

JMD CME Program in Molecular Diagnostics 2007

Association for Molecular Pathology *and the*
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<http://jmd.amjpathol.org>
www.asip.org/CME/jmdCME.htm

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CME Questions # 21-30

(See February and April 2007 Examination Sheets for Questions #1-20)

21. The CpG island methylator phenotype (CIMP) refers to the notion that some tumors exhibit widespread methylation of CpG islands, leading to epigenetic inactivation of tumor suppressor genes by promoter methylation. Based on the referenced article and its related Commentary, select the ONE statement that is NOT true: [See J Mol Diagn 2007 9:281-283 and J Mol Diagn 2007 9:305-314]

- Transcriptional inactivation by cytosine methylation at promoter CpG islands of tumor suppressor genes is an important mechanism in human carcinogenesis.
- CIMP-positive colorectal tumors have a distinct clinicopathological and molecular profile, including proximal tumor location, female sex, mucinous and poor tumor differentiation, microsatellite instability (MSI), and high mutation rates of both *BRAF* and *TP53*.
- The instability of CIMP-positive colorectal tumors is due to promoter methylation of *hMLH1*.
- MSI in tumors associated with hereditary nonpolyposis colorectal cancer (HNPCC) is due to a germline mutation in one of the mismatch repair genes.
- It is imprudent at the present time to regard all microsatellite unstable, CIMP-negative tumors as necessarily HNPCC associated.

22. The choice of markers for CIMP has been controversial. Based on the referenced article and its related Commentary, select the ONE statement that is NOT true: [See J Mol Diagn 2007 9:281-283 and J Mol Diagn 2007 9:305-314]

- Eight loci were selected from 195 loci throughout the human genome because methylation in these loci was a predictor for CIMP-high, and PCR showed excellent amplification efficiency in methylation-positive samples.
- KRAS mutations have been shown to be more common in CIMP-high colorectal tumors than in CIMP-low tumors.
- The best individual marker to predict the CIMP status was *RUNX3*.
- A panel of four markers including at least *RUNX3*, *CACNA1G*, *IGF2*, and *MLH1* can serve as a sensitive and specific marker panel for CIMP-high.
- In addition to quantitative DNA methylation analysis (MethyLight), other techniques such as methylation-specific PCR and pyrosequencing have been used for CIMP determination.

23. Disorders of large granular lymphocytes (LGLs) encompass a broad spectrum ranging from polyclonal reactive proliferation to aggressive leukemia. Based on the referenced article that describes a clonality assay for natural killer (NK) cell proliferations, select the ONE statement that is NOT true: [See J Mol Diagn 2007 9:337-344]

- NK cells normally account for more than 50% of mononuclear cells in peripheral blood.
- NK cells constitute the majority of the morphologically recognizable LGLs in the blood.
- Only 15% of LGL proliferations are derived from NK cells, whereas 85% are of T-cell origin.
- Clonality in LGL proliferations derived from T cells can be detected via use of T-cell receptor gene rearrangement studies.
- In LGL disorders of NK cell lineage, detection of clonality is problematic because of the absence of specific molecular markers.

24. X-chromosome inactivation assays allow the evaluation of clonality in female patients. Based on the referenced article, select the ONE statement that is NOT true: [See J Mol Diagn 2007 9:337-344]

- a. Loci that have been used to assess skewed X-chromosome inactivation include the *PGK* gene, the *HPRT* gene, the *DXS255* locus, and the *HUMARA* gene.
- b. The first exon of the *HUMARA* gene contains a hypervariable CAG short tandem repeat (of 9 to 36 copies) that is polymorphic in 90% of females.
- c. Several cleavage sites for methylation-sensitive restriction enzymes are in close proximity of the CAG repeat of the *HUMARA* gene.
- d. A limitation of the *HUMARA* assay is that it suffers from a lack of standardization.
- e. The *HUMARA* assay was sufficiently robust and specific that it was not necessary to use pure cell populations of NK and control cells.

25. Extranodal marginal zone B-cell lymphoma of mucosa-associated lymphoid tissue (MALT) is the most common primary lymphoma at extranodal sites. Based on the referenced article, select the ONE statement that is NOT true: [See J Mol Diagn 2007 9:351-357]

- a. The most common organs involved in MALT are the stomach, skin, and orbit.
- b. It is sometimes difficult to differentiate between cases of gastric MALT lymphoma and chronic gastritis.
- c. Polymerase chain reaction (PCR) of the immunoglobulin heavy chain (IgH) gene rearrangement at the complementary-determining region III can be performed on formalin-fixed, paraffin-embedded (FFPE) tissue from endoscopic biopsies with high specificity and sensitivity.
- d. Flow cytometry immunophenotypic studies for light chain restriction are not applicable to FFPE tissues.
- e. The diagnosis of gastric MALT lymphoma based on PCR analysis is made when at least 3 sharp bands are seen on size fractionation by gel electrophoresis.

26. The development of fluorescence-based measurements during PCR reactions or at post-PCR using melting curve analysis has made real-time analysis possible. Based on the referenced article describing peak area analysis of the melting curve in the LightCycler system to detect IgH rearrangements in gastric lymphoid infiltrates, select the ONE statement that is NOT true: [See J Mol Diagn 2007 9:351-357]

- a. A training set of 10 gastric MALT lymphoma surgical specimens and 10 chronic gastritis endoscopic biopsies was used to define a discriminate function of the one-peak area analysis.
- b. The analytical approach was based on the use of seminested IgH gene rearrangement amplification.
- c. A potential disadvantage of seminested IgH gene rearrangement amplification is that it requires a manual transfer with the risk of PCR contamination by product carryover.
- d. In the statistical analysis, the receiver operating characteristic (ROC) curve was found to be preferable to the likelihood ratio (LR) in converting continuous data to binary data in diagnostic tests.
- e. Analytical detection of IgH gene rearrangement in gastric lymphoid infiltrates by one-peak area analysis correctly distinguished gastric MALT lymphomas from chronic gastritis.

27. Glaucoma is a heterogeneous group of optic neuropathies. If left untreated, glaucoma leads to blindness. Based on the referenced article that describes a mutation profile of Iranian primary congenital glaucoma patients (PCG), select the ONE statement that is NOT true: [See J Mol Diagn 2007 9:382-393]

- a. PCG is a severe subgroup of the disease characterized by an anatomical defect of trabeculodysgenesis and an age of onset in the neonatal or infantile period, generally before the age of 3 years.
- b. PCG occurs in both sporadic and familial patterns.
- c. The incidence of PCG is geographically and ethnically variable, estimated at 1:10,000 in Western countries and higher in inbred populations.
- d. Linkage analysis in multiply affected families has identified three PCG loci and one gene located on the Y chromosome, *CYP1B1*, a member of the cytochrome P450 superfamily.
- e. The *CYP1B1* gene has three exons and is expressed in the posterior segment of the eye.

28. The proportion of PCG patients whose disease is attributable to CYP1B1 varies among populations. Based on the referenced article, select the ONE statement that is NOT true: [See J Mol Diagn 2007 9: 382-393]

- a. *CYP1B1* plays a greater role in the disease status of African PCG patients as compared with Europeans.
- b. The worldwide profile of variations thus far reported is heterogeneous and includes ~70 alterations.
- c. In Slovak Roma patients, the E387K allele constitutes all *CYP1B1* mutated alleles.
- d. The V364M mutation has been found in all PCG patients of Indonesian descent.
- e. Differences in mutated alleles among populations are likely attributable to variations in frequencies of consanguineous marriages and gene pools among the different populations.

29. Iran, having been a major gateway in human history, has encountered many populations and is expected to have a rich genetic legacy. Based on the referenced article that analyzes the *CYP1B1* mutation profile of Iranian PCG patients, select the ONE statement that is NOT true: [See J Mol Diagn 2007 9:382-393]

- a. Of the 29 sequence variations identified in the *CYP1B1* gene of Iranian PCG patients and controls, 10 have been previously reported in the literature to be disease-associated mutations.
- b. Criteria used to determine if novel variations found in the Iranian population were mutations associated with PCG included frameshift mutations, introduction of stop codons during translation, absence in control individuals, presence in more than one unrelated patient, occurrence at the same site as a previously reported mutation, the nature of the amino acid change, and the degree of conservation during evolution.
- c. Putative disease-causing mutations were identified in more than half of the chromosomes investigated, indicating a *CYP1B1* mutation allele frequency of at least 66% among the Iranian PCG patients.
- d. The four most common mutations detected among the Iranian PCG patients constituted 51% of the Iranian *CYP1B1* alleles studied and more than three quarters of the mutated *CYP1B1* alleles observed.
- e. In Iranians, *CYP1B1* was more often found to be causative of PCG disease among sporadic cases as compared with familial cases.

30. The potential presence of maternal cell contamination in chorionic villus or amniotic fluid samples poses a serious preanalytical risk for prenatal misdiagnosis. Based on the referenced article, select the ONE statement that is NOT true: [See J Mol Diagn 2007 9:394-400]

- a. Maternal cell contamination in chorionic villus or amniotic fluid samples is less common with clinicians who perform at least 50 amniocenteses annually.
- b. The risk of maternal cell contamination is reduced if the first few ml of an amniotic fluid sample are discarded together with the first syringe, and the actual sample is obtained after attachment of a second syringe.
- c. Cultured amniotic fluid cells have a higher chance of maternal cell contamination than uncultured (direct) cells.
- d. Chorionic villus sample cultures present the highest level of potential maternal cell contamination.
- e. Although maternal cell contamination testing of fetal samples is recommended in guidelines by the American College of Medical Genetics, only 60% of surveyed laboratories performed it without exception.

Disclosures: No authors of referenced articles disclosed any potential conflicts of interest.

SEE EXAMINATION ANSWER SHEET – NEXT PAGE

TO REGISTER:

<http://www.asip.org/CME/jmdCME07.htm> or <http://www.amp.org/CME/jmdCME2007.htm>

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CME Questions # 21-30

Examination Answer Sheet #3, Questions #21-30					
Answer	a	b	c	d	e
Question #21	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Question #22	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Question #23	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Question #24	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Question #25	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Question #26	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Question #27	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Question #28	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Question #29	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Question #30	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
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3. Enter your name and email address.
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5. Keep a copy of your Examination Answer Sheet for your records to compare with correct answers.
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