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Balkan Endemic Nephropathy and Associated Urothelial Cancer: Current Status and Future Research

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Summary
Fifty years after BEN discovery, etiology remains its major unresolved problem. Etiology of BEN is extensively studied, especially the possible role of genetic factors, environmen tal agents and immune mechanisms. Evidence has accumulated that BEN is an environmentally-induced disease. The similarity of the morphological and clinical pattern of BEN and Chinese herb nephropathy has raised the possibility of a common etiologic agent, aristolochic acid. Genetic studies have landed support for genetic predisposition to BEN. Genetic expression analysis can identify candidate nephropathy pathogenesis genes and gene networks, which eventually could play role in BEN development.
An increased incidence of tumors of renal pelvis and ureter as well as the bladder tumors, in patients with BEN and in population from endemic settlements, has been observed. The geographic correlation between BEN and urinary tract tumors supports the speculation that these diseases share a common etiology.

Key words: Balkan endemic nephropathy, Urothelial cancer, Epidemiology, Etiology, Genetics, Prevention, Treatment

Balkan endemic nephropathy (BEN) is a chronic tubulointerstitial kidney disease encountered in high rate among the population of settlements along the tributaries of the Danube River in Serbia, Bosnia, Croatia, Bulgaria and Rumania [1-3]. We would like to give a survey of the recent studies and a prospect for future research. Major emphasis will be given to the etiology of BEN and associated urothelial cancer.

Epidemiology

The incidence and prevalence of BEN were stable over many years, but recently the incidence of BEN has been decreasing in some areas [4, 5]. BEN is still a major health problem in some endemic regions, Kolubara River in Serbia and Bijeljina Region in Bosnia, where up to 70 per cent of patients on dialysis are with BEN, and the incidence of BEN is very high.

Genetic predisposition to BEN

A familial aggregation of BEN has been described almost fifty years ago. Development of BEN in emigrants from the endemic region, who left their native villages in early childhood and settled hundreds, sometimes thousands of miles away, is in support of the role of the inheritance in the development of BEN [2]. A specific chromosome marker has been established on chromosome 3. It was suggested that the genetic factor is located in 3q25 [6]. Three of the additional five bands with increased frequencies of lesions in BEN patients were found to contain oncogenes: 1q36-c src, 3p25-raf-1, and 6q23-myb. The frequent association of BEN and cancer could be explained by the chromosomal hypothesis of oncogenesis [7]. Certain BEN relatives carry chromosomal anomalies that have already described in BEN patients, and it is proposed that they are at high risk for developing the disease [8]. Epidemiological data of patients and their healthy relatives are lacking, however, the follow-up studies of healthy relatives could answer the hypothesis that a genetic mechanism might be involved in the development of the disease even in the absence of the exposure to a BEN environment.
Genetic polymorphisms in environmentally regulated genes (such as those involved in the distribution, metabolism, and disposal of toxic compounds, and DNA repair pathways and their regulators) may also account for differential susceptibility to BEN in the endemic region population and particularly to upper urinary tract oncogenesis [3]. Indeed, CYP2D6 allele distribution differed in the group of BEN patients from that of healthy individuals and might be used as a possible marker for the BN susceptibility. Both cytochrome P450 and N-acetyl-transferases (NATs) activate many procarcinogens and chemicals whereas the multidrug resistance gene (MDR) codes for P-glycoprotein and is associated with xenobiotic resistance [9]. A study of polymorphisms in CYP3A4, CYP3A5, NAT1, NAT2 and MDR genes in Bulgarian BEN patients and controls suggested that NAT2 and MDR variants are part of the genetic background of BEN [10]. Genetic variants in xenobiotic metabolizing enzymes and transporters might thus augment the susceptibility of BEN patients to exogenous factors.

Environmental disease

Evidence is presented that environmental rather than genetic factors play a decisive role in the etiopathogenesis of BEN. Two genetically different populations, natives and immigrants from the Ukraine who settled in the endemic regions near Slavonski Brod, Croatia, have been demonstrated to develop BEN [11]. There are three actual theories attempting to explain the environmental cause of this disease: 1) the aristolochic acid (AA) hypothesis, which considers that the disease is produced by chronic intoxication with Aristolochia, 2) the mycotoxin hypothesis, which considers that BEN is produced by ochratoxin A (OTA), and 3) the Pliocene lignite

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hypothesis, which proposes that the disease is caused by long-term exposure to polycyclic aromatic hydrocarbons and other toxic organic compounds leaching into the well drinking water from low rank coals in vicinity to the endemic settlements.

A recent study of home-produced food (beans, potatoes, corn, wheat, flour) and feed from households in villages from the BEN region (Vratza district) of north-western Bulgaria. BEN households consistently had a higher proportion of OTA-positive samples than BEN free households, but similar (for some foods) or lower (for other foods) proportions to households from BEN free villages.

The results indicate that OTA may not alone cause BEN; only synergistically with other environmental toxicants and/or predisposing genotypes may do so [12].

The similarity of the morphological and clinical pattern of BEN and Chinese herbs nephropathy (CHN) has raised the possibility of a common etiologic agent [13] aristolochic acid. DNA adducts can be considered both as markers of the biologically effective dose and as markers of cancer risk. AA-DNA adducts may trigger the carcinogenic process in AAN patients. Indeed, the dA-AAI adduct is a premutagenic lesion and is associated with mutations in genes involved in carcinogenesis, such as the H-*ras* protooncogene and the p53 gene. The detection of specific AA-DNA adducts by 32P-postlabelling in kidney and ureter tissue of CHN patients unambiguously demonstrated the exposure to AA in CHN [14]. Several ochratoxin-DNA adducts were detected in kidney and bladder cancer tissues in two out of three patients from Bulgaria [15].

Immune mechanisms

Increased neopterin concentration in urine is a marker for activated Th1 immune response and for acute virus infections. Malignant tumor diseases were also frequently associated with increased neopterin concentrations. Fifty percent or urinary samples from 48 Bulgarian BEN patients presented with elevated neopterin concentrations compared to healthy controls (16). The patients with elevated neopterin levels did not have a higer degree of renal insufficiency, and there were no differences in other variables studied between the patients with increased neopterin and those with normal values. The background of this elevation, whether it is infectious or of other origin, remains unclear.

Urothelial tumors

An increased incidence of tumors of renal pelvis and ureter in patients with BEN and in population from endemic settlements has been observed. Recently the frequency of urinary bladder tumors in endemic settlements was also found increased compared with the nonendemic villages and cities [17]. The incidence of upper urothelial tumors (UUT) and bladder tumors associated to BEN, in the region of South Morava River in Serbia, in the 30-year period, was recently studied [18]. UUT had 11.2-fold increased incidence in endemic than in nonendemic areas during the last 10-year period (1989–1998), however, this was far less than in the period from 1969 to 1988, when they were 57.1 times more frequent. Bladder tumors were 2.3 times more frequent in endemic than in nonendemic areas, but 11.9 times in the previous study from 1969 to 1988. Similarly to the previous reports that BEN looks like a disease that disappears, this is demonstrated also for urothelial tumors, which are the most frequent associated disease. The geographic correlation between BEN and urinary tract tumors supports the speculation that these diseases share a common etiology [2].

Prevention and treatment

Since the etiological factor is unknown, effective prevention is not possible. Treatment of BEN is similar to that of all chronic interstitial nephropathies: detection and treatment of potentially reversible aggravating factors, limited protein intake, and kidney replacement therapy in end-stage kidney disease patients. Hemo- and peritoneal dialysis as well as kidney transplantation have been used with success. BEN does not recur after kidney transplantation. With longer survival on kidney replacement therapy, patients develop urothelial tumors which should continuously searched for [3].

Kidney disease beyond the Balkans

Fifteen years ago we have suggested that BEN is possibly a kidney disease affecting population beyond the Balkans [1]. Although traditionally described only in a few Balkan countries, kidney diseases similar in etiology to BEN could occur unrecognized in other regions of the world. BEN-like kidney diseases could remain unrecognized as there is at present no specific biologic marker of the disease. As soon as the etiologic agent(s) is discovered diagnosis of isolated cases or clustered BEN cases, as well as of associated urothelial carcinomas, will be made outside of the endemic regions in the Balkans, similar to the CHN, not restricted to the attendants of the Belgian slimming clinic. An increasing number of similar cases due to the consumption of herbs in which AA has been identified or was reputedly contained have been subsequently reported throughout the world [3].

Conclusion

Research on BEN is hampered by the lack of pathognomonic clinical and pathological characteristics. There is at present no specific biologic marker of the disease. Epidemiologic studies point to the causal role of still to be identified environmental factors although genetic, predisposing abnormalities are not fully elucidated. Genetic expression analysis can identify candidate nephropathy pathogenesis genes and gene networks, which eventually could play role in BEN development. Thus, gene expression profiling using microarrays is a high thruoutput method for investigation of renal function and nephrotoxicity. Moreover, the investigation of gene-gene and gene-environment interactions could be content on further studies to determining the precise risk for BEN.

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