

DNA repair genes to improve efficiency for gene targeting involved in designed single base change by short oligonucleotides for functional genomics in plants.

**F818** Caspase-3-mediated cleavage of the NF- $\kappa$ B subunit p65 at the NH2-terminus potentiates naphtoquinone analog-induced apoptosis

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The transcription factor NF- $\kappa$ B plays a crucial role in immune and inflammatory response, and protects cells from apoptosis. In this report, we investigate whether the NF- $\kappa$ B signaling pathway is blocked during apoptosis induced by 2,3-dichloro-5,8-dihydroxy-1,4-naphtoquinone (NA), an analog of naphtoquinone. It is observed that NA triggers apoptotic cell death in HeLa cells and destroys resistance to apoptosis caused by TNF-. Data presented in this study establish that p65/RelA, a subunit of NF- $\kappa$ B, is cleaved at Asp97 by caspase-3 during apoptosis. Caspase-3-cleaved p65 loses transcriptional activity and potentiates NA-induced apoptosis, in contrast to an uncleavable mutant of p65, which protects the cell from apoptosis. Caspase-3, which is responsible for the cleavage of p65, is activated via the cytochrome c/caspase-9 signaling pathway rather than Fas/caspase-8 pathway during NA-induced apoptosis. Our results suggest that NA induces apoptosis by the negative regulation of cell survival through caspase-3-mediated cleavage of p65

**F819** Detection of Trinucleotide Repeat Disease Using Micro-Capillary Electrophoresis Chip

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Several major human single gene disorders are attributed to the expansions of highly unstable trinucleotide repeat (TR) sequences. TR numbers are closely related to not only the onset age and severity but also diagnosis and prognosis of disease. Therefore, it is very effective and essential to estimate the accurate TR size for screening or confirmation of trinucleotide repeat disease (TRD). DNA testing such as southern blotting or silver staining has been commonly used in the diagnosis of TRD. However, such methods require laborious steps for the diagnosis and lack accuracy in detection of carriers and estimation of TR numbers. Therefore, we have developed a new method for the detection of TRD using micro-capillary electrophoresis chip (micro-CE chip). Amplified target sequences by polymerase chain reaction using specific primers designed for TR were separated in micro-CE chip and determined the size of TR. We evaluated the method by analysis of samples from normal subjects, 9 HD patients, 13 SBMA patients, 2 DRPLA patients, 6 FX carriers and 3 DM carriers. Southern blotting method was simultaneously performed to confirm our results. The data obtained from the micro-CE chip and southern blotting were highly comparable. The estimated TR number was also confirmed by sequencing. This study suggests that micro-CE chip is very useful for the detection of TRD and the determination of TR numbers. This new method could be very helpful in early diagnosis, carrier testing, and proper genetic counseling.

**F820** The distribution of Actinobacillus actinomycetemcomitans, Hemophilus aphrophilus and Hemophilus paraphrophilus in subgingival plaque and saliva from Korean periodontitis patients using PCR

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The closely related species *Actinobacillus actinomycetemcomitans*, *Hemophilus aphrophilus* and *Hemophilus paraphrophilus* are common findings in oral microbiota. The aims of this study were to compare the distribution of three species in healthy subjects and periodontitis patients using PCR for 16s rRNA gene. The DNA was extracted from the subgingival plaque and saliva in 112 subjects for restriction enzyme analysis with *HinfI* and *HhaI*. By the compare of restriction enzyme pattern for each pathogen, we found that *Actinobacillus actinomycetemcomitans* was the major distributor in healthy and periodontitis patients. In general, periodontal pathogens were distributed more frequent in subgingival plaque than in saliva. We confirmed that the PCR method for 16s rRNA gene was important for screening and monitoring of periodontal disease.

#### **F821** Microsatellite Polymorphism of D12S391 and D12S67 Genes in the Korean Population

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The highly polymorphic microsatellite loci D12S391 and D12S67 were investigated in healthy Korean subjects. Using the polymerase chain reaction and polyacrylamide gel electrophoresis, we examined the allele frequency and genotype distribution. We observed 13 alleles were identified for D12S391 (n=359) and 11 alleles for D12S67 (n=279) in this study. Heterozygosities were 0.811 at D12S391 and 0.733 at D12S67, respectively. The p values of two loci were calculated, 0.637 and 0.149, by Exact test. Therefore no deviation from Hardy-Weinberg equilibrium was observed. The most frequent allele at D12S391 loci was allele 18 (0.298) followed by allele 19 (0.245), allele 20 (0.152), and minor alleles

(0.335). The size of PCR fragments at D12S67 locus ranged from 233 bp to 273 bp. The genotype distribution and allele frequency of D12S391 and D12S67 in the Korean subjects were similar to those of the Japanese population. This study was demonstrated that the D12S391 and D12S67 was a useful tool for forensic identification and parentage testing.

#### **F822** Developmental and Genetic Characterization of Drosophila pleiohomeotic genes

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Polycomb group (PcG) gene products function in maintaining the expression pattern of the homeotic genes. Pleiohomeotic (Pho) protein is a unique protein containing a DNA binding motif among PcG proteins. *pho* mutation produces a relatively weak phenotypes compared to a zygotic mutation of *Polycomb* and a maternal mutation of *extra sex comb*. PcG proteins form a few complexes with a different combination. Pho was recently found to be a member of a complex. However, its interaction with other PcG genes was not investigated genetically. Pho shows pleiotropic effects including the roles in the formation of cuticle, head, nervous system, the regulation of the homeotic genes and the segmentation. *pho* was expressed uniformly at early embryonic stage and its expression was concentrated in the central nervous system at later embryonic stage. *pho* also enhanced the mutant phenotypes of the *Polycomblike* in embryonic and adult cuticle, but the expression of the homeotic genes was little affected. This results indicate that *pho* has various roles during Drosophila development and works with other PcG proteins. We will also present the effect of *pho* ectopically expressed by GAL4/UAS system.