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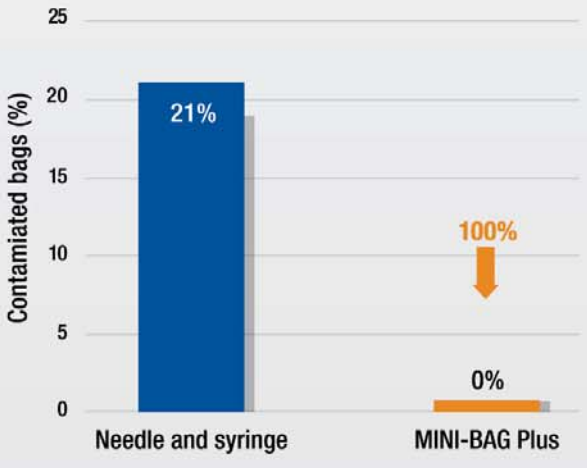

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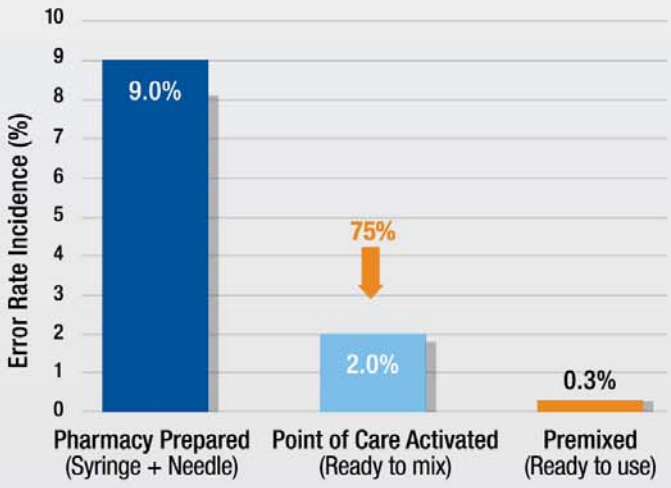

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* On ward comparison of reconstitution methods in a British Hospital

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Eradication of Infectious Diseases by Means of Systematic and Scientific Approaches

Microbial organisms are beneficial to other life not only because they can re-cycle wastes and stuffs but also produce numerous useful substances or materials for our daily consumption and promotion of a healthy life. However, they are also responsible for many infectious diseases and food spoilage.

Epidemics of infectious diseases have been well documented in history. In most incidents that lead to the death of thousands of life, it was because of the spread or the uncontrollable propagation of a pathogenic microorganism. For examples, epidemics of smallpox, leprosy, tuberculosis, meningococcal infections, diphtheria and pandemic flu etc. in ancient Greece and Egypt were well recorded in history. In recent years, we have witnessed the outbreak of Asian flu (1957-58), HIV/AIDS pandemic (1960-present), SARs (2002-3), bird flu (2009), Ebola virus diseases (2014-present), Swine flu (2015-present) etc. in different regions. Emerging of these devastating diseases was attributed to failure in implementing a good strategy to prevent outbreak or developing an effective treatment if someone were infected.

Since infectious diseases are non-discriminatory, they can lead to tremendous loss of resources or life upon individual as well as the whole society. It is always a major public health concern and top priority of any government. Pharmacists, who are part of the healthcare professionals, should be familiar and equipped with adequate knowledge and skills on how to prevent transmission of infectious agents before their outbreak and how to apply an effective treatment if a person were unfortunately infected. Hence, some training in good preventive strategy and treatments are vital to all practicing pharmacists. In this issue, LAI's article (p7) addresses the importance of implementing immunization in workplace for all workers.[1] She pointed out that vaccines not only protect vaccinated individual from a disease but also create herd immunity if a sufficient proportion of the community is immune to a infectious disease. As a result of this approach, unvaccinated individuals could be benefited from exposure to a lower risk environment. The author reminds us what can be achieved and what can't be achieved.

Although prevention is a better approach than post-infection treatment, sometime advance immunization or even some treatments such as bathing with 2% Chlorhexidine (p5) may not work or effectively prevent infection.[2] Thus, it is necessary to apply drugs for effective treatment of infected person. In an article (p17) contributed by LEE and CHEUNG on hurdles of pharmaceutical development faced by many scientists in last year for effective prevention and treatment of the Ebola virus diseased, it summarizes the latest state of their development.[3] The article is informative and up-to-date. This editorial believe that infectious diseases could only be effectively eradicated by means of systematic approaches and in depth scientific explorations of its epidemiology, etiology and new drug discovery.

When I started thinking how to write this editorial after having spent the last day of the 2015 Hong Kong Pharmacy Conference, which was held from March 28 to 29 in the Hong Kong Convention and Exhibition Centre, I was convinced by the hot debate from both for and against side in the plenary session on an issue about the establishment of a local pharmacy council. I told myself a detailed report on this issue should be included for wider circulation and comments as it will have fundamental impacts on pharmacy practice in Hong Kong. I conveyed my request to some panel members and with their support, a report on this plenary discussion was eventually written and passed on to me for publication in this issue (p38).[4] If you missed this hot yet important discussion in the conference, it is the chance for you to aware what have been happening. You are encouraged to go through it and I guarantee whoever closely follows up this issue will be beneficial in their pharmacy practice in long run as you know and respect thyself and thy adversary.

Finally, after a long overdue I am proud to present the first issue of volume 22 of Hong Kong Pharmaceutical Journal to your hand. I hope all of you not only enjoy reading all the articles but don't mind to share your knowledge or experiences with other colleagues through sitting down to write article to this journal. Because we have too many articles, some articles are not printed in this issue. To those affected authors, I have to apologize. Hopefully your article could be included in next issue.

Cheung Hon-Yeung
Editor-in-Chief
11th May, 2015

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(Prepared by Kwan Him Shek, Leung Ho Cheung Brian, Tse Wai Tak Marco, Wong Chi Lap Raymond & Wong Lai Yan Janet)

Asthma Increases Risk of Developing Obstructive Sleep Apnea

Date: January 13, 2015

Obstructive sleep apnea is generally more prevalent among patients with asthma. Whether asthma is associated with the development of Obstructive Sleep Apnea (OSA) remains unknown.

In order to examine the relationship of asthma with incident OSA, a population-based prospective epidemiologic study has been run by the Wisconsin Sleep Cohort Study since 1998. Adult participants were recruited from a random sample of Wisconsin state employees to attend overnight polysomnography studies at 4-year intervals. Asthma and covariate information were assessed during the studies through March 2013. Eligible participants were identified as free of OSA (apnea-hypopnea index [AHI] of <5 events/h and not treated) by 2 baseline polysomnography studies. There were 1105 4-year follow-up intervals provided by 547 participants (52% women; mean [SD] baseline age, 50 [8] years). Questionnaires were employed for assessing presence and duration of self-reported physician-diagnosed asthma. Meanwhile, the associations of both presence and duration of asthma and OSA concomitant with habitual daytime sleepiness were estimated.

Twenty-two of 81 participants with asthma experienced incident OSA over their first observed 4-year follow-up interval compared with 75 of 466 participants without asthma. Using all 4-year intervals, participants with asthma experienced 45 cases of incident OSA during 167 4-year intervals and participants without asthma experienced 160 cases of incident OSA during 938 4-year intervals; the corresponding adjusted relative risk (RR) was 1.39, controlling for sex, age, baseline and change in body mass index, and other factors. Asthma was also associated with new-onset OSA with habitual sleepiness (RR, 2.72 [95% CI, 1.26 - 5.89], $P = .045$). Asthma duration was related to both incident OSA (RR, 1.07 per 5-year increment in asthma duration [95% CI, 1.02 - 1.13], $P = .01$) and incident OSA with habitual sleepiness (RR, 1.18 [95% CI, 1.07 - 1.31], $P = .02$).

By and large, asthma was statistically associated with an increased risk of new-onset OSA.

Source: jama.jamanetwork.com

Daily Bathing of Critically ill Patients with 2% Chlorhexidine Doesn't Reduce Infections

Date: January 27, 2015

Daily bathing of critically ill patients with the broad-spectrum, topical antimicrobial agent chlorhexidine is widely performed and may reduce health care–associated infections. A large-scale study of 9340 patients in 5 adult ICU for a year.

Units performed once-daily bathing of all patients with disposable cloths impregnated with 2% chlorhexidine or non antimicrobial cloths as a control. Bathing treatments were performed for a 10-week period followed by a 2-week washout period during which patients were bathed with non antimicrobial disposable cloths, before crossover to the alternate bathing treatment for 10 weeks. Each unit crossed over between bathing assignments 3 times during the study.

During the chlorhexidine bathing period, 55 infections occurred while in the control bathing period, 60 infections occurred. The primary outcome rate was 2.86 per 1000 patient-days during the chlorhexidine and 2.90 per 1000 patient-days during the control bathing periods. After adjusting for baseline variables, no statistically significant difference between groups in the rate of the primary outcome was detected. These findings do not support daily bathing of critically ill patients with chlorhexidine.

Source: jama.jamanetwork.com

Septrin and Spironolactone Increase Risk of Sudden Death, Study Finds

Date: February 2, 2015

The risk of sudden death, as a result of severe hyperkalemia, was investigated amongst periods aged 66 years or above. We identified cases those died of sudden death within 14 days after receiving a prescription for trimethoprim-sulfamethoxazole or one of the other study antibiotics (amoxicillin, ciprofloxacin, norfloxacin or nitrofurantoin) were followed.

Of the 11 968 patients who died of sudden death while receiving spironolactone, 328 died within 14 days after antibiotic exposure. Compared with amoxicillin, trimethoprim–sulfamethoxazole was associated with a more than twofold increase in the risk of sudden death. Nitrofurantoin was also associated with an increased risk of sudden death, although the risk with nitrofurantoin was not apparent in a sensitivity analysis.

In the view that the antibiotic trimethoprim–sulfamethoxazole was associated with an increased risk of sudden death among older patients taking spironolactone, alternative antibiotics should be considered in these patients when clinically appropriate.

Source: www.cmaj.ca

NSAID Use (Even Short-term) Increases Bleeding and Thrombotic Events in Post-MI Patients, Study Finds

Date: February 24, 2015

Although the usage of antithrombotic treatment for post-myocardial infarction (MI) patients is indicated, there are safety concerns if non-steroidal anti-inflammatory drugs (NSAIDs) are being concomitantly used.

A test was set up to examine the risk of bleeding and cardiovascular events when using NSAIDs for patients after first-time MI with current antithrombotic treatment. By monitoring the occurrence of hospitalization-required bleeding or mixed cardiovascular outcomes like stroke, non-fatal recurrent MI and cardiovascular death, the risks are calculated using adjusted time-dependent Cox regression models according to current NSAIDs and antithrombotic therapy.

61 971 patients with 63% being men and with mean age being 67.7-year-old are included. Comparing patients being given concomitant NSAIDs treatment and without NSAID treatment, the crude incidence rates of bleeding were 4.2 and 2.2 events per 100 person-years respectively. Furthermore, cardiovascular events' rates were 11.2 and 8.3 events per 100 person-years respectively. It is found that bleeding and cardiovascular risks are both increased with NSAID treatment comparing to no NSAID treatment by the multivariate-adjusted Cox regression analysis. Regardless of antithrombotic treatment, the types of NSAIDs being used, or usage duration, there was evidence showing that the risks of bleeding and cardiovascular events were increased with concomitant use of NSAIDs.

In conclusion it was found that use of NSAIDs was associated with elevated risk of bleeding and over-increased thrombotic events even only given in short term. Physicians should be cautious when prescribing NSAIDs for patients with recent MI despite more studies have to be done to confirm the above findings.

Source: jama.jamanetwork.com

Air Pollution is Costing Our Children's Health

Date: March 11, 2015

Air pollution has been a problem in many areas. Association between the resulted better air quality and improved children lung function was assessed in a cohort study in Southern California. The study included 3 separate cohorts of children in separate 3 calendar periods, 1664-1998, 1997-2001, and 2007-2011. The mean ages of children were 11 years and 15 years at the beginning and the end of the study period in each cohort. Lung function of the total 2010 children was measured annually. The increase of forced expiratory volume in 1 second (FEV1) and forced vital capacity (FVC) from 11 to 15 years of age was referred as 4-year growth.

Long term reductions in air pollution were found to be associated with positive effects on lung function growth in children. After adjusted for several potential confounders, improvements in 4-year growth of FEV1 and FVC were found to be associated with declining levels of nitrogen dioxide ($P<0.001$ for FEV1 and FVC). Associations were also observed between improved 4-year growth and reduced levels in particulate matter with an aerodynamic diameter of less than 2.5 μm ($P= 0.008$ for FEV1 and $P<0.001$ for FVC) and less than 10 μm ($P<0.001$ for FEV1 and FVC). As the air quality improved, the proportions of children with clinically low FEV1 (defined as <80% of the predicted value) at 15 years of age decreased from 7.9% to 6.3% to 3.6% across the three periods ($P=0.001$). The associations were observed in both boys and girls, with and without asthma.

Source: www.nejm.org

Culmulative Anticholinergics Use Increase Risk of Developing Dementia in the Elderly, Study Claims

Date: March, 2015

Anticholinergic-induced cognitive impairment is one of causes of anticholinergic therapy discontinuation. A possible association between anticholinergics use and increased risks for dementia has been suggested.

A 10-year cumulative dose-response relationship was observed for dementia and Alzheimer disease (test for trend, $P<.001$). 797 participants (23.2%) developed dementia (637 of 797 [79.9%] developed Alzheimer disease) during the mean 7.3 years of follow-up. Comparing cumulative anticholinergic use with nonuse, adjusted hazard ratios for dementia were 0.92 (95% CI, 0.74-1.16) for TSDDs of 1 to 90; 1.19 (95% CI, 0.94-1.51) for TSDDs of 91 to 365; 1.23 (95% CI, 0.94-1.62) for TSDDs of 366 to 1095; and 1.54 (95% CI, 1.21-1.96) for TSDDs greater than 1095. For Alzheimer disease, results of similar pattern were observed.

The increased risk for dementia was found to be associated with higher anticholinergic use. It is suggested that, when prescribing anticholinergics for the older adults, the potential risk should be taken into account. Possible alternatives and lowest effective doses of anticholinergic therapy should be considered. The aged should be educated about this potential medication-related risk for the aged.

Source: archinte.jamanetwork.com

A Review of Establishing a Workplace Immunization Program

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ABSTRACT

The workplace and the health of its workers are inseparably linked. Workplace is one of the priority settings for health promotion and workplace immunization programs are such examples of a workplace health promotion program. In 2010, the Department of Health launched a workplace health promotion program, "Health@work.hk Pilot Project". However, this pilot project does not include the immunization program. The steps to establish a workplace immunization program to fit the local setting are explored.

Keywords: *workplace, health promotion, immunization program, cost benefit*

INTRODUCTION

A vaccine is any preparation that can develop immunity to a disease by stimulating the production of antibodies in the body.[1] It develops immunity by imitating an infection. This type of infection causes the immune system to produce antibodies and T-lymphocytes without causing any illness.[2] Once the body recovered from the imitated infection, a supply of "memory" B-lymphocytes that produce antibodies can be found in the body. Then the body can remember how to fight that disease in the future.[2]

Traditionally, vaccination is designed to protect the children as their immune system is in development. However, there are too many adults who have become ill and are disabled or even die from diseases that can be easily prevented by vaccine.[3] Vaccine not only protect vaccinated individual from vaccination-preventable diseases but also create herd immunity if a sufficient proportion of the community is immune to a infectious disease. In this circumstance, even unvaccinated individuals are at lower risk of disease as the disease has limited opportunity to spread within the community.[3] Both younger adults and older adults can benefit from vaccination.

Workplace is one of the best candidates to promote the adult immunization. Establishing a workplace immunization program requires a systemic approach. According to the US Centers for Disease Control and Prevention, influenza and pneumococcal vaccination are involved in the program.[4] In this article, the steps to establish a workplace immunization program will be discussed.

INFLUENZA

Background

Influenza is an acute viral infection that spreads easily from person to person through droplets when the infected person coughs, sneezes or talks. Direct contact with the secretion of the infected people can also spread the disease.[5] Influenza can affect anybody in any different group worldwide. Sudden onset of high fever, cough, headache, muscle and joint pain, severe malaise, sore throat and running nose are the symptoms of influenza. Most patients can recover from the symptoms within one week without any medical attention. But it can cause severe illnesses such as chronic heart, lung or metabolic disease, or weakened immune systems.[5] Influenza epidemics occur annually during autumn and winter in temperate regions. The annual epidemics cause about 3 to 5 million cases of severe illnesses and about 0.25 to 0.5 million deaths.[5] Besides, it can cause high levels of absenteeism and productivity losses and eventually result in economic problems.[5]

According to the result of sentinel surveillance (Season 2011-2012) conducted by the Centre for Health Protection (CHP) (Fig. 1), the average monthly consultation rate of influenza-like illness reported by General out-patient (GOPC) clinics and general practitioner (GP) are 4.6 and 40.7 per 1000 consultations respectively.[6]

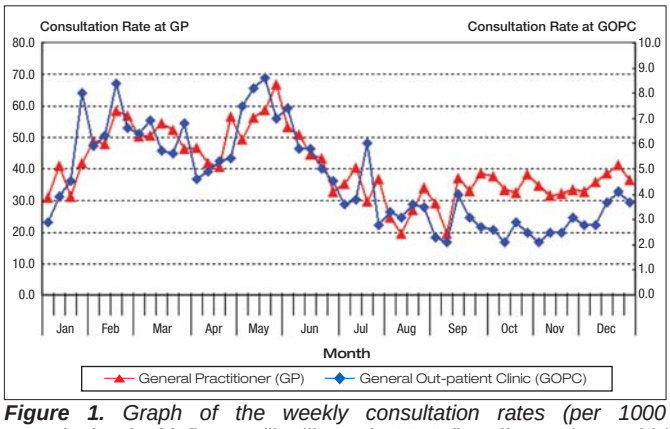

Figure 1. *Graph of the weekly consultation rates (per 1000 consultations) of influenza-like illness in 2012 (http://www.chp.gov.hk/en/data/1/26/44/292/640.html)*

Influenza vaccine

Vaccination is the most effective way to prevent influenza.[5, 7] In Hong Kong, there are two types of registered seasonal influenza, inactivated trivalent influenza vaccine (TIV) and live attenuated influenza vaccine (LAIV). TIV can be given

intradermally to adults aged 18 years or above. LAIV can only be given intranasally to healthy, non-pregnant and non-immunocompromised people aged 2-49 years old.[7] As there will be emergence of new strains and change of circulating strains from time to time, it is recommended to get vaccinated against influenza every year. The World Health Organization (WHO) recommends appropriate formulation of influenza vaccine for every influenza season.[8]

A study conducted by O. Yu At'kov in 2005 demonstrated that influenza vaccination in healthy working adults had good outcomes.[9] In the study, influenza vaccination was offered to employees of Russian Railways Public Corporation. The vaccination demonstrated the effectiveness of reducing the incidence of influenza-like-illness (ILI) by 70.4%.[9] It also resulted in 80.8% reduction in absenteeism and 74.4% reduction in total days with ILI.[9] Besides, the annual cost saving per vaccinated employee ranged from €2.13 to €5.43 depending on the productivity while sick.[9]

PNEUMOCOCCAL DISEASE

Background

Pneumococcal diseases are caused by a type of bacteria called *Streptococcus pneumoniae* which can be frequently found in the upper respiratory tract of healthy children and adults.[10] These bacteria can cause a range of pneumococcal infection ranging from mild otitis media to fatal invasive pneumococcal disease (IPD). Pneumonia, meningitis and sepsis are examples of IPD.[10] The pneumococcal disease can be transmitted by direct person-to-person contact via the respiratory droplets or oral contact from patients and healthy, asymptomatic carriers.[11]

In 2005, according to the WHO, around 1.6 million deaths were caused by the pneumococcal diseases. In the USA and Europe, *streptococcus pneumoniae* account for most of the community-acquired bacterial pneumonia cases in adults. In these regions, the annual incident rate of IPD ranges from 10 to 100 cases per 100 000 population.[12] Since 2007, a laboratory surveillance system on IPD has been set up by the Public Health Laboratory Services Branch (PHLSB) of the Centre for Health Protection (CHP). According to the PHLSB, from 2007 to September in 2011 (Fig. 2), the average numbers of invasive pneumococcal isolates analyzed each year were around 120 for persons of all age groups.[13]

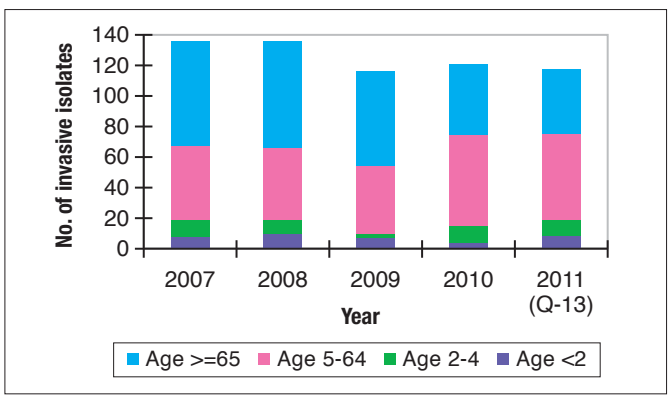

Figure 2. *Graph of the yearly number of invasive pneumococcal isolates by age group, January 2007 to September 2011. (Communicable Diseases Watch 2012 Compendium)*

Pneumococcal vaccine

It is now more difficult to treat pneumococcal infections as bacteria become resistant to some commonly used antibiotics. The antibiotic resistance has both economic and clinical consequences. Therefore, there is a need for the use of vaccine to prevent the pneumococcal disease.[10] In Hong Kong, there are two types of pneumococcal vaccines indicated for healthy adult aged 50 or above: 23-valent pneumococcal polysaccharide vaccine (23vPPV) and 13-valent pneumococcal conjugated vaccines (PCV13).[14-16]

WORKPLACE HEALTH PROMOTION

Background

Along with the school, hospital, city, island, and marketplace, the workplace has been recognized as one of the priority settings for health promotion.[17] The workplace and the health of the workers within it are inextricably associated.[18] It has direct influence on the physical, mental, economic and the social well-being of workers and in turn the health of their families, communities and society.[17] The workplace should not only maintain and protect the safety and well-being of the employees but also provide them opportunities for better long term health and quality of life.[18]

The idea of workplace health promotion (WHP) is to improve the health and well-being of people at work through the combined efforts of employers, employees and society.[19] The WHP programs have many potential benefits for both employees and employers. Employees can benefit from not only reducing risk of life threatening diseases and the associated costs but also improving everyday quality of life. Besides, employees can also gain knowledge, develop self management and coping skills through participating in the health promotion activities.[20] For employers, they can benefit from enhanced employee productivity, reduced employee absenteeism and lower insurance and workers compensation costs. The WHP program can be regarded as a recruitment and retention tool to attract and keep high quality employees and maintain the morale. In addition, the public image of the company can also be enhanced.[20]

Health@work.hk Project

In Hong Kong, with over three million people in the workforce, spending most of the time in the workplace, workplace is an ideal setting to engage people in healthy lifestyles.[21] In 2010, the Department of Health (DH) launched the "Health@work.hk Pilot Project" to call on employers and employees to work together to create a working environment supporting, guiding, motivating and encouraging everyone to live in a better health (Fig.3). The first phase of the project was completed with positive response and outcome from users. In 2012, DH launched the second phase of this project in order to have better benefit for a larger workforce.[21] The five main health modules of the pilot projects are: Physical activity, Healthy eating, Avoidance of alcohol use, Promoting the baby friendly workplace and Smoking cessation.[22] Although immunization program is not involved in this project, the component of the project is a valuable reference as it reflects the local situation in Hong Kong.

Figure 3. *Website of Health@work.hk Project (http://www.cheu.gov.hk/eng/healthatwork/index.asp)*

WORKPLACE IMMUIZATION PROGRAM

In order to build a workplace immunization program, a coordinated, systematic and comprehensive approach should be involved. This should consist of planned, organized and all-round sets of programs, policies, benefits, and environmental supports to promote the staff immunization.[23] Although there are different sets of workplace health promotion models recommended by different health organization, all the models involve a systematic approach. In this article, five main steps are emphasized in the systematic approach of building a workplace immunization program.

STEP 1: IDENTIFY AND ESTABLISH THE PROGRAM TEAM

Components of the team

Setting up a program team is the critical and essential first step to initiate a workplace immunization program.[24] The program team is responsible for providing strategic direction, leadership and organization necessary to operationalize the program.[25] In reality, the program team plays a role in helping the employer understand the critical health problems and cost drivers, assessing the barriers and the work environment, developing and promoting the program.[26] The program team should comprise members from different departments. As a result, more employees will take the ownership of the program and in turn support it.[27]

First of all, a team leader is decided. The team leader is required to lead, advocate and promote the program.[24] For the purpose of facilitating the job of the team leader, the team leader should have support from the senior management and the program team. The leader is responsible for overseeing the annual budget and implementing the program.[26]

Next, the senior managers play a key role in the program team. They can be the strongest support for the program. Their commitment and support can communicate the purpose and the process of the program to all levels of the company which may further gain support from other managers.[27] Furthermore, they can serve as role models being vaccinated in the first line and hold some informal gatherings to educate the employees in order to encourage the participation in the program.[28]

Then, there should be an administrative support in the program team. Taking meeting minutes and maintaining a master file of resource materials are the roles of administrative support.[24]

Besides, the role of communication is also influential. The team member can assist in development and implementation of the immunization program by producing or acquiring educational materials. They can also organize or promote different events during the implementation, for example, campaign kick-off ceremony.[24]

Last but not least, representative from information technology and statistician should also be involved. The information technology representative can help with the development and maintenance of the database system.[24] The statistician can assist with determination of what information should be collected, monitored, reported and evaluated and how the immunization rate will be calculated.[24]

The identified roles of program team members are not mutually exclusive. The size of the company and availability of resources should be considered when establishing the team.[24]

Case Study 1 - Health@work.hk pilot project

Although the Health@work.hk pilot project does not involve the workplace immunization program, the report of the pilot project is still a valuable reference to set up an adoptable workplace health promotion program in local setting. In the pilot project, the participated companies are encouraged to establish a Wellness Committee (WC) having a similar function with the program team mentioned above. The WC comprises senior management (for endorsement), middle management (for planning and executing health policy in the company) and frontline staff (for organizing and encouraging participation in the events).[29] Besides, as it is a pilot project conducted by the Central Health Education Unit (CHEU) of DH, the WC also acts as a contact point with the CHEU. The WC is an essential group in the project as it coordinates different items related to the workplace health promotion program.[29]

Case Study 2 – HK Government Vaccination Programme: Seasonal Influenza Vaccination for DH Staff 2011/2012

In Hong Kong, the uptake rate of seasonal influenza vaccination among the health care workers was low. In an effort to encourage the staff of DH to receive seasonal influenza vaccine, Dr Thomas Tsang, the Controller of the CHP (Fig. 4), has been invited to appear in a short promotion video. Besides, he also received the vaccination for the purpose of role modeling to appeal the participation.[30]

Figure 4. *Dr Thomas Tsang, the Controller of the CHP (Centre for Health Protection. CHP newsletter Issue No. 30)*

STEP 2: ASSESSMENT

Assessment of the barriers

It is necessary to do a thorough assessment to get a snapshot of the company situation to identify the readiness and outline the immunization program objectives.[24] Assessment can be done by different tools such as survey.[31] There are many identified barriers to the program implementation (Table 1).[24]

Each company will have a unique set of barriers to overcome. By assessing and understanding the barriers, the program team is responsible to find the workaround. Therefore, assessment for each company is needed as early and accurate recognition of barriers may improve the immunization program.[24]

Case Study 3 – Baystate Health

At the beginning of the program, the occupational team held forums in which the employees can receive the information and ask questions about the immunization program. After that, the program team continued to spread the news of the program through emails and newsletter articles. Then the team conducted a survey to address the employees' concerns. One of the barriers is the thought of a healthy individual not needing a flu vaccine.[27] To counter this barrier, the program reminded them that they may be a carrier of virus without any symptoms and they may spread the disease unknowingly. The program team found that the concept of getting the vaccination to help protecting those have contacted with the employee resonated with the employee. This case demonstrates the importance of early assessment of the barriers.[27]

STEP 3: PLANNING

Identify goals and objectives

After completing the assessment for the barriers, the next step is to clearly identify the goal and objective of the program. The goal and objective of the program are recommended to be linked and integrated with the company's mission. As a result, the company's commitment and processes to implement the program can be described.[26]

Goals will vary from company to company, but it is necessary to have a target to work towards.[27] Goal is defined as a general statement of what a project is trying to do while objective is defined as a specific, measurable of the wanted change(s) that a project proposed to accomplish with a given period of time.[24] "SMART" objective is more likely to be achieved (Table 2).

After identifying the goals and objectives, the evaluation of the program can be started. Evaluation is not only carried out at the end of the program but should continue throughout the life of the program.[24] Evaluation can involve baseline, process and outcome measures which will be discussed later.

Table 2. Description of "SMART" objective[24]

Key Words	Description
S	**S**pecific (Be specific about what the project is going to achieve)
M	**M**easurable (The objective can be quantified)
A	**A**ttainable (Make sure what the project is going to do can be achievable)
R	**R**ealistic (Make sure the objective is measurable and have necessary resources to support)
T	**T**ime-Bound (State a timeframe to achieve the objective)

Draft a timeline

When drafting a timeline, it is important to set the launch date of the immunization program first. After that, the program team can start to set target date for each of the specific tasks. The program team should avoid tight schedules and unrealistic expectation when drafting the timeline.[24]

Develop communication strategies

Successful implementation of the program mostly depends on how the employees react to it. Even a mild misunderstanding can result in major disruptions such as low participation.[32] Therefore, regular and consistent communication is an essential factor of the program. The employees are one of the key stakeholders. So they should be informed of the program's purpose, the actions taken, the reasons and results of the action through the communication.[32]

If employees are unaware of the available health promotion opportunities, they are not likely to participate. The goal of the program cannot be achieved without sufficient participation of employees.[32]

The aims of developing a communication plan are to increase awareness and recognition of the program; increase awareness of the disease; increase trust between management and employees; increase program participation. A well designed communication plan is able to increase the employee's knowledge and awareness of the disease or solution, refute myths and misconceptions and increase support for the program and events.[32]

In practice, the immunization program should be branded, including designing a logo and used in all communication materials. Different communication channels should be used to ensure the employees receive the information. Emails, bulletin boards, newsletters, intranet, presentations and direct communication from managers can be considered.[32] If the program team consists of members from multiple department,

Table 1. Barriers to implementation and Immunization[24]

Internal (within the company)		External (outside the company)	
Individual	**Organizational**	**Individual**	**Organizational**
- Incorrect information provided - Fear of side effects - Fear of needles - Perceived low benefit-to-risk ratio - Inadequate knowledge about vaccine - Inadequate knowledge about the diseases	- Lack of program resources - Lack of interest from the management - Lack of understanding the roles of program team	- Incorrect information from the internet - Vaccine-influenza strain mismatch - Inadequate vaccine supply	- Negative or confusing media coverage - Inadequate vaccine supply

this plays a role in the communication strategy as it is a way to have broad support within the company.[27]

Besides, there are several considerations when developing the communication plan. The optimum timing and frequency of message delivery should be considered. Revising the content of messages should also be considered as there may be more updated information. It is also critical to ensure the messages delivered are understood and applicable to all individuals.[32]

Dedicate resources

A lack of resources can be challenging to an immunization program. The program team should reallocate and maximize the available resources to maximize the efficiencies and minimize duplication. Resources such as promotional items (e.g. print resources), communication costs (e.g. promotional videos), information technology costs (e.g. database) and vaccine and immunization supply costs (e.g. antiseptic) may be required.[24] Staff time for planning and implementation, space and employee's time to participate are also considered as a kind of resources. The program team can also consider bringing additional resources such as experts from local hospitals or universities to conduct promotion seminars which a program cannot be accomplished by the company alone.[33]

All the necessary resources should be itemized in the annual budget. Tracking of the program costs can be a part of the evaluation to compare the investment with goals and outcomes.[33]

Utilize informatics

The collection, analysis and interpretation of related data are essential to the immunization program. Continuous monitoring of the data is a kind of process.[24] The program team can make use of the data for tracking the participation rate. It is useful to determine and understand participation rates by employee background characteristics (e.g. age group) and job category in order to identify the area for improvement.[33] By continuous monitoring and analyzing the data, the team can brainstorm solutions to the barriers which may not be considered before.[24]

Consider issue of convenience

The program team should think of strategies to allow easier and more convenient access to immunization for the employees. Providing on-site vaccination, mobile vaccine carts and extending the period of vaccination can be considered.[24] Employees can get vaccination at a convenient time if the program allows appointment of the time of vaccination for individuals.

Confirm order

It is necessary to ensure adequate supply of the vaccine. For example, if the vaccination of the staff will be held in October, the vaccine should be ordered no later than the end of March. The size of the workplace and expected participation should be considered when ordering the vaccine.[27]

Case study 4 – American Express

A strong communication strategy is one of the keys to the successful workplace influenza vaccination program of American Express. *"Got Your Shot?"*, the company's 2009 marketing campaign was effective in reaching all employees. This campaign brought the message "To get the shot if you want to stay healthy and protect yourselves and others" to the employees.[27] This message is spread through multiple communication channels such as company-wide emails, posters at all locations and television monitors displaying the message. Besides, the employees can be vaccinated conveniently as mobile flu carts are available in the workplace.[27]

STEP 4: IMPLEMENTATION

After the assessment and planning, the next step is implementation. The overall workplace immunization program should contain a combination of individual and organizational level strategies and interventions.[34]

Education

The workplace immunization program may not be successful because the employees do not understand the infectious and serious nature of the diseases. So education and communication about the severity of the diseases and need for immunization are keys to launch the program.[27] In the assessment phase, the barriers of the employee immunization are identified. The information obtained can assist in launching the education tool to the employees to overcome the barriers, misconceptions and also concerns.[3] The education tool can be ranging from an informal discussion to a formal education talk.

Health-related policies

Health-related policies are supportive written statements that are designed to protect or promote employee health. The policy can affect a large group of employees simultaneously and make them adopting healthy behaviors much easier.[31] Encouraging sick employees to stay home is one of the examples. The employers can structure time and attendance policies to encourage this act. As a result, the healthy employees are less likely to become sick as their exposure to sick co-workers is limited by this supportive policy.[3]

The health-related policies also have a role to promote a corporate "culture of health".[3] The culture of health is the creation of a working environment where the health and safety of the employee are valued, supported and promoted. Promoting a culture of health establishes the workplace immunization program as a routine part of the company operations aligned with overall company goal.[35] The culture can demonstrate the employer's concerns for the health and well-being of the employees, enhance the business's competitiveness and present the company as an attractive place to attract and retain employees.[28]

Incentives

The purpose of offering incentives is to motivate employees to participate in program activities.[33] First, reducing out-of-pocket costs to employees for immunization can be implemented by paying for the vaccine, providing insurance coverage or reducing co-payments for vaccination or service.[36] On-site immunization service is an effective strategy to reduce out-of-pocket cost. It can reduce the cost barriers such as transportation to a clinic and the need for child care.[3] Apart from this, incentives can also come in other forms, such as a memento and recognition by coworkers or supervisors.[33]

Environmental supports

Environmental support offers a workplace physically designed to promote and encourage good health. Providing hand washing stations for employees and encouraging the use can be considered.[3] Hand washing is considered one of the good hygiene practices. Adults who wash their hand regularly are less likely to get sick.[37] Employers can consider organizing comprehensive hand washing campaigns for promoting and educating the importance of regular hand washing. If sinks are unavailable in the workplace, hand sanitizer wipes or gel should be provided. Besides, the employees can be reminded about regular hand washing by placing signs, reminder or posters in all public spaces such as washrooms and break rooms.[37]

Case Study 5 – Intel

The company has a long history of building a culture of wellness in the USA. Flu immunization is an important part of the culture.[27] In recent years, the company reevaluated the participation rate of the on-site flu immunization program. The company's goal is to increase the rate from 19% at 2009 to at least 30% at 2010.[27] To achieve this, the culture of wellness has evolved into a comprehensive prevention focused initiative, called Health for Life. As a result, the flu vaccination program is integrated into the company's benefit model and design. The program is not just built through evidence based medicine but also customized for the company's culture.[27]

Case study 6 - HK Government Vaccination Programme: Seasonal Influenza Vaccination for DH Staff 2012/2013

In order to encourage the staff of DH to receive seasonal influenza vaccination, Dr Constance Chan (Fig 5.), the Director of Health is invited to appear in a short video. The short video has an educational purpose clarifying the flu vaccine is safe and effective. The vaccine is not only suitable for healthy people but also for those with chronic disease. Besides, a badge will be given to the staff who get a flu shot in appreciation for their support.[38]

Figure 5. *Dr Constance Chan, the Director of Health (Centre for Health Protection. CHP newsletter Issue No. 32)*

STEP 5: EVALUATION

An effective program evaluation is able to systematically examine the implementation and results of strategies and interventions in the purpose of using the findings to improve the actions. Ideally, the development of the evaluation plan should be done in the planning phase and before the beginning of implementation but not just the end of the program. In the Centers for Disease Control and Prevention (CDC) framework for program evaluation, six steps and four standards are involved (Fig. 6).[39]

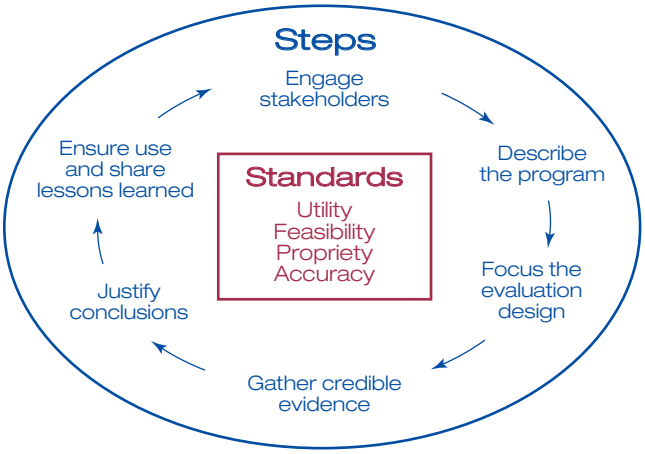

Figure 6. *CDC framework for program evaluation[39]*

First, stakeholders must be engaged in the enquiry to ensure their perspectives are understood. Next, to ensure understanding of program goals and strategies, the program should be described in details. Stakeholders may have different ideas about the program goals and purposes. Without agreement in the program definition, there is only a limited use of evaluation. Then, the evaluation must be emphasized on assessing the issue of greatest concern to stakeholders while using time and resources efficiently.[40] After that, a program evaluation should aims to collect information that will reflect a well-rounded picture of the program. Before using the evaluation results with confidence, the stakeholders must agree that conclusions are justified. Finally, it is necessary to ensure the findings are used and disseminated appropriately.[40] The evaluation approach must be useful, feasible, ethical and accurate in order to meet the four standards.[39]

Evaluation helps define the value of the workplace immunization program

The level of company investment in the workplace immunization program depends on the perceived value of the program.[39] The framework includes stakeholder engagement as the first step because the decision about the value of the program varies not just from company to company but also between the stakeholders within a single company. So it is necessary to first identify the difference in preferences and needs that stakeholders will bring to assessing the value and success of the program.[39]

The value of the program consists of three dimensions: merit (was it a quality program), worth (cost effectiveness) and significance (did it accomplish something important). The program may score well on all the dimensions or just some.[39] Whether the program is success or not, it depends on the agreement on the perceived value of the stakeholders.

Measurement

The development of the evaluation plan should be done before the start of the implementation. Evaluation is a continuous process throughout the program. Three kinds of measurement may be required for evaluation: baseline measures (before the program), process measures (during the program) and outcome measures (after the program).

Baseline measures

Baseline measures are essential for developing the evaluation before the implementation phase as they are able to establish a starting point and frame of reference for the workplace immunization program. The baseline measures can be developed from the data collected during the assessment phase.[39] For example, the program team is able to determine the number of employee being vaccinated before the program by the baseline measures. By making use of the data, the program team can set a target which is able to assess the workplace immunization program performance. In general, data from the previous 12 months will provide sufficient baseline information.[41]

Process measures

Process measures examine all the steps taken in implementing a program and the outputs generated, for example, the number and the type of educational materials developed for the employees. The process measures can help keeping the implementation on track.[39] If the program fails to achieve its intended goal and outcomes, it can be due to wrong approach of the program or just the program is implemented incorrectly. The process measures can give a clue on it. The process can also assess the details of the activities such as the number of employee reached.[39]

Outcome measures

Outcome measures are able to determine the effectiveness of the program. The outcome measures can be further divided into short-, intermediate- or long-term. Long-term measures are closely related to the goals and objectives of the program which often take a year to observe.[39] On the other hand, short- and intermediate-term measures are associated to the intermediate steps taken to achieve the long term outcomes. These measures focus on the individual outcomes of each intervention. The health related policies are the examples of the intermediate steps.[39]

Metrics for evaluation

Metrics for worker productivity, health care costs, health outcomes and organizational change allow the baseline, process and outcome measurement of the workplace immunization program. These are the suggested approach for the evaluation. However, it is not necessary to use all these metrics for the evaluation as some information may be difficult or costly to obtain.[41]

Worker productivity measures

Healthier employees are less likely to be sick. By assessing the number of sick days associated with influenza and/or pneumococcal infection, it can determine whether the workplace immunization program can increase the worker productivity (Table 3).[41]

Health care costs measures

The health care costs refer to the actual costs of providing services associated with the delivery of health care, including the procedures, therapies and medications (Table 4).[42]

Table 4. Health care costs measures[41]

Baseline Measures	Process Measures	Outcome Measures
1. Determine health care use and costs.	1. Periodic repeats of baseline measures.	1. Compare the health care use and costs of employees before and after the program such as an education program or vaccination campaign.

Health outcomes measures

The effectiveness of workplace immunization program depends on the intensity of the program effort and also the use of multiple interventions (Table 5).[41]

Organizational change measures

The organizational change measures are the assessment of the company-initiated programs and policies. Regular measures of employee attitudes and program development are critical to determine whether the new programs or policies are effective

Table 3. Worker productivity measures[41]

Baseline Measures	Process Measures	Outcome Measures
1. Determine the number of sick days per employee due to influenza and/or pneumococcal infection in the past 24 months. As there may be variation in the severity and spread of influenza, it is necessary and beneficial to conduct measurement over 24 months. 2. Conduct surveys of employee reports of influenza and/or pneumococcal infection episodes in the past 24 months. 3. Determine the cost of worker absenteeism due to influenza and/or pneumococcal infection. The costs include costs of replacement workers, costs in training replacement workers and loss and delay in productivity. 4. Determine the time employees spend during working hours on participating in the programs.	1. Periodic repeats of baseline measures.	1. Assess the change in the number of sick days per employee due to influenza and/or pneumococcal infection. 2. Assess the change in the cost of worker absenteeism due to influenza and/or pneumococcal infection. 3. Assess the change in the time employees spend during working hours on participating in the programs.

Table 5. Health outcomes measures[41]

Baseline Measures	Process Measures	Outcome Measures
1. Determine the current rate of employee immunization by using survey. 2. Determine employee knowledge, attitudes, concerns and beliefs about the immunization. This part may include assessing the employee awareness of existing workplace immunization programs, policies and benefits.	1. Periodic repeats of baseline measures.	1. Assess the changes in the rate of employee immunization. 2. Assess changes in employee knowledge, attitudes, concerns and beliefs about the immunization.

or require time for adaptation to prevent continuing investment in ineffective efforts (Table 6).[41]

Other measures

Throughout the program, make sure there is ongoing discussion within the program team to note what is going well and what needs to be changed. The program should also develop a list of recommendations to improve the program. The employees should also be encouraged to provide suggestion to the program team.[27]

It is also beneficial to determine who did and did not receive the vaccine. This may help to find out the defect of the program as it may fail to reach certain type of employees. As a result, the program can think of alternative way to reach that type of employees.[27]

A SUCCESSFUL IMMUNIZATION CAMPAIGN

In 2003, Sanofi Pasteur launched a successful workplace immunization program. In 2002, only 34% of employees were vaccinated.[43] The company was committed to build and conduct a well-planned worksite immunization program to boost employee immunization rates. As a result, the employee immunization rates increased to 76% in 2003.[43] The following table (Table 7) summarizes key components of the campaign that contributed to the success.

COST BENEFIT MODEL

Workplace immunization program is a company-initiated program. Most companies want to conclude that the money they have invested in the program has resulted in some degree of savings. The cost benefit model can assist the company to obtain the result.

Through the literature review, the principle of a cost benefit model of the workplace vaccination program is similar which has only slight difference in the type of costs included. The net financial benefit of the vaccination program, in the form of cost saving can be calculated using the following formula (Table 8):[44-48]

Cost Saving = Costs Averted Due to Vaccination - Costs of vaccination

Table 8. Details about the costs[44-48]*

Costs Averted Due to Vaccination	Costs of vaccination
Avoided costs of lost work days due to vaccine-preventable diseases	Costs of vaccine
Avoided costs of reduced work effectiveness due to vaccine-preventable diseases	Costs of program implementation (Vaccine administration, promotional materials, educational materials etc.)
Avoided costs of replacement workers	Costs of loss of work time for vaccination
	Costs of loss of work time due to serious adverse reactions of the vaccine
	Costs of reduced work effectiveness due to mild adverse reactions of the vaccine

* This cost benefit model is only a basic one. This model can be modified for each company, for example, depending on the perceived value of the program.

CONCLUSION

Establishing a workplace immunization program requires a systematic approach. Steps involved in the establishment are identified and supported by using both overseas and local examples. Besides, a basic cost benefit model is defined to assist the company in evaluating the program.

Table 6. Organizational change measures[41]

Baseline Measures	Process Measures	Outcome Measures
1. Determine the barriers to employee's ability to obtain immunizations. 2. Conduct surveys of employee attitude towards the workplace immunization program.	1. Reassess the barriers to employee's ability to obtain immunizations. 2. Document steps taken and progress toward implementing each intervention, for example, the number of educational materials prepared and number of employees participated in the education talk. Besides, the timeline for each intervention should be described. Moreover, budget for each intervention such as education classes or organizing an on-site clinic should also be documented. 3. Reassess the employee attitude and also the satisfaction with the workplace immunization program.	1. Measures the decrease in the barriers to employee's ability to obtain immunizations. 2. Assess the details for each intervention in the implementation such as cost and length of time. 3. Assess change in employee attitude and also satisfaction with the workplace immunization program.

Table 7. Key components to success[43]

Step 1: Identify and establish the program team	Multiple departments were involved in the program team in an effort to achieve widespread support
	Upper management endorsed the immunization initiative as an important program and supported the effort
Step 2: Assessment	Barriers were assessed and identified. Perceived lack of need of vaccination is the most common identified barrier. Other barriers include concerns about reactions to vaccination and skepticism about vaccine efficacy.
Step 3 and Step 4: Planning and Implementation	Detailed "to do" tasks were assigned to ensure the publicity, promotion and advocacy were underway
	Mobile immunization cart was introduced. This made immunization more convenient and accessible.
	Multiple communication channels were used for promotion. Frequent announcements were sent via email and the internal newsletter. Web-based communications included automatic reminders of clinic dates and times. Print communications such as tent cards, posters and banners were also available.
	Fact cards and posters were used to dispel myths and misconceptions related to immunization. The program team held two "Lunch and Learn" programs to offer detailed information and the opportunity for employees to ask questions and express their concerns.
Step 5: Evaluation	A post-campaign survey was distributed to employees via email. According to the survey, 42% of them wanted more information on influenza before getting immunized. The program should consider additional ways to communicate on this issue in the future campaign.

Author's background

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AmBisome Abbreviated Prescribing Information

Presentation: Each vial contains a sterile, yellow lyophilized cake or powder of 50 mg amphotericin B (50,000 units) encapsulated in liposomes. **Indications:** For the treatment of (1) systemic mycotic infections due to organisms susceptible to this anti-infective; (2) fever of unknown origin (FUO) in neutropenic patients; (3) as primary therapy of visceral leishmaniasis in immunocompetent patients including both adults and children, and in immunocompromised patients. AmBisome should not be used to treat the common clinically inapparent forms of fungal disease which show only positive skin or serologic tests. **Dosage:** Dosage must be adjusted to the specific requirements of each patient. Recommend iv infusion at 0.20-2.00 mg/ml. Adults: (1) Systemic mycotic infections: Instituted at a daily dose of 1.0 mg/kg body weight, and increased stepwise to 3.0 mg/kg as required; (2) FUO: Initiated at 1.0 mg/kg/day, the dose may be raised to 3 mg/kg/day if indicated; (3) Visceral leishmaniasis: a dose of 1.0-1.5 mg/kg/day for 21 days or alternatively a dose of 3.0 mg/kg/day for 10 days. Pediatrics: Dosage should be calculated on the same per-kg body weight basis as for adults. Safety and efficacy have not been established in infants under 1 month old. Elderly: No alteration in dose or frequency of dosing is required. Renal impairment: No adjustment in dose or frequency of administration was required during clinical trials conducted in patients with pre-existing renal impairment administering 1-5 mg/kg/day AmBisome. Hepatic impairment: No data available on dose recommendation. **Contra-indications:** Hypersensitivity (unless in the opinion of the physician that the condition requiring treatment is life-threatening and amenable only to AmBisome therapy). **Warnings and Precautions:** Anaphylaxis and anaphylactoid reactions and other severe infusion-related reactions; Renal adverse reactions; Acute pulmonary toxicity has been reported in patients given amphotericin B (as sodium deoxycholate complex) during or shortly after leukocyte transfusions; Regular laboratory evaluation of serum electrolytes, renal, hepatic and haematopoietic functions should be performed; Patients with rare hereditary problems of fructose intolerance, glucose-galactose malabsorption or sucrase-isomaltase insufficiency should not take AmBisome; For treatment of diabetic patients: AmBisome contains approximately 900 mg sucrose per vial. **Interactions & Side effects:** refer to Package Insert.

Before prescribing, please consult full prescribing information which is available upon request.

AmBisome is a registered trademark

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Recent Efforts and Achievements on the Development of Pharmaceuticals for Prevention and Treatment of Ebola Virus Diseases

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ABSTRACT

The 2014 outbreak of Ebola virus disease (EVD) that caused more than 10,000 people's deaths has accentuated an urgent need to develop effective vaccine(s) to prevent the spread of this fatal virus, and effective drugs to improve survival rates among those infected with the virus. To date three DNA vaccine candidates have been developed and are clinically evaluated for their effectiveness. A few investigational drugs based on monoclonal antibodies (Mabs), SiRNA and antiviral small molecules are also being explored and used in clinical trials to treat patients suffering from the viral hemorrhagic disease. Although none has been fully tested for safety or effectiveness, these experimental pharmaceutical products for the treatment of EVD are being clinically screened. Preliminary data indicated that combination of several Mabs gave best therapeutic results while expression of subunit surface antigen is the main approach that enables to rapidly develop new vaccines at a lower cost and fewer manufacturing obstacles than traditional vaccines.

Keywords: *Ebola virus, Epidemiology, hemorrhage, muscle pain and fever, Vaccines, monoclonal antibodies, cocktail therapy, glycoprotein expression gene*

INTRODUCTION

Ebola virus disease (EVD), formerly known as Ebola hemorrhagic fever is a severe, often fatal illness in humans if untreated. The disease is due to the transmittance of Ebola virus (EBOV), which belongs to the genus *Ebolavirus* of the Filoviridae family. The name of this disease derives from a remote village along Ebola River in Yambuku in the Democratic Republic of Congo, where the virus was first identified in 1976 when EVD had simultaneous outbreaks in Nzara, Sudan, and Yambuku, Congo. The most recent outbreak of EVD in 2014 in West Africa was the largest and most complex epidemic since the Ebola virus was discovered. It caused more than 10,000 deaths in only about nine months since March 2014. The disease spread between countries starting in Guinea; then across land borders to Sierra Leone and Liberia; by air to Nigeria; and by land to Senegal while a separate, unrelated Ebola outbreak began in Boende, Equateur Province, an isolated place in the Democratic Republic of Congo. Because infected subjects can remain asymptomatic for a period of up to three weeks, it is difficult to effectively implement travelers' screening for the prevention of travel-associated spreading of the disease. Hence, for the first time that last year's EDV outbreak in West Africa, it spread to some Western countries e.g. USA, Spain, UK when a few infected healthcare workers had to stop their voluntary African services and go home for treatment. Consequently it worsened the epidemic situation and the fatality rate of EVD reached between 50% to 90% in patients.[1]

There are five species of Ebola viruses identified so far; namely Zaire ebolavirus (EBOV), Bundibugyo ebolavirus (BEBOV), Sudan ebolavirus (SEBOV), Taï Forest virus (TFEBOV), and Reston ebolavirus (REBOV).[2] Researchers found that three species of fruit bats, which are *Hypsignathus monstrosus, Epomops franqueti,* and *Myonycteris torquata*, are the carriers of these viruses. It is believed that these fruit bats are the natural reservoir host of the Ebola viruses.[3] Yet these bats are immune to the disease and show no symptoms.[4]

BIOLOGICAL FEATURES OF EBOLA VIRUSES

Figure 1 shows the morphological and genomic structure of an EBOV. It is a single-strand RNA (ssRNA) virus. Its' genome is approximately 19,000 nucleotides long. It encodes seven proteins which are nucleoprotein (NP), polymerase cofactor (VP35), (VP40), GP, transcription activator (VP30), VP24, and RNA-dependent RNA polymerase (L).[6] It carries a negative-sense RNA genome in the virion and consist of a viral envelope, matrix, and nucleocapsid components. The glycoproteins (GPs) are 7-10 nm long spikes from its lipid bilayer surface.[7] The outer viral envelope of the virion comes from the host cell membrane into which the GP spikes have been inserted during their biosynthesis.

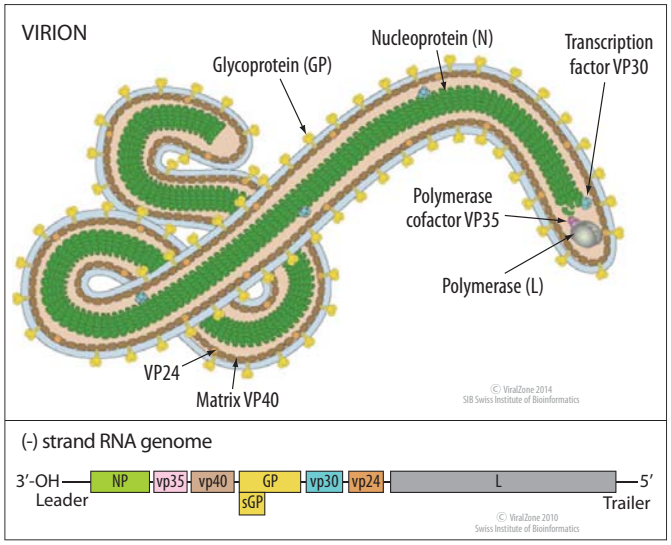

Figure 1. *Morphological (top) and genomic structure (bottom) of Ebola virus.*

SYMPTOMS AND SIGNS OF EBOLA VIRAL INFECTION

The incubation period from infection with the virus to the onset of symptoms is around 2 to 21 days. Humans are not infectious until they develop symptoms. The first symptoms include malaise and sudden onset of acute fever fatigue, chills, abdominal pain, headache, cough and sore throat. This is followed by vomiting, diarrhea, rash, symptoms of impaired kidney and liver functions as explicit in Figure 2, and in some cases, both internal and external bleeding; i.e. oozing from the gums, blood in the stools, and dehydration may be noted. In severe cases, low white blood cell and platelet counts and elevated liver enzymatic activities are commonly found through laboratory tests.

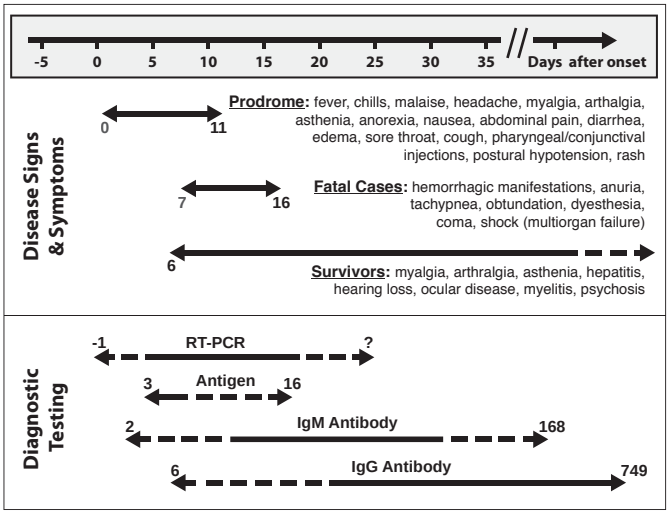

Figure 2. *Occurrence of symptoms and different diagnostic tests of Ebola viral infection in sequential order.*

DIAGNOSTIC TESTS OF EVD

From the symptoms alone as described above it is difficult to distinguish EVD from other infectious diseases such as malaria, typhoid fever and meningitis. Confirmation that symptoms are due to Ebola virus infections can be achieved by further biomedical tests as shown in Figure 2. These tests include antibody-capture enzyme-linked immunosorbent assay (ELISA), antigen-capture detection tests, serum neutralization test, reverse transcriptase polymerase chain reaction (RT-PCR) analysis, electron microscopy and virus isolation by cell culture.

Whenever carrying out a biomedical test, non-inactivated samples from patients should be handled with great care and should be conducted in maximum biological containment environment, as they are an extreme biohazard.

EPIDEMIOLOGY OF EBOLA DISEASE

Since the outbreak of the hemorrhagic fever caused by ebolaviruses in the early 90's in Africa, it has become an annual event. In the past, these outbreaks had been successfully controlled within few weeks. The situation for the March 2014 outbreak was much different from the past. It became the largest Ebola epidemic in history. As of 18 March 2015, the World Health Organization and respective governments have reported a total of 24,788 suspected cases and 10,251 deaths.[5] The 2014 Ebola outbreak in West Africa which started in March 2014 was caused by Zaire EBOV.

Ebola virus disease can only be spread after the symptoms begin. There are two main postulates on the mechanisms of evasion of the Ebola virus. One of the two mechanisms of immune system evasion is the effect that infection exerts on the maturation of dendritic cells, where it has been shown that the Ebola VP35 protein expressed in infected immature dendritic cells prevents the expression of key activating molecules, including CD40, CD80 and CD86, as well as MHC Class II molecules, and desensitizes dendritic cells to toll-like receptor 'danger' signals.[8,9] This mechanism is expected to cause a down regulation of the T cell responses, especially the CD4+ T cell response, during the early challenge with the virus (Figure 3). This reduces both the cellular immune response (cytotoxic CD8+ T cells) as well as the humoral immune response (B cells), which would produce high-affinity neutralizing and opsonising antibodies against the virus. In non-human primate studies, the counts of CD4+ T cells were particularly low during virus evasion. This indicates the importance of specifically activating CD4+ T cells during vaccination.[10] These findings highlight the importance of inducing a strong CD4+ T cell response, and this can/should be a part of the vaccination strategy.

Another important mechanism of immune system evasion that has been proposed for Ebola is the production of a secreted, truncated version of soluble glycoproteins by infected cells.[11] Their production and secretion is believed to redirect the immune response towards the production of antibody for the epitopes that are shared between the soluble and membrane-bound GP antigens. Molecular mimicry further allows the secreted, soluble glycoproteins to absorb cross-reactive anti-GP antibodies, thereby enabling the virus to escape a strong antibody response.[11] This postulate has also important implications for vaccine design. It highlights the importance of developing a broad-spectrum antibody against different viral proteins, and suggests that strong T cell-mediated cellular responses against infected cells may enhance antibody-driven immunity.

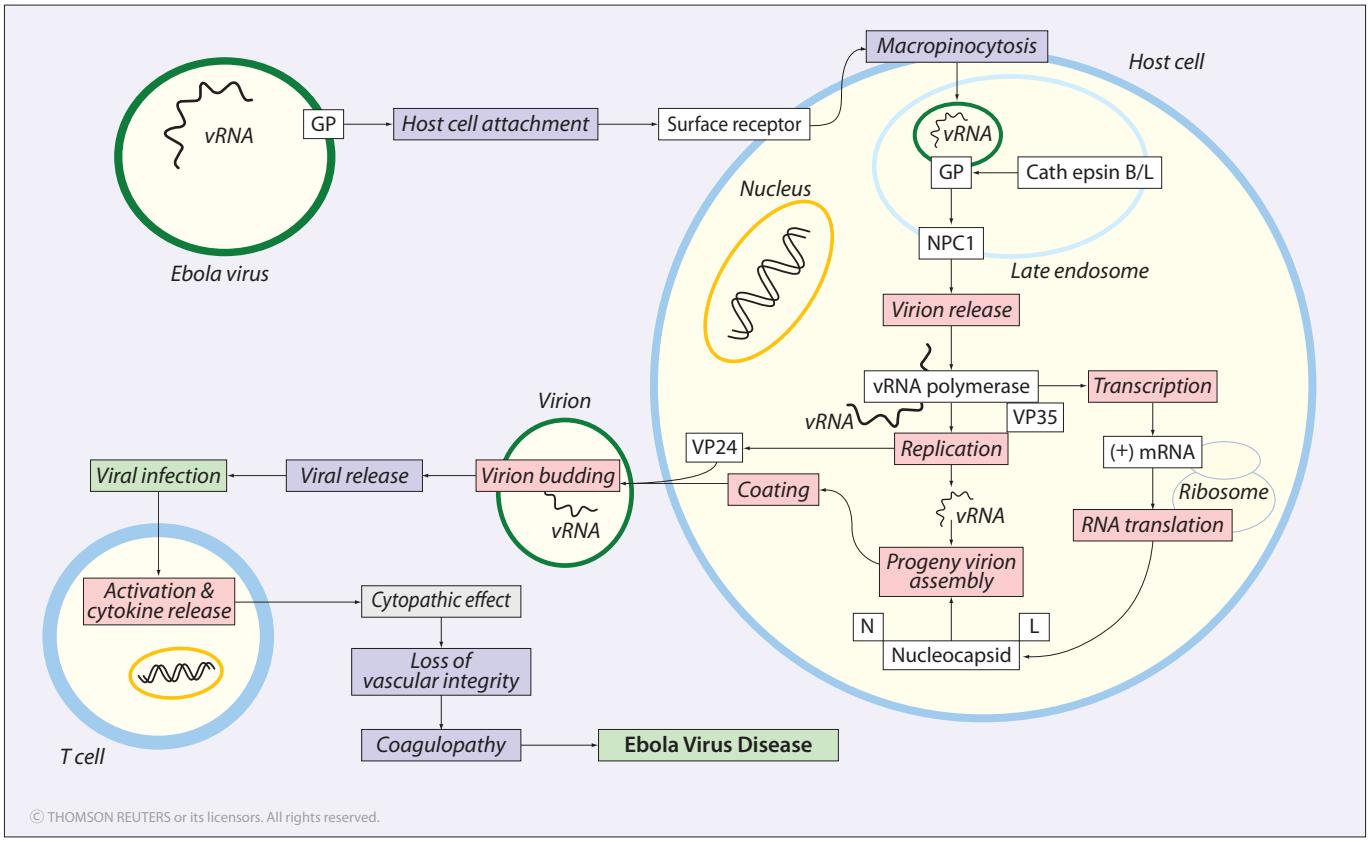

Figure 3. *Route of Ebola virus transmission and infection at the molecular level.*

PROPHYLAXIS AND TREATMENT OF EBOLA VIRUS DISEASE

Gene synthesis is the main tactic used by most research groups to develop vaccine(s) against the viruses. Since the recent cases of Ebola virus disease were caused by Zaire ebolaviruses, most efforts are devoted to develop vaccines targeting Zaire ebolaviruses to prevent further infections and epidemics. Because the predominant expression of glycoproteins over the surface of a virus is vital, various vaccines targeting the glycoproteins of Zaire ebolaviruses have been the focus.[12] Several vaccine platforms based on this approach have shown effectiveness in protecting against viral diseases. Among those vaccines developed for ebolaviruses, two of them (cAd3-ZEBOV and rVSV-ZEBOV) are the most promising candidates and have entered phase I clinical trials. They have been shown to be effective to protect against ebolaviruses in the cynomolgus macaque model.[13]

Ebola Vaccines

The goal of an Ebola vaccine is to stimulate a person's immune system to develop antibodies against the disease and, as a result, protect that person from Ebola disease. A number of potential Ebola vaccines have been tested in animals, and human trials are ongoing. A few of them are described below.

cAd3-ZEBOV Vaccine

cAd3-ZEBOV is an experimental vaccine for Zaire ebolavirus. It is being developed by GlaxoSmithKline and tested by the National Institute of Allergy and Infectious Disease (NIAID, USA). It is a vaccine containing a recombinant vector. The virus used as vector is a chimpanzee adenovirus called Chimp Adenovirus type 3 (ChAd3). It is originated from a human-derived adenoviral vector. Previous evidence has supported that ebolavirus glycoproteins expressed by human-derived adenoviral vectors (i.e., rAd5, Ad26, and Ad35) provide protection against ebolavirus.[14,15] However, the pre-existing immunity of humans against adenoviruses is a major problem. It has lead to future development of similar vaccine platforms. More recently, Stanely *et al*. developed an EBOV GP-based vaccine platform using chimpanzee-derived adenovirus (ChAd). It eliminates the problem of pre-existing vector immunity.[16] In the ChAd3, one of the genes is replaced by a glycoprotein-expressing gene of Zaire ebolavirus. The modified ChAd3 will not replicate but will induce an immune response against itself in human cell lines.

rVSV-ZEBOV Vaccine

rVSV-ZEBOV is being developed by the Public Health Agency of Canada. It is an experimental recombinant vesicular stomatitis virus-Ebola vaccine specifically against Zaire ebolavirus. Vesicular stomatitis virus, that usually causes illness in animals, has been weakened, modified and used as the host. In the vesicular stomatitis virus, the glycoprotein-expressing gene of Zaire ebolavirus is inserted into a recombinant vector. As the recombinant vector is replication-competent, the modified virus will replicate in human cell lines and induce an immune response against itself.

When comparing cAd3-ZEBOV with rVSV-ZEBOV, both have low level of pre-existing immunity and are able to provide

immunity to Zaire ebolavirus among Non-human primates (NHPs). NHPs have yielded complete protection against Ebola in experimental animal studies when cAd3-ZEBOV and rVSV-ZEBOV were injected. The results of successful phase-I clinical trials of cAd3-ZEBOV on 20 participants were published recently.[17] The report shows results associated with vaccine-induced protective immunity in studies involving non-human primates. The participants showed no severe side effects.[17] However, in some cases the vaccine has become ineffective 10 months after injection. The results of phase-I clinical trials of rVSV-ZEBOV on 59 participants were published. Initial results showed that the vaccination is very well tolerated. In the hours and days after the injection, 4 volunteers had muscle pains -which was expected.[18] The cAd3-ZEBOV cannot replicate in cells while rVSV-ZEBOV can replicate. The replication-competent rVSV-ZEBOV may provide longer lasting immunity than the cAD3-ZEBOV, although definitive data are still lacking. One possible reason is that a replication-competent virus vector can achieve efficient and persistent gene delivery as the virus replicates. The virus will stay in the human body for a longer time, and the immune response will be triggered earlier to prevent the evasion of virus.

Table 1. Comparison of cAd3 to rVSV as vaccine against Ebola viruses.

Parameter	cAd3-ZEBOV	rVSV-ZEBOV
Effectiveness	• Provide immunity to Ebola virus • Low level of pre-existing immunity • Yielded complete protection against Ebola in experimental Non-human primates (NHPs) studies	
Side Effects	No serious side effects	Pain in the joints of figures or toes
Replicability in mammalian cells	Cannot be replicated	Can be replicated
Protection Period	Ineffective 10 days after injection	Varying degrees of protection post-exposure in NHPs
Stage of Clinical Study	Phase-I clinical trial on 20 NHPs completed.	Phase-I clinical trial of on 59 NHPs completed. Results indicated vaccination was very well tolerated.

According to the NIH, two experimental vaccines have been successfully administered to over 600 volunteers in Liberia in the stage 2 (safety) phase of PREVAIL trials as of March 14, 2015. These two experimental vaccines will enter stage 3, which checks how effective the vaccines are in protecting people from Ebola infection. The two vaccines are the cAd3-EBOZ developed by NIAID scientists and GlaxoSmithKline, and the VSV-ZEBOV candidate vaccine developed by the Public Health Agency of Canada and licensed to NewLink Genetics Corporation and Merck. The trial might hit a snag, since Liberia's transmission rate is very low right now. This will make it difficult to determine how well the participants are protected from the disease, since too few are exposed to to the virus right now. The trial may have to be extended to other Ebola-affected countries.

MVA-BNFilo and Ad26.ZEBOV vaccines

These vaccines are proprietary and patented products of Oxford Vaccines Group along with Bavarian Nordic and Janssen. They were originally developed in collaboration with the National Institute of Allergy and Infectious Disease, National Institutes of Health. Their development is based on the MVA-BN® vector, which is a robust and adaptable platform available for addressing a wide variety of infectious diseases including a number of biological threats. This platform offers flexibility to design a multivalent vaccine candidate for protection against Ebola Zaire, Ebola Sudan and Marburg virus. As Figure 4 shows synthesized genes encoding Filo virus glycoproteins or surface antigens of Ebola could be mass-produced on permissive primary chicken embryo fibroblasts; afterwards these synthetic genes are inserted into the vector.

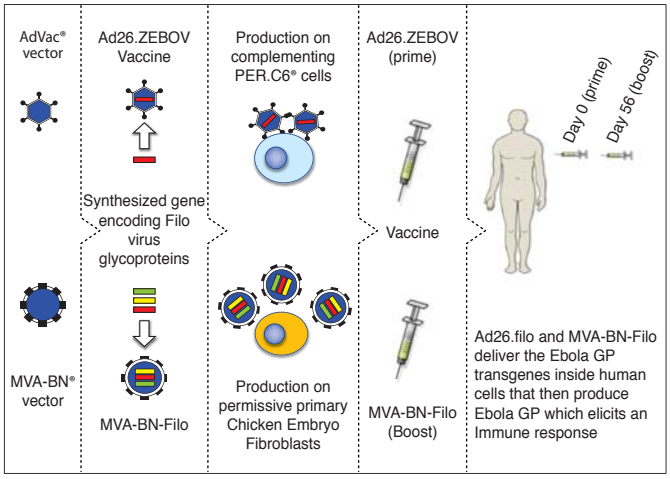

Figure 4. *Technology platform of Janssen Ebola Vaccines*

VesiculoVax Vectored Ebola Vaccines

They are a type of recombinant vaccines developed by Profectus Biosciences, Inc. of the University of Texas Medical Branch at Galveston based on a form of vesicular stomatitis virus (rVSV) which has been engineered to express the surface protein from the Zaire and Sudan variants of the Ebola virus. Results of a study recently published in Nature revealed that a single dose of this vaccine protected non-human primates against the "Makona" strain of Ebola virus, which was the strain responsible for the 2014 Ebola outbreak in West Africa. The research group reported that the VesiculoVax technology platform provides the flexibility to construct vaccine delivery vectors with different levels of attenuation; a capacity that enables the identification of vaccines to provide complete protection with the highest level of safety. Hence, this vaccine may have pre- and post-exposure protection against the Ebola viruses.

SynCon® DNA Vaccines

T-cell immune responses are considered vital to fighting cancerous cells and chronic infections. While live virus vaccines are able to generate T-cells, this approach is not viable for many diseases, such as HIV. Inoviio's SynCon® DNA Vaccine is a synthetic vaccines designed to provide powerful immune responses by not only preventing but treating diseases. It can produce strong T-cell response (both CD4+ and CD8+) while other "conventional" vaccine technologies, such as recombinant proteins, virus-like particles, subunit vaccines and peptides can at best produce only weak T-cell responses. It is a DNA fragment whenever placed into cells of the body where it does nothing but give instructions to the cells to produce the designed non-infectious antigen. The novel genetic sequences, which do not exist in nature, could be synthetically created for the selected antigen and then inserted into DNA plasmid

following by transformation of a host for mass production. The developer claims that this synthetic vaccine can be designed in weeks instead of months or years. They are manufactured by a fast and efficient fermentation process (Figure 5). The developer claims that SynCon® DNA vaccine helps the immune system to recognize and break tolerance of cancer antigens/ cells or achieve universal protection against multiple existing or newly emergent virus strains. It has a better stability profile at room temperature compared to conventional vaccines and may not require a cold-chain for storage and transport.

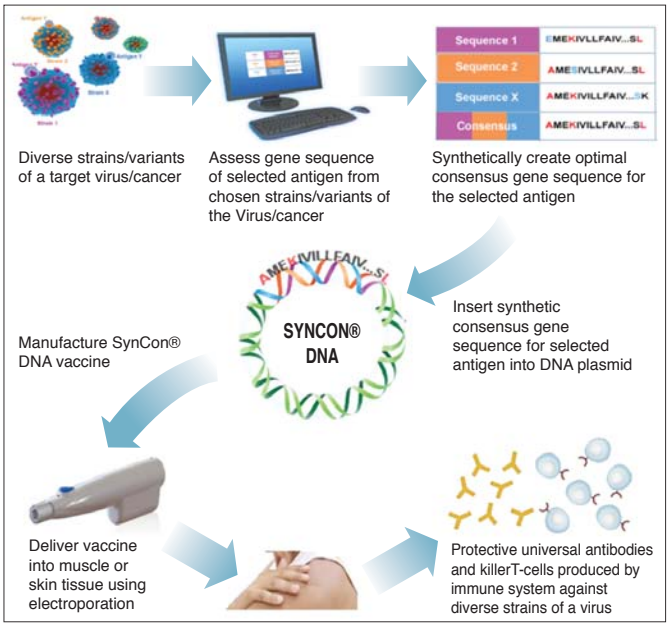

Figure 5. *Inovio's technology platform for production and applications of SynCon® Vaccines.*

Besides what have been mentioned above, a novel recombinant adenovirus type-5 vector-based Ebola vaccine called MIL77 has recently developed by researchers in China based on the expression of the glycoprotein of the 2014 epidemic Ebola strain. The Phase I trial of this vaccine has been successfully completed on 120 healthy volunteers from Taizhou of Jiangsu province. The vaccine was administered at low-dose and high-dose. According to a recent report dated March 29, 2015 in The Lancet, we were told that the high-dose vaccine was found safe and produced an immune response in the participants within 14 days.

Potential Drugs for Treatment of Ebola Virus Diseases

Developing vaccines against Ebola virus disease is not enough to control the epidemic since most vaccines only prevent the infection of Zaire ebolavirus, but cannot treat the disease when a person is infected. Once a person is infected, the destruction or neutralization of the virus provided by drugs may enable a person to overcome the infection. Therefore, developing drugs is also necessary in order to control the epidemic. The following three drugs are the most common and in most advanced stage of introduction.

ZMapp

ZMapp is a drug developed to fight against ebolaviruses. It is being developed by cooperation of Public Health Agency of Canada, Dreyfus, the U.S. Army Medical Research Institute of Infectious Diseases, Kentucky BioProcessing, and Mapp Biopharmaceutical. It is a cocktail drug made up of three highly purified monoclonal antibodies; namely c13C6, c2G4 and c4G7. The first one, c13C6, is selected from MB-003 while c2G4 and c4G7 are derived ZMAb.[19] It is produced by the leaves of a particular species of tobacco plant called *Nicotiana benthamiana* . The tobacco plants are grown in hydroponic system. Mapp Biophamaceutical states that the plant is suitable to express those proteins using indoor cultivation under tightly controlled conditions and this process is rapid and capable of mass production.[1] It targets the glycoproteins of Zaire ebolavirus. When ZMapp is injected into an infected patient,, two monoclonal antibodies of ZMapp bind near the base of the virus. Then the remaining monoclonal antibody induces the immune response like $CD8^+$ and T cell activation.[20] This cocktail drug is now in Phase I clinical trial. According to a recent report published in Nature, the efficacy of ZMapp was regarded to exceed any other therapeutic. The authors indicated that these findings warrant further development of this cocktail approach for clinical use.[28]

TKM-Ebola

TKM-Ebola is another Zaire ebolavirus-killing drug developed by Tekmira Pharmaceuticals Corp. It is a compound therapeutic composed of different small interfering RNAs (siRNAs), which belong to the class of double-stranded RNA molecules. They are 20 to 25 base pairs in length.[21] The targets are the genes encoding three of seven proteins of Zaire ebolavirus which are Zaire Ebola L polymerase, Zaire Ebola membrane-associated protein (VP24), and Zaire Ebola polymerase complex protein (VP35). Zaire Ebola L polymerase is responsible for mRNA formation. VP24 and VP35 are responsible for the inhibition of type 1 interferon response of host. The mechanism of treatment is called RNA interference. Under this mechanism, the siRNAs bind to the mRNAs to degrade these mRNAs.[22] Therefore, this results in termination of translation. The expression of the above three proteins is inhibited, and then the replication of Zaire ebolavirus in the cells is halted. It is in phase I clinical trial, but only allowed to be used in Ebola patients.[23]

Brincidofovir

Brincidofovir is a drug targeting viral DNA. It is being developed by Chimerix. It is a prodrug of cidofovir (Figure 6) and could be taken orally. This drug is a molecule conjugated to lipids originally designed as a cure against DNA-containing viruses such as smallpox. *In vitro* tests showed that it inhibits Zaire ebolavirus[24] although the mechanism of inhibition of Zaire ebolavirus is still unknown. It has fewer renal side-effects and is more potent than cidofovir. The success in *in vitro* tests makes it a potential drug to treat Zaire ebolavirus infection. Its mechanism against DNA-containing virus is that when brincidofovir enters the cells, the lipid component is removed to deliver cidofovir. Then two phosphates are added to cidofovir to give cidofovir diphosphate which is similar in structure to cytidine by enzymes; hence it is used by viral DNA polymerases. As a result, viral DNA replication is halted.[25] It is in phase II clinical trial but the trial had been halted due to a significant drop in the number of new Ebola cases.[26] As cytidine is also present in the human body, it also inhibits human polymerases but the action is 8-600 times weaker than its actions on viral DNA polymerases.[27]

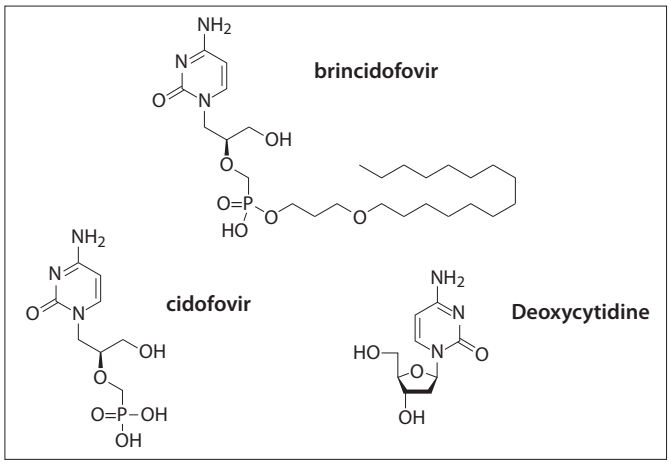

Figure 6. *Analogues of nucleic acid for treatment of Ebola Virus Diseases.*

Comparison of Different Drugs

Comparison between ZMapp, TKM-Ebola and brincidofovir reveals that all these compounds can treat Zaire ebolavirus albeit mechanisms of activity are different. ZMapp targets the glycoproteins on the virus surface. It binds to glycoproteins of Zaire ebolavirus to induce an immune response. TKM-Ebola targets the protein-encoding genes. It destroys the mRNA to terminate protein synthesis. The mechanism for treating Zaire ebolavirus by brincidofovir is still not clear since the Zaire ebolavirus is a RNA virus, while brincidofovir targets DNA viruses.

All of them are capable of providing protection against Zaire ebolaviruses in NHPs or in *in vitro* test. A study of 21 rhesus macaques showed that ZMapp is able to rescue 100% of macaques infected with a lethal dose of Zaire ebolavirus.[28] In a 2010 study of TKM-Ebola, researchers tested the drug on four monkeys by giving them seven doses of TKM-Ebola after they were infected with high doses of Ebola. The drug worked to protect them from the disease.[29] The *in vitro* of brincidofovir results show that it can provide protection against Zaire ebolaviruses.

ZMapp and brincidofovir are proven to provide protection against Zaire ebolaviruses in Phase I clinical trial while limited data are available for the action of TKM-Ebola in humans. In early August 2014, two American medical workers who were infected by the Zaire ebolaviruses while treating Ebola patients in Liberia were treated with ZMapp. Both of them claimed that they felt stronger after receiving ZMapp and even recovered from Ebola virus disease.[1] In early 2014, a freelance cameraman who was infected with the Zaire ebolaviruse recovered after being treated with brincidofovir.[30]

CONCLUSION

In an article published recently in Science, analysis of the Ebola virus isolated in Mali refuted fears that it had mutated to become even more threatening. Eight people were infected in Mali between October and November 2014 and the analysis of the virus there showed that it had not changed significantly from the one that infected people in Guinea. The team involving researchers in Mali and colleagues from the National Institute of Allergy and Infectious Diseases in Hamilton, Montana sequenced four recent Ebola virus samples in Mali. They found the virus largely unchanged and conclude that "on the basis of our data, it is unlikely that the types of genetic changes observed thus far would impair diagnostic measures or affect the efficacy of vaccines or potential virus-specific treatments." Needless to say, monitoring of the situation remains paramount to ensure that this continues to be the case. Nevertheless, the search for effective prophylactic vaccines or therapeutic agents against ebolavirus infection is still a paramount task for most drug discovery scientists.

In conclusion, among all potential vaccines mentioned above two experimental vaccines, i.e. rVSV-ZEBOV and cAd3-ZEBOV, show quite promising protective effect against Zaire ebolavirus due to its replication mechanism in nature. Among the three drugs described above, ZMapp seems to give more effective results than TKM-Ebola and brincidofovir in treating Ebola virus disease since it has a known mechanism against Zaire ebolavirus while the later ones have not. Also it shows satisfactory results in pre-clinical trial and in phase I clinical trial while TKM-Ebola has only limited data of a phase1 clinical trial, and brincidofovir has limited data of pre-clinical trials.

Author's background

Mr. LEE Kin Ho *is an undergraduate student currently taking Dr. Cheung's Pharmaceutical Biotechnology Course. This is a literature research project assigned to him as part of his continuous assessment in the last five months of the course.* ***Dr. CHEUNG Hon-Yeung****, who is an Associate Professor of Pharmaceutical Analysis, Microbiology & Biotechnology at the City University of Hong Kong since 1989, is a manufacturing pharmacist and biotechnologist. He has more than 40 years of working experience in industries, academic and consultancy jobs. He was an expert witness in court and a member of the Biotechnology Committee for Hong Kong and Shenzhen Government. Dr. Cheung has published more than 220 papers and articles in many prestigious international journals. His email address: cheung.honyeung@cityu.edu.hk*

Table 2. Comparison of three drugs for treatment of Ebola Virus Diseases.

Parameter	ZMapp	TKM-Ebola	Brincidifovir
Effectiveness	• All were designed to treat Zaire ebolavirus • All show protection against Zaire ebolavirus in pre-clinical stage		
Target Site of Action	Glycoproteins of the viral surface	Glycoprotein encoding genes	Mechanistic action remained unknown
Side Effects	No known side effect	Headache, dizziness, chest tightness, raised heart rate at high dose	Diarrhea
Results of Clinical Trial	Successfully rescued 21 Rhesus macaques (100%) from lethal dose of Zaire ebolavirus infection	The drug protected 4 monkeys from Ebola virus diseases	In vitro test showed it provided protection against Zaire ebolavirus
Successful cases of Treatment in Human	In August 2014, two American medical workers recovered from Zaire ebolavirus infection	Limited available data	After given the drug to a freelance cameraman, he recovered from the Zaire ebolavirus infection

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Warfarin and Analgesics – How Safe is Concurrent Use?

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ABSTRACT

Warfarin is a widely used anticoagulant known for its interaction with numerous drugs. Analgesics, such as paracetamol, non-steroidal anti-inflammatory drugs (NSAIDs) and tramadol are commonly prescribed in hospital and community settings for pain control. Some case reports and studies have reported that concurrent use of these analgesics and warfarin have an effect on the International Normalized Ratio (INR). The interaction between NSAIDs and warfarin is well-documented. Whilst NSAIDs do not affect warfarin's anticoagulant effect, concurrent use is associated with an increased risk of bleeding. Excessive use of topical methyl salicylate ointment can also lead to an increase in INR. For paracetamol, a number of cases reported an increase in INR when used with warfarin. This is possibly due to two pathways – via CYP450 or by a toxic paracetamol metabolite N-acetyl-p-benzoquinone imine (NAPQI). Interaction between warfarin and tramadol is uncertain, however the rare possibility of such an interaction is still highlighted in product monographs and package inserts. It is best to avoid NSAIDs as much as possible in patients who are on warfarin and topical methyl salicylate ointment should be prescribed with care. Caution should be exercised when prescribing tramadol and paracetamol with warfarin. Frequent monitoring of INR is needed when two to four grams of paracetamol are consumed. Patients should also be encouraged to inform healthcare professionals about their medication profile prior to purchase of over-the-counter medications.

Keywords: *Warfarin, analgesics, NSAIDs, paracetamol, tramadol, interaction*

INTRODUCTION

Warfarin has a narrow therapeutic index so it is crucial to keep within the recommended range of and it is crucial to keep the international normalized ratio (INR) within the recommended range. Warfarin's interaction with many medications, herbs and food, which affect the INR, are well documented. Caution should be taken when on prescribing concurrent medications and monitoring of drug effects is required. Analgesics such as paracetamol, non-steroidal anti-inflammatory drugs (NSAIDs) and tramadol are widely prescribed in hospital and community settings. This makes it difficult to avoid concurrent use with warfarin despite concerns about the safety of the combination.

WARFARIN

Warfarin is an anticoagulant used worldwide for the prevention and treatment of venous and arterial thromboembolism and embolism related to atrial fibrillation. It is a vitamin K antagonist that acts by inhibiting vitamin K epoxide reductase (VKOR), thereby interrupting the replenishment of reduced vitamin K (VKH2) and the conversion of glutamic acid (glu) to gamma-carboxyglutamic acid (gla) by the enzyme gamma-carboxylase (Figure 1)[2]. This conversion is responsible for the activation of clotting factors II, VII, IX and X and proteins C and S.[1-3]

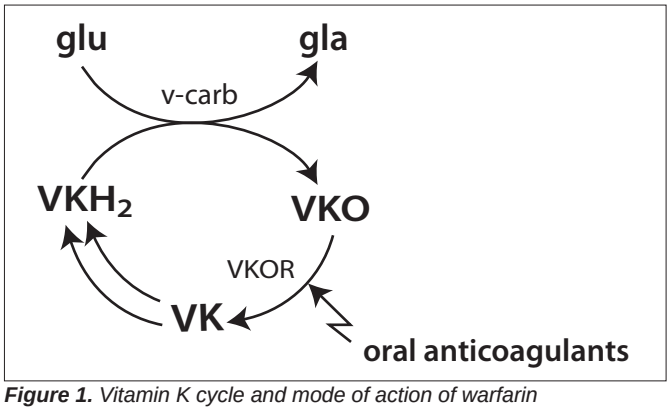

Figure 1. *Vitamin K cycle and mode of action of warfarin*

Concurrent use with *NSAIDs*

The risk of serious gastrointestinal bleeding increases when NSAIDs and warfarin are used concurrently.[4] This interaction is reported in different studies and is well documented in various references, including Lexicomp and Stockley's Drug Interactions. The interaction is classified under category D (consider therapy modification) in Lexicomp, and "Give guidance about possible adverse effects and/or consider some monitoring" in Stockley's Drug Interaction.[5,6] Although non-selective NSAIDs such as ibuprofen, naproxen, diclofenac and indomethacin do not affect warfarin's anticoagulant effect, concurrent use with warfarin is associated with an increased risk of both gastrointestinal and non-gastrointestinal bleeding.[6] Some case reports also noted an increase in prothrombin time and other coagulation parameters.[6] Selective COX-2 inhibitors such as celecoxib and rofecoxib (now withdrawn from market) have a reduced risk of causing gastrointestinal ulcers. However, it was noted that the risk of bleeding and the INR increased when taken with warfarin.[6,7]

The following mechanisms may explain the interactions:
1) NSAIDs can impair platelet function, giving rise to a

risk of bleeding. Such risk is further increased when co-administered with warfarin.[6]

2) NSAIDs interfere with prostaglandin's protective effect on the gastric mucosa, thereby causing direct injury to the gastrointestinal mucosa. This irritation is dose- and duration-dependent and can lead to gastric bleeding. Such risk is heightened in patients on warfarin.[6,8,9]
3) Some NSAIDs, e.g. ibuprofen and celecoxib, and warfarin are substrates of CYP2C9. Competition for the isoenzyme can inhibit metabolism and elimination of warfarin, therefore increasing the anticoagulant activity.[7,10]
4) Celecoxib and warfarin are both highly protein-bound. Hence, concurrent use of celecoxib may displace warfarin from protein-binding sites, thus increasing the anticoagulant activity.[7,10]

For these reasons NSAIDs and COX-2 inhibitors should be avoided as far as possible in patients on warfarin.

Methyl salicylate ointment, e.g. Analgesic Balm, is a topical preparation for pain and inflammation. It is as widely prescribed as paracetamol in general out-patient clinics (GOPCs) and in the elderly to relieve chronic pain, especially for osteoarthritis in the knees. Although it is only used topically, a local case series study has shown that excessive use can lead to an increase in INR and bleeding or bruises.[11] Prior to admission to the study, patients had been using topical methyl salicylate ointment at least 2 to 4 g daily for 2 weeks. Of the 11 patients, 3 had bleeding manifestations, where 2 had bruises and 1 had gastrointestinal bleeding. The authors concluded that topical methyl salicylate ointment should be prescribed with care and excessive usage is to be avoided.

Concurrent use with *paracetamol*

Paracetamol is the preferred and commonly prescribed analgesic for patients on warfarin therapy due to the well-documented drug interaction between NSAIDs and warfarin. Although co-administration of paracetamol with warfarin is perceived to be relatively safe compared with NSAIDs, several case reports and prospective, randomized, double-blind and placebo-controlled studies have demonstrated an increase in INR when used concurrently.[12-14] In these studies, patients stable on warfarin therapy were recruited and assigned randomly to receive placebo or paracetamol. These studies generally have small sample sizes, and paracetamol dose was at least 2g/day. It is arguable to apply the observation that concurrent use of paracetamol and warfarin causes an increase in INRto a wider population and to patients who are on lower or intermittent dose of paracetamol , therefore it is debatable to unanimously conclud that paracetamol interacts with warfarin. Despite this, the possible effect of such a combination cannot be ignored. A review of published literature on paracetamol-warfarin interaction was also conducted in 2011.[15] The authors concluded that co-administering warfarin and paracetamol may result in a significant elevation of INR although several studies do not support this observation.[16-18]

Although the first report regarding a possible interaction between warfarin and paracetamol appeared as early as 1968,[19] the exact mechanism of action of such an interaction is still unknown. However, the following two mechanisms have been postulated:

1) Inhibition via the CYP450 pathway[15]
   Paracetamol is metabolized in the liver in several ways, one of which is by the cytochrome P450 enzymes CYP1A2, 2E1 and 3A4, whilst warfarin is metabolized by the enzymes CYP1A2, 2C9 and 3A4. It was suggested that paracetamol interacts with warfarin via both CYP3A4 and 1A2. In the setting of old age or hypoxia, the metabolism of paracetamol could shift towards the CYP pathway, thus competing with warfarin for these enzymes and subsequently increasing the concentration and anticoagulation effect of warfarin.
2) Inhibition of VKOR and gamma-carboxylase by N-acetyl-p-benzoquinone imine (NAPQI).[2,20] NAPQI is a toxic paracetamol metabolite formed mainly via the CYP2E1 pathway. Under normal conditions, NAPQI is further metabolized by glutathione. However, when depletion of glutathione or induction of CYP2E1 occurs, the accumulation of NAPQI inhibits VKOR and gamma-carboxylase, which are both required in the vitamin K cycle and synthesis of clotting factors. This mechanism was proposed in an in-vitro study2 and it remains to be seen whether the results can be replicated.

Increased monitoring of INR is recommended in patients on warfarin who commence and cease regular medium-to-maximum recommended doses of paracetamol (ie. 2 to 4 g/day).[12,15] Warfarin dose may need to be adjusted accordingly.

Concurrent use with *tramadol*

Whilst there are no documented interactions between warfarin and opioid analgesics such as codeine, dextropropoxyphene and hydrocodone,[6] there have been a number of reported cases on the effect of warfarin while on tramadol.[21,22] Tramadol is a synthetic opioid for the treatment of mild to moderate pain and is often an alternative for patients on warfarin when paracetamol alone is ineffective. Reports on the drug interaction between warfarin and tramadol, which can lead to an increase in INR and hemorrhagic effects, are comparatively scarce, yet product monographs and package inserts still highlight the rare possibility of such an interaction,[22] The exact mechanism of this drug interaction is unclear, but there are suggestions it may be related to the metabolism of both drugs via the CYP450 pathway. Tramadol is a substrate of both CYP2D6 and CYP3A4, whilst warfarin is a substrate of CYP3A4. It was proposed that in patients with CYP2D6 polymorphisms, tramadol might metabolized less extensively by CYP2D6, therefore competing with warfarin for CYP3A4 metabolism.[23,24] Caution should be exercised when tramadol and warfarin are used concurrently. The first case of successful continuation of tramadol therapy after an observed tramadol-warfarin drug interaction was reported in 2006. Adjustments were made to the patient's warfarin and tramadol doses over three weeks, and with successful continuation of both warfarin and tramadol and a stable INR. Although this was not a large-scale study, it is worth considering the authors' suggestion to reduce the warfarin dose by 25% initially if tramadol is to be taken long-term, along with repeated INR measurements within one week.[25] The warfarin dose may need to be increased upon discontinuation of tramadol.

CONCLUSION

Paracetamol, NSAIDs and tramadol are commonly-prescribed analgesics, and interactions between these analgesics and warfarin should not be overlooked. It is important that healthcare professionals are aware of these possible warfarin interactions. The choice of analgesics, their dose and duration, and the warfarin dose should be,carefully considered whilst INR should be monitored closely. Thorough drug-use history should be sought from patient or patient's carer upon hospital admission or consultation to preclude prescribing medications that may interact with warfarin and to rule-out any possible interactions between over-the-counter medications and warfarin. Patients should also be encouraged to inform healthcare professionals of their medication profile prior to any self-purchase of medications. Patient counselling on possible side effects of warfarin and their management should be offered.

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Questions for Pharmacy Central Continuing Education Committee Program

(Please be informed that this article and answer sheet will be available on PCCC website concurrently. Members may go to PCCC website (www.pccchk.com) to fill in their answers there.)

2 CE Units

Warfarin and Analgesics – How Safe is Concurrent Use?

1. ***Warfarin***
   a) inhibits the conversion of glutamic acid (glu) to gamma-carboxyglutamic acid (gla) by inhibiting enzyme gamma-carboxylase.
   b) interrupts the replenishment of reduced vitamin K (VKH_2) by inhibiting vitamin K epoxide reductase (VKOR)
   c) activates the synthesis of vitamin K-dependent clotting factors II, VI, IX, and X, and the anticoagulant proteins C and S
   d) does not interact with common analgesics.

2. ***Interaction between warfarin and NSAIDS cannot be explained by the following mechanisms***
   a) NSAIDS impair platelet function, therefore leading to risk of bleeding. Co-administration with warfarin can further increase such risk.
   b) Direct injury to the gastrointestinal mucosa is caused by NSAIDs due to interference of prostaglandin's protective effect on the gastric mucosa. Such risk is heightened in patients on warfarin.
   c) NSAIDs are inhibitors of the isoenzyme CYP2C9, of which warfarin is a substrate, therefore inhibit the metabolism and elimination of warfarin.
   d) Celecoxib may displace warfarin from protein-binding sites, therefore leading to an increased anticoagulant effect.

3. ***Which of the following is correct?***
   a) Interaction between NSAIDs (both selective and non-selective) and warfarin is documented in various references with strong evidence. Concurrent use is best to be avoided or monitor any emergence of adverse effects.
   b) The risk of bleeding is not affected with concurrent administration of non-selective NSAIDs and warfarin, because non-selective NSAIDs do not affect warfarin's anticoagulant effect.
   c) Due to lower risk in causing gastric ulcers, selective NSAIDs such as celecoxib do not affect the risk of bleeding with taken with warfarin.
   d) Methyl salicylate ointment is used topically. Since there is limited systemic absorption of the active ingredient, its use in patients on warfarin need not be a main concern.

4. ***Which of the following is incorrect?***
   a) The first report regarding a possible interaction between paracetamol and warfarin appeared in the 1960s.
   b) The exact mechanism of the possible interaction between paracetamol and warfarin is still unknown.
   c) Several studies and literature review support the observation that coadministration of paracetamol and warfarin can result in significant elevation of INR with no opposing views.
   d) Metabolism of paracetamol can be shifted towards the CYP patheway when patient is hypoxia or at an older age.

5. ***In studies and literature review concerning an interaction between paracetamol and warfarin, which of the following is incorrect?***
   a) These studies were prospective, randomized, double-blinded and placebo-controlled.
   b) It was observed that INR was significantly increased when consumed with a daily paracetamol dose of 2-4g. Frequent INR monitoring is needed.
   c) It is debatable to draw a conclusion that paracetamol interacts with warfarin due to the limitations of the studies.
   d) Since a conclusion of such interaction cannot be drawn, it is safe to prescribe paracetamol in patients on warfarin.

6. ***Paracetamol is metabolized in the liver via CYP450 pathway and the following enzymes are involved:***
   I. CYP1A2
   II. CYP2CP
   III. CYP2E1
   IV. CYP3A4
   a) I, II, III
   b) I, II, IV
   c) I, III, IV
   d) I, II, III, IV

7. ***In view of the prescribing of common analgesics with warfarin concurrently, which of the following is least important?***
   a) Check patient's concurrent diseases.
   b) Dose and duration of analgesic therapy
   c) Dose of warfarin therapy
   d) Obtain thorough drug-use history from patient or patient's carer (including self-purchased medications)

8. ***N-acetyl-p-benzoquinone imine (NAPQI) is:***
   a) a non-toxic paracetamol metabolite.
   b) metabolized via the CYP2E1 pathway.
   c) accumulated when inhibition of CYP2E1 occurs.
   d) an inhibitor of VKOR and gamma-carboxylase.

9. ***Which of the following regarding the use of tramadol in patients on warfarin is incorrect?***
   a) Although there are limited reports and studies on the interaction between tramadol and warfarin, product monographs and product inserts highlights the rare possibility of such interaction.
   b) The exact mechanism of this drug interaction is well-established.
   c) Tramadol may compete with warfarin for CYP3A4 in patients with CYP2D6 polymorphism, therefore inhibiting warfarin metabolism and increasing its anticoagulant effect.
   d) If tramadol is prescribed to be taken with warfarin long-term, consider reducing warfarin dose 25% initially with close monitoring of INR within one week of change of warfarin dose.

10. ***Which of the following common analgesics is absolutely safe in patients stable on warfarin?***
    I. Paracetamol
    II. NSAIDs
    III. Methyl salicylate
    IV. Tramadol
    a) I only
    b) I, and III
    c) I and IV
    d) None of the above.

Answers will be released in the next issue of HKPJ.

CE Questions Answer for 214(D&T)

Treatment Updates on the Hepatitis C Infection

1. A 2. D 3. B 4. B 5. C 6. B 7. D 8. A 9. D 10. C

SUCRATE® gel

(Sucralfate 1g/5ml)

Actively treat GERD & Gastritis with lesser early relapse
Heal damaged G.I. lesions & promote complete recovery

Indication
Gastro-esophageal reflux disease (GERD), gastritis and peptic ulcers of various origin

Composition
Per 5ml sachet containing 1 gram of sucralfate gel

Product mechanism and features
Not offered by any Proton Pump Inhibitors, H2-blockers or other acid suppressing agents, Sucrate Gel uniquely forms a cyto-protective layer on the inflamed and damaged mucosae of the G.I. tract. This layer prevents stomach acid, pepsin and bile salts from further eroding the ulcerated tissues. Also, Sucrate Gel stimulates the production of endogenous tissue growth factors (epidermal growth factor, fibroblast growth factor, transforming growth factor alpha, platelet derived growth factor), which promote cell regeneration and angiogenesis.

Active ulcer healing is achieved through better reconstruction of mucosal architecture and thus prevents early relapse.

- Patented gel form with double surface area of bio-adhesion to ulcerated G.I. tissues
- Does not affect acid secretion - no influence on digestion and micro-organism killing in the stomach (especially relevant for the weak elderly)
- Easily swallowed with good tolerance

Dosage
One sachet 2-4 times a day, according to physician's judgement.

Manufacturer & origin
Product of Lisapharma S.p.A., Italy.
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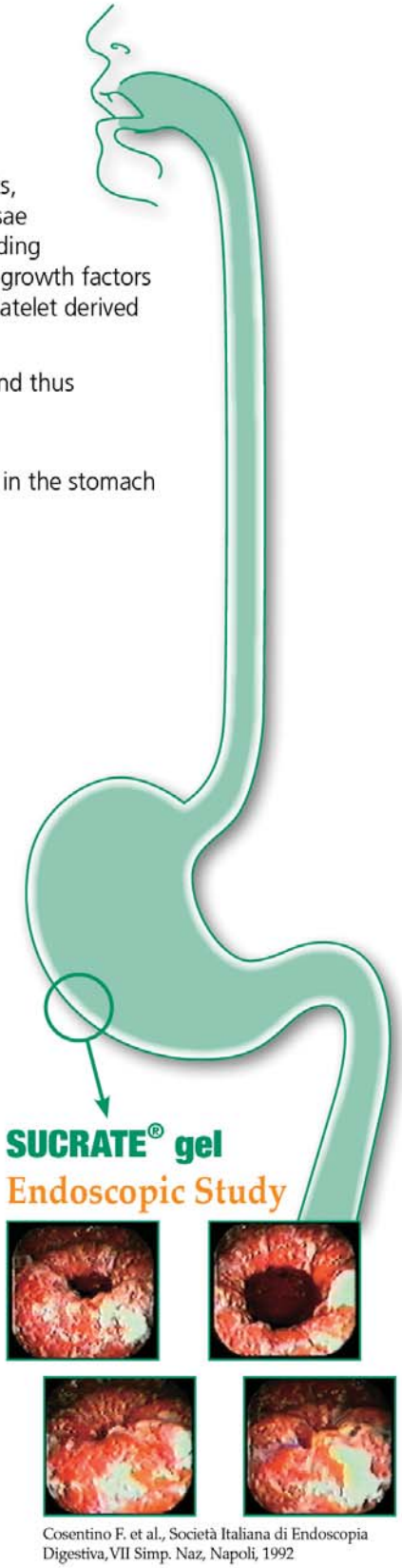

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Chemical Constituents and Bioactivities of Plantaginis Herba

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Botanical Name: *Plantago asiatica* L.; *Plantago major* L.; *Plantago depressa* Willd.; *Plantago lanceolata*; *Plantago ovata* Forsk.

Plant Family: Plantaginaceae

Chinese Name: 車前[che qian (*P. asiatica* L.)]; 大車前[Da che qian (*P. major* L.)]; 平車前[Ping che qian (*P. depressa* Willd.)]; 長葉車前[Chang ye che qian (*P. lanceolata L.*)]; 卵葉車前[Luan ye che qian (*P. ovata* Forsk.)]

Pharmacopoeia Name: Plantaginis Herba; Plantaginis Semen

Part Used: Dried whole plant, seeds

Other Name: che lun cai, plantain, Asian plantain, common plantain, great plantain, Japanese plantain, ribwort plantain, broadleaf plantain, waybread, white man's foot, Mu de, Xa tien, Daun Sendok

ABSTRACT

Plantago, which is derived from the dried whole plant of *Plantago* species (Plantaginaceae), is a herbal medicine distributed and utilized worldwide for centuries. *P. asiatica* L. and *P. depressa* Willd. are the endorsed official sources of the herb in China. The former is also an official medicinal herb in Japan while *P. lanceolata* L. is commonly used in Europe. Apart from these official species, many other species such as *P. major* L. and *P. ovolata* L. are used in folk medicines for the treatment of a variety of diseases such as hematuria, chronic inflammation and constipation. In addition to common primary metabolites, a great number of bioactive constituents have been isolated from the herb, *e.g.* phenylethanoid glycosides, flavonoids, iridoids, triterpenoids, phenolic acids and so on. A variety of useful biological activities such as diuretic, anti-inflammatory, antioxidative, antiviral, antibacterial and immunomodulatory effects was reported, while very few contraindications have been described. This article aims to review (1) the complexity of this herb; (2) what components of this herb promote human health and (3) what sorts of scientific evidences are available.

Keywords: *plantago herb; Plantago species; herbal medicine; hematuria; diuresis; phenylthanoid glycosides; iridoids*

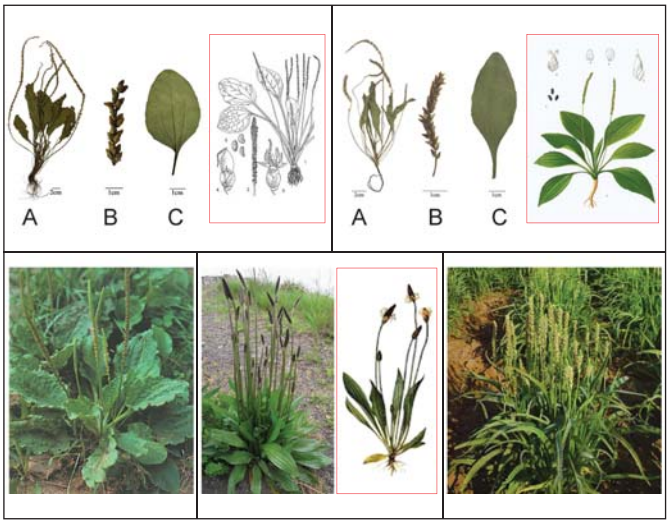

Figure 1. *Photographs of some Plantaginis herbs. Plantago asiatica L. (top left), P. depressa Willd. (top right), P. major L. (bottom left), P. lanceolata L. (bottom centre) and P. ovata Forsk. (bottom right).* ***A*** *= whole plant;* ***B*** *= Capsules;* ***C*** *= Leaf. Inserted red frame is a sketch of the plant.*

Contraindications

Use with caution in individuals who have known hypersensitivity or allergy to the plant; faecal impaction or intestinal obstruction; diabetes mellitus where insulin adjustment is difficult; contraindicated in pregnancy or lactation may also happen.

Undesirable Effect

May cause transient adverse effects such as nausea, vomiting, GI distress and diarrhea. The plant may also cause low blood pressure.

Interaction with Conventional Drugs

Combine use of plantaginis herb with certain drugs may change their therapeutic effects; such as digoxin, beta blockers (Inderal, Procardia), lithobid and tegretol. People using plantain should avoid bulk laxatives.

INTRODUCTION

Plantago herb, called as *che qian* in Chinese, is a herbal medicine with a long application history throughout the world particularly in Asia, Europe and America.[1] It was firstly described in an old Chinese medicinal book *The Classic of Herbal Medicine (also Shen-nung Pen-tsao Ching)* and is famous for the curative effects in promoting urination and treating stranguria [difficulty in micturition due to urethra spasm] in traditional Chinese medicine.[2] This herb originates from the dried whole plant of Plantaginaceae family. There are approximately 275 *Plantago* species, which are widely distributed in both the temperate and tropical areas worldwide. Amongst these only 20 species are found in China. In the 2010 edition of Chinese Pharmacopoeia, only two species, namely *P. asiatica* L. and *P. depressa* Willd. (Figure 1) of the Plantago genus are recommended for medicinal uses.[3] However, *P. major* L., *P. ovata* Forsk., *P. psyllium* L. and other species of *Plantago* are also used as folk medicine and have been traded as commodities in other countries. In Japan, *P. asiatica* L. is listed in their Pharmacopeia and *P. lanceolata* L. is described as ribwort plantain in European Pharmacopeia.[4,5] Besides the whole plant, the seed of the *Plantago* plant can be individually used as herbal medicine and is recorded in both the Chinese Pharmacopeia and Japanese Pharmacopeia.[3,4]

A wide array of primary and secondary metabolites comprise the chemical constituents of plantago herb, *e.g.* phenylethanoid glycosides, flavonoids, iridoids, triterpenoids, monoterpenoids, phenolic acids, polysaccharides and fats.[1,6-8] Among these identified components, the active secondary metabolites take charge of various biological activities (*e.g.* diuretic, antiinflammatory, antiasthmatic, antitussive, immunomodulatory and antileukemic activities) reported by numerous pharmacological investigations using the extracts or purified constituents of plantago herbs.[1,6,8] In this article, the chemical constituents and bioactivities of plantago herbs from different species of *Plantago* are reviewed and discussed.

DESCRIPTION AND IDENTIFICATION

P. asiatica L. is a perennial plant (Figure 1, top left photo). Roots are numerous and fibrous. Leaf blades are 4-12 cm long and 2.5-6.5 cm wide, broadly ovate to broadly elliptic, thinly papery to papery, and sparsely pubescent. Petiole is 2-15 cm long and sparsely pubescent. Spikes are 3-40 cm long, narrowly cylindric, loosely to densely flowered, and sometimes interrupted basally. Peduncle is 5-30 cm long and white pubescent. Bracts are 2-3 mm long, narrowly ovate-triangular to triangular-lanceolate, glabrous or pubescent at apex, and keel thick. Sepals are 2-4 mm long, keel extending or not extending to apex, and obtuse at apex, rounded or acute. Lower sepals are elliptic and keel broad. Upper sepals are broadly obovate-elliptic to broadly obovate. Corolla is 1-1.5 mm, white and glabrous. Lobes are narrowly triangular, patent to reflexed, and acuminate to acute at apex. Stamens are adnate only to near base of corolla tube and exserted. Anthers are about 1-1.2 mm ovoid-ellipsoid and white. Pyxis are 3-6 mm long, fusiform-ovoid, ovoid, conic-ovoid, or narrowly conic-ovoid, circumscissile near base, and containing 5-15 seeds. Seeds are 1.2-2 mm long, ovoid-ellipsoid to ellipsoid, angled, ventral face prominent to slightly flat and blackish brown. Cotyledons parallel to ventral side. The plant blooms from April to August and fruits from June to September.

P. depressa willd. is an annual, winter annual or perennial plant (Figure 1, top right photo). Taproot is long, fleshy or becoming woody when oldens. Leaf blades are 3-15 cm long and 1-5.5 cm wide, elliptic, elliptic-lanceolate, obovate-elliptic, or ovate-lanceolate, and papery. Petiole is 2-7 cm long. Spikes are 6-12 cm longm, narrowly cylindric, densely flowered, and interrupted basally. Peduncle is 5-18 cm long and white pubescent. Bracts are 2-3.5 mm long, triangular-ovate, glabrous, and keel extending near or to apex. Sepals are 2-2.5 mm long, glabrous, and keel not extending to apex. Lower sepals are narrowly obovate-elliptic to broadly elliptic and upper sepals are obovate-elliptic to broadly elliptic. Corolla is white and glabrous. Lobes are 0.5-1 mm long, elliptic to ovate, patent to reflexed. Stamens are adnate to near apex of corolla tube and exerted. Anthers are 0.6-1.5 mm long, ovoid-ellipsoid to broadly ellipsoid and white. Pyxis is 4-5 mm long, ovoid-ellipsoid to conic-ovoid, circumscissile near base, and containing 4 seeds. Seeds are 1.2-1.8 mm long, ellipsoid, yellowish brown to black and ventral face prominent to slightly flat. Cotyledons parallel to ventral side. The plant blooms from May to July and fruits from July to September.

MICROSCOPIC APPEARANCE

Root

As shown in Figure 2A, the root of *P. asiatica* L. has one layer of subround epidermal cells with cutin on the surface. Cortex is broad, and cells are subround to elongate. Endodermis is distinct. Phloem is narrow and surrounding the xylem tissue. Xylem is broad and vessels are arranged radially.

In Figure 2B, the cork in the root of *P. depressa* Willd. consists of several layers of rectangular to elongated cells, which are tangentially extended and closely arranged. Cortex is relatively narrow. Phloem is broad with clefts. Cambium is distinct and undulating. Xylem is broad and vessels are arranged radially.

Leaf

As shown in Figure 3A, upper epidermis in the leaf of *P. asiatica* L. consists of one layer of subsquared to rectangular cells. Mesophyll cells are rounded and subrounded. Xylem vessels are radially arranged and surrounded by the phloem tissue. Vascular bundle belongs to amphicribral type. Collenchyma is sub-rounded and found beneath the lower epidermis. Lower epidermis consists of one layer of squared to subsquared cells which are orderly arranged.

In Figure 3B, the microscopic features of leaf of *P. depressa* Willd. are very similar to *P. asiatica* L. except that upper epidermis consists of one layer of rectangular to elongated cells.

Powder

The powder of *P. asiatica* L. or *P. depressa* Willd. is greyish-green or brownish-yellow. In Figure 4, upper epidermal cells of *P. asiatica* L. are subrectangular with cuticle striations. Lower epidermal cells are subrectangular with wall deeply undulantly curved. Stoma belongs to anomocytic type with 3-4 subsidiary cells. Glandular hairs are yellowish-brown or colourless with a unicellular stalk and two cells in the head. Abundant non-glandular hairs are on the petiole and bract, and usually broken.

Intact hairs are composed up of dozens of cells. Pericarp cells are irregular shape. Anticlinal walls of epidermis are sepal deeply undulant and usually extended. Pollen grains are pale yellow or colourless, subrounded, 17-33 μm in diameter, and with warty sculptures on the surface. Fragment of endothecium are polygonal with dense cylindrical striations. Fibres are slender with relatively thickened walls ; mostly reticulated. Spiral vessels are 8-47 μm in diameter.

In Figure 5, upper epidermal cells of *P. depressa* Wild. are sub-rectangular with walls undulantly curved. Abundant non-glandular hairs are on the head of root and usually broken. Intact hairs are composed up of dozens of cells. Non-glandular hairs on scape and the surface of leaf are rare. Pollen grains are 16-30 μm in diameter. Endothecium is rare. Vessels are 6-55 μm in diameter.

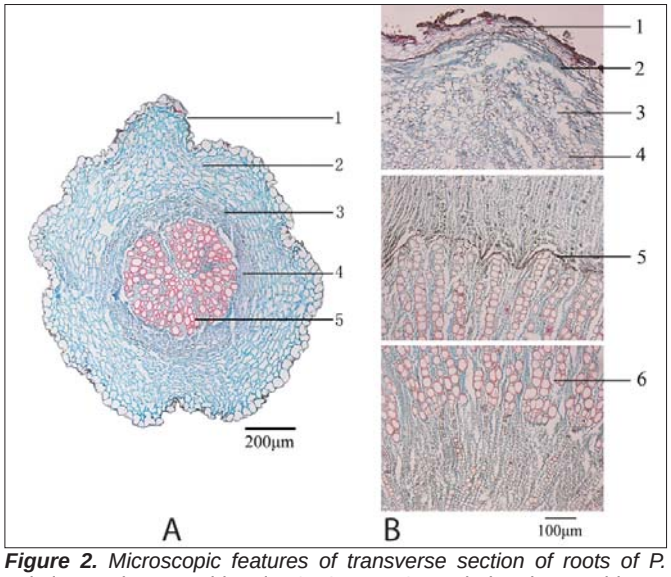

Figure 2. *Microscopic features of transverse section of roots of P. asiatica L. (A: 1. Epidermis; 2. Cortex; 3. Endodermis; 4. Phloem; 5. Xylem) and P. depressa Willd. (B: 1. Cork; 2. Cortex; 3. Clefts; 4. Phloem; 5. Cambium; 6. Xylem)*

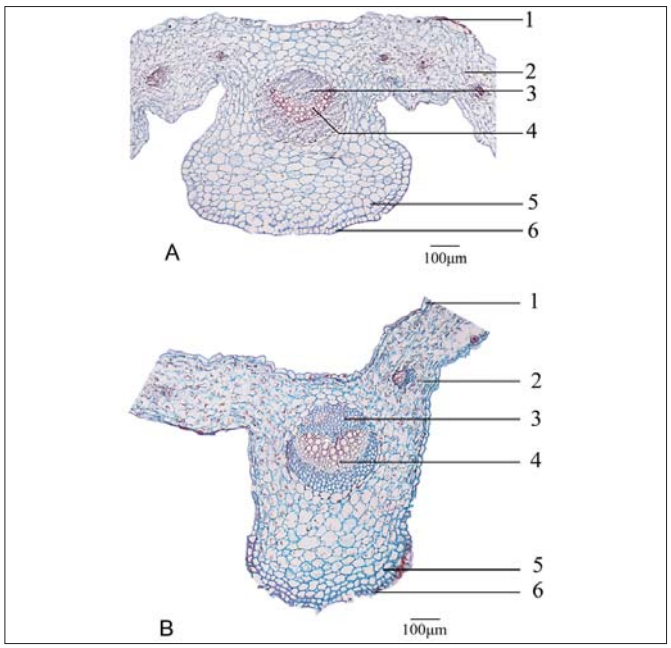

Figure 3. *Microscopic features of transverse section of leaves of P. asiatica L. Plate (A) and P. depressa Willd. (B) (1. Upper epidermis; 2. Mesophyll cells; 3. Phloem; 4. Xylem; 5. Collenchyma; 6. Lower epidermis)*

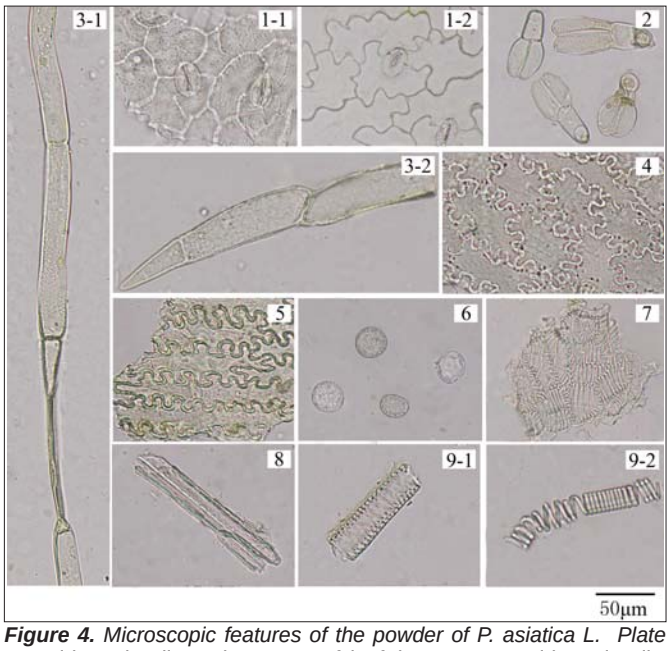

Figure 4. *Microscopic features of the powder of P. asiatica L. Plate 1. Epidermal cells and stomata of leaf (1-1. Upper epidermal cells; 1-2. Lower epidermal cells); 2. Glandular hairs; 3. Non-glandular hair (3-1. On the petiole and bract; 3-2. On the scape); 4. Pericarp cells; 5. Epidermis of sepal 6. Pollen grains; 7. Fragment of endothecium; 8. Fibre; 9. Vessel (9-1. Reticulated vessels; 9-2. Spiral vessels*

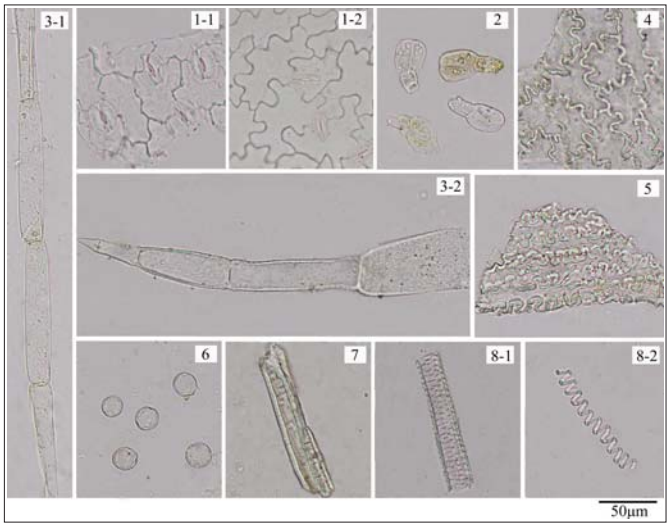

Figure 5. *Microscopic features of the powder of P. depressa Willd. Plate 1. Epidermal cells and stomata of leaf (1-1. Upper epidermal cells; 1-2. Lower epidermal cells); 2. Glandular hairs; 3. Non-glandular hair (3-1. On the head of root; 3-2. On the scape and the surface of the leaf); 4. Pericarp cells; 5. Epidermis of sepal; 6. Pollen grains; 7. Fibre; 8. Vessel (8-1. Reticulate vessels; 8-2. Spiral vessels)*

CHEMICAL CONSTITUENTS

Apart from common essential primary metabolites, various secondary metabolites related to a variety of biological activities have been isolated and identified from *Plantago* species. Up to now, more than 60 phytochemicals have been identified from pantago herbs, which can be divided into phenylethanoid glycosides, flavonoids, iridoids, triterpenoids, polysaccharides, phenolic acids and some other compounds.[1,6-8]

Phenylethanoid glycosides

Phenylethanoid glycosides (PGs) are derived phenolic compounds with a phenethyl alcohol structure widely distributed

in plants. Up to 15 species of PGs have been isolated from the whole plant of *P. asiatica* L. based on previous research.[9,10] PGs are generally considered as the most important type of bioactive metabolites and main antioxidants in plantago herb. Plantamajoside and acteoside (Figure 6) are the prominent PGs with a relatively higher content.[11-13] In *P. asiatica* L. and *P. major* L., plantamajoside is the chief PG and related to significant inhibitory activities of angiotension-converting enzyme,[14] while in *P. lanceolata* L. and *P. depressa* Willd., acteoside which has been shown to have anti-inflammatory, hepatoprotective and neuroprotective activities is the most abundant PG.[15,16] Plantamajoside and acteoside are also used as the quality evaluation standards in Chinese Pharmacopoeia and European Pharmacopoeia respectively.[3,5]

Flavonoids

Flavonoids, which are rich in foods of plant origin such as vegetables, fruits, teas and wine, are a class of polyphenols which have anti free radical properties toward reactive oxygen species. This class of biosynthetic metabolites may contribute to the prevention of several human diseases, and also the ageing process, by reducing the number of endogenous free radicals, without exhibiting any inherent toxicity themselves. A great number of flavonoids have been found in plantago herbs. Expressed as milligram of quercetin equivalent (QE) per gram of dw, the total content of flavonoids in *P. asiatica* L. was determined as 5.31 mg of QE/g of dw.[17] Apigein, luteolin, scutellarein, 6-hydroxyluteolin (Figure 6) and other corresponding glucosides were isolated from *P. asiatica* L. and *P. hostifolia* NAKAI et KITAGAWA. Subsequent comparative studies of flavonoids in other seven plantago herbs including *P. major* L., *P. depressa* Willd. and *P. lanceolata* L. concluded that these flavonoids could only be isolated from *P. major* L. which contained plantaginin, homoplantagin and glycosides of scutellarein as major flavonoids.[18] Associated with the PGs contents in these herbs, an interesting conclusion was reached that *P. asiatica* L. and *P. major* L. abundant in plantamajoside have similar distribution of flavonoids, and these flavonoids could not be found in acteoside abundant herbs, *P. depressa* Willd. and *P. lanceolata* L..

Iridoids

Iridoids, a class of monoterpenoids possessing a cyclopentan-(c)-pyran moiety, are usually found as glycosides in plantago herbs. This group of phytochemicals, it is thought act primarily as defense substances for plants and are thus are generally the active components of herbs. Iridoids are accepted as the other major bioactive metabolites of plantago herbs besides the PGs. Of the present iridoids in plantago herbs, aucubin, catalpol and geniposidic acid (GA) (Figure 6) are generally regarded as the main constituents and subject to the greatest scientific interest.[16,19-22] Catalpol is general known as an active diuretic agent and aucubin can stimulate the removal of uric acid from tissues and kidneys.[19] The total content of these iridoids have been utilised for quality control purposes. The final contents of aucubin, catalpol in *P. asiatica* L. were determined at 9.4, 1.3 and 0.1 mg/g respectively.[20] The quantification of catalpol and aucubin in *P. lanceolata* L. found that the content of catalpol was lower than 1 mg/g in most of the examined samples, whereas the content of aucubin ranged from 9.8 mg/g to 41.8 mg/g.[21] In other studies, the aucubin content of seven *Plantago* species was determined at 0.22-9.33 mg/g with different quantification methods and GA was detected in 28 *Plantago* species with a content ranged from 0.05 to 10.04 mg/g.[8, 22]

Triterpenoids

Triterpenoids belong to another group of important secondary metabolites in *Plantago* species. Oleanolic acid (OA) and ursolic acid (UA) (Figure 6), two primary triterpenoids of *P. major* L., have received most attention and have shown selective prostaglandin biosynthesis inhibition via cyclooxygenase-2 (COX-2).[24] Using conventional high-performance liquid chromatography (HPLC) methods, 1.5-1.1% of OA and 0.8-1.8% of UA in P. major L. were detected.[23] The contents of OA and UA in leaves and seeds of *P. major* L. were investigated with Soxhlet extraction method using a variety of solvents also.[24]

Polysaccharides

Polysaccharides are polymeric carbohydrate molecules widely found in plants. Up to now, several polysaccharides have been found in *P. asiatica* L.. These polysaccharides may promote defaecation and induce maturation of murine dendrite cells. Determined by high-performance gel permeation chromatography (HPGPC) method, a water-soluble polysaccharide with molecule weight about 1894 kDa was isolated and showed antioxidant activities.[25] In another study, three fractions of polysaccharide from the seed of *P. asiatica* L. were obtained and elucidated with various analytical methods.[26]

Phenolic Acids

Phenolic acids are types of aromatic acid compound. Caffeic acid, chlorogenic acid, ferulic acid, p-coumaric acid and vaillic acid (Figure 6) have been detected in plantago herb *P. major* L.[6] Among these acids, chlorogenic acid could potently enhance human mononuclear cell proliferation and interferon-gamma (IFN-γ) production, and ferulic acid could reduce the murine interleukin-1-alpha or interleukin-8 production in response to influenza virus infections *in vitro* and *in vivo*. Using an ELISA assy, chlorogenic acd, ferulic acid, p-coumaric acid and vanillic acid were found to enhance the activity of human lymphocyte proliferation and secretion of IFN-γ.[27]

Other phytochemicals

Many other phytochemicals, *e.g.* alkaloids, fats, oils, monoterpenoids, coumarins, sterols and volatile substances, have been identified from plantago herb.[1,6,27] At the same time, various biological activities derived from these compounds were investigated.

BIOLOGICAL ACTIVITIES

Owing to the constituent diversity of plantago herb, a wide range of biological activities have been reported so far.[1,6] In traditional Chinese medicine, plantago herb was commonly used as a diuretic and anti-inflammatory agent, majorly for patients with urination problems such as hermaturia and odynuria.[3] In the folk medicine of Egypt and India, these herbs have been used for the treatment of diarrhea and constipation. A great number of clinical applications relating to this herb have been recorded worldwide, e.g. treatments of diseases related to respiration, infection, reproduction, circulation, cancer prevention, digestion and so on.[2,3,6] Some definite biological activities using modern pharmacological methods have been listed as follows.

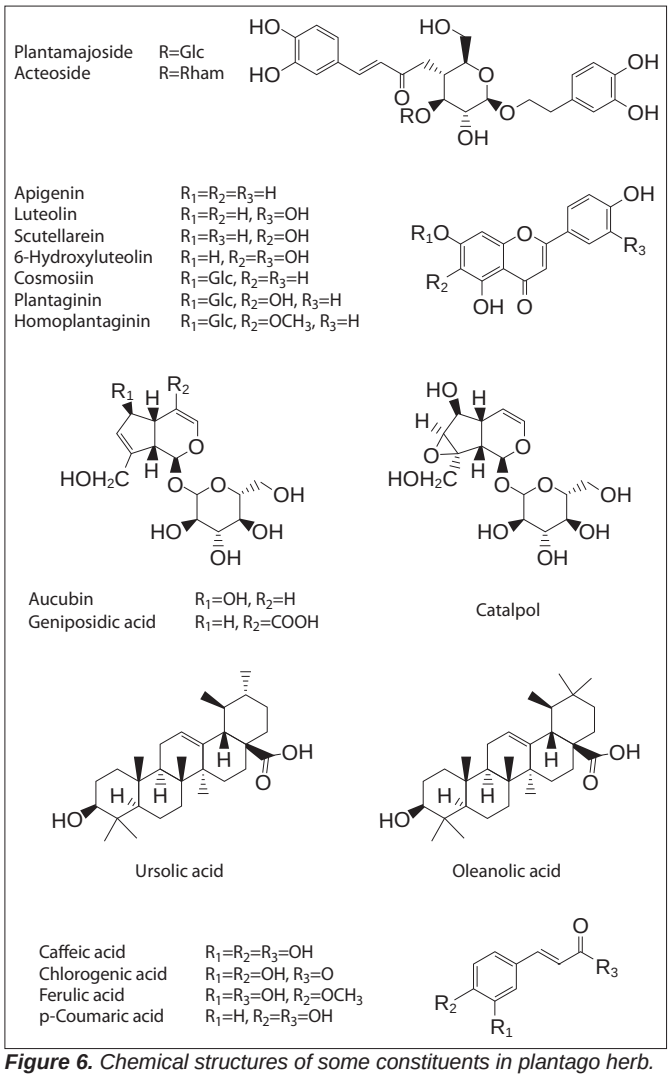

Figure 6. *Chemical structures of some constituents in plantago herb.*

Diuretic activity

Generally, the diuretic action is considered to be the most important function of plantago herbs in traditional Chinese medicine. In a famous Chinese material medica book *Compendium of Materia Medica* (Bencao Gangmu), plantago herb was described to promote the excretion of urine and toxins from the human body. So it has been applied to treat painful or difficult urination in history. The latest Chinese Pharmacopoeia listed the diuresis and detoxification effects as the major functions of plantago herb.[3] It was also listed as one of urinary system improvement herbs to treat urinary problems of animals and human in an United States patent.[28] The study carried out by Geng *et al.* found oral administration of the ethanol extract of *P. asiatica* L. increased the urine volume and concentrations of sodium, potassium and chloride ions in urine using the typical water-sodium retention rat model.[29] The diuretic effect could be induced by the component of iridoids. As the major bioactive constituents of plantago herbs, aucubin stimulates the removal of uric acid from kidneys and catalpol is the active diuretic agent in some fruits.[30]

Anti-inflammatory activity

Chronic inflammation is a symptom of a number of disease such as rheumatoid arthritis and autoimmune diseases. The anti-inflammatory effect of plantago herb is another significant biological activity. Topical application of two phenylethanoids, namely acteoside and plantamajoside isolated from *P. lanceolata L.* has been found to have significant anti-inflammatory activity on mouse ear edema.[31] Antinephritic and immunosuppressive effects have also been reported with aceteoside. To determine the anti-inflammatory effect of the extract of *P. lanceolata* L. on the production of nitric oxide (NO) and prostaglandin E2, NO synthase (NOS) type II, cyclooxygenase-1 (COX-1) and COX-2 mRNA expressions in a murine macrophage cell line J774A1 were examined.[32] The final results suggested that the anti-inflammatory effect may reflect decreased NO production, which is possibly due to inhibitory effects on inducible NOS gene expression or to NO-scavenging activity. According to comparative result of COX-1 inhibitory activity, *P. lanceolata* L. showed better inflammatory potential than *P. altissima* L.. In the examination of the anti-inflammatory activity of plantago seeds from 12 *Plantago* species including *P. asiatica* L., *P. depressa* Willd., *P. major* L. and *P. lanceolata* L., all of the tested seeds showed potential anti-inflammatory effect, especially seeds from *P. depressa* Willd..[8] Among the present constituents in plantago herb, OA and UA have been shown to inhibit COX-2 related biosynthesis and aucubin has been proved to suppress the inflammation induced by the injection of carrageenan.[33,34] All these results will contribute to the eventual determination of the actual anti-inflammatory agents found in these herbs.

Antioxidant activity

There are reports indicating that the effectiveness to prevent diseases, such as cardiovascular problems, ageing process and diabetes, and particularly when associated with anti-free radical activity in cancer and atherosclerosis, it is presumably attributed to the antioxidant properties of a herbal food. In order to examine the antioxidant properties of plantago herbs extracts, various assays measuring free radical scavenging ability were conducted, *e.g.* DPPH, hydroxyl radical, superoxide anion and nitric oxide scavenger capacity tests.[8,17,25] All of the tested extracts from different *Plantago* species including *P. major* L. showed potent antioxidant effect compared with a commonly used antioxidant.[8,35] In general, polyphenol compounds, phenylethanoid glycosides and flavonoids, which act as scavengers of free radicals, were considered as the major reason for their antioxidative effect. In another study, the scavenging activities of polysaccharides of *P. depressa* Willd. to hydroxyl free radical and superoxide anion free radical revealed polysaccharides were responsible for the antioxidation effect of the herb also.[36]

Antiviral activity

Because of the presence of some water-soluble phenolic acids, such as caffeic acid, chlorogenic acid, ferulic acid and p-coumaric acid, the aqueous extract of *P. major* L. showed moderate antiviral activity against herpes simplex virus HSV-2 and adenovirus ADV.[37] Other research has investigated, the antiviral activity of pure flavonoids from *P. asiatica* L. towards human immunodeficiency virus (HIV), the causative retrovirus of acquired immune deficiency syndrome (AIDS).[38] Scutellarein and 6-hydroxyleution demonstrated better HIV-reverse transcriptase inhibition (leading to the end of virus production) than a known inhibitor baicalein, while plantaginin and 6-hydroxyluteolin 7-glucoside had equivalent anti-HIV activities to baicalein.

Antibacterium activity

The growth inhibition of *Bacillus subtilis*, *Mycobacterium tuberculosis*, *Escherichia coli* and *Mycobacterium phlei* by different kinds of solvent extracts from plantago herbs were investigated in several studies.[39,40] The aqueous extract inhibited the growth of *Bacillus subtilis* and *Mycobacterium tuberculosis*, wheras the methanol extract significantly inhibited the growth of *Mycobacterium phlei*. The antibacterial activities of *P. major* L. extracts obtained by classical (maceration) and ultrasonic extraction were examined against two Gram-positive bacterial species and two Gram-negative bacterial species.[35] Similar antibacterial activity for both extracts were detected which were comparable with two controlled antibiotics, erytromycin and tylosin tartarate.

Immunomodulatory activity

The immunomodulatory activities of flavonoids, iridoids, monterpenoids, triterpenoids and phenolic acids extracted and isolated from plantago herb were evaluated on human peripheral blood mononuclear cells (PBMC).[27] results showed that all of the tested pure compounds showed immunomodulatory activity and some compounds exhibited immunostimulatory activity, which indirectly supported the application of plantago herbs in the treatment of cancer and many infectious diseases. When the aqueous extracts of *P. asiatica* L. and *P. major* L. were tested on various human leukemia cells, both species exhibited effects of immunodulatory activity by enhancing lymphocyte proliferation and secretion of IFN-γ.[41]

Glycation inhibitory activity

The glycation reaction involves a series of non-enzymatic reactions leading to the formation of advanced glycation end-products (AGEs), which have been implicated in the pathogenesis of many diseases, e.g. diabetic vascular disease, connective tissue disease, neurodegenerative Alzheimer's disease and end-stage renal disease. Current research found that the extract of *P. asiatica* L. and pure plantamajoside showed significant inhibitory effects on *in vitro* AGEs formation.[42] Compared with a standard anti-glycation agent aminoguanidine, the glycation inhibitory activity of plantagmajoside was comparable.

Other reported uses

In addition to the biological activities described above, the leaf extracts of *P. major* L. and *P. lanceolata* L. have been used topical as an astringent for burns and wounds, for treatment of poison by ivy, and for inflammation of mucous membranes and skin. herb also exhibited cytotoxic activities to various cancer cells, *e.g.* bladder, bone, cervix, kidney, lung and stomach, and the essential oil of *P. asiatica* L. showed hypocholesterolaemic effects *in vitro* and *in vivo* by altering the expression of 3-hydroxy-3-methylglutaryl-co-enzyme A (HMG-CoA) reductase.[41,43] More pharmacological activities of this valuable herb need to be further investigated in the future.

SAFETY EVALUATIONS/CONTERAINDICATIONS

As a herbal medicine with a long application history, there are few reports relating to the safety of plantago herbs,[44] although there are a few reports describing some contraindications associated with plantago use. A few undesirable effects relating to individual allergic response has also been reported.[45] Hence, some countries restrict its use for both pregnant and breastfeeding women. However, there is not any proved scientific document to support this restriction. To date there appears very few contraindications associated with the use of this plant.

CONCLUSIONS

Plantago herb is widely used in herbal medicine, especially in China and some developing countries. This herb originating from the dried whole plant of *Plantago* family has remarkable effect on the treatment of hematuria and inflammation.[2,3,6] Of all the *Plantago* species, *P. asiatica* L., *P. depressa* Willd., *P. major* L. and *P. lanceolata* L. are the major species used in the clinic, and have attracted most scientific attention. The herb contains several bioactive chemical constituents (*e.g.* phenylethanoid glycosides, flavonoids, iridoids, triterpenoids, phenolic acids) which exhibit the corresponding biological activities (*e.g.* diuresis, anti-inflammation, antioxidation, antivirus, antibacterium and immunomodulatory).[1,6,8,9] Generally, plantamajoside in *P. asiatica* L. and *P. major* L., and acteoside in *P. lanceolata* L. and *P. depressa* Willd. are considered to be the main bioactive constituents of the herb and are used as a quality control measure with respect to proposed biological activity.[2-4] Aucubin and catalpol, which are also major active constituents of the herb, are regarded as the bioactive secondary metabolites in charge of the diuretic effect.[16,29,30] As a valuable herbal medicine with long history, further investigation of plantago herb should be conducted in the future.

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Author's background

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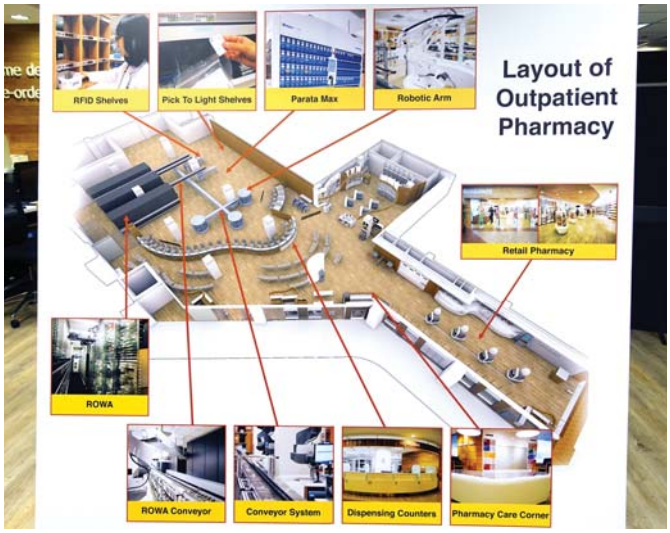
SHPHK Delegation to Singapore Pharmacy Congress 2014 and Hospital Visits

The Singapore Pharmacy Congress is an annual event organized by the Pharmaceutical Society of Singapore. During the past years, many Hong Kong Pharmacists have attended the Congress with very good feedback regarding the progress Singapore has made during the past decades on the Pharmacy profession and the Pharmaceutical service. It has also been a great opportunity for hospital visits and to learn about their state-of-the-art practices in Singapore.

This year (2014) the Congress was organized in conjunction with the International Medication Safety Network (IMSN). Apart from having Medication Safety in the main stream, the Congress also featured other interesting workshops in clinical pharmacy, legislation and community pharmacy practice.

The Society of Hospital Pharmacist of Hong Kong (SHPHK) has organized a delegation of 20 for a 4-day visit on 17-20 October, 2014. The delegates came from hospital and community pharmacists in Hong Kong and Macau. We visited the National University Hospital, the Tan Tock Seng Hospital, the Singapore General Hospital and the ST Logistics company.



Automated pharmacy layout

With Congress Chairman Mr Wu

ICM Pharma (ST Logistics)

Inside look of strip pack dispenser

The first place we went to right after we landed was a repacking factory call ICM Pharma, contracted under the company ST Logistics. They provide repackaging of unit dose medications for hospitals. Dosage forms that could not be dispensed using automatic machines by hospitals, e.g. blister pack, ampoule, vial, etc, would be repackaged in this factory. The hospitals provided the bulk products, and the company would repack, label (with labeling instruction provided by the hospitals) and deliver back to the hospitals. They believe that it is more cost effective to contract out this job. There are other companies doing similar service, e.g. DHL.

National University Hospital

Our next stop was the National University Hospital (NUH). The Outpatient Pharmacy was recently renovated with full sets of automation. Rowa Vmax Box Picking Machine was used for products either came in as original boxes or repacked as boxes in the pharmacy. For loose tablets and capsules, Parata Max Pill Counter was used. Conveyer belt system running above and below the counters was installed to deliver the medications to the check points. Radio Frequency Identification (RFID) was extensively used to ensure the right medications were picked and checked. The pharmacists would take the baskets and give to the patients with appropriate counselling. Automation was shown to improve waiting time and accuracy of dispensing and allow more time for the pharmacists to talk to the patients.

In the inpatient setting, Omnicell Automated Dispensing Cabinets (ADC) were installed. Pharmacy Technicians would periodically refill the ADCs. Unit dose repackaging was prepared by the JVM machine. A manual deblistering machine was available for recovering tablets from push-thru blisters before putting the tablets into the JVM system. Ampoules and vials were repacked by the ICM Pharma as mentioned above.

Our delegation were given a concise overview of the clinical pharmacist services they provided in the NUH, including Anticoagulation Consultation service (ACC), Cardiac Clinic (Medication Therapy Management-Heart failure), Kidney Transplant Pharmacist, Paediatric Antimicrobial Stewardship Programme (ASP), Adult ASP, DM Integrated Care Team, ICU Clinical Pharmacy Service, as well as Pharmacy outreach programmes. Most of these programs were chargeable to

the patients and they have shown convincing data on both cost recovery and cost-benefit outcomes.

Tan Tock Seng Hospital

After two days of Congress, we visited the Tan Tock Seng Hospital. Tan Tock Seng Hospital is one of the biggest hospitals in Singapore and is well known for their innovative services. Like the NUH they used Rowa and Parata Max for outpatient dispensing. What is different is that they had installed a fascinating Robotic Arms System for "grabbing' the baskets, which are contains the boxes and bottles of dispensed medications, to the pigeon holes. This spectacular system is the first one of its kind. Their patient service counters are aesthetically arranged in to align with the semi-circular Robotic Arms System. (See diagram)

Singapore General Hospital

The Singapore General Hospital (SGH) has another first of its kind technology. The humbly named Drug Dispensing System (DDS) is a battery of automatic dispensing machines for counting blister packs. This solves the problem of having to deblister packages. For the small number of non-blister packs, manual dispensing was carried out using the LED-guided Picking system to ensure accuracy.

Robotic Arm Dragon Claw

For inpatient, the SGH uses the Swisslog system for dispensing unit dose of virtually all kinds of dosage forms. The Swisslog system can pick tablets, capsule, vials, ampoules, etc, leaving a small proportion on products to be hand picked, e.g. ointment, inhalers, etc.

Apart from the use of technology and having safety as their priority, the hospital pharmacies in Singapore were noted to be highly client focused. There were appropriately posted signages, numerous (20+) service counters, open design counters, short waiting time, disable access counters, etc.

The SHPHK delegates totally enjoyed this eye opening trip, not to mention the knowledge learned from the Congress and the nice food and environments enjoyed from the hawker stalls and elegant restaurants. The SHPHK will continue to organize visits to overseas countries. Please stay tuned and join us if you can.

Reported by Michael Ling, SHPHK

(For more details, please visit the www.shphk.org.hk website)

Proven safety

Unitdose prepacking

Parata for loose tablets

Visiting Tan Tock Seng Hospital

Many drug issue counters

Plenary Session of the 2015 Hong Kong Pharmacy Conference held on 29 March 2015 – Pre Pharmacy Council Task Force

This year's final plenary session was devoted to examining the fundamental issues integral to establishing a pharmacy council in Hong Kong (HKPC). The HKPC shall be an independent statutory body of licensed pharmacists dedicated to setting and implementing professional standards of practice. The ultimate goal is to advance pharmacy practice for better therapeutic outcomes in patient care. A 22-member Pre-Pharmacy Council Task Force with representation from all sectors of the pharmacy profession in Hong Kong was appointed by the Pharmacy Conference Committee in December 2014. The task force held 3 meetings before the conference to set the agenda for the plenary session. Ms. S.C. Chiang was named the session's moderator.

Ms S C Chiang

In her opening remarks, Mrs. Mary Cheng reminded everyone that the notion of a pharmacy council was explored on and off for almost three decades. The closest we were to the finishing line was in 1998. Regrettably, the bill to create the pharmacy council was derailed by a competing bill of a higher order of complexity that concerned the dissolution of the Urban Council. Now almost two decades later, when there are promising signs of support by government for establishing a pharmacy council, all eyes must be focused on upgrading pharmacy practice and elevating its professional stature in Hong Kong.

The second talk by Ms. Chiang on the current functions of the Pharmacy and Poisons Board (PPB) set the stage for the final talk on possible functions of the pharmacy council by Mr. William Chui. The model pharmacy council was synthesized following a meticulous study, as well as a lively discussion at the task force meetings, of the medical and nursing councils of Hong Kong as well as the General Pharmaceutical Council of the United Kingdom. The HKPC was envisaged to perform core as well as operational functions, 9 in total. Many of these 9 functions were intended to capture the PPB's existing role in ensuring the rigor of pharmacy education and practice. Consequently, the reconfigured PPB will be able to focus on the supply chain of pharmaceutical products, spanning

Mrs Mary Cheng

Mr William Chui

Ms Iris Chang

The panelists gave their views on the establishment of Pharmacy Council in Hong Kong

manufacturers, wholesalers, and distributors, as related to the assurance of drug product quality. The Drug Office will be able to spend more time on drug product registration, thereby making timely access to life-saving drug products by those needy patients a reality.

Mr. Chui's talk was followed by a session of invitees from various pharmacy sectors and then by an open session from the floor. The invitees were Ms. Iris Chang (community pharmacist), Mr. Philip Chiu (community chain pharmacist), Mr. William Chui (hospital pharmacist), Ms. Annette Chiu (pharmaceutical industry pharmacist), Mr. Michael Yim (pharmacist from the Department of Health), Dr. May Lam (HKU) and Mr. Allan Chan (consumer). They were asked to share their views of the pharmacy council as articulated by the task force. While the majority of the panelists supported the formation of the Pharmacy Council, Ms. Iris Chang felt there was no urgent need to establish the HKPC. A community pharmacist suggested a public forum be organized for the community pharmacists to understand more about the functions of the Pharmacy Council.

Establishing a pharmacy council is a powerful public statement of the coming of age of the pharmacy profession --- a healthcare profession that is ready to set high standards and self-regulate. The council serves as a bridge to other health related councils in our collective commitment to improve the efficiency of our health-care system. By not seizing this rare opportunity of external support and consensus among leaders in the pharmacy profession for establishing a pharmacy council, the pharmacy profession in Hong Kong will forever be relegated to be in the long shadow of the medical profession in Hong Kong. The pharmacy profession in Hong Kong has historically been underutilized and the public has therefore been underserved in drug utilization. By speaking out on pharmacist manpower and pharmaceutical care, the council is our only hope of making an impact in enhancing job satisfaction for pharmacists and the care we are capable of delivering.

Given the generally favorable sentiment for the pharmacy council at the plenary session, we hope that the Pre-Pharmacy Council Taskforce, to be led by CUHK and PSHK, will move forward in stride to establish the Pharmacy Council to the next phase. There are 5 strategic tasks in this phase, as follows:

a. to sharpen the vision and mission statements of the proposed Pharmacy Council, to propose standard of pharmacist practice, and to discuss means to bring all pharmacists to meet those standards;

b. to expand the breadth and depth of grassroots, peer to peer, and governmental support;

c. to determine the optimal membership of the council, terms of reference, and by-laws;

d. to determine the infrastructure, personnel, budget, and revenue streams required to launch and sustain the HK PC; and

e. to establish teams and time-lines to execute the action plan as articulated above.

The preliminary plan was to have a report to the pharmacy community in Hong Kong by the time of the 2016 pharmacy conference with a final draft to the Food and Health Bureau for action by July 1, 2016. For additional information on the current phase, please refer to the presentation material at www.pharmacyconference.org before the 2016 Pharmacy Conference.

Vincent H. L. Lee

Chair, Pre-Pharmacy Council Task Force

April 3, 2015

Ms Annette Chiu

Mr Philip Chiu

Mr Michael Yim

Dr May Lam

Mr Wing-Kai Chan

Professor Vincent Lee

TRAJENTA® DUO

(Boehringer Ingelheim)
(Eli Lilly)

Edited by Ms. Ivy Chan

Active Ingredient:
Linagliptin and Metformin Hydrochloride

Presentation:
Trajenta Duo 2.5mg/500mg - Each film-coated tablet contains 2.5 mg of linagliptin and 500mg of metformin hydrochloride
Trajenta Duo 2.5mg/850mg - Each film-coated tablet contains 2.5 mg of linagliptin and 850mg of metformin hydrochloride
Trajenta Duo 2.5mg/1000mg - Each film-coated tablet contains 2.5 mg of linagliptin and 1000mg of metformin hydrochloride

Pharmacological Properties:
Linagliptin is an inhibitor of the enzyme DPP-4 (Dipeptidyl peptidase 4) an enzyme which is involved in the inactivation of the incretin hormones GLP-1 and GIP (glucagon-like peptide-1, glucose-dependent insulinotropic polypeptide). These hormones are rapidly degraded by the enzyme DPP-4. Both incretin hormones are involved in the physiological regulation of glucose homeostasis. Incretins are secreted at a low basal level throughout the day and levels rise immediately after meal intake. GLP-1 and GIP increase insulin biosynthesis and secretion from pancreatic beta cells in the presence of normal and elevated blood glucose levels. Furthermore GLP-1 also reduces glucagon secretion from pancreatic alpha cells, resulting in a reduction in hepatic glucose output. Linagliptin binds very effectively to DPP-4 in a reversible manner and thus leads to a sustained increase and a prolongation of active incretin levels. Linagliptin glucose-dependently increases insulin secretion and lowers glucagon secretion thus resulting in an overall improvement in the glucose homoeostasis. Linagliptin binds selectively to DPP-4 and exhibits a > 10,000 fold selectivity versus DPP-8 or DPP-9 activity *in vitro*.
Metformin hydrochloride is a biguanide with antihyperglycaemic effects, lowering both basal and postprandial plasma glucose. It does not stimulate insulin secretion and therefore does not produce hypoglycaemia.
Metformin hydrochloride may act via 3 mechanisms:
(1) reduction of hepatic glucose production by inhibiting gluconeogenesis and glycogenolysis,
(2) in muscle, by increasing insulin sensitivity, improving peripheral glucose uptake and utilization,
(3) and delay of intestinal glucose absorption.
Metformin hydrochloride stimulates intracellular glycogen synthesis by acting on glycogen synthase.
Metformin hydrochloride increases the transport capacity of all types of membrane glucose transporters (GLUTs) known to date.
In humans, independently of its action on glycaemia, metformin hydrochloride has favourable effects on lipid metabolism. This has been shown at therapeutic doses in controlled, medium-term or long-term clinical studies: metformin hydrochloride reduces total cholesterol, LDL cholesterol and triglyceride levels.

Indications:
Treatment of adult patients with type 2 diabetes mellitus:
Trajenta Duo is indicated as an adjunct to diet and exercise to improve glycaemic control in adult patients inadequately controlled on their maximal tolerated dose of metformin alone, or those already being treated with the combination of linagliptin and metformin.
Trajenta Duo is indicated in combination with a sulphonylurea (i.e. triple combination therapy) as an adjunct to diet and exercise in adult patients inadequately controlled on their maximal tolerated dose of metformin and a sulphonylurea.
Trajenta Duo is indicated in combination with insulin (i.e. triple combination therapy) as an adjunct to diet and exercise to improve glycaemic control in adult patients when insulin and metformin alone do not provide adequate glycaemic control.

Dosage & Administration:
The dose of antihyperglycaemic therapy with Trajenta Duo should be individualised on the basis of the patient's current regimen, effectiveness, and tolerability, while not exceeding the maximum recommended daily dose of 5 mg linagliptin plus 2,000 mg of metformin hydrochloride.
<u>For patients inadequately controlled on maximal tolerated dose of metformin monotherapy</u>
For patients not adequately controlled on metformin alone, the usual starting dose of Trajenta Duo should provide linagliptin dosed as 2.5 mg twice daily (5 mg total daily dose) plus the dose of metformin already being taken.
<u>For patients switching from co-administration of linagliptin and metformin</u>
For patients switching from co-administration of linagliptin and metformin, Trajenta Duo should be initiated at the dose of linagliptin and metformin already being taken.
<u>For patients inadequately controlled on dual combination therapy with the maximal tolerated dose of metformin and a sulphonylurea</u>
The dose of Trajenta Duo should provide linagliptin dosed as 2.5 mg twice daily (5 mg total daily dose) and a dose of metformin similar to the dose already being taken. When linagliptin plus metformin hydrochloride is used in combination with a sulphonylurea, a lower dose of the sulphonylurea may be required to reduce the risk of hypoglycaemia.
<u>For patients inadequately controlled on dual combination therapy with insulin and the maximal tolerated dose of metformin</u>
The dose of Trajenta Duo should provide linagliptin dosed as 2.5 mg twice daily (5 mg total daily dose) and a dose of metformin similar to the dose already being taken. When linagliptin plus metformin hydrochloride is used in combination with insulin, a lower dose of insulin may be required to reduce the risk of hypoglycaemia.
For the different doses of metformin, Trajenta Duo is available in strengths of 2.5 mg linagliptin plus 500 mg metformin hydrochloride, 2.5 mg linagliptin plus 850 mg metformin hydrochloride, and 2.5 mg linagliptin plus 1,000 mg metformin hydrochloride.
Trajenta Duo should be taken twice daily with meals.

Forensic Classification:
P1S1S3

Hong Kong Pharmaceutical Journal: For Detailed Instructions for Authors

INTRODUCTION

Hong Kong Pharmaceutical Journal (HKPJ) is the official publication of the Pharmaceutical Society of Hong Kong, the Practising Pharmacists Association of Hong Kong and the Society of Hospital Pharmacists of Hong Kong. It is a journal of the pharmacists, for the pharmacists and by the pharmacists. The Journal is currently divided into several sections: ***Editorial Comment; News & Short Communications; Pharmacy Practice; Over-the-Counter & Health; Drugs & Therapeutics; Herbal Medicines & Nutraceuticals; Pharmaceutical Technology*** and ***New Products***. It publishes review articles or original papers relevant to these different fields of pharmacy. In addition to the regular four issues of the Journal per year, there are issues dedicated solely to reports on special function of the society. The Aims and Scope of the Journal are published on the inside back cover of each issue.

Submission of Manuscript

Submission of a paper implies that it has not been published previously, that it is not under consideration for publication elsewhere, and that if accepted it will not be published elsewhere in the same form, in English or in any other language, without the written consent of the publisher. Authors are specifically discouraged from submitting papers as fragmented studies of a particular topic. A manuscript must be indicated which section it is belonged. Upon received, it will be screened by a **Sectional Editor** of HKPJ for initial consideration before it is sent out for further review or comment.

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Abbreviations
About, approximately: ca.
Anhydrous: dry (not anhyd.)
Aqueous: aq.
Circular dichroism: CD
Concentrated (or mineral acids): conc.
Concentrations: ppm (or ppb), μM, mM, M, %, mol
Dry weight: dry wt; fresh weight: fr. wt
Electricity: V, mA, eV
Force due to gravity (centrifugation): g; rpm (revolutions min^{-1})
Gas chromatography: GC
Gas chromatography-mass spectrometry: GC-MS Trimethylsilyl derivative: TMSi (TMS cannot be used as this refers to the internal standard tetramethylsilane used in ^{1}H NMR)
High performance liquid chromatography: HPLC
Infrared spectrophotometry: IR
Length: nm, μm, mm, cm, m
Literature: lit.
Mass spectrometry: m/z $[M]^{+}$ (molecular ion, parent ion)
Melting points: uncorr. (uncorrected)
Molecular mass: Da (daltons), kDa
Molecular weight: M_r
Nuclear magnetic resonance: ^{1}H NMR, ^{13}C NMR, Hz, δ
Numbers: e.g. 1, 10, 100, 1000, 10000; per or $^{-1}$
Optical rotatory dispersion: ORD
Paper chromatography: PC
Precipitate: ppt.
Preparative thin-layer chromatography: prep. TLC
Radioactivity: dpm (disintegrations per min), Ci (Curie), sp. act (specific activity), Bq (1 becquerel = 1 nuclear transformation sec^{-1})
Repetitive manipulations: once, twice, x3, x4, etc.
RR_t (relative retention time), R_1 (Kovat's retention index), ECL (equivalent chain length- term frequently used in fatty acid work)
Saturated: satd.
Solution: soln.
Solvent mixtures including chromatographic solvents: abbreviate as follows n-BuOH-HOAc-H_2O (4:1:5)
Statistics: LSD (least significant difference), s.d. (standard deviation), s.e. (standard error)
Temperature: (with centigrade), mp, mps, mmp, bp
Temperature: temp.
Thin-layer chromatography: TLC, R_f
Time: s, min, h, day, week, month, year
Ultraviolet spectrophotometry: UV, A (absorbance, not aD-optical density)
Volume: 1, (litre), μ1, ml
Weight: wt, pg, ng, μg, mg, g, kg

Inorganics, e.g. $AlCl_3$ (aluminum chloride), BF_3 (boron trifluoride), Cl., CO_3, H_2, HCl, $HClO_4$ (perchloric acid), HNO_3. H_2O, H_2O_2, H_2SO_4, H_3B0_3 (boric acid), He, $KHCO_3$ (potassium bicarbonate), $KMn0_4$ (potassium permanganate;), KOH, K-Pi buffer (potassium phosphate buffer), $LiAlH_4$ (lithium aluminium hydride), Mg^{2+}, $MgCl_2$, N_2, NH_3, $(NH_4)_2S0_4$, Na^{+}, $NaBH_4$ (sodium borohydride), NaCl, $NaI0_4$ (sodium periodate), NaOH, Na_2S0_3 (sodium sulphite), Na_2S0_4 (sodium sulphate), $Na_2S_20_3$ (sodium thiosulphate), 0_3, PPi (inorganic phosphate), SO_4^{2-}., Tris (buffer).

Organics, e.g. Ac_20 (acetic anhydride), n-BuOH (butanol), C_6H_6 (benzene), CCl_4 (carbon tetrachloride), CH_2Cl_2 (methylene chloride), $CHCl_3$ (chloroform), CH_2N_2 (diazo-methane), CM (carboxymethyl), DEAE (diethylaminoethyl), DMF (dimethylformamide), DMSO (dimethyl sulphoxide), EDTA (ethylene-diaminetetra-acetic acid), Et_20 (diethyl ether), EtOAc (ethyl acetate), EtOH (ethanol), $HC0_2H$ (formic acid), HOAc (acetic acid), iso-PrOH (iso-propanol), Me_2CO (acetone), MeCOEt (methyl ethyl ketone), MeOH (methanol), NaOAc (sodium acetate), NaOMe (sodium methoxide), petrol (not light-petroleum or petroleum ether), PhOH (phenol), PrOH (propanol), PVP (polyvinylpyrrolidone), TCA (trichloroacetic acid), TFA (trifluoroacetic acid), THF (tetrahydrofuran). ^{1}H NMR solvents and standards: $CDCl_3$ (deutero-chloroform), D_2O, DMSO-d_6 [deuterodimethylsulphoxide not $(CD_3)_2S0$], pyridine-d_5 (deuteropyridine), TMS (tetramethylsilane).

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First-line treatment for chronic hepatitis B (CHB) in adults[1-3]

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tenofovir disoproxil fumarate 300 mg

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References: 1. Lok ASF and McMahon BJ. Hepatology 2009;50:1-36 2. EASL. Journal of Hepatology 2012;57:167-185 3. Liaw YF et al. Hepatol Intl 2012;6:531-56 4. Corsa A et al. AASLD 2014. Poster #1707 5. Marcellin P et al. AASLD 2014. Poster #229 6. Marcellin P et al. The Lancet 2013;381(9865):468-475

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