

and accounts for the second leading cause of death among malignant tumors. Surgery is the mainstay of treatment for gastric cancer. As an adjuvant therapy after surgical resection, chemotherapy has received considerable attention in the treatment of gastric cancer. However, development of insensitivity or resistance to chemotherapy occurs frequently in tumor cells.<sup>2</sup> Novel agents are, therefore, needed to sensitize the resistant cancer cells. Akt is a 60kD serine/threonine kinase and functions downstream of phosphatidylinositol 3-kinase (PI-3K). Activation of PI-3K generates phosphatidylinositol 3,4-bisphosphate, which may induce the membrane translocation of Akt coincident with its phosphorylation and activation. Akt is activated in response to insulin and growth factors, and upon activation, it phosphorylates several substrates including glycogen synthetase kinase-3 (GSK-3), Bax, Caspase-9. Activation of Akt kinase activity is inhibited by wortmannin or LY294002, inhibitors of PI-3K.<sup>3</sup> Over activation of Akt has been demonstrated in gastric cancer, and is correlated with clinicopathological parameters and poor outcome.<sup>4</sup> In our study, we determined the basal activities of Akt/PKB in 4 gastric cancer cell lines with different differentiation degrees, and found that Akt/PKB activity was inversely correlated with the degrees of differentiation of cancer cells, which may account for the reason why over activation of Akt is associated with poor outcome in gastric cancers.

Akt/PKB plays an important role in cell survival.<sup>5</sup> In our study, treatment with etoposide within 24 hours resulted in time-dependent inhibition of cell survival rates and induction of cell apoptosis rates in cancer cell lines. However, there was a marked induction of Akt/PKB activity in a time-dependent manner during the course of etoposide treatment. The induction of Akt/PKB activity may confer protection against etoposide-induced death because inhibition of Akt/PKB by wortmannin enhanced the effects of etoposide. Cell survival rates and apoptosis rates were significantly decreased and elevated respectively in SGC-7901, BGC-823, and HGC-27 cell lines after treatment with etoposide + wortmannin compared with etoposide alone. However, we found that the chemotherapy-sensitizing effect of wortmannin was related to the degree of differentiation of cancer cells. There was no significant difference in cell survival rates and apoptosis rates between treatments of etoposide + wortmannin and etoposide alone in MKN-28, a well-differentiated cell line. The MKN-28 had less activity of Akt/PKB than other cell lines, which may explain the lack of chemotherapy-sensitizing effect by wortmannin.

In conclusion, Akt/PKB activity is inversely correlated with the degree of differentiation of gastric cancer cells, and inhibition of Akt/PKB activity

may sensitize moderately- to undifferentiated cancer cells to chemotherapy, which maybe extrapolated to clinical practice in selecting the patients for combined chemotherapy with Akt/PKB inhibitor.

Received 26th December 2006. Accepted 24th March 2007.

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## Random amplified polymorphic DNA typing of nosocomial *Candida albicans* isolates

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The incidence of candidiasis is dramatically on the rise due to the increase in the immuno-suppressed population, especially related to HIV infection, chemotherapy, and organ transplantation.<sup>1</sup> *Candida albicans* is the most frequent pathogenic species of candidiasis, and the mortality rate in candidiasis varies from 38-50%.<sup>2,3</sup> Because of its clinical importance, several typing schemes were developed to assess the strain identity. These typing methods are generally considered too variable to be of any practical value in epidemiological investigations.<sup>4</sup> With the advent of molecular genetics,

several DNA-based typing methods are being used for analyses, such as karyotyping, restriction enzyme analysis, pulse field gel electrophoresis, and multilocus enzyme electrophoresis; however, these methods are laborious and time consuming.<sup>5</sup> The analysis of *Candida albicans* by random amplified polymorphic DNA (RAPD) is rapid, convenient, as well as reliable, and helps better in understanding the epidemiological aspects of candidiasis.<sup>6</sup> In this study, isolates of *Candida albicans* from 2 hospitals, Sir Gangaram Hospital and St. Stephen's Hospital, in Delhi were analyzed by different random primers to establish the suitability of the RAPD typing for *Candida* strains.

Ten nosocomial isolates of *Candida albicans* were recovered from various clinical specimens, such as, bronchoalveolar lavage, blood, urine, pus, and plastic devices, and confirmed by Analytical Profile Index *Candida*. The genomic DNA was extracted according to the method described by Ausubel with minor modifications.<sup>7</sup> After ribonucleotidase treatment, 2 µl of extracted DNA template of 50ng/ml concentration was added into the mixture of 2.5 µl 10X buffer, 0.5 µl dNTPs mix, 0.2 µl Taq DNA polymerase, 1 µl RAPD primer, and 18.8 µl standard water to prepare the assay reaction mixture. Amplification parameters consisted of 45 cycles of denaturation at 94°C for 60 seconds, annealing at 35°C for 90 seconds, and extension at 72°C for 90 seconds. In the first cycle, the denaturation was carried out for 3 minutes, and 10 minutes for the final extension. Amplification reaction was carried out in an assay mixture of 25 µl using 6 different RAPD primers from Operon OPB-11 (5'-GT AG AC CC GT-3'), OPB 14 (5'-TC CG CT CT GG-3'), OPB 15 (5'-GG AG GG TG TT-3'), OPB 18 (5'-CC AC AG CA GT-3'), OPB 19 (5'-AC CC CC GA AG-3'), and OPB 20 (5'-GG AC CC TT AC-3') in Perkin Elmer Gene Amplifier 2400. Agarose gel electrophoresis of the polymerase chain reaction (PCR) product was carried out in 1.4% gel concentration, with 2 µl of DNA samples for 2 hours at 2 V/cm electric circuits. The molecular weight marker used in this study was 1 kb DNA ladder (Lambda DNA EcoRI/Hind III Digest, Sigma). Genomic DNA with A 260/A 280 ratio in between 1.8 to 2.1 was used, and the RAPD patterns generated was analyzed using Diversity Database software version 1.1 incorporated into gel documentation system from PDI, USA. Cluster analysis of the 10 *Candida albicans* was carried out using the unweighted-pair group method with arithmetic average, and the phylogenetic tree was constructed for all samples in each database.

The application of RAPD technology for strain delineation of *Candida albicans* has proven to be a valuable tool for clinico-epidemiological studies.<sup>6,8,9</sup>

Gyanchandani et al,<sup>10</sup> reported that RAPD of 19 *Candida albicans* showed non-identical profile when tested with 21 primers. The present study with oligonucleotide primers OPB-18, OPB-19, and OPB-20 did not produce any RAPD profile with the strains. However, the RAPD patterns of *Candida albicans* exhibiting intraspecific polymorphisms were obtained with OPB-11, OPB-14, and OPB-15 primers. However, unlike other primers, OPB-14 exhibited RAPD profile with all the 10 isolates. The OPB-15 showed profiles against 7 isolates only, while OPB-11 showed profiles to 6 isolates only. The range of molecular weights of DNA obtained by using OPB-11 was between 1.91 kb and 0.85 kb, while with OPB-14 it was 728 kb to 278 kb, and with OPB-15 it was between 1800 bp and 250 bp.

In the last few decades, there has been increasing reports of *Candida* infections in India.<sup>11</sup> In our study we found that 3 primers OPB-18, OPB-19, and OPB-20 were ineffective in producing profiles, but OPB-11 primer when used to generate RAPD profile, 3 clusters were clearly represented. One band of 1.4 kb was shared by all the strains when OPB-11 was used. Non-randomization was found in isolates from blood, urine, and pus of 3 unrelated patients; and between isolates of plastic device and skin of 2 unrelated patients in the same hospital. However, when OPB-15 was used to amplify the same isolates, it exhibited randomness in these strains. The OPB-15 primer therefore has better discriminatory power, with higher detection of strain variability, and is more efficient in reflecting intra-specific variation.

Received 16th December 2006. Accepted 17th March 2007.

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### Assessment of nutritional status and lifestyle pattern among Saudi Arabian school children

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Though the prevalence of obesity varies greatly in different populations of the world, various studies have shown a significant increase in the prevalence of overweight and obesity among children.<sup>1</sup> The epidemiologic transition in developing countries as a result of urbanization, migration, sedentary life styles, and changing dietary habits are recognized as some of the factors in the development of overweight and obesity in childhood.<sup>2</sup> As health-related behaviors and beliefs established during childhood have been linked to patterns of behavior in adulthood, therefore, the schools can play an important role in the health promotion of children and represent the best opportunity to acquire a healthy lifestyle not only through knowledge, but also attitude and behavior.<sup>3</sup> The purpose of the study was to describe the nutritional status among school children in healthy cities in the Kingdom of Saudi Arabia (KSA), using the 85th and 95th percentiles of body mass index (BMI) for age and gender as cut-off points.

This was a cross-sectional study carried out in the provinces of KSA from December 2004 to January 2006. Using a stratified random sampling technique, the sample subjects were chosen from 3 selected provinces, namely, Western, Northern, and Eastern of the 5 provinces. The ongoing 'healthy cities' program in these provinces made it convenient to carry out a study in these regions and give recommendation for its expansion. From each selected province, 3 cities each from Western and Northern provinces, and 2 cities from the Eastern province were selected randomly from a list of cities. Four schools (2 boys and 2 girls intermediate schools) from each city were randomly selected, except for one city in the Eastern province, where the study was conducted in 2 girl schools only due to certain administrative reasons. Two classes from each school were randomly selected and thereafter, each class was considered as a cluster in which all students (average 27 students per class) were enrolled in the study. Thus, a total of 30 schools were surveyed, wherein a total of 60 classes, namely, 28 boys and 32 girls (grades 1-3) were included in the sample. Approval was taken from the Ministry of Education and the Principal of the school to carry out the survey in the chosen schools and classes. The health survey questionnaire used in this study was modified and adapted from the Global School Health Survey (GSHS), which basically assesses the overall health of school students. This instrument included questions on food consumption, daytime physical inactivity, and smoking habits and, was subsequently translated to Arabic vernacular by the investigators. Furthermore, the questionnaire was pilot tested separately on boys and girls and, accordingly adapted for implementation in KSA. In each randomly chosen class, all students were briefed on the survey health questionnaire, and subsequently administered and supervised while filling the questionnaire by the investigators. Their basic demographic details were initially recorded. Subsequently, anthropometric measurement, namely, one trained male and female nurse carried out height and weight of participating school children for boys and girls. For each child, BMI was estimated by age and sex, and compared to the BMI latest World Health Organization/National Center for Health Statistics (WHO/NCHS) 2000 reference.<sup>3</sup> The cutoff percentiles used to classify the nutritional status of the children were underweight, BMI  $p < 10$ ; normal, BMI  $p \geq 10$  to  $p < 85$ ; overweight,  $p \geq 85$  to  $p < 95$ ; and obese, BMI  $p \geq 95$ .

The study group included a total of 1454 children in the 3 provinces, in the age range from 12-19 years, with the mean and median age as 15 years. Out of all participating children, 45.2% were boys, and 54.8%