

HEALTH SCIENCES

1. MUTAGEN-INDUCED CHROMOSOME DAMAGE: IMPLICATION IN CANCER SUSCEPTIBILITY

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The objective of this study is to determine if bleomycin-induced chromosome aberrations can predict which individuals are at risk of developing environmentally-induced cancers. Peripheral blood samples were collected from thirty (30) head and neck cancer patients from two hospitals in Metro Manila. Twenty noncancer, normal individuals, age and sex-matched with the cancer patients constituted the control group. Lymphocytes were cultured by the microculture method, incubated for 72 hours at 37°C, and treated with bleomycin (30ug/mL) 5 hours before harvest. Metaphase chromosomes were homogenously stained with Giemsa. From each individual, 50-100 metaphase cells were screened for the presence of chromatid-type and chromosome-type aberrations. Preliminary data reveal that the mean frequency of chromatid breaks in the cancer group is 1.11 breaks per cell (b/c). This is above 0.8 b/c, established to be the indicator of sensitivity to bleomycin-induced chromosome damage. It is likewise higher than 1.0 b/c which is considered to indicate hypersensitivity in many toxicology cytogenetic studies. The mean frequency of chromatid breaks in the control group is 0.48 b/c.

Key words: Mutagen, chromosome damage, cancer susceptibility, head and neck cancer, metaphase, bleomycin, microculture method, lymphocytes, chromatid break, environmentally-induced

2. BREAST CANCER SUSCEPTIBILITY GENE 1 MUTATION IN HIGH RISK FILIPINO FEMALES

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BRCA 1 is a tumor suppressor gene whose mutation is associated with susceptibility to breast cancer. Women who have an inherited mutation in *BRCA1* gene have an 85% chance of developing breast cancer during their lifetime compared to only 12.5% for the general population [Zsabo and King 1995]. The aim of this study is to identify *BRCA1* gene mutations among Filipino breast cancer patients who have any two of the following criteria: (1) At least two members of the family have breast or ovarian cancer; (2) The patient has bilateral breast cancer; and (3) The patient developed cancer before the age of 35. A total of 51 breast cancer patients have been screened. Ten patients satisfy the inclusion criteria. The polymerase chain reaction was performed on the ten subjects using forward and reverse primers of *BRCA1* exons 2, 11a, 11b, and 20. These four exons have been shown to contain the most commonly identified germ-line mutations by complete screening of the *BRCA1* gene [Couch et al. 1996]. Initial results showed the absence of mutations in exons 11a, 11b, and 20 of 4 patients included in the study. The absence of mutations was confirmed by sequence analysis. Currently, exon 2 and the samples from 6 remaining patients are being evaluated.

Key words: *BRCA1* gene, tumor suppressor gene, mutation, breast cancer, PCR, exons 2, 11a, 11b, 20, sequence analysis

3. DEVELOPMENT IN TRANSFORMED TISSUES: BREAST CARCINOMA IN FILIPINO FEMALES

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The normal human mammary gland consists of lobes surrounded by loose connective tissues containing adipose cells. The dense cellular connective tissues divide the lobes into lobules. A lobule consists of several intralobular ducts with a simple cuboidal epithelium that rests upon the basal lamina. Fat droplets are observed in the cytoplasm and lumen of the alveoli in active glands.

Histological analyses done on twenty seven (27) clinical cases revealed three types of pathological observations. The mucinous type has mucus secretions in the lumen. Epithelia consist of small, multilayered, light-staining cells. Secretions are observed in the lumen. The ductal type exhibits hyperplasia of the intralobular duct with small cells arranged in several layers. The basal lamina is absent. In the medullary type, the cells are lightly staining.

Key words: Histopathogenesis, mammary gland, breast carcinoma, connective tissues, adipose cells, intralobular ducts, cuboidal epithelium, hyperplasia

4. DEVELOPMENT IN TRANSFORMED TISSUES: CERVICAL CARCINOMA IN FILIPINO PATIENTS

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A histological study of six cases of cervical carcinoma in Filipinos was undertaken. Changes noted under light microscopy were the following: erosion and various thickenings of the epithelial layer in some portions of the cervical epithelium facing the vaginal canal; loss of the orderly cellular patterns of the epithelium; invasion of the underlying lamina propia with tumor cells from the epithelial layer; presence of whorled masses of various sizes in the deeper portions of the lamina propia.

Key words: Transformed tissues, cervix, cervical carcinoma, epithelial layer, cervical epithelium, vaginal canal, lamina propia, tumor cells light microscopy.

5. FACTORS INFLUENCING DELAY IN DIAGNOSIS AMONG COLON AND RECTAL CANCER PATIENTS

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This prospective, descriptive study was conducted on all histologically proven cases of colon and rectal cancer admitted at our institution from January 1 to June 30, 1997, in order to determine the frequency of delayed diagnosis (defined as time interval from onset of symptoms to establishment of diagnosis of more than three months) and the factors that influence the delay. An open-ended interview was conducted on all patients with delayed diagnosis. A total of 78 cases of colon and rectal cancer were admitted, of which 46 (58%) were presented with delayed diagnosis. Majority of patients had low educational attainment, originated from outside Metro Manila, and did not immediately consult a physician mainly for financial reasons. Only 56% initially consulted a physician, at an average interval of five months from onset of symptoms. A significant number of physicians were perceived by patients to have performed inadequate assessments, and only 47% considered the possibility of malignancy on consult. Amoebiasis and hemorrhoidal disease were the most common initial considerations. Nearly 90% of patients with delayed diagnosis presented with advanced stage of disease (Stage III or IV) on surgical exploration. Resectability was 88%. This study showed that socio-economic factors contributed to patient-related delay while physician-related delay was also significant. It demonstrates the need to further improve public awareness and primary physician education in order to enhance earlier detection of colon and rectal cancer and consequently improve results of treatment.

Key words: Colon and rectal cancer, delayed diagnosis

6. BINDING PATTERNS OF PROSTATE TISSUES TO SOME LECTIN AND NEOGLYCOPROTEON PROBES

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As a source of corroborative data of medical diagnosis and prognosis, a lectin from *Viscum album* (VAA) was tested for the presence of binding sites in 21

cases of benign hyperplasia and eight cases of adenocarcinoma of the prostate. Some neoglycoproteins (simple sugars conjugated with unreactive Bovine Serum Albumin), as well as fucoidan and heparin were also tested. Of the probes used, VAA, mannose, lactose, N-acetylglucosamine, and heparin were negative in both benign hyperplastic tissues (Benign Prostatic Hyperplasia) and adenocarcinoma. Towards N-acetylgalactosamine there was a strong positive reaction in adenocarcinoma found specifically in the connective tissues surrounding the prostate glands. The gland epithelia themselves were negative. Benign Prostatic Hyperplasia gave a weak positive response to N-acetylgalactosamine.

Responses to fucoidan are described.

Key words: Lectin, neoglycoprotein probes, prostate cancer

7. GENOTYPE ANALYSIS OF HIV-1 ISOLATES FROM RISK GROUPS IN METRO MANILA

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The human immunodeficiency virus (HIV), the causative agent of AIDS, has infected 12-13 million people worldwide, and continues to infect many more. There are types of HIV which can be distinguished genetically and antigenetically, the HIV-1 and HIV-2. HIV-1, which is most common in North America, Europe, and parts of Asia has been shown to be genetically diverse, comprising at least nine distinct genetic subtypes, A through I, plus a highly divergent subtype O. The presence of such genetically and antigenetically diverse HIV-1 subtypes is of major concern for three important reasons: (1) implications in vaccine development, (2) differences in sensitivity of antiretroviral drugs, and (3) certain subtypes have higher heterosexual transmission efficiency, and therefore, major implications in transmission and epidemics.

From August 1995-December 1996, 37 blood samples from HIV (+) individuals were collected for genotyping, using the Polymerase Chain Reaction (PCR) technique and Heteroduplex Mobility Assay (HMA) method. The most common subtype identified was subtype B (B1,2,3) in 19 samples (51.35%), followed by subtype E (E1, 2,3) in 11 samples (29.73%), and Subtype C/A in 7 samples (18.92%). These findings presumably reflect the modes by which the virus strain was transmitted,

and would give an idea of the risk group of the patient. Of special concern is the fact that subtype E is circulating in our country, the subtype which has higher heterosexual transmission efficiency, and therefore, has higher probabilities for epidemics. Knowledge of the most prevalent subtypes in our country will be helpful in coming up with potential candidate vaccine molecules most suitable for our population, and in determining appropriate therapeutic agents for each patient. These also have implications in the development of antibody-based reagents for laboratory diagnosis.

Key words: AIDS, HIV-genotypes, subtypes, vaccines, HIV-transmission

8. IGM-CAPTURE ELISA OF SERUM SAMPLES FROM FILIPINO DENGUE PATIENTS*

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Dengue infection, an arthropod viral disease, is a major health problem in the Philippines. It is caused by four antigenically distinct dengue virus serotypes namely, D1, D2, D3, and D4. The present study is an attempt to identify the most useful assay antigen to detect dengue patients. Viral antigens for four dengue serotypes were produced in C6/36 *Aedes albopictus* cells. These were used as assay antigens for IgM-capture ELISA to detect IgM antibodies in sera of dengue patients from three hospitals in Metro Manila, Philippines. A total of 376 serum samples came from the National Children's Hospital (NCH), San Lazaro Hospital (SLH), and St. Luke's Medical Center (SLMC), from January to November 1995. Three hundred and three (303) out of 376 serum samples, or 80.58% showed positive IgM ELISA titer against at least one of the four assay antigens. D4 antigen detected antibodies in 62.5% (235/376) respectively. The results indicate that D4 is significantly the best antigen to use in identifying dengue infections in the batch of samples used in this study.

*Best Poster Paper Award in the Health Sciences Division.

Key words: Filipino dengue patients, C/636 *Aedes albopictus* cell lines, IgM-capture ELISA, antigen, arthropod viral disease, antibody, serotype

9. USEFULNESS OF SANDWICH ELISA IN PREDICTING THE DETECTION OF DENGUE VIRUS BY RT-PCR FROM INFECTED CULTURED FLUIDS OF C6/36 *Aedes albopictus* CELLS

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Serum samples of patients from two hospitals in Metro Manila, Philippines, were collected within five days from onset of fever and inoculated into C6/36 cells. Infected cells were cultured for one week and 282 infected culture fluids were harvested and subjected to sandwich ELISA for virus detection. Identification of dengue virus in the same culture fluid samples was done by RT-PCR using universal dengue primers. Optical densities (P) obtained from the fluid samples were compared with that of the culture media (N), which served as negative control. The ratio P/N was correlated with RT-PCR results. In this study, a P/N ratio of at least 4.08 gave a positive RT-PCR. The value of the P/N ratio obtained by sandwich ELISA provides a convenient way of predicting the possibility of isolating the virus from a given serum sample.

Key words: Sandwich ELISA, dengue virus, reverse transcription-polymerase chain reaction, universal dengue primers, C6/36 *Aedes albopictus* cells, optical density, virus detection, infected culture fluid

10. COMPARATIVE NUCLEOTIDE AND AMINO ACID SEQUENCES OF THE NONSTRUCTURAL (NS1) GENE OF DENGUE VIRUS SEROTYPE 3 ISOLATED DURING A 1995 OUTBREAK IN METRO MANILA, PHILIPPINES

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The nonstructural (NS1) gene of dengue virus type 3 isolates from a 1995 outbreak in Metro Manila, Philippines was sequenced and compared to the prototype H 87 strain isolated in 1956. Nucleotide sequence homology shows greater than 94% homologies whereas deduced amino acid sequence indicated greater than 96% homologies, suggesting that the NS1 remains to be a highly conserved gene. Phylogenetic analysis indicates that the 1995 dengue outbreak was due to the re-emergence of an existing circulating strain rather than the introduction of a new variant virus. Of the nucleotide changes observed, 76% did not lead to any amino acid sequence change while 24% resulted in missense mutations. Functional regions such as the 2 N-glycosylation sites, 3 casein kinase II (CK2) phosphorylation sites, 7 protein kinase C (PKC) phosphorylation sites, 7 myristoylation sites, and the tyrosine phosphorylation site were not affected. An amino acid change from glutamic acid (E) to lysine (K) was observed between dengue hemorrhagic fever (DHF) and dengue fever (DF) isolates and a change in the amidation site from glutamine (Q) to proline (P) was observed in one DF isolate. This has brought about changes in the secondary conformation in this region of the protein molecule.

Key words: NS1 gene, dengue virus, dengue hemorrhagic fever, prototype H 87 strain, missense mutation, amino acid sequence, nucleotide sequence, sequence homology

11. ISOLATION, CULTURE, AND CHARACTERIZATION OF PHILIPPINE ISOLATES OF *Helicobacter pylori*

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Helicobacter pylori is a Gram-negative bacterium found to be the cause of approximately 95% of gastric ulcers and 100% of chronic gastritis. In the Philippines, gastritis caused by *H. pylori* is always confirmed histologically or by CLO (Campylobacter Like Organism) test and never by culture. In this study, the Microbiology Section of the Research and Biotechnology Division of St. Luke's Medical Center, using biopsies from the antrum of the duodenum, isolated *H. pylori*. Out of 26 gastric biopsies processed and cultured using Dent's Selective Medium, three strains of *H. pylori* were isolated. One isolate, HP-03 is spiral in form while another HP-06 is coccoid with few bacilli. HP-10 is bacilloid in form and metronidazole-resistant. All isolates give positive urease, oxidase, and catalase tests. Culture of these isolates is under microaerophilic conditions (5% O₂, 10% CO₂, and 85% N₂). This paper is the first report of *H. pylori* isolates in the Philippines

Key words: *Helicobacter pylori*; culture; CLO test; microaerophilic; gastritis; gastric biopsies; Dent's selective medium; spiral, coccoid, and bacilloid forms; metronidazole-resistant; urease positive

12. SURVEY OF BACTERIA ASSOCIATED WITH DRINKING WATER IN THE PROVINCE OF CAVITE

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Samples of drinking water were taken from faucets, deep wells, and springs. Counts of variable bacteria were higher in spring water than in water obtained from deep wells. Water samples taken directly from springs have a higher percentage of fecal contamination compared to water taken from deep wells.

About 257 bacterial isolates were obtained from water samples collected. These bacterial isolates were tentatively classified under the genera *Micrococcus*, *Flavobacterium*, *Sarcina*, and *Bacillus*.

Key words: Variable count, fecal contamination

13. THE ANTIDOTAL EFFECT OF ORALLY ADMINISTERED 50% *Amaranthus spinosus* (URAI) LEAF EXTRACT IN PARALYTIC SHELLFISH POISONING IN MICE

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Paralytic shellfish poisoning (PSP), is caused by the ingestion of mussels infected with the red tide organisms. A search for a safe, available, and inexpensive home remedy for the poisoning is ongoing. Coconut milk is reportedly being used in the coastal towns to counteract the effects of the poison but it has not been very effective. Urai, a weed, commonly found throughout the Philippines was tested for its antidotal effect on paralytic shellfish poisoning in mice. Poisoning was induced in four groups of the experimental animal by forced feeding each mouse with 2 mL crude toxin which was extracted from the contaminated shellfish obtained from sampling stations in Limay, Bataan. Three dose levels, 6, 15, and 30g/kg of the 50% water extract of urai leaves (LD50=60.7 g/kg), were administered to three groups of mice by oral feeding. The control group received distilled water. Treatments were given at 0, 15, 30, and 60 minutes after poison induction. The survival time was recorded and analyzed using survival analysis. Mice treated with 15g/kg of urai extract 15 minutes after poisoning survived longer ($p=0.05$). Using the Cox regression model, the 15g/kg dose was found protective and statistically significant ($p=0.022$). The effective dose was computed as 13.7 g/kg at the 15-minute interval. From the results of the study, urai extract, with a large margin of safety of 4.4 (LD50/ED50=60.7g/kg/13.7 g/kg), can be a safe and effective household remedy for red tide poisoning.

Key words: PSP, red tide, antidote, *urai*, LD50, ED50, survival time

14. HEALTH SOCIAL SCIENCE CONCEPTS AND METHODS IN THE PREVENTION OF HIV/AIDS/STD

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Health social science is a holistic multi- and interdisciplinary approach to health care in its preventive, promotive, and maintenance aspects. Health is not just a biomedical as much as a psychosocial concern. In the case of HIV/AIDS, the disease currently afflicts 22.6 million people all over the world as of 1997. The daily infection rate is 7,000. These numbers are expected to rise from 10 to 15% by the year 2000. This study was undertaken in an effort to help in HIV/AIDS/STD prevention by using health social science concepts and methods. Social scientists in UP Manila closely collaborated with the medical doctors in preventive work among prostituted men and women, seafarers, and their sex partners.

Social science methods and approaches such as environmental and person sensing, Knowledge, Attitudes and Practice (KAP), pre- and post-test assessments, iterative ethnographic interviews, focus group discussions, key informant interviews, seminar workshops and lectures, peer education, and value formation were some of the social science concepts and approaches used in preventive work against HIV/AIDS/STD among prostituted men and women, seafarers, and their sex partners for the past 5 to 6 years now.

Among the results documented were the following: marked improvement of KAP; since intervention work is always preceded by environment and person sensing, iterative ethnographic interviews, the results have been appropriate and culture sensitive intervention measures. Noticeable behavior changes have also been documented, among them, increased use of condoms, better negotiation skills, better locus of control and heightened self-esteem, greater care in choice of partners, and many times, refusal to barfine. Some left their kind of trade altogether. There were also documented cases of ability to share information and to influence peers.

HIV/AIDS for which there is no known cure yet cannot be prevented by biomedical approaches alone. Only a close collaboration between the biomedical doctors and the social scientists, in what we call health social science, will lead to prevention or minimization of the disease through improved and accurate knowledge of the etiology of the disease leading to behavior modifications.

Key words: HIV/AIDS prevention, health social science, prostituted men and women, holistic health, behavior modification, health social science techniques, peer education, environment sensing, person sensing, health intervention, behavior change

