5 and Acorus calamus ethanol extracts were prepared to detect α-azarone. In a n-hexane-acetone (10:2) solvent system α-azarone was detected with an iodine giving blue spot.

Costunolid from Saussurea lappa in Garidi-5 was detected a toluene-ethylacetate (9.5:0.5) solvent system by spraying with a vanillin sulfuric acid reagent giving crimson fluorescence

Garidi-5 and musk of Moschus moschiferus were extracted in acetone-benzene (4:1) mixture. Chloroform-acetone solvent system (9:1) and vanillin sulfuric acid reagent were used to determine testosterone.

DPPH radical scavenging activity assay. The free radical scavenging activity of fractions was measured in vitro by a DPPH assay as described in the literature [6]. Garidi-5 was extracted in methanol for 24 h at room temperature. Aliquots (2.5 ml) of the Garidi-5 extract at various concentrations (10-50 mg/ml) were mixed with 1 ml of methanol solution of 3x10^-4 M DPPH. The control was prepared as above without Garidi-5. The reaction mixture was shaken well and incubated in the dark for 30 min at room temperature. Absorbance of sample and control solutions was determined at 517 nm using spectrophotometer. The scavenging activity was estimated based on the percentage of DPPH radical scavenged as the following equation:

\[
\text{Scavenging effect} = \frac{\text{control absorbance-sample absorbance}}{\text{control absorbance}} \times 100
\]

Acetic acid-induced writhing test. The test was carried out as described previously. Mice were fasted overnight and divided into 6 groups with six animals in each group. Separate groups of animals received vehicle, Garidi-5 (20 mg/kg, 80 mg/kg, and 200 mg/kg) or aspirin (100 mg/kg) 1 h before intraperitoneal injection of acetic acid (0.6 %, 10 ml/kg). The pain response was recorded for 20 min, starting immediately after acetic acid administration.

Carrageenan induced rat paw edema. The method of Winter et al. (1962) was used to study acute inflammation. Wistar rats in groups of five each were treated with vehicle, Garidi-5 (30, 80 and 200 mg/kg, p.o.) and Indometacin (10 mg/kg) one hour prior to Carrageenan injection. 0.1 ml of 1% Carrageenan was injected into the subplantar tissue of left hind paw of each rat. Swelling of carrageenan injected foot was measured at 0, 0.5, 1, 2, 4 h using Plethysmometer (UGO Basile, Italy) (Vogel, 2002). The right hind paw was injected with 0.1 ml of vehicle.

RESULTS

Bioactive compounds. Gallic acid, total alkaloids, α-azarone, costunoloid, and testosterone, were determined in Terminalia chebula, Aconitum Kusnezoffii, Acorus calamus, Saussurea lappa, and musk of Siberian musk deer respectively. All these bioactive compounds except testosterone were also detected in Garidi-5 (Table-1).

<table>
<thead>
<tr>
<th>Sample</th>
<th>Solvent system</th>
<th>Detect substances</th>
<th>Detect blot</th>
<th>R&lt;sub&gt;1&lt;/sub&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Garidi-5,</td>
<td>benzene-ethylacetate-formic acid-acetone (5:5:2:0.5)</td>
<td>gallic acid</td>
<td>dark blue color fluorescence</td>
<td>0.81</td>
</tr>
<tr>
<td>Terminalia chebula</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Garidi-5,</td>
<td>toluene- acetone -methanol-ammonia (49.5:41.5: 8:5)</td>
<td>alkaloid</td>
<td>orange color fluorescence</td>
<td>1.0</td>
</tr>
<tr>
<td>Aconitum kusnezoffii</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Garidi-5,</td>
<td>hexane-acetone (10:2)</td>
<td>α-azarone</td>
<td>blue color fluorescence</td>
<td>0.34</td>
</tr>
<tr>
<td>Acorus calamus</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Garidi-5,</td>
<td>toluene-ethylacetate (9.5:0.5)</td>
<td>costunolide</td>
<td>crimson color fluorescence</td>
<td>0.48</td>
</tr>
<tr>
<td>Saussurea lappa</td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Moschus moschiferus</td>
<td>chloroform-acetone (9:1)</td>
<td>testosterone</td>
<td>bluish violet color fluorescence</td>
<td>0.87</td>
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</table>

Table 1. Bioactive compounds of Garidi-5 and its components
By our pharmacological study Garidi-5 LD$_{50}$ = 2.35 ± 0.1 which shows low toxicity by Sidrov’s classification.

**DPPH radical scavenging activity of Garidi-5.** The highest (95.11%) DPPH radical scavenging activity of methanol extract of Garidi-5 was determined at a concentration of 0.05 µg/ml. This result indicates that Garidi-5 may possess a high anti-oxidative activity.

**Effect of Garidi-5 on acetic acid-induced writhing in mice.** Garidi-5 (20 mg/kg, 80 mg/kg, and 200 mg/kg) significantly (p<0.05) reduced abdominal writhes induced by acetic acid in mice by 40.4-47.9% (Figure 1). This result suggests that the water extract of Garidi-5 possesses significant peripheral analgesic effect.

**Carrageenan induced rat paw edema**

The Garidi-5 (20, 80 and 200 mg/kg) significantly (P < 0.05) inhibited carrageenan induced rat paw edema as compared to control group. Maximum inhibition of paw edema was observed with Garidi-5 (80, 200 mg/kg) at 4 h when compared to the control group. Aspirin inhibited paw edema by 49.54%. The observations are given in Table 2.

**DISCUSSION**

The preliminary phytochemical screening of Garidi-5 showed the presence of gallic acid, alkaloids, α-azarone, costunolide and testosterone in our laboratory. Table 1. This study was performed to evaluate the putative anti-inflammatory and antinociceptive activities of Garidi-5, using various animal models to clarify the pain and inflammation relieving effects.

The classification of antinociceptive drugs is usually based on their mechanism of action either on the central nervous system or on the peripheral nervous system. Thus, both the acetic acid-induced abdominal constriction and hotplate methods were used to analyze peripheral and central activity, respectively. It has been proposed that acetic acid acts indirectly by inducing the release of endogenous mediators which stimulate the nociceptive neurons sensitive to nonsteroidal anti-inflammatory drugs and opioids. Acetic acid-induced writhing is a visceral pain model which, despite its low specificity, has been widely used for the evaluation of peripheral antinociceptive activity because of its high sensitivity. Oral administration of Garidi-5 significantly reduced the number of writhings induced by acetic acid in mice (Fig. 1) and the activity was comparable to that observed with 100 mg/kg Aspirin (used as a reference drug, po). Figure 1 shows that Garidi-5 inhibited acetic acid-induced abdominal contractions in mice, thereby exhibiting an antinociceptive effect.
The injection of Carrageenan into mice produces a typical biphasic edema associated with the production of several inflammatory mediators, including bradykinin, prostaglandins and cytokines. The first phase peaks at 3 h and the delayed phase peaks at 48 h after Carrageenan injection.\(^{15,\ 16}\) Development of edema induced by Carrageenan is commonly correlated with the early exudates stage of inflammation, which is one of the important processes of inflammatory pathology.\(^{17,\ 18}\)

It was found that the injection of Carr into the mice paw induced the liberation of bradykinin, which later induced the biosynthesis of PGs and other autacoids that are responsible for the formation of the inflammatory exudates.\(^{19}\)

We also evaluated the anti-inflammatory effects of Garidi-5 on paw edema induced by Carr in mice and detected the levels of TNF-\(\alpha\) the paw edema 4 h after Carrageenan injection.

It is clear from Table 2, that Garidi-5 produced a dose-dependent inhibition of Carrageenan induced paw edema. Statistical analysis revealed that Garidi-5 and Indomethacin significantly inhibited the development of edema 1–4 h after treatment (Fig. 2) and they both showed anti-inflammatory effects in Carrageenan induced mice paw edema. In the current study, the levels of TNF-\(\alpha\) were decreased significantly in a dose dependent manner by treatment with 20, 80 and 200 mg/kg of Garidi-5.

**Conclusion:** Biological activity substances were detected in the Traditional drug Garidi-5. Traditional drug Garidi-5 has low toxicity, anti-inflammatory and analgesic effects.

**REFERENCES**


Systematic Review of the Association Between Volume and Outcome of Surgeons

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¹Nomun hospital
²Health Sciences University of Mongolia

ABSTRACT
The volume-outcome relationship has been widely discussed over past decades. Although previous studies that examined the volume-outcome relationship show that the majority of these works support the reverse relationship of volume and outcome on various procedures, other studies reported inconsistent results and arguments if volume is robust enough to predict outcome. Therefore, extending volume outcome association to measuring efficiency as well as process quality of care, and linking the performance with outcome have been important issues not only to health purchasers, but also providers and consumers when pursuing cost containment and quality improvement in health care industry. The purpose of this study is to review studies on the relationship between volume and outcome, and to identify possible measurements and methods. The large quantity of literature reported surgical volume-outcome relationship, but several limitations of those studies also have criticized and should be explored and re-verified. First, surgical volume was argued as a proxy of experience of surgeon and higher volume cannot naturally guarantee better outcome. Second, since the health care purchasers have drawn attention to the concept of pursuing value in health care, the efficiency of providers and the quality of care delivered should be measured instead of cost or volume alone. In addition, some specific possible outcome indicators should be considered in studies of the relationship between surgeons volume and outcome in future.

Key words: Surgical volume, outcome, health care quality

INTRODUCTION
Over the past several decades, there has been a growing interest among health care researchers to improve health care quality and to lower hospital expenditures. Neither of the two could be neglected, but the way to enhance efficiency and improve quality has been a challenging journey for health care providers. The good outcome can contribute the patients needs and some surgical volume-outcome relationship studies had found that the majority of these studies support the reverse relationship of volume and outcome of varied procedures and also on cancer treatment⁴-⁸. Some researchers emphasize that hospital volume alone effects health care quality⁴,⁹-₁₂, but Weinberg and colleagues compared hospitals in California in terms of their Medicare spending, resources input, and utilization, the results showed that the hospitals that use more resources do not provide better quality¹³. Therefore, extending the idea of a volume-outcome associations into measuring efficiency as well as process quality of care, and linking the performance with outcome have been important issues not only to health purchasers, but also providers and consumers. This study proposes to review the relationship between volume and outcome, and to identify possible measurement and methods.

MATERIALS AND METHODS
We reviewed studies published since 1975 which aimed at examining the relationship between hospital of physician volume and clinical outcomes in international scientific peer-reviewed journals summarized.

RESULTS AND DISCUSSIONS
The relationship between a hospitals volume and patient mortality was first evaluated by Lee et al in 1957, and after that this issue was raised by Luft and colleagues article, published in 1979, which examined 12 surgical offered by 1498 hospitals in 1974 and 1975 to explore the relationship between hospitals surgical volume and mortality. This study showed that part of the 12 surgeries, for instance, cardiac, vascular and orthopedic, had volume-outcome correlation, which indicated that mortality rates could be improved through regionalization’s. They also mentioned that physician volume may be more important than hospital volume¹⁴. Before that, there were hypotheses which identified that the enrichment of surgical volume could develop physicians professional capacity. Shapley et al reviewed 36 articles published between 1986 and 1998.
which were concerning volume-outcome relationship in peripheral vascular surgery and they found positive volume-outcome relationships. Helm et al reviewed 135 studies published between 1980 and 2000 which aimed at examining relationship between hospital or physician volume and clinical outcomes among 27 procedures or clinical conditions. The review showed that among all studies 71% of all studies of hospital volume and 69% of studies of physician volume reported statistically significant correlations between higher volume and better outcomes, but the magnitude of the association varies greatly. For studies included in this review which relate to cancer care, surgery on pancreatic cancer and esophageal cancer were found with stronger correlations than other type of treatments.

Another review done by Chow bury et al showed that among 163 articles covering 42 different procedures, 74.2% of studies showed high-volume hospitals and 74% of studies showed high-volume surgeons had significantly better outcomes. Specializations was also associated with improved patient outcome in 91%of the studies that reported positive hospitals volume-outcome relationship and 90 out of 13 reported positive surgeon volume-outcome relationship for cancer related surgeries. Therefore, most researchers emphasized that the physicians volume standards should consider the different types of outcomes and other outcome indicators such as mortality in the future study.

CONCLUSIONS

Although large quantities of literature report surgical volume-outcome relationship, several limitations of those studies have been criticized and should be explored and revivified. First, surgical volume had been argued as a proxy of quality or experiences of surgeon at higher volume but cannot necessarily guarantee better outcome. Second, since the health care purchasers have drawn attention to the concept of pursuing value in health care, but the efficiency of providers and the quality of concept of care delivered should be measured instead of cost or volume alone. Third, former studies often failed to consider the clustered data which may over-estimate the magnitude of volume-outcome relationship. To investigate the relation volume-outcome, there is need for clustered data, including hospital, efficiency, volume etc, which are more optimal, and concrete indicators and multilevel modeling approach is adopted to properly handle the statistical concerns of clustered data. Moreover, past researches often focused on short-term outcomes. /e.g. mortality/and were inadequate in reflecting long-term outcome of care.

REFERENCES

The Prevalence, and Risk Factors of Hepatitis C Virus Infection Among Population of Mongolia Over 40 Years of Age

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2Division of Virology, Department of Infection and Immunity, Jichi Medical University, Japan

ABSTRACT

WHO estimates 130-170 million (2-3% of world population) people with hepatitis C virus infection in the world. Around 70% or 130 million of those with hepatitis C viral infection develop chronic forms with further progress to liver cirrhosis and liver carcinoma.1 Number of studies confirm the high prevalence of hepatitis C among Mongolian population.2,3,4 This study aimed to evaluate prevalence of hepatitis C virus (HCV) infection and its risk factors among population of Mongolia aged over 40. The results suggest that overall prevalence of HCV infection is 36.7%, prevalence among female is higher than male (p=0.0001). Risk factors related to medical procedures such as transfusion of blood and blood products, (OR=2.082, 95% CI 1.629-2.660), surgery (OR=1.289, 95% CI 1.087-1.529), injections (OR=1.4, 95% CI 1.093-1.893) were statistically significant risk factors.

Key words: HCV, prevalence, risk factors

INTRODUCTION

Hepatitis C is endemic in most parts of the world. 3% of world population is infected with HCV which demonstrate that about 170 million people are at high risk of developing liver carcinoma. WHO classifies the prevalence of HCV among population according to presence of antibody to HCV as high (over 10%), intermediate (2.5-10%) and low (1-2.5%). [3] In Mongolia mortality from health indicators had decreasing tendency in 2006 but it had risen sharply by 2010. Hepatitis morbidity level is high in 2007,2008, decreased in 2009 and increased abruptly in 2010. Our country belongs to zone with high prevalence of hepatitis viral infection and its consequences like liver cirrhosis and liver carcinoma are one of the concerns of public health. This study aims to evaluate the prevalence of hepatitis C virus infection and its risk factors among population of Mongolia aged over 40.

MATERIALS AND METHODS

This is a population-based cross sectional study. Multistage random sampling method was employed to select 2313 observations above 40 years old. This is a nationally representative sample from 4 districts of Ulaanbaatar city, 13 soums from 4 aimags with equivalent sex and locality. Approval obtained from Ethical Review Board. HBsAg was determined by ELISA (enzyme-linked immunosorbent assay) method in virology laboratory of Jichi Medical University, Japan. Descriptive and advanced statistical analysis was executed using SPSS statistical package version 17.0. After ensuring normal distribution of variables Pearson’s chi-square test performed to compare proportions, t-test to compare means, univariate and multivariate regression analysis. 2-tailed p-value of less than 0.05 was considered as statistically significant.

RESULTS

There are 858 (37.1%) male, 1455 (62.9%) female recruited for the study. Of total participants 1289 (55.7%) sampled from Ulaanbaatar, 328 (14.2%) from aimag centers, 696 (30.1%) from soums. 16.6% are aged 40-44, 21.8% 45-49 and 23.4% aged above 60.

41.2% of soums residents, 32.2% of Ulaanbaatar and 35.4% of aimag residents had HCV infection. Prevalence among soum residents were relatively higher (p=0.008). Prevalence of HCV among female (41.9%) is higher than male (28.0%) (p=0.0001). Prevalence in age group above 60 (54.7%) is highest amongst other age groups: 38.2% in age group 55-59, 30.9% in 45-49, 26.0% in age group 40-44 which illustrates statistically significant increase of prevalence HCV infection with increasing age groups. (p=0.0001)
Table 1.

Prevalence of HCV infection among population of Mongolia aged above 40 by locality and sex

<table>
<thead>
<tr>
<th>Variables</th>
<th>Total</th>
<th>Anti-HCV (+)</th>
<th>Anti-HCV (-)</th>
<th>P-value</th>
</tr>
</thead>
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<tr>
<td></td>
<td>n</td>
<td>n</td>
<td>%</td>
<td>n</td>
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<tr>
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<td>Aimag</td>
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<td>105</td>
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<td>Soum</td>
<td>689</td>
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<tr>
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<td>239</td>
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<td>604</td>
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<td>839</td>
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<td>40-44</td>
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<td>325</td>
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<td>55-59</td>
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<td>136</td>
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<td>Above 60</td>
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<td>255</td>
<td>54.7</td>
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Table 2.

Regression analysis of HCV by medical procedure related risk factors

<table>
<thead>
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<th>P value</th>
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<td>Yes</td>
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<td>37.9</td>
<td>1.4</td>
<td>1.093</td>
<td>1.893</td>
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<td>Injection at home</td>
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</tr>
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<td>40.9</td>
<td>1.326</td>
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<td>1.582</td>
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<td>1995</td>
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<td>52.2</td>
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<td>46.2</td>
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<td>34.5</td>
<td>1.124</td>
<td>0.901</td>
<td>1.402</td>
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</table>
Of total people affected by HCV infection 40.0% had undergone surgery ($p=0.004$), 40.9% had acupuncture ($p=0.002$), 37.9% had injections in hospital ($p=0.008$) which are statistically significant factors that impact HCV infection. Cohorts who had transfusion of blood and blood products had increased odds of infecting HCV to compare with those with no transfusion. OR=$2.082$, 95% CI 1.629-2.660.

**Table 3.**

<table>
<thead>
<tr>
<th>Variables</th>
<th>P-value</th>
<th>OR</th>
<th>95% confidence interval</th>
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<td>Age group</td>
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<td>0.694-0.794</td>
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<td>Family history of viral hepatitis</td>
<td>0.866</td>
<td>1.000</td>
<td>0.997-1.002</td>
</tr>
<tr>
<td>Blood procedure</td>
<td>0.0001</td>
<td>0.530</td>
<td>0.404-0.695</td>
</tr>
<tr>
<td>Acupuncture</td>
<td>0.809</td>
<td>0.976</td>
<td>0.803-1.187</td>
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<td>Injection in hospital</td>
<td>0.094</td>
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<td>Surgery</td>
<td>0.145</td>
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<td>Education</td>
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<td>1.050-1.201</td>
</tr>
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</table>

Multiple regression analysis demonstrate that female have 47.5% higher risk of HCV infection than male (OR=$0.525$, 95% CI 1.046-1.853) and it is statistically significant ($p=0.001$). Age category also have statistically significant differences. Having injection in-home raises HCV infection by 26.9% (OR=$0.731$, 95% CI 0.605-1.040, $p=0.033$).

**DISCUSSION**

According to study by Chong-Shang, Ting Ting Chan 27.3% of Taiwan rural residents aged above 60 had anti-HCV antibody. In our study prevalence of anti-HCV among rural elderly population aged above 60 was 41.2%. Chong Shen Wang et al. study suggest that low educational level, blood transfusion, smoking, age above 50 were major determinants of HCV infection. Our findings are also highlights blood transfusion (OR=14.0), age above 60 (prevalence is 54.7%) being risk factors of HCV infection. In study by Tsatsralt-Od B., Baatarkhuu O, 2006 the prevalence of HCV infection was 15.6%, Davaalkham et al. estimated as 10.7%, 2006. In our study prevalence of HCV infection among population aged above 40 is high as 36.7%.

**CONCLUSION**

Prevalence of HCV infection among population aged above 40 is 36.7%, female having higher rate than male (p=0.0001). Transfusion of blood and blood related products (OR=2.082, 95% CI 1.629-2.660), surgery (OR=1.289, 95% CI 1.087-1.529), injection (OR=1.4, 95% CI 1.093-1.893) were statistically significant risk factors related to medical procedures. It is necessary to prevent from medical procedures that could be a threat to HCV infection.

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The Detection of Bacterial Meningitis Pathogens and Their Serotyping in Ulaanbaatar, 2002-2011

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1National Center for Communicable Diseases
2Health Sciences University Mongolia

ABSTRACT

Childhood bacterial meningitis is severe and largely preventable by vaccination. Little data on childhood meningitis exist in Asia. The objective of study was to determine serotypes of this bacterial meningitis caused by N. meningitidis, H. influenzae, S. pneumoniae in Mongolia. We conducted prospective, active, hospital-based surveillance for bacterial meningitis in children up to 5 years of age. Clinical data, blood and cerebrospinal fluid were collected according to a standard protocol. Multiplex and RT-PCR were used for serotyping.

From February 2002 to December 2011, 577 suspected meningitis cases were enrolled. We estimated that the incidence of bacterial meningitis during 10 years of study in children younger than 5 years was 20.4 cases per 100,000. The most dominant bacterial pathogens are N. meningitidis A, 6A/B/C, 2, 14, 7F serotypes of S. pneumoniae, H. influenzae type b. Among children younger than that of 5 years of age, S. pneumoniae, N. meningitidis and H. influenzae were the predominant causes of bacterial meningitis and the morbidity rate was higher than other Asian countries.

Keywords: bacterial meningitis, N. meningitidis, S. pneumoniae, H. influenzae type b, real time PCR

INTRODUCTION

Twenty three outbreaks of meningococcal infection have been reported in some countries of Europe, South America, Africa, and Asia and latest largest outbreak was reported in Russian Federation, January, 2006, caused N. meningitidis A. 90% of the meningococcal outbreaks were caused by serotypes for A, B, C, N. meningitidis and 10% were caused by serotypes for N. meningitidis Y, W135 and main serotypes were B, C in Europe, Latin America, and serotype A in Asia1,2. Most meningococcal outbreaks were reported among children aged under 4 years.3 During last 5 years 77.8% of meningococcal disease mortality was reported among children aged under 2 years and 47.5% were reported in Ulaanbaatar and Choibalsan cities.4

Meningococcal outbreaks have been reported since 1951 and started to be diagnosed using clinical symptoms since 1960 in all suspected cases in Mongolia. 0.1% of respiratory disease cases including measles, diphtheria, pertussis were meningococcal diseases in Ulaanbaatar during 1960-1968.5

The study of Tsend N et all has shown that two big outbreaks occurred between 1951-1975 in Mongolia. The first one was during 1951-1959 and the second one was 1969-1975 the outbreak continued for 16 years, and the interval period between outbreaks was 10 years (1958-1974). The first meningococcal disease outbreak continued for 10 years and 9.8-30.2 per 10,000 people in peak period and second peak occurred in 1974 with 194.1 per 10,000 people. 50.7% of meningococcal diseases cases was reported from Ulaanbaatar in 1974.6

75%-83.2% of cases were reported in the winter and spring and 78% of cases occurred among children aged under 10 years. Overall mortality was 2.5-40% and 10.3-19.7% of mortality occurred among children under one during the second meningococcal diseases outbreak peak.3,4,5,6

Most meningococcal infection outbreaks are large and by different microbial agents.7

There are one million of meningococcal disease cases with 20 000 deaths in the inter epidemic period in worldwide.8 A previous study has shown that there are 68 suspected and confirmed bacterial meningitis cases out of every 100,000 people, 28 confirmed Hib meningitis cases out of every 100,000 people, 11 cases of pneumococcal meningitis out of every 100,000 people 13 cases of meningococcal meningitis out of every 100,000 people.9,10
The vaccine is important for prevention and control of the bacterial meningitis. Mongolia conducted nationwide supplementary immunization against meningococcal diseases among children aged 16 using meningococcal polysaccharid vaccine A,C during outbreak years 1974-1975, 1994-1995.10 The vaccine can establish immunity against bacteria and serotypes which components of that vaccine such as H.influenzae, N.meningitidis, S.pneumoniae. Hib vaccine does not prevent other serotype of the H.influenza. The bacteriology laboratory has played an important role for testing bacterial agents and serotypes of the meningococcal diseases.11

Preventive and control measures against bacterial meningitis still a problem in Mongolia as well as the study of disease burden and bacterial agents identification. Therefore the study objective was to determine serotypes of bacterial meningitis caused by N.meningitidis, H.influenzae, S.pneumoniae in our country.

**MATERIALS AND METHODS**

*Design.* Prospective, active, hospital-based surveillance for bacterial meningitis was initiated in February 2002. We analyzed surveillance data until December 31, 2011.

Children 2 months to 5 years of age who were hospitalized in Ulaanbaatar for suspected meningitis were identified according to World Health Organization (WHO) guidelines12. Residency was determined by review of identification cards and interview of family members. Surveillance was performed at 6 hospitals in Ulaanbaatar that admit children: The National Centre for Communicable Diseases (NCCD), the National Centre for Maternal and Child Health (NCMCH), KhanUul District Hospital, Songinokhairkhan District Hospital, Sukhbaatar District Hospital, and Bayanzurkh District Hospital. Clinical data were collected by surveillance personnel using a structured form.

*Case definitions.* For surveillance purposes, suspected meningitis was defined as clinically suspected meningitis with at least 1 of the following: fever, headache, stiff neck, bulging fontanelle, or mental status change. Possible bacterial meningitis was defined as the presence of a WBC count of 10–99 cells/mL in CSF with either ≥80% neutrophils or abnormal CSF protein and glucose levels. Probable bacterial meningitis was defined as the presence of a WBC count of ≥100 cells/mL in CSF or turbid appearance of CSF (if the WBC count was not measured). Confirmed bacterial meningitis was defined as the isolation of a bacterial pathogen from CSF culture, detection of bacterial antigen in CSF by latex agglutination, detection of bacterial nucleic acid by PCR (if criteria for probable bacterial meningitis were met), or isolation of a bacterial pathogen from blood culture (if CSF was not obtained or if criteria for probable bacterial meningitis were met). Detection of bacterial DNA inclinical samples using real-time PCR is a sensitive technique, and false-positive PCR results can be generated by cross-contamination of samples.13,14 Therefore, our a priori case definition required that a PCR result would only be taken as the sole method of confirming an infection if the case also satisfied the criteria for probable bacterial meningitis. If there were any discrepancies between laboratory tests, an expert in pediatric infectious diseases who was not involved in the surveillance project was asked to determine the final diagnosis. Common skin flora, such as coagulase-negative staphylococci or diphtheroids, were considered contaminants. Aseptic meningitis was defined as suspected meningitis with a WBC count ≥10 cells/mL of CSF that did not meet the criteria for possible, probable, or confirmed bacterial meningitis. Patients who had WBC counts of less than 10 cells/mL were categorized as not having meningitis.15

*Laboratory methods.* The CSF and blood specimens were collected at each treating hospital. At least 1 mL of blood per patient was inoculated into a Bactec® Peds Plus blood culture bottle (BD). Specimens from the MCHRC were tested in a laboratory in that hospital.

For other hospitals, neat CSF and blood culture bottles were transported to the NCCD with warm packs and thermometers to ensure that the specimens were transported at 20°C–25°C. Specimens were transported on the day of collection or in the morning of the following day if collected at night. On arrival at the laboratory, the blood culture bottles were placed in an Bactec 120 machine at 35°C–37°C. They were then subcultured onto locally manufactured sheep blood and chocolate agar plates along with growth factors and incubated with a candle jar. In the laboratory, CSF specimens were immediately plated onto supplemented sheep blood and chocolate agar plates and incubated for 48 h at 35°C–37°C with a candle jar. All plates were checked for growth after 18–24 h of incubation, and any organisms isolated were identified according to standard laboratory procedures. Detection of bacterial antigen in CSF was performed using the Meningite 5 latex agglutination test kit (Wellcogen) according to the manufacturer’s instructions.16

*PCR analysis.* DNA was extracted from 200mL of each CSF specimen using the MagNA Pure LC TotalNucleic Acid Isolation Kit (Roche) and a MagNA Pure LCrbotic workstation (Roche), eluting the DNA in a final volume of 100 mL of buffer.16 Each DNA sample (volume of 5 mL) was assayed for the presence of bacterial DNA encoding the H.influenzæ gene bexA (specific to serotypes b and c only), the N.meningitidis gene sodC, and the S. pneumoniae gene lytA using multiplex real-time PCR method of Corless
and Carvalho.\textsuperscript{17,18} Results with a cycle threshold value of 138 were considered negative. Because contaminants that interfere with PCR are sometimes purified along with DNA from CSF, each DNA sample was tested both neat and after 1/10 dilution in water. Successful amplification of target DNA at either of these 2 concentrations was considered a positive result. We used Latin America multiplex PCRs for the determination of \textit{S. pneumoniae} serotypes.\textsuperscript{20}

\textbf{RESULTS}

In this study, we selected 577 patients of the age 0-5 with suspected meningitis from 2002 to 2011 identified by the surveillance system (Figure 1).

In our study, 399 (69.1\%) of total cases were diagnosed as meningitis by clinical features, 100 (17.3\%) as meningococemia, 24 (4.1\%) cases with meningoencephalitis, 8 (1.4\%) cases registered convulsion and non-meningococcal was detected in 46 (8.0\%) cases. Of these 577 patients 385 were age 0-1 year (66.7\%), 20 aged 4-5 years (3.5\%) and 327 (56.7\%) of the total number of patients were male. Of all patients 577, 80 (63.5\%) of 126 (21.8\%) satisfied the criteria for unconfirmed probable bacterial meningitis, 174 (69.9\%) of 249 (43.2\%) satisfied the criteria for unconfirmed possible bacterial meningitis, 25(28.4\%) of 88(15.3\%) confirmed aseptic meningitis and 58 (50.9\%) of 114 (19.8\%) meningococcemia case had been detected the etiology of bacterial meningitis (Table 1).

\begin{table}[h]
\centering
\caption{Causes and cases definition of bacterial meningitis}
\begin{tabular}{|c|c|c|c|c|c|c|c|c|}
\hline
Cases definition & Total & \textit{N.meningitis} & \textit{H.influenza} & \textit{S.pneumonia} & others & Total \\
\hline
Unconfirmed probable & 126 (21.8) & 22 & 17.5\% & 18 & 14.3\% & 29 & 23.0\% & 11 & 8.7\% & 80 & 63.5\% \\
Unconfirmed possible & 249 (43.2) & 58 & 23.3\% & 58 & 23.3\% & 47 & 18.9\% & 11 & 4.4\% & 174 & 69.9\% \\
Aseptic meningitis & 88 (15.3) & 5 & 5.7\% & 0 & 0.0\% & 12 & 13.6\% & 8 & 9.1\% & 25 & 28.4\% \\
Meningo-coccemia & 114 (19.8) & 22 & 19.3\% & 8 & 7.0\% & 22 & 19.3\% & 6 & 5.3\% & 58 & 50.9\% \\
Total & 577 (100.0) & 107 & 18.5\% & 84 & 14.6\% & 110 & 19.1\% & 36 & 6.2\% & 337 & 58.4\% \\
\hline
\end{tabular}
\end{table}

Out of the total 577 cases Hib was responsible for 84 (14.5\%) of the confirmed cases, \textit{S.pneumoniae} in 109 (19.1\%), and \textit{N.meningitidis} in 107 (18.5\%). In 2002-2005 the prevalent causative agent of bacterial meningitis was \textit{Hib}, instead from 2006 to 2008 were detected mostly \textit{N.meningitidis} and \textit{S.pneumoniae} and in 2009, 2010, 2011 was identified \textit{S.pneumonia} (Figure 2).

29.5\% of bacterial culture and 36.1\% of latex agglutination tests were positive for \textit{N.meningitidis}, \textit{H.influenzae}, \textit{S.pneumoniae}. In 225 CSF cases confirmed bacterial meningitis by culture, latex agglutination test for real time PCR were detected in 100\% of \textit{H.influenzae} spp., \textit{N.meningitidis} in 92.6\% and \textit{S.pneumoniae} in 100\% respectively (Table 2).
In the negative culture test by real time PCR 39 (23.8%) cases were detected pathogens. In one case, the real time PCR result was different from the culture and/or antigen detection tests. However, in one sample positive for *H. influenzae* the culture and latex agglutination test was detected *N. meningitidis B* in real time PCR and in another one case which was positive for *N. meningitidis B* latex agglutination detected Hib in real time PCR test.

**Table 2. Detection of *H. influenzae*, *N. meningitidis*, *S. pneumoniae* in CSF by rt-PCR**

<table>
<thead>
<tr>
<th>Primer</th>
<th>Detected by latex and culture tests in CSF, blood samples Non detected in CSF and blood Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Hi</td>
</tr>
<tr>
<td>Positive rtPCR</td>
<td>61</td>
</tr>
<tr>
<td>hpd #3</td>
<td>61</td>
</tr>
<tr>
<td>bexA</td>
<td>60</td>
</tr>
<tr>
<td>sodC</td>
<td>1b</td>
</tr>
<tr>
<td>lytA</td>
<td>0</td>
</tr>
<tr>
<td>RNase P</td>
<td></td>
</tr>
</tbody>
</table>

**Table 3. The detection of *S. pneumoniae* by rt-PCR**

<table>
<thead>
<tr>
<th>Step</th>
<th>Function</th>
<th>Temperature</th>
<th>Time</th>
<th>Repeat</th>
</tr>
</thead>
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<td>1</td>
<td>INCUBATE</td>
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<td>0.0:10:0</td>
<td>0</td>
</tr>
<tr>
<td>2</td>
<td>INCUBATE</td>
<td>95.00</td>
<td>0:0:0:15</td>
<td>0</td>
</tr>
<tr>
<td>3</td>
<td>INCUBATE</td>
<td>60.00</td>
<td>0:1:0</td>
<td>0</td>
</tr>
<tr>
<td>4</td>
<td>SCAN</td>
<td></td>
<td></td>
<td>0</td>
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<tr>
<td>5</td>
<td>GOTO</td>
<td>Step2</td>
<td>40</td>
<td></td>
</tr>
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</table>

<table>
<thead>
<tr>
<th>Well</th>
<th>Sample Name</th>
<th>Probe</th>
<th>Type</th>
<th>Flu. Dye</th>
<th>Quencher</th>
<th>Ct</th>
<th>Concentration</th>
<th>Ct Threshold</th>
</tr>
</thead>
<tbody>
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<td>Std_FAM</td>
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<td>FAM</td>
<td>BHQ</td>
<td>25.58</td>
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</tr>
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<td>D3</td>
<td>SB-11-0301 1:100</td>
<td>Std_FAM</td>
<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
<td>26.45</td>
<td>0</td>
<td>1483</td>
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<tr>
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<td>SK-11-0111 1:100</td>
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<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
<td>determined</td>
<td>-</td>
<td>1483</td>
</tr>
<tr>
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<td>Std_FAM</td>
<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
<td>determined</td>
<td>-</td>
<td>1483</td>
</tr>
<tr>
<td>H1</td>
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<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
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<td>0</td>
<td>1483</td>
</tr>
<tr>
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<td>FAM</td>
<td>BHQ</td>
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<td>1272</td>
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<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
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<td>0.00E+00</td>
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</tr>
<tr>
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<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
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<td>1272</td>
</tr>
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<td>Std_FAM</td>
<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
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<td>0.00E+00</td>
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<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
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<tr>
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<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
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<td>0.00E+00</td>
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<tr>
<td>H7</td>
<td>pos hpd</td>
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<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
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<tr>
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<td>Sample</td>
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<td>645</td>
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<tr>
<td>H3</td>
<td>POS CLLYTA</td>
<td>Std_FAM</td>
<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
<td>22.89</td>
<td>0</td>
<td>645</td>
</tr>
<tr>
<td>B3</td>
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<td>Std_FAM</td>
<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
<td>24.28</td>
<td>0</td>
<td>700</td>
</tr>
<tr>
<td>F1</td>
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<td>Std_FAM</td>
<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
<td>25.27</td>
<td>0</td>
<td>700</td>
</tr>
<tr>
<td>H3</td>
<td>pos et LytA</td>
<td>Std_FAM</td>
<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
<td>20.13</td>
<td>0</td>
<td>700</td>
</tr>
<tr>
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<td>Std_FAM</td>
<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
<td>36.02</td>
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<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
<td>17.63</td>
<td>0.00E+00</td>
<td>1272</td>
</tr>
<tr>
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<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
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<td>789</td>
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<td>Sample</td>
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<td>BHQ</td>
<td>25</td>
<td>0.00E+00</td>
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<tr>
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<td>Sample</td>
<td>FAM</td>
<td>BHQ</td>
<td>25.18</td>
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<td>Sample</td>
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<td>BHQ</td>
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<td>Std_FAM</td>
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<td>FAM</td>
<td>BHQ</td>
<td>24.28</td>
<td>0</td>
<td>637</td>
</tr>
</tbody>
</table>
Serotyping of *N. meningitidis*. During our study we detected 107 *N. meningitidis* spp., from total number we identified A serogroup at 42.9%, B serogroup at 35.8% and non-defined at 15.8% respectively. Although at Mongolia started to use polysaccharide A, B vaccine the incidence of meningococcal meningitis among children ages 0-5 52 (A serogroup 46 cases, C serogroup 6 cases) cases were registered. The mortality of meningococcal meningitis was 13.8% (Table 4).

### Table 4.

<table>
<thead>
<tr>
<th>Pathogens and Serogroups</th>
<th>Result</th>
<th>The rate of mortality</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Not detected</td>
<td>240</td>
<td>18</td>
<td>7.0%</td>
</tr>
<tr>
<td>N. meningitidis serogrupes</td>
<td>81</td>
<td>13</td>
<td>13.8%</td>
</tr>
<tr>
<td>Non-defined</td>
<td>10</td>
<td>3</td>
<td>23.1%</td>
</tr>
<tr>
<td>A</td>
<td>42</td>
<td>4</td>
<td>8.7%</td>
</tr>
<tr>
<td>B</td>
<td>33</td>
<td>4</td>
<td>10.8%</td>
</tr>
<tr>
<td>C</td>
<td>6</td>
<td>2</td>
<td>25.0%</td>
</tr>
<tr>
<td>W135</td>
<td>3</td>
<td>0</td>
<td>0.0%</td>
</tr>
</tbody>
</table>

Serotyping of *H. influenzae*. *H. influenzae* type b (Hib) was responsible for 83 (96.3%) of the confirmed 84 *H. influenzae* cases. *H. influenzae* type b conjugate vaccine was introduced into the routine immunization schedule in 2005 for the children ages 0-5 years and the incidence rate of Hib infection dramatically reduced in Mongolia. The study showed that 33.1% of children received conjugate vaccine compare with 66.9% of children who did not receive vaccine completely. Among children who did not receive vaccine completely meningitis due to Hib registered in 41.4% and in vaccinated case only 1.2% (p<0.001) children were infected by Hib (Table 5).

### Table 5.

<table>
<thead>
<tr>
<th>Pathogens and serotypes</th>
<th>Result</th>
<th>The mortality rate</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Not detected</td>
<td>240</td>
<td>18</td>
<td>7.0%</td>
</tr>
<tr>
<td>H. influenzae serotypes</td>
<td>71</td>
<td>13</td>
<td>15.5%</td>
</tr>
<tr>
<td>Hib</td>
<td>69</td>
<td>12</td>
<td>14.8%</td>
</tr>
<tr>
<td>Non Hib</td>
<td>2</td>
<td>1</td>
<td>33.3%</td>
</tr>
</tbody>
</table>

Serotyping of *S. pneumoniae*. In our study we identified totally 110 of *S. pneumoniae* spp., and detected serotypes of 72 species of *S. pneumoniae*. We detected 33 serotypes of *S. pneumoniae* by multiplex PCR method using Latin American standard (Figure 3). We detected from a total of 72 species in 16 (14.5%) cases serotype 6A/B/C, serotype 2 in 9 (8.2%) cases and other serotypes as 14, 7F, 17F, 19F identified as minority (Table 6).

### Table 6.

<table>
<thead>
<tr>
<th>Serotypes of <em>S. pneumoniae</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Serotypes</td>
</tr>
<tr>
<td>-----------------------------</td>
</tr>
<tr>
<td>Non-defined</td>
</tr>
<tr>
<td>6A/B/C</td>
</tr>
<tr>
<td>2</td>
</tr>
<tr>
<td>14</td>
</tr>
<tr>
<td>7F</td>
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<td>17F</td>
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<tr>
<td>Total</td>
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</table>
Figure 3. Multiplex PCR serotyping. Lanes represent the different patterns generated by multiplex PCR on clinical isolates.
DISCUSSION
We estimated that the incidence of bacterial meningitis during 10 years of study in children younger than 5 years was 20.4 cases per 100,000. The incidence of meningitis caused by Hib and S.pneumoniae increased markedly between epidemics of meningococcal meningitis. Routine immunization in 2005 of H.influenzae type b conjugate vaccine reduced infection due to Hib starting from 2007. In our study we detected pathogens in some CSFs but the number of white blood cell was not high. Also, the parameters of assays were similar in confirmed bacterial meningitis and in unconfirmed probable samples. In other countries studies the difference between parameters of assays between confirmed and unconfirmed probable samples of bacterial meningitis were high, probably because of low usage of antibiotics before treatment hospital. 52% of cases of using antibiotics before visits the hospital was identified by Oyungerel. Therefore the low detection of pathogens of bacterial meningitis in the clinical samples may be related usage of antibiotics before to visiting the hospital. According to our study in infants and young children less than 5 years S.pneumoniae, N.meningitidis, and Haemophilus influenzae type b (Hib) were the most common causes of bacterial meningitis and the morbidity rate was higher than in many other Asian countries.

The most common serotype that caused invasive disease in Poland during 1997-2004 was Hib which detected in 233 cases from total 245 isolates, H.influenzae type f -2 and non capculate in 10 cases. The most common serotypes of S.pneumoniae in developing countries were 14, 19F, 3, 7F, 6A/6B, 10A, 18C, 23F, 5, 1 but serotypes 1 and 5 were prevalent in North America. In our study we detected the domination of following serotypes: for N.meningitidis serogroup A in 35.4%, for H.influenzae type b in 96.3% and for S.pneumoniae serotypes 6A-15.9%, serotype 7 in 9.5%, serotype 14 in 12.7% and serotype 19F detected in 11.1% respectively, and this incidence rate was similar to the report studied in Poland.

There are 5 different vaccines against S.pneumoniae and the most widely used vaccine is 13 cent polysacharide conjugated vaccine consisting the following types: 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F, 23F. Our detected serotypes are same as the conjugated vaccine types.

CONCLUSION
Among children younger than 5 years of age, S.pneumoniae, N.meningitidis and H.influenzae were the predominant causes of bacterial meningitis and the morbidity rate was higher than other Asian countries. The predominate pathogens were N.meningitidis serogroup A, Hib and 6A/B/C,2,14,7F serotypes of S.pneumoniae.

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The Prevalence of Asthma and Its Association with Body Mass Index in Adults

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ABSTRACT

Obesity and asthma are two important public health problems in the worldwide. Over the last two decades, there has been a significant increasing in the prevalence of asthma and obesity in different parts of the world. The aim of this study was to investigate the association between bronchial asthma and body mass index. The height and weight of all subjects were measured by standard methods. Body mass index (BMI) was calculated by dividing the weight in kilograms by the square of the height in meters (kg/m²). We interviewed subjects using a questionnaire developed on the basis of WHO Protocol for Assessment of Prevalence of Major Respiratory Diseases and modified by local risk factors assessment and by other international survey approach including Global Initiative for Asthma (GINA) (9) and European Community Respiratory Health Survey (ECRHS). The questionnaires included 79 questions with 11 different groups. We randomly chose 1201 adults. Of the respondents, 606 were males (50.5%) and 595 were females (49.5%). 3.8% (n=46) out of all subjects were classified as underweight, 49.1% (n=590) were classified as normal weight, 30.2% (n=363) were classified as overweight and 16.8% (n=202) were classified as obese. The prevalence of wheezing was higher in overweight and obese subjects than in those of a normal weight. The BMI was higher in the group with wheezing than in the group without wheezing (p<0.01). The prevalence of physician-diagnosed asthma was higher in obese subjects than in normal weight subjects (p<0.01). This study suggests that BMI is associated with bronchial asthma in adults.

Key words: Adults, asthma, body mass index, prevalence

INTRODUCTION

Obesity and asthma are two important public health problems in developing countries. Over the last two decades, there has been a significant rise in the prevalence of asthma and obesity worldwide1,2.

Bronchial asthma affects around 300 million people throughout the world1. Over the past 20 years, the morbidity and prevalence of asthma have increased in different parts of the world4. A decade ago, the prevalence of atopic diseases in Mongolia was in the lowest range as reported from previous studies. Particularly, there was a quite good population-based epidemiological study result in 1999-2000. In this study, the prevalence of asthma, allergic rhinoconjunctivitis and allergic sensitization with 95% confidence intervals were 1.1%, 9.3% and 13.6% in Mongolian villages and 2.1%, 18.4% and 31.0% in Ulaanbaatar city, respectively4.

Obesity is capable of reducing pulmonary compliance, lung volumes, and peripheral airway diameter, as well as an increase in airway hyperreactivity, alteration in pulmonary blood volume and ventilation-perfusion mismatch5.

The association between asthma and obesity has been widely reported in many adult studies7-9. Studies have suggested that obesity is a risk factor for adult asthma10-12. Recently, several studies have also demonstrated a positive correlation between body mass index (BMI) and asthma, with an emerging consensus that the association is much stronger among women than men13-15. A meta-analysis of seven prospective epidemiological studies in which BMI was self-reported showed that the incidence of asthma increased by 50% in overweight/obese individuals.

The main aim of this study was to investigate association between bronchial asthma and body mass index.

MATERIALS AND METHODS

Study was carried out in the city of Ulaanbaatar, Mongolia (Total population of the survey area was 1.1 million in 2009) 10 out of 52 district family clinics were randomly selected for the study. We randomly chose 1201 adults aged 20 years and over who live in different districts of
Ulaanbaatar city. This city was chosen because it is the most populated city in Mongolia, and so a high response rate could be obtained.

We interviewed subjects using a questionnaire for subjects was developed on the basis of WHO Protocol for Assessment of Prevalence of Major Respiratory Diseases\textsuperscript{16} and modified by local risk factors assessment and by other international survey approaches including Global Initiative for Asthma (GINA)\textsuperscript{17} and European Community Respiratory Health Survey (ECRHS)\textsuperscript{18}. The questionnaires included 79 questions with subjects height, weight, clinical history, respiratory symptoms, prior diagnosis, risk factor exposure including smoking and treatments.

The height and weight of all subjects were measured by the standard methods. Body mass index (BMI) was calculated by dividing the weight in kilograms by the square of the height in meters (kg/m\(^2\)). According to the WHO classification, a BMI of <18.5 kg/m\(^2\) is underweight, 18.5–24.9 kg/m\(^2\) is normal, 25.0–29.9 kg/m\(^2\) is overweight and >30 kg/m\(^2\) is obesity.

Completed questionnaires were scanned and exported to SPSS version 17. Chi-square testing was used to assess the trends in the prevalence of asthma symptoms in overweight and obese children compared to normal weight children. P-values (two-sided) less than 0.05 were regarded as statistically significant. The study was approved by the Medical Ethics Control Committee of the Ministry of Health, Mongolia.

**RESULTS**

The study was conducted from June to July of 2009. In total, 1201 subjects aged 20 and over participated in this study. Of the respondents, 606 were males (50.5\%) and 595 were females (49.5\%). These data are shown in Table 1.

<table>
<thead>
<tr>
<th>Age group</th>
<th>Male n(%)</th>
<th>Female n(%)</th>
<th>Overall n(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>20-29</td>
<td>184 (15.3)</td>
<td>161 (13.4)</td>
<td>345 (28.7)</td>
</tr>
<tr>
<td>30-39</td>
<td>121 (10.1)</td>
<td>134 (11.1)</td>
<td>255 (21.2)</td>
</tr>
<tr>
<td>40-49</td>
<td>153 (12.7)</td>
<td>152 (12.7)</td>
<td>305 (25.4)</td>
</tr>
<tr>
<td>50-59</td>
<td>101 (8.4)</td>
<td>107 (8.9)</td>
<td>208 (17.3)</td>
</tr>
<tr>
<td>60-69</td>
<td>40 (3.4)</td>
<td>34 (2.8)</td>
<td>74 (6.2)</td>
</tr>
<tr>
<td>≥70</td>
<td>7 (0.6)</td>
<td>7 (0.6)</td>
<td>14 (1.2)</td>
</tr>
<tr>
<td>Overall</td>
<td>606 (50.5)</td>
<td>595 (49.5)</td>
<td>1201 (100.0)</td>
</tr>
</tbody>
</table>

3.8\% (n=46) out of all subjects were classified as underweight, 49.1\% (n=590) were classified as normal weight, 30.2\% (n=363) were classified as overweight and 16.8\% (n=202) were classified as obese (Figure 1).

![Figure 1. BMI groups of subjects](image)

The prevalence of obesity and overweight among females was 31.1\% and 19.7\%, among males was 29.4\% and 14.0\%, respectively. The BMI was higher in females than in males (p=0.011) (Figure 2).

![Figure 2. Comparison of the frequencies of BMI by sex segregated](image)

The prevalence of current asthma (wheezing or whistling in the last 12 months) and physician-diagnosed asthma was 15.6\% [95\% CI, 13.5-17.6], 4.8 [95\% CI, 3.6-6.0] among adults in Ulaanbaatar, respectively.
The prevalence of current and physician-diagnosed asthma among obese adults was 33.7% and 51.7%, among overweight adults was 42.2% and 36.2%, among normal-weight adults was 23.0% and 10.3% respectively, and there was a statistical association between prevalence of asthma and obesity (p< 0.001). The prevalence of physician-diagnosed asthma was directly associated with BMI (Figure 3).

The prevalence of obesity and overweight among men with current wheezing was 34.1 % and 37.5 %, among women with current wheezing was 33.3% and 46.5%, respectively. Our analysis showed that 11.8% were normal weight, 29.4% were overweight, 58.8% were obese among males with diagnosed asthma and 9.8% were normal weight, 39.0% were overweight, 48.8% were obese among females with diagnosed asthma (Table 3). There was no statistical association between obese and overweight subjects with current asthma in either sex (p = 0.289).

### Table 3.

<table>
<thead>
<tr>
<th>BMI group</th>
<th>Current asthma</th>
<th></th>
<th>Diagnosed asthma</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Male, n (%)</td>
<td>Female, n (%)</td>
<td>Male, n (%)</td>
<td>Female, n (%)</td>
</tr>
<tr>
<td>Underweight</td>
<td>0 (0.0)</td>
<td>2 (2.0)</td>
<td>0 (0.0)</td>
<td>1 (2.4)</td>
</tr>
<tr>
<td>Normal</td>
<td>25 (28.4)</td>
<td>18 (18.2)</td>
<td>2 (11.8)</td>
<td>4 (9.8)</td>
</tr>
<tr>
<td>Overweight</td>
<td>33 (37.5)</td>
<td>46 (46.5)</td>
<td>5 (29.4)</td>
<td>16 (39.0)</td>
</tr>
<tr>
<td>Obese</td>
<td>30 (34.1)</td>
<td>33 (33.3)</td>
<td>10 (58.8)</td>
<td>20 (48.8)</td>
</tr>
<tr>
<td>Overall, n</td>
<td>88</td>
<td>99</td>
<td>17</td>
<td>41</td>
</tr>
</tbody>
</table>

### DISCUSSION

The results of this study showed that the prevalence of physician-diagnosed asthma was statistically significantly higher among obese subjects than subjects with normal weight (51.7% vs. 10.3%). Also, the prevalence of physician-diagnosed asthma among overweight adults was higher than in normal-weight adults (36.2% vs. 10.3%). These associations are consistent with other studies12,19,20.

In our study, the relationship between current asthma and obesity and overweight was independent of sex (p = 0.289). This finding was consistent with other reports7,21-23, but some reports have noted the association only in women, or that it was stronger in women13-15. This inconsistency in results for males and females between studies may be attributed to differences in study populations, the age distribution of participants, and different definitions of asthma used.

On the basis of these findings, overweight and obesity seem to be significant risk factors for asthma. If these can be considered to be modifiable risk factors, interventions that affect weight loss could be associated with a decrease in asthma symptoms. Thus, adults with co-morbid asthma and obesity should be encouraged to increase physical activity and lose weight.

### CONCLUSION

The results of this study suggest that in our region there is a strong association between prevalence of asthma and BMI (in overweight and obese subjects) in both sexes among adults.

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Use of Mercury-Based Medical Devices in Health Care Organizations of Mongolia


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ABSTRACT

Health-care facilities are one of the main sources of mercury releases into the atmosphere because of emissions from the incineration of medical waste. The most common potential mode of occupational exposure to mercury is via inhalation of liquid mercury vapors. If not cleaned up properly, spills of even small amounts of mercury, such as from breakage of thermometers, can contaminate indoor air above recommended limits and lead to serious health consequences.

The overall objective of the study is to conduct mercury-based medical devices used in health care organizations and develop strategy and recommendations on further activity. A cross-sectional study design was used. Totally 578 units of 38 governmental and private health care organizations in Ulaanbaatar, Darkhan, Erdenet cities and Uvurkhangai provinces were conducted in the survey.

Conclusion: Mercury containing devices such as thermometer, blood pressure sphygmomanometer, energy saving fluorescence lamp and thermostats were used in urban and rural hospitals. There are not any regulations for safe handling, storage, and transportation and disposal system of mercury containing devices. Knowledge on handling, storage and disposing mercury based devices are not enough among the medical personals. The current situations for inappropriate disposal system can be posed to increase risks of environmental pollution with mercury.

Key words: mercury-based medical device, thermometer, sphygmomanometer, health care facility

INTRODUCTION

In the United States, according to US Environmental Protection Agency (EPA) in a 1997 report, medical waste incinerators may have been responsible for as much as 10% of all mercury air releases. Thermometers and sphygmomanometers are frequently used in health care. These both add to the global burden of mercury removed from its below ground repository and spread about on the surface to form highly neurotoxic organomercury compounds. Further, these devices break or leak with regularity, exposing health care workers to the acute effects of the inhalation of the metal itself1-4. Dental amalgam is the most commonly used dental filling material. It is a mixture of mercury and a metal alloy. The normal composition is 45-55% mercury; approximately 30% silver and other metals such as copper, tin and zinc. In 1991, the World Health Organization confirmed that mercury contained in dental amalgam is the greatest source of mercury vapor in non-industrialized settings, exposing the concerned population to mercury levels significantly exceeding those set for food and air 5,6.

In Mongolia recently has been registered intoxication case associated with mercury contamination released from illegal gold mining. Human health and environmental health impact assessment for mercury intoxication had done by International and Mongolian investigation team. But there is no data on usage, storage and disposal of mercury-based medical equipment, for that reason it is difficult to know whether mercury releases from health care organizations or not, if yes what effect will be posed to health workers, patients as well as community and environment. Some countries have restricted the use of mercury thermometers or have banned them without prescription. For our country, mercury tools have been used in all sections of health care. Therefore, it needs to list all mercury-based medical devices used in health care organizations and to investigate the conditions of handling, storage, transportation and disposal of mercury containing equipment. The overall objective of the study is to conduct mercury-based medical devices used in health care organizations and develop strategy and recommendations on further activity. The specific objectives of the demonstration project are:

1. To list mercury-based medical devices used in governmental and private health care organizations;
2. To assess the current situations with regards to storage and disposal management for mercury-based medical
devices used in selected health care organizations;
3. To investigate dental amalgam use in dental hospitals and determine mercury concentrations in samples of dental amalgam;
4. To develop recommendations for further activities based on survey results.

MATERIALS AND METHODS
A cross-sectional study design was used. Totally 578 units of 38 governmental and private health care organizations in Ulaanbaatar, Darkhan, Erdenet cities and Uvurkhangai aimags were conducted in the survey. The survey was conducted by means of a questionnaire given to the medical workers and doctors to complete. There were 3 parts of questions. The first part of the questionnaire dealt with the use of mercury-based medical devices, working, transportation and storage conditions, and waste management. The second section was concerned with knowledge, attitude and practice (KAP) of medical personals for safety handling, storage and disposal of mercury containing devices. The third part of the questionnaire dealt with the dental amalgam. Mercury concentration of dental amalgam samples were detected by portable mercury vapor analyzer RP-91, PYRO-915+ in the Poison Information Center of Public Health Institute. Data processing was done by using statistical program SPSS-10.

RESULTS
Mercury-based medical devices which we found were divided into 5 groups by applications; 1) devices for measuring temperature (room and body temperature) and pressures including blood pressure sphygmomanometer 2) esophageal dilators and other tubes, 3) electrical devices, 4) Lighting bulb and 5) mercury containing laboratory reagents and chemicals. As a result of the survey, the devices for measuring temperature and pressures were used frequently in the tertiary level hospitals (p=0.016) and esophageal dilators and lighting tools were used more in the secondary level hospitals (p=0.009, p=0.0068). No statistical significant differences in use of fever thermometers, blood pressure sphygmomanometers, dilatators and electrical devices were found between urban and rural hospitals (p=0.218, p=0.916, p=0.516). But the use of fluorescence lamps and UV lamps in rural hospitals were significantly high (p=0.003) compared with urban hospitals.

Thermometers and blood pressure sphygmomanometers were accounted for 38% and 24% among the total of 797 devices that categorized by mercury-based devises for measuring pressures and temperatures used in health care organizations (Figure 1).
Unsafe storage conditions were dominated in the rural hospitals. 40.4% of the rural medical personals answered that they did not exactly know how to store mercury devices in a safe way (p<0.0001). For laboratory reagents and tools with mercury, 37.3% of the survey units were not known how to keep them and 30.3% were kept on the special shelves of laboratory tables. 58.5% of the rural hospital units were not known the proper storage of mercury-based reagents and devices and 33.7% of the urban hospital units were not known the special shelves of the laboratory tables (p<0.0001). Generally, 46.8% of the total hospital units (n=408) were transported the mercury-based medical devices with original containers or with special plastic containers. 56.6% of the tertiary level hospitals were carried mercury devices with original containers and 51.6% of the primary level hospitals were transported the mercury-based devices in a safe way. However, no significant statistical differences in transportation status (p=0.011, p=0.007, p=0.005) were observed between health care facilities of the three levels.

When questioned what you will do if mercury containing large equipment had broken, 41% of the total (425) survey hospital units answered that put in plastic bags and wrap up the top of bags tightly then place safe areas, 23% said that inform to the National Emergency Management Organization (NEMO), 14% do not know what should do and 13% said dispose with hospital wastes (Figure 4).

When had broken mercury-based medical small devices such as fever thermometer and sphygmomanometers, 67.1% of the total (414) survey hospital units were disposed to hazardous waste landfills, 12.8% were burnt it with other hospital wastes, 7.5% were put in trash cans and 12.6% were answered do not know how to dispose. Especially, medical workers of the primary and secondary level hospitals were answered that they do not have appropriate disposal and treatment methods for broken small mercury-based devices. 98.3% of the total medical personals were not involved screening analysis for mercury in blood, hair and urine.

**DISCUSSION**

Mercury, one of the world’s most ubiquitous heavy metal neurotoxicants, has been extensively used in health care since antiquity. It has been an integral part of many medical devices, most prominently thermometers and sphygmomanometers. In Mexico City, the 250-bed “Federico Gomez” Children’s Hospital is a medical service, teaching, and research hospital affiliated with the National Autonomous University of Mexico. This prestigious children’s hospital documented a thermometer breakage rate of 385 per month, or well over 4,000 per year. The total number of estimated broken thermometers in this one hospital between 2002 and early 2007 is nearly 22,000 – the equivalent of 22 kilograms of mercury. In a study of New Delhi hospitals, the NGO Toxics Link found dangerously high levels of mercury in a series of indoor air samples. They found the “substantial presence of mercury in ambient air of both the hospitals” studied. These levels, which ranged from 1.12 microgram per cubic meter to 3.78 microgram/m³, were all higher than numerous international standards. One of the biggest mercury hot spots that Toxics Link found in its study was the room used to calibrate blood pressure devices (sphygmomanometers), which contain 80-100 grams of mercury or 80-100 times the amount found in a single fever thermometer. As a result of our study we found that thermometers, light sources and sphygmomanometers were used frequently in Mongolia. 38% of fever thermometer and 24% of blood pressure sphygmomanometers of the total (797) investigated temperature and pressure measuring devices was indicated that it is possible to contaminate indoor air of hospitals when release mercury from broken medical devices. Particularly, improper disposal of mercury containing broken tools, burning with other medical wastes in open areas, putting directly in trash can, leads to increase health and environmental risks in medical personals, patients and community. Environmental Protection Agency (EPA), in 1996, prior to the mercury phase-out in U.S. health care, medical waste incinerators was the fourth largest source of mercury emissions to the environment. Hospitals were also known to contribute 4-5% of the total wastewater mercury load. And mercury fever thermometers alone contributed...
about 15 metric tons of mercury to solid waste landfills annually. In 2005, Transande et al. using national blood mercury prevalence data from the US Centers for Disease Control estimated that between in this century 316,588 and 637,233 US children each year have cord blood mercury levels > 5.8 μg/L, a level associated with loss of IQ 8–11. European parliament made a decision to ban use of fever thermometer in European countries in 2007. But any regulations related to ban or phase out mercury-based medical device application are developed in Mongolia. Based on our study we found out that to organize training and develop promoting material is one of the important issues to increase awareness of medical workers, doctors for handling, storage, transport and disposal principles when work with mercury equipment.

**CONCLUSIONS:**

1. Mercury containing devices such as thermometer, blood pressure sphygmomanometer, energy saving fluorescence lamp and thermostats were used in urban and rural hospitals.
2. There are not any regulations for safe handling, storage, and transportation and disposal system of mercury containing devices.
3. The current situations for inappropriate disposal system can be posed to increase risks of environmental pollution with mercury.
4. Knowledge on handling, storage and disposing mercury based devices are not enough among the medical personals

**ACKNOWLEDGEMENT**

We would like to thank Sh.Enkhtsetseg, environmental officer of WHO and B.Tsetsegsaikhan, officer of Ministry of Health, Mongolia for supporting and advising of our survey to be success. Also we would like to express our gratitude to head of province, district and cities hospitals, head of medical units and quality managers for collaborating and assisting the survey and study team members for hard working to achieve the study goals.

**REFERENCES:**

Tumor Infiltrating T lymphocyte Pattern in Mongolian and Han Nationals with Uterine Cervix Cancer

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2: Department of Oncology, School of Medicine, HSUM;
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ABSTRACT

Uterine cervix cancer (UCC) or cervical cancer is a malignant neoplasm arising from cells originating in the cervix uteri. Worldwide, UCC is second most common and the fifth deadliest cancer in women. In many solid tumors the presence of tumor-infiltrating immune cells correlates with better overall survival. In this patient based descriptive-retrospective study were investigated stored surgery tissue materials obtained from total 150 patients (78 Han and 72 mongolian nationals) with UCC, including 41 patients with in situ carcinoma, 50 patients with microinvasive carcinoma and 59 patients with invasive carcinoma. Immunohistochemical staining techniques using anti-human CD4, CD8, and CD25 primary monoclonal antibodies, was performed in 150 dehydrolyted and stored as paraffinized block tissue materials. Mean age of patients of Mongolian nationals was (51.5±5.5 year; CI 95%: 40-64) similar to patients of Han nationals (49.3±6.9; CI 95%: 36-69) and majority of patients (53.3%) belongs to age group of 45-54 year. Percentage of Han patients with invasive cancer (38.7% vs 0.7%) was notably higher (c²=84.3, p=0.000) than Mongolian patients. Count and infiltration degree of anti-CD4, anti-CD8, anti-CD25 antibody stained cells and CD4/CD8 ratio have tends to decrease in advanced stages of the UCC. Mean value of CD4+, CD8+, CD25+ cells and CD4/CD8 ratio and percent of tumor infiltrated CD4+ and CD25+ T cells significantly elevated in Mongolian nationals comparing to Han nationals (p<0.05). However portion of Han patients with advanced tumor stage was much greater than Mongolian nationals. Comparison of mean count and percentage of Tumor infiltrating T cells of patients in same stage of tumor was demonstrated no nationality related significant difference (p>0.05) in count and percentage of CD4+ T cells, CD8+ T cells, CD25+ T cells and CD4/CD8 ratio. Comparison of mean values and percentage of CD4+ T cells, CD8+ T cells, CD25+ T cells and CD4/CD8 ratio of Mongolian and Han patients with different degree of cancer cells had demonstrated no significant difference (p>0.05) between two nationals.

Key words: Tumor infiltrating T lymphocytes, uterine cervix cancer, immunohistochemical staining

INTRODUCTION

Uterine cervix cancer (UCC) or cervical cancer is a malignant neoplasm arising from cells originating in the cervix uteri. Worldwide, UCC is second most common and the fifth deadliest cancer in women. Worldwide, in 2008, it was estimated that there were 473,000 cases of cervical cancer, and 253,500 deaths per year. In the United States, it is only the 8th most common cancer of women. In 2008 in the US an estimated 11,000 new cases were expected to be diagnosed, and about 3,870 were expected to die of cervical cancer. Among gynecological cancers UCC ranks behind endometrial cancer and ovarian cancer.

In Mongolia UCC prevalence for 100,000 populations were rated as 6.6 cases of morbidity and 3.4 cases of mortality in 2000, but it increased dramatically and rated 15.5 cases of morbidity and 4.0 cases of mortality in 2008.

In People’s Republic of China 75434 new cases (9.6 cases for 100,000 population) and 33914 deaths (5.2 cases for 100,000 population) of UCC were registered.

Conventional cancer therapeutic modalities, such as chemotherapy and radiation of tumor, and small molecule targeted therapies, have been thought of as agents acting

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directly on tumor cells and/or the tumor vasculature. Patients are believed to respond to these therapies based on the inherent susceptibility of their tumor to the specific therapeutic modality versus dose-limiting toxicities. Little attention has been given to the host immune system in terms of prognosis and/or potential response to therapy. On the other hand, the role of the immune system in cancer has long been known.6

It is known that patients with immunosuppressive disorders such as HIV/AIDS, or undergoing immunosuppressive regimens for transplantation, have been shown to have a higher incidence of certain cancers. In recent years, numerous studies have reported a link between prognosis and/or response to conventional therapy, and tumor immune infiltrate in several different solid tumor types.6

In many solid tumors the presence of tumor-infiltrating immune cells correlates with better overall survival.6 However, it is necessary to know the phenotype of the immune cells; for example, in colorectal cancer the number of CD8+ T-cells and a high ratio between CD8+ and CD4+ T-cells have been correlated with increased survival, whereas high numbers of mature dendritic cells predicted shorter survival.5 Regulatory T-cells have been shown to have both a positive and a negative impact in colorectal carcinoma, possibly depending on the location of the infiltrate and on the definition of ‘regulatory T-cells’ used. These cells were also shown to be a positive predictive factor in head and neck cancer, but a negative predictor in hepatocellular, pancreatic, ovarian, endometrial, cervical and breast carcinomas. However, as mentioned above, one must be extremely careful in distinguishing true regulatory T-cells with suppressive functions from activated T-cells.7-15

Our objective was to compare tumor infiltrating T lymphocyte count and infiltration degree in Mongolian and Han patients with uterine cervix cancer in relation with cancer stage and cell differentiation profiles.

MATERIALS AND METHODS

Patient based descriptive-retrospective study design was used.

Stored surgery tissue materials obtained from total 150 patients (78 Han and 72 mongolian nationals) with UCC, treated in Department of gynecological surgery of Affiliated Hospital of Inner Mongolian Medical University, including 41 patients with in situ carcinoma, 50 patients with microinvasive carcinoma and 59 patients with invasive carcinoma were investigated.

Immunohistochemical staining techniques using anti-human CD4, CD8, and CD25 primary monoclonal antibodies, was performed in 150 dehydrolyted and stored as paraffinized block tissue materials according to diagnostic kit protocol attached by manufacturer (Mai Xin Biological, Fujian, China). Mean number (M) of antibody stained or not stained cells for a microscope field of vision (from 5 fields of vision) counted using Smart scape 2002 Biological microscopic image analysis system/ S/N:SV-0002393 software in images from light microscope transferred into personal computer was calculated. Percentage of tumor infiltrating cells subset was calculated were calculated from total number of mononuclear cells counted in a field of vision.

Degree of T cell infiltration was calculated using cut-off value for each cell subset infiltration percentage (18% for CD4+ and CD25+ cells and 22% for CD8+ cell). Statistical calculation was performed using independent T test, one-way ANOVAs test and Pearson’s Chi-square (c²) test.

RESULTS

Mean age of patients of Mongolian nationals was (51.5±5.5 year; CI 95%: 40-64) similar to patients of Han nationals (49.3±6.9; CI195%: 36-69) and majority of patients (53.3%) belongs to age group of 45-54 year. In table 1 shown cancer stage of Mongolian and Han patients with UCC, attended to the study.

<table>
<thead>
<tr>
<th>Stage</th>
<th>Mongolian nationals</th>
<th>Han nationals</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>In situ carcinoma</td>
<td>30/20.0</td>
<td>11/7.3</td>
<td>41/27.3</td>
</tr>
<tr>
<td>Microinvasive carcinoma</td>
<td>41/27.3</td>
<td>9/6.0</td>
<td>50/33.3</td>
</tr>
<tr>
<td>Invasive carcinoma</td>
<td>1/0.7</td>
<td>58/38.7</td>
<td>59/39.4</td>
</tr>
<tr>
<td>Total</td>
<td>72/48</td>
<td>78/52.0</td>
<td>150/100.0</td>
</tr>
</tbody>
</table>
As shown in table 2 percentage of Han patients with invasive (38.7% vs 0.7%) was notably higher ($\chi^2=84.3$, $p=0.000$) than Mongolian patients. In table 2 shown mean value of tumor infiltrating T lymphocytes of patients with UCC, in relation with cancer stage.

**Table 2**

<table>
<thead>
<tr>
<th>Tumor infiltrating immune cells</th>
<th>In situ carcinoma (n=41)</th>
<th>Microinvasive carcinoma (n=50)</th>
<th>Invasive carcinoma (n=59)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>CD4</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Count</td>
<td>26.7±0.3</td>
<td>21.1±0.3$^f$</td>
<td>15.8±0.3$^{ch}$</td>
</tr>
<tr>
<td>Percentage</td>
<td>19.6±0.2</td>
<td>18.1±0.2$^f$</td>
<td>15.8±0.3$^{ch}$</td>
</tr>
<tr>
<td><strong>CD8</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Count</td>
<td>31.2±0.4</td>
<td>29.6±0.9$^f$</td>
<td>24.9±0.7$^{ch}$</td>
</tr>
<tr>
<td>Percentage</td>
<td>22.5±0.2</td>
<td>21.6±0.1</td>
<td>21.7±0.2</td>
</tr>
<tr>
<td><strong>CD25</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Count</td>
<td>22.9±0.1</td>
<td>21.1±1.1$^f$</td>
<td>16.8±0.3$^{ch}$</td>
</tr>
<tr>
<td>Percentage</td>
<td>18.1±0.1</td>
<td>18.1±0.2</td>
<td>15.5±0.2$^{ch}$</td>
</tr>
<tr>
<td><strong>CD4/CD8</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Count</td>
<td>0.86±0.01</td>
<td>0.71±0.00$^f$</td>
<td>0.63±0.01$^{ch}$</td>
</tr>
</tbody>
</table>

$^a$ – Mean and standard error  
$^{b,c,d}$ – Antibody stained cells;  
$^e$ – Ratio of anti-CD4 and anti-CD8 antibody stained cells;  
$^f$ – Statistical significance when compared with mean value of in situ carcinoma patients $p<0.05$;  
$^{ch}$ – Statistical significance when compared with mean value of microinvasive carcinoma patients $p<0.05$

As shown in table 2 count and infiltration degree of anti-CD4, anti-CD8, anti-CD25 antibody stained cells and CD4/CD8 ratio have tends to decrease in advanced stages of the UCC.

**Table 3**

| Mean number of tumor infiltrating T cell subsets and their ratio (M±SE)$^a$ |
|-------------------------------|-------------------|-------------------|-------------------|
|                              | CD4$^{+b}$        | CD8$^{+c}$        | CD25$^{+d}$       | CD4$^{+}/CD8^{+e}$ |
| **Han nationals**             |                   |                   |                   |                   |
| Count                        | 17.9±0.5          | 26.0±0.3          | 18.0±0.3          | 0.7±0.01          |
| Percentage                   | 16.6±0.2          | 21.7±0.1          | 16.2±0.2          |                   |
| **Mongolian nationals**       |                   |                   |                   |                   |
| Count                        | 23.4±0.4$^f$      | 30.5±0.2$^f$      | 21.8±0.2$^f$      | 0.8±0.01$^f$      |
| Percentage                   | 18.8±0.2$^f$      | 22.1±0.1          | 18.5±0.1$^f$      |                   |
| **Total**                    |                   |                   |                   |                   |
| Count                        | 20.5±1.1          | 28.2±0.3          | 19.9±0.3          | 0.72±0.01         |
| Percentage                   | 17.6±0.2          | 21.9±0.1          | 17.3±0.2          |                   |

$^a$ – Mean and standard error;  
$^{b,c,d}$ – Antibody stained cells;  
$^e$ – Ratio of anti-CD4 and anti-CD8 antibody stained cells;  
$^f$ – Statistical significance when compared with mean value of Han nationals

As shown in table 3 mean value of CD4$^+$, CD8$^+$, CD25$^+$ cells and CD4/CD8 ratio and percent of tumor infiltrated CD4$^+$ and CD25$^+$ T cells significantly elevated in Mongolian nationals comparing to Han nationals ($p<0.05$).

However portion of Han patients with advanced tumor stage was much greater than Mongolian nationals (table 2). So we have compared mean count and percentage of Tumor infiltrating T cells of patients in same stage of tumor. There was demonstrated no nationality related significant difference ($p>0.05$) in count and percentage of CD4$^+$ T cells (Figure 1), CD8$^+$ T cells, CD25$^+$ T cells and CD4/CD8 ratio of patients with same stage of cancer.
**Table 4**

<table>
<thead>
<tr>
<th>Cell differentiation degree</th>
<th>Mongolian nationals</th>
<th>Han nationals</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Well differentiated</td>
<td>29/13.3</td>
<td>23/7.3</td>
<td>52/27.3</td>
</tr>
<tr>
<td>Moderately differentiated</td>
<td>19/27.3</td>
<td>29/6.0</td>
<td>48/33.3</td>
</tr>
<tr>
<td>Low differentiated</td>
<td>30/0.7</td>
<td>20/38.7</td>
<td>50/39.4</td>
</tr>
<tr>
<td>Total</td>
<td>72/48</td>
<td>78/52.0</td>
<td>150/100.0</td>
</tr>
</tbody>
</table>

As shown in table 3 distribution of Han and Mongolian patients with different degree of cancer cell differentiation had no significant difference (p=0.1).

Comparison of mean values and percentage of CD4+ T cells, CD8+ T cells, CD25+ T cells and CD4/CD8 ratio of Mongolian and Han patients with different degree of cancer cells had demonstrated no significant difference (p>0.05) between two nationals.

**DISCUSSION**

There are many reports highlighting correlation between T cell infiltration of tumor tissue and cancer stage, survival of patient with different types of solid cancers, particularly cervical cancer. Piersma et al. (2007)\(^1\) investigated human papilloma virus (HPV)-induced cervical cancer and found a significantly stronger CD8+ T-cell infiltration, a higher CD8/CD4 T-cell ratio and a higher CD8+/regulatory T-cell ratio in patients with no metastases to the draining lymph nodes, which is associated with better prognosis (n = 59, p<0.05).

Badoual C. et al. (2006)\(^1\) found that only two variables influenced overall survival probability: T stage (P = 0.036) and CD4(+)CD69(+) T-cell infiltration (P = 0.017).

Yi-Lan Zhang et al. (2010)\(^1\) has investigated 106 biopsy specimens from newly diagnosed nasopharyngeal cancer patients of Han nationality and found the density of CD8+ TIL was positively correlated with lymph node metastasis (p<0.05) meanwhile a low density of CD8+ TIL was correlated with better progression free survival in early stage patients (stages I and II, p<0.05).

Zhang Y et al. (2006)\(^1\) have compared lymphocyte-population bearing surface markers in anticoagulated peripheral blood samples of 75 Uygur and 104 Han normal healthy people in Xinjiang province of Peoples Republic of China. Were established the helper T cell percentage and CD4/CD8 ratio of Uygur male were lower than those in Han male. And in female, Uygur people had higher percent of NK cell (p<0.01), but lower CD25+ cell than those in Han’s (p<0.01). They have concluded the nationalities and gender could influence the reference value of lymphocyte immunophenotype, the reference values of blood lymphocyte immunophenotype in the adults of Uygur and Han nationalities in Xinjiang.

But there were no reports indicating the difference in tumor infiltrating lymphocytes in different nationalities.

**CONCLUSIONS**

1. Count and infiltration degree of anti-CD4, anti-CD8, anti-CD25 antibody stained cells and CD4/CD8 ratio have tends to decrease in advanced stages of the uterine cervix cancer.
There were no differences in count and infiltration degree of tumor infiltrating T cells in Han and Mongolian patients with uterine cervix cancer

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15. Piersma E, Mariette L.E. van Poelgeist E, et al. High number of intraepithelial cd8+ tumor-infiltrating lymphocytes is associated with the absence of lymph node metastases in patients with large early-stage cervical cancer Cancer Res. 2007:354
Usage of E-Library Among Branch Schools’ Teachers of Health Sciences University of Mongolia

1Health Sciences University of Mongolia
2Central Medical Library

ABSTRACT

In an age where information technology is developing rapidly, more and more people are turning to digital books rather than paper books. Digital books have become the main form of literature in many developed countries, and in other countries such as Mongolia, it is quickly becoming the main source of information for young adults, specially students and teachers.

This research evaluates the state and usage of e-library among HSUM professors, identifies the level of knowledge and trend of e-library among university students and teachers, and reflects the opinions of professors in the development of e-library.

This was a cross-sectional design of analytical study and retrospective directional descriptive design to study current activity of e-library of HSUM by evaluating documents and reports of Central Medical Library as well as survey which consisted of 28 questions, divided into 4 groups from 133 teachers from branch schools of HSUM. The data analysis was processed using SPSS 20.0 program.

Based on the results, the usage of e-library is increasing each year among HSUM professors. Majority of the professors support the development of e-library, however, are unenthusiastic about publishing their works in digital form. 81.8% of professors use Google search engine to obtain free information, which reflects the inadequate knowledge professors have of other free literature databases.

Although the usage of e-library is increasing, there are complications for reading electronic materials. We suggest the following solutions that will support the growth of e-library: provide information of e-library to professors and researchers, ensure the stability of internet connection in rural areas and expand the e-library fund to include learning materials required for some courses being taught at HSUM.

Keyword: Online information database, e-library, e-book

INTRODUCTION

As rapid growth in information technology continues, the demand for electronic information data base and publication of electronic literature is high. More and more professors and students are using the internet to acquire knowledge, and the digital database of materials and literature used for research are growing exponentially.¹

E-book funds started increasing due to electronic publications since 1980, and since 1990, electronic publications started including academic literature.²

Use of e-library is just becoming a trend in Mongolian universities, and many libraries have begun to use e-library programs to search, read and expand their library database. There are many electronic publication companies; however, the publication of academic literature among them is very low. Therefore, many university libraries often scan Mongolian academic literature into the system and search for free foreign literature to expand their catalog. There are many challenges to appeal to readers in Mongolian universities, including a lack of policy or guideline on e-library, absence of legal framework on copy right, and inadequate training on e-library.

With the support of WHO, HSUM created its own e-library in 2001 with information database mainly in audio and video format. In 2005, the implementation of HINARI
international electronic medical database created the foundation for the e-library fund, which has expanded its catalog with 3203 e-books and 11 online information databases.

Although e-library usage in Mongolia is in its early stage, this research aims to focus on evaluating the usage of e-library about teachers and students, identify any complications in using e-library and define a solution to complications to further develop e-library system in Mongolia.

MATERIALS AND METHOD
This was a cross-sectional design of analytical study and retrospective directional descriptive design to evaluate documents, reports of Central Medical Library (CML) as well as survey.

For this research, a survey was taken from 133 (n=133) professors working at branch schools in Darkhan-Uul, Gobi-Altai and Dornogobi provinces as well as School of Health Technology (SHT) of HSUM. The survey consists of 28 open and closed-ended questions that fall into 4 categories. The questionnaire result was processed using SPSS 20 software.

RESULTS
From the 133 teachers that participated in the study 22.6% (30) were from SHT, 21.8% (29) from Darhan-Uul, 23.3% (31) from Gobi-Altai, and 32.3% (43) Dornogobi provinces. By age groups 16.7% (22) were under 25, 34.6% (46) were 26-35, 24.8% (33) were 36-45, 15.8% (21) were 46-55, and 7.5% (10) were over 55. Gender wise 23.3% (31) were male and 76.7% (102) were female teachers. By degree 31.6% (42) has acquired a bachelor’s degree, 61.7% (82) has master’s degree and 6% (8) has PhD.

Table 1.

<table>
<thead>
<tr>
<th>In use</th>
<th>Used in trial</th>
</tr>
</thead>
<tbody>
<tr>
<td>• HINARI</td>
<td>• Ebsco Host</td>
</tr>
<tr>
<td>• New England Journal Of Medicine</td>
<td>• Nursing Education in Vide</td>
</tr>
<tr>
<td>• OECD</td>
<td>• AMED</td>
</tr>
<tr>
<td>• IMF</td>
<td>• Taylor &amp; Francis Group</td>
</tr>
<tr>
<td>• Royal Society Journal Of Archive</td>
<td>• Springer</td>
</tr>
<tr>
<td>• BioOne</td>
<td></td>
</tr>
<tr>
<td>• Oxford University Press</td>
<td></td>
</tr>
<tr>
<td>• Cambridge University Press</td>
<td></td>
</tr>
<tr>
<td>• PubMed</td>
<td></td>
</tr>
<tr>
<td>• CochraneCollaboration</td>
<td></td>
</tr>
</tbody>
</table>

Table 2.

<table>
<thead>
<tr>
<th>Year</th>
<th>2007</th>
<th>2008</th>
<th>2009</th>
<th>2010</th>
<th>2011</th>
<th>2012</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 Online database access</td>
<td>439</td>
<td>420</td>
<td>315</td>
<td>588</td>
<td>983</td>
<td>7651</td>
</tr>
<tr>
<td>2 E-book usage</td>
<td>163</td>
<td>268</td>
<td>293</td>
<td>398</td>
<td>1814</td>
<td>1491</td>
</tr>
</tbody>
</table>

From Table 2 it is clear that from 2007-2012, online database usage has increased 8-10 times. HINARI online database access has increased 8 times from 2011-2012.

Even though 97.7% of the teachers involved in the research answered that e-library is mandatory in university libraries and 94.7% believed that online material is significant to education, there aren’t any teachers that have uploaded their textbooks as an e-book. 81.8% of teachers use free online information databases, 6.8% of them use paid databases and 11.4% do not use databases.
In the study, 58.3% of the participants read textbooks, 52.5% are journal articles, 35.7% dissertation/thesis, 49.2% are conference reports, 30% are essays, 40% are lectures, 10.8% new information.

42.9% of teachers use online information on a weekly basis while 36.1% use it daily. 80.5% of teachers use it for tutoring and 72.2% use it for research.

**Figure 2. The usage of online information /by degree/**

From the above figure, 76.9% of teachers with PhD use the online database weekly.

**Figure 3. Difficulties of using online database**

21.06% of teachers answered that when using an electronic material there were some difficulties, 78.94% replied that there weren’t any.

**DISCUSSION**

Many national and international studies on development of e-library were conducted, including “JISC National E-books Observatory Project (2008)”, “208 Global Student E-book Survey”, and “E-book Collections: ARL Spec Kit 313 (2009)” which studied the usage of e-book in libraries. The results of such studies show that usage of e-books are increasing which showed similar result to our study.3

Based on a nationwide survey by Mongolian Librarian Consortium in 2013, 41% of 17 libraries in Mongolia have transitioned to digitalized system; however, due to weak legal framework on copyrights, many textbooks and learning materials in Mongolian have not been converted to electronic forms.4

**CONCLUSION:**

1. There is an inadequate collection of e-books in Mongolian. Majority of the participants support the development of e-library in universities believing that it benefits the learning process. However, none of the participants have placed their publications on an e-library.

2. The usage of e-library by teachers has increased in 2012 and the majority of them use e-library fund as weekly for their tutoring, as well as for research.

3. 78% of teachers said that there are no difficulties to use e-library, but minority has problems related to internet connection, language barrier, no materials in Mongolian., etc.

We suggest the following solutions that will support the development of e-library: provide information of e-library to professors and researchers, ensure the stability of internet connection in rural areas and expand the e-library fund to include learning materials required for some courses being taught at HSUM.

**REFERENCES:**


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Text should consist of introduction including a brief review of the literature; details of Research design, Subjects, Materials and Methods, Ethics, Statistics; Results and Discussion. Long articles should provide sub-headings. Abbreviations and Nomenclature List in an alphabetical order non-standard abbreviations contained in the manuscript (excluding references) with definitions after the keywords. Use abbreviations sparingly and only when necessary to save space, and to avoid repeating long chemical names or therapeutic regimes. In a figure or table, define the abbreviations used in a footnote.

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3. Book

4. Book chapter

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