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A factor V mutation G1691A (FV Leiden) was found to destroy the vital cleavage site R506Q of the factor V, which is necessary for factor V deactivation, and thus causing thrombophilia. Mutation in prothrombin (factor II, G20210A) was discovered to be often associated with mutation in factor V (G1691A), thereby indicating an additional risk for thrombosis elevated to factor II levels. This experiment investigates the prevalence of these two mutations among two hundred nineteen unrelated Korean subjects using multiplex PCR-RFLP. Data revealed two heterozygous subjects for the factor V Leiden without prothrombin G20210A mutation. Both variations yield significant differences in relation to ethnic origins: the FV Leiden occurs at a frequency rate of 2.3% among Koreans, 11.0% among Swedish, 6.1% among Swiss people, and 19.0% among Jordanian people. Prothrombin (G20210A) mutation occurs at 0% frequency rate among Koreans, 1.2% among English, and 1.1% among Swiss people.

F806

Identification of the Mutation in the Patient with Dihydrolipoamide Dehydrogenase (E3) Deficiency

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The mammalian pyruvate dehydrogenase complex (PDC) plays a pivotal role in the decarboxylation of pyruvate derived from glucose and amino acids to acetyl-CoA, which is further utilized for energy production or for biosynthetic processes. This enzyme complex contains multiple copies of components including pyruvate dehydrogenase (E1), dihydrolipoamide acetyltransferase (E2), dihydrolipoamide dehydrogenase (E3), E1-kinase, phospho-E1 phosphatase, and E3-binding protein. Mammalian E3 is a common flavoprotein component of a-keto acid dehydrogenase complexes and catalyzes the reoxidation of dihydrolipoyl group; two cofactors, NAD and FAD, are utilized for the reoxidation. E3 deficiency leads to lactic acidosis, increased concentrations of branched-chain amino acids in the plasma and increased urinary excretion of a-keto acids. E3 deficiency also causes neurological degeneration due to the sensitivity of the central nervous system to defects in oxidative metabolism. In this study, E3 mutant cDNA from a patient showing PDC deficiency was amplified by RT-PCR and subcloned into pBlusscript SK- for further analysis. The clone was sequenced for the identification of mutation(s). Two substitutions were found. One was a single point mutation of the interface domain (CCT →CCC) which encodes Proline, but there was no change in amino acid sequence. The other was missense mutation due to substitution of thymine for guanine (GGT→ TGT), causing an Gly→Cys substitution at amino acid 229 of the mature protein. This substitution may interfere with the proper folding of E3 dimer by forming disulfide bridge with the other cystein(s) in NAD-binding domain. This mutation, therefore, causing diminished production of E3 protein and a possible structural change in the E3 dimer, probably leads to loss of activity.

F807

Construction of ESTs and Identification of a Stress-induced Gene in Chinese Cabbage

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Expressed sequence tags (ESTs) are short, single pass cDNA sequences generated from randomly selected library clones. The ESTs are widely used to clone genes and/or elucidate their structures and/or functions. Chinese cabbage (*Brassica* spp.) is economically important crop in orient. Its genome size is relatively small ($7.7X10^8$), so that it has been served as a favorite model system in studying plant genome. In this study, 30 ESTs were generated from the chinese cabbage (*Brassica rapa* L ssp. *pekinensis*) as follows. Poly A+ RNAs were isolated from 10-day-old seedlings germinated in dark. Randomly selected cDNA clones were sequenced by the Sanger and/or by using ALFexpress™ DNA Sequencer (Phamacia Biotech, Sweden). A number of ESTs showed similarity to the protein coding sequences in Genbank and EMBL databases. Four clones had high levels of similarities in the reported amino acid compositions. DII#215 clone showed very high level of homology to a gene for homeodomain-leucine zipper protein in *Arabidopsis thaliana*. It has been repoted that homeodomain-leucine zipper protein has a role of responsibility to stress condition such as dehydration and higher salt. Sequence of a homeodomain-leucine zipper in Chinese cabbage has been determined and full clone of the gene has been isolated by screening cDNA library from Chinese cabbage.

F808

Genetic Analysis of *Prunus yedoensis*, Native and Cultivar, Based on Nucleotide Sequences of *rbcL* and *psbA* in Chloroplast DNA

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Expressed sequence tags (ESTs) are short, single pass cDNA sequences generated from randomly selected library clones. The ESTs are widely used to clone genes and/or elucidate their structures and/or functions. Chinese cabbage (*Brassica* spp.) is economically important crop in orient. Its genome size is relatively small ($7.7X10^8$), so that it has been served as a favorite model system in studying plant genome. In this study, 30 ESTs were generated from the chinese cabbage (*Brassica rapa* L ssp. *pekinensis*) as follows. Poly A+ RNAs were isolated from 10-day-old seedlings It has long been disputed whether *Prunus yedoensis* was originated from Mt. Halla in Cheju or Cheju originatied taxon is the same as the cultivated one from Japan. By comparing the base sequences of *rbcL* and *psbA* in chloroplast DNA (cpDNA), genetic analysis was assesed between *P. yedoensis*-Native and Cultivar. The cpDNA sequences were amplified from the total DNA using polymerase chain reaction (PCR). The oligonucleotide primers used to amplify the *rbcL* and *psbA* coding regions by PCR were designed by referring to the sequences of tobacco cpDNA. In the comparing of the nucleotide sequences, base substitutions in twenty sites (8 transition and 12 transversion) were found from 1398 base pairs (bp) of *rbcL* and twenty sites of base substitution (6 transition and 14 transversion) were observed from 1062 bp of *psbA*. The value of amino acid sequences divergence between two taxa was 3.66% (17/465) in *rbc* and 3.97% (14/353) in *psbA* coding region, respectively. These results being same as those of the previous studies on the basis of random amplified polymorphic DNA and internal transcribed spacer region, we can suggest that *P. yedoensis*-Native should be distinguished from *P. yedoensis*-Cultivar, and at the same time, the scientific name of *P. yedoensis*-Native should be changed.

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