Microsatellite Instability, Clinical Implications, and New Methodologies

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Boland et al. (1) noted that the discovery of microsatellite instability (MSI) (2) in colorectal cancers and its linkage to hereditary nonpolyposis colon cancer (HNPCC) in 1993, “. . . opened new chapters in tumor biology and in the clinical management of patients with heightened cancer susceptibility” (1). The scope of this discovery has now been expanded as a unique form of genomic instability embracing a litany of cancer types, some related to HNPCC and some not. The National Cancer Institute workshop from which the publication by Boland et al. (1) evolved endorsed a new terminology for MSI. Tumors were classified as high-frequency MSI when two or more of a panel of five microsatellite markers demonstrated instability by insertion/deletion mutations and as low-frequency MSI when only one of the five markers showed instability. Importantly, those high-frequency MSI tumors, about 15% of all colorectal cancers, showed a unique clinical and pathologic phenotype in contrast to low-frequency MSI and microsatellite stable tumors, which appeared to be phenotypically similar. Boland et al. (1) stressed the importance of identifying colorectal cancer patients with high-frequency MSI tumors because some harbor germline mutations in mismatch repair (MMR) genes that will have a direct impact on the management of the patient and family members. In addition, the biology of high-frequency MSI tumors will differ from that of low-frequency MSI or microsatellite stable tumors.

Implications of the article by González-García et al. (3) in this issue of the Journal abound, and several clinical correlations of MSI are discussed in this editorial. Cancer risk factors, aided by molecular genetic findings, including MSI, provide important indications for germline MMR mutation testing. When a mutation is identified, cancer predisposition exists, posing a problem for physician and patient alike. This is particularly difficult when cancer-control measures are limited, as for pancreatic carcinoma. Such molecular clues may prove to be exceedingly more rewarding when dealing with the risk for colorectal cancer and the major lesion in HNPCC, given the opportunity for early initiation of colonoscopy (4). The clinical implications of the relationship between MSI and MMR mutations are varied and complex; thus, a better understanding of this relationship and rational guidelines for standardizing testing practices are needed.

The high-frequency MSI phenotype is associated with a predominance of proximal colorectal cancer and histopathologic findings consonant with those observed in HNPCC, namely, an increased frequency of mucinous and signet-cell carcinomas and also an increase in diploid tumors detected by flow cytometry (4). These studies include investigations of sporadic cases (5,6), gastric cancers (7), and transitional-cell carcinomas of the bladder (8). Sidransky (9) has indicated that microsatellite analysis is important in detecting clonal cell populations that may facilitate early cancer diagnoses. Gleeson et al. (10) identified novel alterations in microsatellite alleles in three patients with short-segment Barrett’s esophagus and concluded that the presence of these genetically altered cell populations may be a tool for early detection of neoplastic changes in Barrett’s esophagus. Furthermore, in HNPCC patients a colonic adenoma may evolve into carcinoma in only 2–3 years, whereas in non-HNPCC patients it takes 8–10 years for cancer to occur (11). Hence, there are further implications for identification of high-frequency MSI patients, since they may show accelerated carcinogenesis and benefit from more frequent colonoscopic screening.

Despite the observations implicating accelerated carcinogenesis, survival appears to be better in patients with MSI tumors. We studied 274 case patients (98 HNPCC families) and 820 consecutive control patients with colorectal cancers (12). HNPCC case patients were at a lower stage at diagnosis than control patients, which may be due to rarer distant metastases at the time of diagnosis. The estimated death rate in HNPCC case patients, adjusted for stage and age differences, was at most two thirds of that in the control patients. We provide two hypotheses: 1) Mutator genes (MLH1 and MSH2) cause genomic instability, leading to an enormous burden of microsatellite disturbance, sending cells to apoptosis; or 2) an immune response (namely, peritumoral lymphocytic infiltration, Crohn’s-like reaction, and tumor-infiltrating lymphocytes) results in immunologic intolerance for the tumor and a subsequent survival advantage. A survival advantage also occurs in patients with sporadic disease (lacking MMR mutations) with the high-frequency MSI phenotype. The data obtained by González-García et al. (3) also support this survival advantage.

Our studies of patients with colorectal cancer indicate a highly statistically significant association of tumor-infiltrating lymphocytes in HNPCC compared with sporadic colorectal cancer. In addition, quantitative assessment of tumor-infiltrating lymphocytes in colorectal cancer appears to be predictive of MSI (Smyrk TC, Watson P, Kaul K, Lynch HT: unpublished data). Thus, observation of tumor-infiltrating lymphocytes may be useful in determining which patients would benefit from further specific testing for MSI. Again, such observations should be useful in the development of MSI-testing algorithms.
Another potential application of MSI testing is in the selection of appropriate chemotherapeutic agents. Carethers et al. (13) investigated the effect of 5-fluorouracil on MMR-deficient colorectal cancer cell lines and found a growth advantage indicative of tolerance to 5-fluorouracil by the MMR-deficient cell lines. Thus, it is conceivable that the MSI status could aid in selection of appropriate chemotherapeutic agents. This hypothesis merits further testing in a well-controlled clinical trial.

However, the relationship between MSI and MMR mutations is not yet fully understood. For example, Wu et al. (14) divided 36 patients with suspected HNPCC into equally sized low-frequency MSI and high-frequency MSI groups. They found that four presumably causative MSH6 mutations (22%) were identified in the low-frequency MSI tumor group. They concluded that low-frequency MSI phenotype cannot be considered an exclusion criterion for mutation testing of MMR genes in general. MSH6 families show other differences from “classical” HNPCC families in that there is often a paucity of colorectal cancer but an excess of gynecologic cancer. Hence, MSI and the infrequent occurrence of colorectal cancer in such MSH6 families may be problematic in their diagnostic assessment. The data reported by González-García et al. (3) supported a two-population model (microsatellite stability and high-frequency MSI); however, since only patients with colorectal cancer were included in the study, an underrepresentation of MSH6-mutated tumors could be expected. It would be interesting to repeat this study on a cohort of patients with endometrial cancer to determine their high- or low-frequency MSI status.

Increasingly, research and clinical molecular pathology laboratories will need to assess MSI in colorectal cancer as well as in other tumor types, making standardization, reliability, and quality control of MSI testing more critical (15). At present, molecular testing is often time consuming and work intensive. Increased standardization of MSI testing is required during the transition to the clinical laboratory. More studies addressing reliability and quality control of MSI testing also are needed (13). Algorithms must be developed for MSI testing that take into account the biologic relationship of the disease and the mutations implicated. González-García et al. have taken a novel approach to the issue of MSI in colorectal cancer. Their work not only provides insight into a rational and cost-effective testing process for MSI but also provides statistical data to underscore a bimodal model, in which tumors are either MSI negative or MSI positive. More such work is needed in the future.

**REFERENCES**


