

17.2% (N = 119), which is higher compared to the 8.29% (N = 208) of bladder cancer from other areas with statistical significance.<sup>31</sup> Clinically, endemic diseases or symptoms were the frequently associated manifestations with urinary cancers in the BFD endemic area. Another analytical study of 74 patients with urinary cancer from the BFD endemic area between 1988 and 1990 showed the prevalence of Bowen's disease, skin cancer, and lung cancer being higher in this group, and 1 patient had simultaneous black foot disease.<sup>32</sup>

### Basic and Molecular Study of BFD Endemic Urinary Cancer

DNA analysis by means of flow cytometry was conducted in a study of 155 patients with primary transitional cell carcinoma of the renal pelvis and ureter, among which, 46 patients came from the BFD endemic area.<sup>33</sup> A retrospective nuclear DNA ploidy analysis by flow cytometry comprised 41 formaldehyde solution-fixed, paraffin-embedded tissue specimens. The preparation of nuclear suspensions from paraffin-embedded tissue blocks and staining was modified by means of the techniques of Hedley and Vindelov. One of the results showed that 79% of the nondiploid patterns of the transitional cell carcinoma were present in patients from the BFD endemic area, whereas only 22% appeared in patients living in other areas. The nondiploid pattern of the transitional cell carcinoma also proved to be more related to the tendency of clinical tumor progression. The conclusion was that patients with renal pelvic cancer and ureteral cancer from the BFD endemic area had a poorer outcome in comparison to those from other areas in Taiwan.

Close examination of the molecular changes in the p53 tumor suppressor gene could contribute to an understanding of the nature of the carcinogenic damage. A study was conducted to determine the pattern of p53 mutation in bladder cancer from the endemic area of BFD cases to compare these results with previously reported mutations in bladder neoplasm.<sup>34-36</sup> Bladder tumors from 12 patients (9 males and 3 females) and 1 tumor of the renal pelvis from a male patient were analyzed. DNA was isolated from archival paraffin-embedded specimens by microdissecting tumor tissue from a single 8-10  $\mu$ m hematoxylin- and eosin-stained

section; it was analyzed by PCR-SSCP (polymerase chain reaction-based single strand conformation polymorphism) followed by direct sequencing. Eight cases (62%) showed mutations, and 9 of the 10 point mutations observed were transitions. The type and position of the mutations were not significantly different when compared with the spectra of p53 mutations previously reported for transitional cell carcinomas (TCCs). However, two of the mutations were CGCCAC base changes at codon 175, a mutational hotspot for many tumor types but previously unreported in TCCs except in cases associated with inflammatory agents. Three of the tumors examined were found to contain double mutations, a relatively rare mutagenic event in human cancers. These results suggest that the agents responsible for the high risk of bladder cancer in the black foot disease region might operate through an inflammation-based mechanism which increases the amount of DNA damage per mutagenic event.<sup>37</sup>

### Future Prospectives in the Study of BFD Urinary Cancer

The high incidence of urinary cancer in the BFD endemic area in Taiwan was one of the new, important resources found in recent years for further investigation of the causes and mechanisms concerning urinary carcinogenesis and other endemic diseases in that area. Since the etiology of BFD, urinary cancer, skin cancer, and many other endemic diseases had been suggested to be associated with drinking water derived in the endemic area from artesian wells with a high concentration of arsenic or other organic compounds such as humic acid, the use of artesian well water as drinking water soon was prohibited by the Taiwanese government. In some parts of these endemic areas, a tap-water supply was provided by the government in 1956, and all areas were almost completely supplied between 1966 and 1975.<sup>38</sup> The withdrawal of arsenic exposure proved to be effectively in decreasing the incidence of skin cancer and internal cancers including urinary cancers according to a recent study. The results also showed significantly declining trends for mortality rate ratios of all malignant tumors with 1971-1973 as the standard and with prominence through the last interval studied (1992-1994). A decrease of mortality rate ratios from malign-