The George Washington University

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Revolutionizing Medicine Through Research and New Technology

Am delighted to present the 2015 edition of Fusion magazine. Fusion is a publication created by the students of the William H. Beaumont Society, a prestigious honor society established at the George Washington University (GW) School of Medicine and Health Sciences (SMHS) to promote student research. The depth and breadth of the work presented in the following pages is a testament to the level of energy and intellectual curiosity of SMHS medical students.

As a recent arrival at GW, I have been extremely impressed with medical student engagement in the research endeavor. Their involvement is fundamental to moving the frontiers of knowledge forward. The world of biomedical research is changing dramatically. New technologies are revolutionizing medicine. They span all aspects of biomedical research ranging from real-time analysis of the interactions of identified cells and even molecules in non-invasive ways through to the rapid genomic interrogation of large cohorts of patients. Such advances allow us to grasp new principles of physiology and pathology that were previously elusive. While much has been learned, it is clear there remains a lot of work to do.

The emergence of the Ebola epidemic over the last year demonstrates our critical needs both for more basic research to understand modes of viral transmission and to develop effective therapies and vaccines, as well as the need to effectively deliver health care worldwide. Such needs will be met in the future by biomedical researchers such as those whose work is represented in this copy of Fusion.

The research presented here by SMHS medical students represents the outcome of hard work and dedication to the scientific enterprise and for this they deserve high recognition. More importantly, the work inspires tremendous confidence that SMHS medical students, through a combination of outstanding research and health care delivery, will be the medical innovators of tomorrow. The faculty of SMHS congratulate the students of the Beaumont Society for their achievements and look forward to supporting their future successes.

Robert H. Miller, Ph.D.
Senior Associate Dean for Research
Vivian Gill Distinguished Research Professor
Professor of Anatomy and Regenerative Biology

Letter from the Beaumont Society:

As your co-presidents of the William H. Beaumont Medical Research Honor Society (Beaumont Society), we are proud to present you with the 2015 edition of Fusion—a student-run research journal that serves as a forum for students to share their basic science, clinical, and health policy research experiences with the GW School of Medicine and Health Sciences (SMHS) medical community.

Our 2015 edition showcases a spectrum of student research projects, including basic science research, clinical research, health policy, and research abroad. This year, we invited medical students to submit abstracts for projects that were completed while attending SMHS. The Beaumont Society’s editorial board, composed of SMHS M.D. program students from the classes of 2015 and 2016, was responsible for the editing and production of this year’s journal.

We have an exciting new addition to the Beaumont Society’s annual activities this year—a Student Research Seminar Series. This series creates an arena for students to present and discuss their past or present research projects. It enables students to better understand the clinical relevance of scientific research and provide them with the chance to critically evaluate their peers’ projects.

The Dean’s Office and the Office of Student Opportunities led by Cynthia Powell, have been paramount in introducing students to research opportunities nationally and internationally through the William T. Gill Summer Fellowship, the GW Health Services Scholarship, and the Research Track Program, which helps students direct their careers toward academic medicine. We are most grateful for the support of Robert H. Miller, Ph.D., senior associate dean for research, Vivian Gill Distinguished Research Professor, and professor of anatomy and regenerative biology, whose guidance helped shape the current mission of the Beaumont Society, as well as the many dedicated faculty members at SMHS who mentored our peers. We would like to thank the SMHS Office of Communications and Marketing for assistance with the production of this journal. Finally, we also thank our editors and artists for their hard work in making Fusion a great success each year.

We invite you to share our enthusiasm for student research, and encourage our peers to continue careers in academic medicine to advance patient care in all dimensions, whether in basic science, knowledge, clinical care, or health policy. Thank you and we hope you enjoy Research Day and Fusion 2015!

Sincerely,
Allison Ikeda and Karoline Jaluba
Co-Presidents of The William H. Beaumont Medical Research Honor Society
Comparison of Adaptive and Innate Immune Responses Induced by Licensed Vaccines for HPV

Douglas Herrin, MSII
ADVISORS: Barry Gurnham and Julie Ledgewood
Vaccine Research
Center, National Institutes of Health

STUDENT CONTRIBUTORS: Emily Costes, Pamela Costner, Troy Kemp, Martha Naso, Kapil Sahania, David Pan, Umarine Sermoir, Lindsay Sullivan, Gaiyna Yarmishchok, Richard Koup, Yi Li, Ying Peng, Robert Sedder, John Schiller, Barney Graham, Liga Pinto, and Julie Ledgewood

The human papillomavirus (HPV) is one of the most common sexually transmitted infections in the United States. There are more than 130 subtypes of HPV, of which 16 and 18 are classified as oncogenic and are important causes of anal, cervical, and oropharyngeal cancers in men and women. HPV-16/18 account for 70% of cervical cancer cases, and 21% of cervical cancer cases are associated with the closely related non-vaccine types within the Alpha-papillomavirus species group. HPV-16: 31/35/39/52/58 and A7 (HPV 18-like: 39/45/68). A7 (18-like: 39/45/68). A7. Globally, there are an estimated 530,000 cases diagnosed with more than 235,000 cervical cancer deaths each year.

Two HPV viruses, vaccine related in patients, HPV-16/18 (Gossin et al., Cervarix®) and HPV-6/11/16/18 (Merck, Gardasil®), are currently licensed in the United States. Aluminum salt is the main adjuvant in the Gardasil® vaccine which promotes a Th2 response and enhances the time for antigen exposure, while Gardasil® contains the AS04 adjuvant which includes aluminium salts and a TLR4 agonist and activates the MyD88/TRIF pathways. Given the similar antigenic content but different adjuvant formulation, in the two vaccines, they provided an efficient method for evaluating adjuvants and comparing the kinetics of the innate and adaptive immune responses. We randomized women to receive either Cervarix® or Gardasil®, followed six month vaccination delivery schedules per manufacturer’s recommendations, and then analyzed the humoral immune response, T cell response, and circulating plasma cytokine levels in response to vaccination.

In this study we observed that Cervarix® and Gardasil® are both highly immunogenic vaccines. Cervarix® recipients had higher anti-HPV-16 antibody and neutralization titers at month seven, and elevated anti-HPV-18 antibody and neutralization titers at months seven and 12 (Figure 1). Peak antibody titers were observed after three doses of both vaccines, whereas peak antibody titers were observed after two doses of Gardasil® indicating a more prominent booster effect from the third dose of Cervarix®. Patterns of antibody avidity were overall similar for the two vaccines, with durable maintenance of plateauing levels up to 24 months after last dose of vaccine. We also evaluated levels of cytokine genes related to non-vaccines neutralizing antibodies and observed that HPV-31 only had evidence of cross-protection and only in response to Cervarix®. Comparing CD4 T cell response and circulating plasma cytokine levels in response to vaccination. This study is the first to report a comprehensive immune comparison between Cervarix® and Gardasil®, with novel data on T cell responses. This is also the first study to measure an extensive time course of circulating plasma cytokine profiles within hours and days after vaccination with the HPV vaccines. The biological implications of the differences in immune responses are unknown, but there is increasing evidence that the antibody response induced after two vaccinations, or even one, may be sufficient for protection. As such, the World Health Organization recently changed the recommendations to two vaccine doses, administered at months one and six, in girls when vaccination is initiated prior to 15 years of age.

Given the potential differences in mechanisms of action of the adjuvants in these two vaccines, the observed cytokine profile may suggest an influence and activation of different cell types that translate into differences in the circulating cytokines and adaptive immune responses. Innate cellular response at the time points studied, including microarray analyses, would aid in interpretation of the differences and the cell components underlying the cytokine profile observed. Understanding the kinetics, breadth, and magnitude of the innate and adaptive immune responses can contribute to identification of candidates for correlates of protection against infection and aid in development of a systems biology approach to vaccine evaluation, and in building predictive models of vaccine immunogenicity and efficacy.

REFERENCES:
Epigenetic Alterations in a Transgenic Alzheimer’s disease Model

Matthew Bake, MSI
ADvisor: Gary D. Sacks, Ph.D.
STUDENT CONTRIBUTORS: Nour Tahir, Michael Canin, Rebecca Haraf, and Amands Hazy, Dept. of Biology and Chemistry, Liberty University, Lynchburg, Va.

The pathological features of Alzheimer’s disease (AD) have been researched and documented extensively, however the causes of these features are still unknown. Genetic contributions leading to the development of AD are mostly obscure, however several lines of evidence point to epigenetic modifications, specifically cytosine methylation, playing a pathological role. Cytosine methylation represses transcription of genes by blocking transcriptional machinery through several different mechanisms, ultimately resulting in down-regulation of genetic products.

First, more than 90% of AD cases are late onset and appear to be sporadic, meaning that no genetic cause has been determined.1 To expound on this factor, a non-Mendelian mode of acquiring AD is proposed due to the fact that twin studies looking at the development of AD show differing results.2,3 These studies suggest that DNA sequence alone does not lead to AD development. Second, mouse and human studies provide evidence that DNA methylation is altered between affected and control counterparts on a genomic level in DNA obtained from regions of the brain affected by AD.4 While there were significant changes in diseased parts of the brain, methylation levels from unaffected regions of the brain remained unchanged between the affected and control counterparts. This specificity appears to be direct evidence that DNA methylation is involved in AD pathology. Also, several genes that have been linked to AD pathology, such as APP, β-APP cleaving enzyme, and neprilysin, are transcriptionally regulated by DNA methylation in their promoter regions.5,6

A final factor providing evidence for epigenetic alterations contributing to AD pathology is a recent expression study that showed over and under expressed mRNA levels from several genes that are associated with processes malfunctioning in AD, which could be the result of methylation changes in the promoter region of these genes.7 Together, these factors provide a strong argument for an epigenetic impact on the development of AD pathology. If determined, these specific pathological alterations could not only give insight into the molecular pathology behind the development of AD, but they could also provide a signature to help in the early determination of AD, as well as in the target and development of future therapies.

Our study compared the methylation levels of DNA promoter regions from transgenic mice, which over express beta amyloid as seen in AD, not of control transgenic mice. Two methodologies were used for determining promoter methylation statuses. The first methodology used was HpaII tiny fragment Enrichment by Ligase-mediated PCR, or more commonly the HELL assay, which uses methylation sensitive

...IThe MeDIP data seemed to suggest that the regions undergoing the most change in methylation were those linked to microRNA genes. MicroRNA genes seem to work by shutting off expression of other genes through translational inhibition.

HELP ASSAY LINKED ONTOLOGICAL FUNCTIONS UNDERGOING METHYLATION CHANGES

<table>
<thead>
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<th>Scale</th>
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*Italic text - Onotologial functions associated with genes becoming hypomethylated

MEDIP ASSAY LINKED ONTOLOGICAL FUNCTIONS UNDERGOING METHYLATION CHANGES

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*Italic text - Onotologial functions associated with genes becoming hypermethylated

have target mRNA involved in neuronal and apoptotic functions. Therefore, the up or down regulation of these microRNA genes through alterations in epigenetic methylation may be inducing neuronal or apoptotic pathology that can only be determined through expression studies. Overall, this data provides vast support for our hypothesis that epigenetic methylation may be leading to AD related pathology.

REFERENCES:
Validation of Genetic Variants Associated with Physical Activity in a Young African American Cohort

Trevor Sleazak, MS

ADVISORS:
Laura Torri, M.D.
Joseph Devaney, Ph.D.
Heather Gordish-Dressman, Ph.D.
Eric Hoffman, Ph.D.

1 Department of Orthopaedic Surgery, Children’s National Health Network
2 Children’s Research Institute

It has been established that the inherent drive to begin or continue exercising is closely linked to one’s genetic make-up. Estimates are that from 41 to 71% of the variance in adult exercise behavior can be explained by genetics with the remaining variance attributed to environmental factors. Until recently, genome-wide association studies (GWAS) examining physical activity have been performed primarily in adult Caucasian cohorts. The objective of this study was to determine whether six single nucleotide polymorphisms (SNPs) previously demonstrated to be associated with leisure-time exercise behavior in an older Caucasian population 1 (mean age 76 years) are also associated with physical activity levels in young African Americans (mean age 7 years).

Our cohort consisted of 143 African American children, aged 5–9 years, who were originally recruited for a study on fracture risk and vitamin D levels. Parents of participants reported the average number of hours their children spent per day in outdoor playtime. Parental reporting has been shown to be a reliable measure of physical activity in children. 2 DNA samples were collected, SNPs were genotyped, and associations between SNPs and phenotypes were tested using a one-way ANOVA. All analyses used a dominant genetic model to compare homozygous common allele individuals to heterozygous and homozygous rare allele individuals.

SNP rs9633417, shown to be associated with physical activity in Caucasian adults, was also found to be associated with physical activity in young African Americans. Rs9633417 is located within the SGIP1 gene; SGIP1 is primarily expressed in the hypothalamus and is implicated in the regulation of energy homeostasis. 127 of the children studied had the homozygous common allele (CC) and were found to play outside an average of 3.2 hours per weekday. Three children in the cohort did not report time played outside. The remaining 11 children had the less common heterozygous allele (AC) and were found to play outside an average of only 1.6 hours per weekday—a 36 minutes less per day than their peers (p = 0.046; Figure). One SNP associated with physical activity in the older Caucasian cohort was not directly associated with physical activity in young African Americans but did correlate with body composition (Table). Rs10946904 is located in the PRSS16 gene, which encodes renin, a serine protease. In our cohort, however, it is associated with total body fat and total percent body fat. The mechanism of the PRSS16 gene’s effect on physical activity and body composition is not yet known. The remaining four SNPs were not associated with physical activity or body composition in our cohort.

This is the first study to look specifically at SNPs associated with physical activity in a young African American cohort. This area of research has significant public health implications because the African American community has been found to suffer from high rates of obesity and obesity-associated health risks, and physical activity has been shown to decrease adiposity. 4,5 To date, most GWAS studies have used cohorts that were of European ancestry, and only recently have researchers paid attention to persons of differing race and ethnicity. If we hope to encourage personalized medicine strategies, more effort will be required to identify the genetic variants specific to diverse populations. At the same time the positive associations found with rs9633417 and rs10946904 deserve further analysis in additional populations as these they may prove important among other communities.

REFERENCES:
The Optimization of Treatment Planning and Ablation Rate Improvements on Feasibility of Pediatric MR-HIFU Applications

Doug Wackerle, MSII
ADVISORS:
Matthew E. Ostien, M.D,1,7 Pavel S. Yarmolenko, Ph.D,7 and Haydar Celik, Ph.D,1,2
1 Georgia Washington University
School of Medicine and Health Sciences
2 Children’s National Health System

Magnetic resonance-guided high intensity focused ultrasound (MR-HIFU) ablation provides a precise, non-invasive treatment for various lesions in adults. This technique uses MR-imaging to guide an ultrasound beam at a frequency high enough to induce hyperthermia in targeted tissue (Figure 1). In children, MR-HIFU’s potential remains largely unexplored, though its non-invasive and non-ionizing nature holds promise for pediatric medicine. Yet, pediatric patients pose challenges affecting treatment: young children require general anesthesia, exhibit wide ranges of anatomy, and have varying lesion sizes and locations. To address these challenges, the overall treatment must be reduced. This may be achievable through standardized treatment approaches and physical aids to optimize patient position and faster repositioning. Further improvement of ablation rate and reduction of risk are also possible via improved monitoring of skin temperature during ablation and mild hyperthermia. Improvements in treatment planning and volumetric rate may save time and allow for treatment of larger lesions, increase patient throughput, and possibly increase efficacy and lower cost. This study aims to examine and quantify how such improvements could increase the portion of anesthesia time spent on direct ablation (the actual treatment) and produce better patient outcomes.

We retrospectively examined 41 pediatric patients with various limb tumors at Children’s National Health System from November 2009 to October 2013 as potential candidates for MR-HIFU ablation therapy. After identifying the tumor location (Figure 1, left), we used software (Arizco Standard Edition 8.0.0, Visualization Sciences Group, SAS, Berlin, Germany) to define its area through axial slices (Figure 2) and create a 3D segmented model to measure its volume (Figure 2). As a reference, we used an estimated ablation rate, obtained from Phillips Healthcare, of 180 cc/hour as the maximum. Anesthesia time was limited to four hours to reduce risks to the children, and to address problems with restraints on surgeon focus and room and machine time, and cost. Within the four-hour anesthesia window, significant time is often spent on treatment planning and patient repositioning. In this study, we seek to show how spending a higher proportion of that time actually ablation can allow the treatment of larger lesions and more patients.

Finally, we graphically combined this tumor volume and ablation rate data to show effects of theoretical improvements. We found that increasing the time available for ablation can substantially increase treatable tumor volume (Figure 3). In the examined 41 patients, using only one hour for ablation (at 180 cc/hour) left 15 patients (37%) untreated.

With more time, up to three or four hours of ablation, only two patients (5%) would be untreated. In addition to treatment time, ablation rate was also found to be a significant factor in patient outcomes. Complete treatment of a lesion was directly related to ablation rate (Figure 3). At the current rate of (180 cc/hour), two patients (5%) were untreated, yet with double the current rate (360 cc/hour), all 41 lesions could be treated. Although there is some risk of hyperthermia and skin burns with an increased ablation rate, new real-time temperature monitoring techniques being developed can allow for faster and safer treatment of these large lesions. Improvements in planning guidelines and treatment rates could have substantial impacts on the effectiveness of MR-HIFU ablation and the size of treatable tumors and number of patients treated with this technique.

REFERENCES:

FIGURE 2:
Susceptibility to Obesity and Bone Mineral Density in Young African American Populations

Michael Liu, MSII; with Elizabeth Hedges, MSII; Zachary Kendrick, MSII; Elizabeth Dominici, MSII; Jacqueline McKeisy, MSII.

ADVISORS: Courtenay A. Sprouse, Heather Cardish Dressman, Ph.D.; Retica M. Ryan, M.D.; David Rowlands, Ph.D.; Kevin Bell, Joseph M. Devaney, Ph.D.; Laura L. Tosi, M.D.1

1Children’s National Health System, 2The George Washington University School of Medicine and Health Sciences, 3Johns Hopkins University School of Medicine, 4Georgetown University School of Medicine, 5School of Sport and Exercise, Massey University, New Zealand, 6Wakeelfeld Sports Medicine

In the United States in 2014, 31.8% of children and adolescents were obese.6 Studies have found that overweight and obese children are at a greater risk to become obese adults.2 This disease further disproportionately affects minority populations including African American (AA) children. Studies show that 20% of AA children ages 2–19 are obese compared to 14% in Caucasians.2 Affected individuals are more likely to suffer from many chronic diseases including type 2 diabetes, cardiovascular diseases, and several types of cancers.

To date, most genome-wide association studies (GWAS) have identified various loci associated with BMI within European origin.4 However, given the disparate effects of obesity on Europeans and AAs, it is crucial to generate a better understanding of the genetic influences on obesity and how they may be modulated within the AA pediatric population.

Monda et al. recently published a GWAS identifying six independent SNPs associated with BMI.1 These six SNPs were selected for genotyping in our pediatric AA cohort to examine the relationship between genetic risk variants for obesity and bone mineral density.

This study includes AA children, ages 6–9. Case patients had an isolated and radiographically demonstrated forearm fracture (radius, ulna or both bones) and control patients had no self-reported history of a prior bone fracture.

Dual energy X-ray absorptiometry scans were obtained, and participants received whole body and lumbar spine scans because these are most accurate and reproducible in pediatric patients.4 Phenotypes analyzed here included total body and lumbar bone mineral density (BMD). All measurements in the BH cohort were calculated without inclusion of the head. DNA was isolated from blood samples and

Given the disparate effects of obesity on non-whites, it is crucial to understand the genetic influences on obesity. This will offer the possibility of better intervention and treatment options in the future.

REFERENCES:
In order to improve quality of care, many Electronic Health Records (EHRs) have recently developed patient portals, which allow healthcare providers to communicate electronically with their patients. As part of the 2009 Health Information Technology for Economic and Clinical Health (HITECH) Act, the Medicare and Medicaid EHR Incentive Program was created to motivate providers to demonstrate “Meaningful Use” of their EHRs in order to receive incentive payments. One such requirement mandates increased patient engagement through electronic communication. This can pose a problem for patients who lack regular access to computers and the Internet or those who have low literacy or English proficiency.

To better understand how to meet the needs of this patient population, a survey was conducted at a network of safety-net clinics in the uninsured in Montgomery County, Maryland. Moreover, patients were multi-ethnic with a majority speaking Spanish only or both English and Spanish. The survey sought to understand patient use of technology and interest in using these technologies to communicate with their healthcare providers. Prior studies have shown that in safety-net populations, there is high use of technology, a strong desire to use patient portals to manage health, and a want for resources to increase technological literacy.1,2

A total of 50 surveys were collected at the three clinic sites over a month-long period. Upon checking in, patients were given a survey in English or Spanish to complete in the waiting area. If they required assistance, the survey was administered verbally by clinic staff. The findings mirrored what exists in the literature: patients have access to the Internet (primarily through their cell phones) and are interested in using patient portals, smartphone applications, and text messaging to communicate with their healthcare providers.

Text messaging ranked as the most preferred method of communication. Among types of text message communications, nine in ten patients were interested in receiving appointment reminders. The second most interested in receiving preventive reminders for an annual flu shot.

Given these findings, it is important to develop appropriate promotional and patient education materials, including both video and written instructions. As a whole electronic forms of communication have the potential to increase patient satisfaction and improve quality of care.

REFERENCES:

In addition to the patient survey, a provider survey was conducted to identify potential provider barriers and facilitators. When asked to identify characteristics of patients who may have difficulty using a patient portal, older age and limited English proficiency ranked highest. This finding is similar to studies showing that providers view younger, more computer-literate patients as likely users of a portal and offered enrollment less frequently to older, non-English speaking and/or uninsured patients. This points to the need to ensure that providers discuss the portal with all of their patients and that the clinic staff offers portal enrollment to all patients.

Given these findings, it is important to develop appropriate promotional and patient education materials, including both video and written instructions. Portals should also be tailored to a variety of platforms, with a special emphasis on smartphones. It may also be worth while to have a computer kiosk in waiting areas with a staff navigator to guide patients through the sign-up process. Secure text messaging may also prove to be an easy way to communicate with patients. As a whole electronic forms of communication...
Can Uganda’s Village Health Teams Endure Without Material Support?

Daniel Mašs, MSIV
ADVISORS: Edward O’Neil Jr., M.D.; Edison Mezarrow, M.D.; and W. Scott Schrott, M.D., M.P.H.*

*The George Washington University School of Medicine and Health Sciences, Onni Med., Makerere University College of Health Sciences

For more than a decade, Uganda’s Ministry of Health has led a community health worker program model in which the Village Health Team (VHT), a cadre of unpaid volunteers, is charged with delivering preventative health services and education to their local community. Studies have demonstrated the effectiveness of Uganda’s VHTs in improving certain health outcomes; however, it is known that VHTs are not optimally supported.

There has been steady VHT attrition in recent years. The Ministry of Health has recognized the inadequate support of VHTs thus far and is aiming to expand VHTs to all local governments and explore ways of sustaining VHTs.

The objective of this study is to evaluate the extent to which “material support” is a deciding factor in the efficiency, scalability, and durability of Uganda’s VHT initiative. Material support may take the form of monetary stipends, regular payment, transportation assistance, or other materials such as mobile phone airtime.

This study will review the existing literature and gather novel data through surveys of VHT members and VHT stakeholders. The quantitative and qualitative survey data will be analyzed for trends that may point to a conclusion in the context of existing health policy discourse on community health worker remuneration. The purpose of this study is to strengthen the knowledge base on whether or not material support is an indispensable and indispensable means of support for Uganda’s VHTs. This information can be used by government and non-governmental organizations in their work to strengthen and sustain VHTs throughout the country.

REFERENCES:

Overview of Bhutanese Traditional Medicine and Observations on the Integrated Western and Traditional Health Care System in Bhutan

Ian Barrows, MSIII
ADVISOR: Bob Morrow, M.D., FACPM, The George Washington University School of Medicine and Health Sciences

Bhutan established a unique national health care system in 1967. The official government policy was to preserve and extend traditional Bhutanese medicine, a 1200 year old Tibetan medical practice and system, while simultaneously offering integrated Western medical services. Both services are provided free of charge to the population, and integration of the two systems provide more options for patients. Additionally, traditional medicine and the use of natural products can be an important resource for future drug discovery.

The data for this project was collected through six weeks of direct observation and interviewing of Bhutan health care professionals and focused on traditional Bhutanese medicine, its integration with Western medicine, and the future possibilities of this assimilated system.

To better understand the medical practice in Bhutan, it is important to understand its origins from Buddhist medicine. From a traditional Buddhist perspective, health entails a fine balance within the body. Disturb this balance, and one’s health is disturbed. It is believed that all imbalances originally stem from ignorance, which in turn leads to apathy, then to attraction, hatred, and destruction (the three poisons). Each of these poisons originates from a different organ within the body and manifests with specific symptoms. For example, attachment, associated with desire, greed, or jealousy, comes from the genital organs and results in neutral or wind disorders such as breathing and psychological disorders. Hatred manifests itself as a hot disorder related to bile and blood sicknesses, headaches, and vision problems, while delusion can affect the stomach and joints. Diagnosis in Bhutanese medicine can involve questioning the patient, observing symptoms, taking one’s pulse, or performing a urine analysis. Pulse taking can inform the practitioner about the underlying poison, and it is a skilled art as the proper interpretation takes years to master.

Currently, Bhutan has hospitals and health clinics throughout the country offering both Western and traditional care. Interestingly, many citizens seek care from both practices, and patients’ preference for one form of medicine over the other cannot be defined by age, socioeconomic status, or any other easily defined classification. Findings indicate however, that the system can hardly be considered integrated. Patients are free to seek care from either traditional or Western physicians, yet care from the two practices is seldom integrated.

Few physicians of one background have adequate understanding of the other practice. This makes it difficult to refer patients, collaborate on treatment plans, or effectively work together with their counterpart colleagues.

To bridge the gap between the two practices of medicine, the model Mental Health Integration program was initiated in 2013. This program has been successful during its short existence in incorporating unique insights, knowledge, and other components from both medical practices into the care of patients. Physicians are trying to assess how each practice treats comparable symptoms in patients and the outcomes associated with each approach. It is hoped that this can be used as a model for integrated improvement in other fields of medicine as well.

At the present time though, the Bhutanese health care system is not truly integrated; therefore it is unable to maximize on the benefits of an integrated system. Further research is needed to follow-up on the effectiveness of the mental health integration effort, as well as to investigate the mechanism of action and efficacy of traditional Bhutanese treatments.