Cancer Screening of HNPCC Family Members Depends on Gene Mutation Types and Cancer Sites in Affected Relatives

Pavel Elsakov[1], Raimundas Meškauskas[2]

[1] *Institute of Oncology of Vilnius University, Vilnius, Lithuania*
[2] *National Centre of Pathology, Vilnius, Lithuania*

The diagnosis of hereditary non-polyposis colorectal cancer (HNPCC) is confirmed by identifying a germline mutation in one of the DNA mismatch repair genes. Tumours of the colon, endometrium, stomach, ovary, urinary collecting system and other sites are related to this disease. Six Lithuanian families with HNPCC diagnoses were followed-up and analysed after the identification of mutations in the DNA mismatch repair genes. Three families had mutations in the hMLH1 genes and three families in hMSH2. Follow-up of HNPCC families' probands and individuals shows that the early diagnosis of cancers in probands and individuals of these families is related to gene mutation confirmation. Mutation of the gene hMLH1 is not a prognostic factor for the development of colorectal cancers in HNPCC families. Mutation of the gene hMSH2 is a prognostic factor for the development of extracolonic cancers in HNPCC families depending on the site of cancers in their affected relatives. From this limited study one can see that the identification of the most well known HLMH1 and HMSH2 gene mutations in HNPCC families helps improve follow-up of these families depending on gene mutation type and thus to improve early cancer diagnosis. The results of this study show that there is a need to organise a national institution that will follow-up/screen high risk families and those with HNPCC for earlier diagnosis, genetic testing, further screening and prevention in other members of these families.

Key words: hereditary nonpolyposis colorectal carcinoma (HNPCC), HMSH2/HMLH1 gene mutation, early hereditary cancer

INTRODUCTION

The recognition of HNPCC begins with a high index of suspicion based on family history. The diagnosis is confirmed by identifying a germline mutation in one of the DNA mismatch repair genes (1). Tumors of the colon, endometrium, stomach, ovary, urinary collecting system and other sites are related with this disease (2). The need arises for complex follow-up of family members to diagnose or prevent cancers in their early stages.

MATERIALS AND METHODS

From 1995 to 2002 at Lithuanian Oncology Center 26 HNPCC families were selected (Amsterdam II criteria). Six Lithuanian families with diagnosed HNPCC were followed-up and studied after identification of mutations in their DNA mismatch repair genes. Three of these families had mutations in the hMLH1 genes and three in hMSH2. Patients with the following probands: 095.01; 092.07; 091.07; 004.01; 118.01; 119.01 after surgery were treated depending on the diagnosis: with either radiotherapy or chemotherapy. Every 2 years follow-up by colonoscopy, clinical examination, laboratory testing, and various specialist consultations was arranged. New cancer cases diagnosed in individuals from these families were analyzed and compared for gene mutation type, cancer site members and stage at diagnosis.

RESULTS

The follow-up and analysis of HNPCC families' probands and individuals is shown in Table 1. There was a correlation between high risk and age at on-

Adress correspondence to: Dr. Pavel Elsakov, Vilnius University Institute of Oncology, Santariškių 1, LT-2021 Vilnius, Lithuania. Tel.: 370-5 278 6804.

Table 1. **Development and diagnosis of cancers in individuals with HNPCC depending on gene mutation type**

Proband(sex) Birth date(BD)	Cancer site/ Year	Stage (TNM)	Gene Mutation/ Year	Cancer site/Year	Stage (TNM)	Individual (sex) BD
004.01 (f) 1948	Ovary/1986 Rectum/1995 Breast/1999	I III II	hMSH2/1997			
091.07 (f)* 1969	Cecum/1994	II	hMHL1/1996	Cecum/1996	III	091.01(m) 1959
092.07 (f) 1959	Cecum/1994	II	hMLH1/1996	Cecum/1998	III	092.01(f) 1962
095.01 (f) 1948	Sigmoid/1990 Cecum/1993 Stomach/2000	III II II	hMSH2/1996			
118.01 (f) 1929	Colon/1996 Colon /1998	III II	hMHL1/2000			
119.01 (f) 1929	Uterus/1966 Rectum/1981	I II	hMSH2/2000			

* Miscarriage (1996), live birth (male) 1998.

set; the most frequent tumor types also tended to occur at a younger age. The median age of the patients at the time of diagnosis was 43.2 years. The youngest patients with colorectal cancer and ovarian/breast or endometrial cancer were aged 38 and 37 years, whereas the respective ages of the youngest patients with colorectal and gastric cancers were 36, 39, 42 and 67 years, respectively. The relations between cancer site, generation and individuals (relatives) of families depending on gene mutations are shown in table 2. Follow-up in the case of proband 004.01 despite CEA and CA-19-9 testing remaining in the normal ranges resulted in a diagnosis of breast cancer (stage II -TNM). For histopathological features see histopathology description. Follow-up in the case of proband 095.01 resulted in a diagnosis of cancer of the stomach (stage II). In case of individual 092.01 colon cancer

Table 2. **Relation among cancer site, generations and individuals (relatives) of HNPCC families depending on gene mutations types**

Generation	Family N and cancer site of affected individuals (relatives) *	
	Gene mutation	
	hMLH1	hMSH2
	N091	N095
I	Colon	No
II	Colon, Colon	Stomach, Stomach
III	Colon, Colon	Colon/Colon/Stomach
	N092	N004
I	Endometrium	Cancer ?
II	Colon	Stomach
III	Colon, Colon	Ovary/Rectum/Breast/Colon polyps, Colon
	N118	N119
II	Colon, Stomach	Rectum, Endometrium, Rectum
III	Colon, Colon/ Colon	Stomach, Endometrium, Endometrium /Colon

* a cell for every affected individual.

(stage III) was diagnosed late, because the patient did not get the necessary screening follow-up at the local hospital. In other cases of individuals 091.07, 118.01, 119.01 and 091.01 colon, rectum and uterine cancers (stage II–III) were diagnosed at different Lithuanian hospitals by retrospective analysis. A case of colon cancer (stage II-replace) was diagnosed in proband 118.01 at our clinic during routine follow-up colonoscopy. The extra-colonic cancer types found in our study fit well with the concept of the tumor spectrum found in HNPCC and which develop in patients' hMSH2 gene mutation.

HISTOPATHOLOGY

The surgical material taken from patient R. C. on several occasions was embedded in paraffin (1995 and 1999 samples) and by cryo section (1986). All sections were stained by hematoxilin and eosin and mucicarmin. Slides were examined by light microscopy under 100× and 400× magnification. Figure 1. comes from the removed uterus and adnexa (1986). Histologically in the ovaries there was a cystic dilatation of the glands with papillary projections covered with slightly pleomorphic mucous epithelium. The diagnosis of well-differentiated papillary mucous adenocarcinoma was established (Fig. 1).

Resection of the rectum (performed in 1995) revealed a moderately differentiated mucous adenocarcinoma 3 cm in diameter, infiltrating the perirectal tissue. The tumor consists of irregular glands and small groups of pleomorphic mucous epithelium (Fig. 2A, 2B) – practically the same as in the ovary tumor. Perirectal tissue examination revealed lymph node metastases.

Mastectomy with axillary lymph node clearance (1999) revealed a 3 cm poorly differentiated invasive ductal carcinoma in the mammary gland. Large pleomorphic cells with central necrosis filled the gland. The same cell complexes were found in the surrounding stroma. There were no metastases in the regional lymph nodes (Fig. 3).

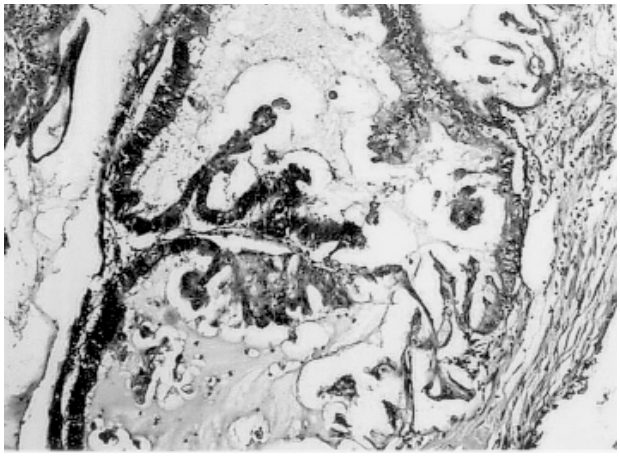

Fig. 1. Mucous papillary adenocarcinoma of the ovary: dilated cystic glands with papillary projections covered with mucous epithelium

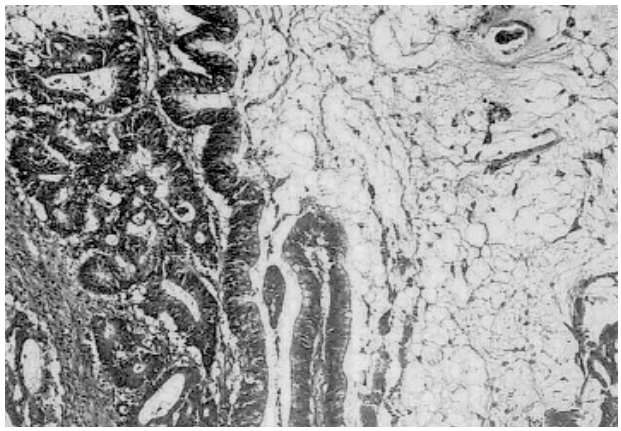

Fig. 2A. Mucous adenocarcinoma of the rectum: irregular glands covered with pleomorphic mucous epithelium

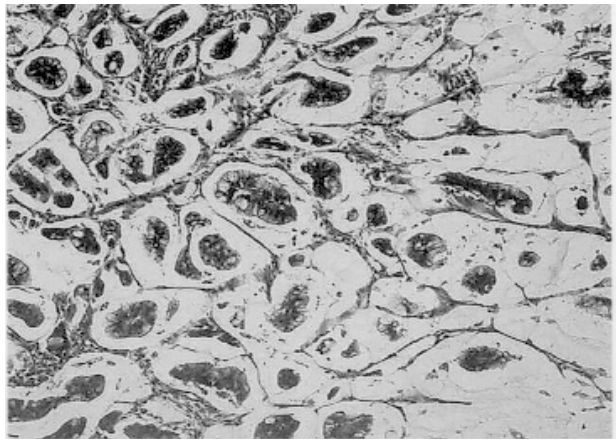

Fig. 2B. Mucous adenocarcinoma of the rectum: epithelial complexes surrounded by abundant mucin

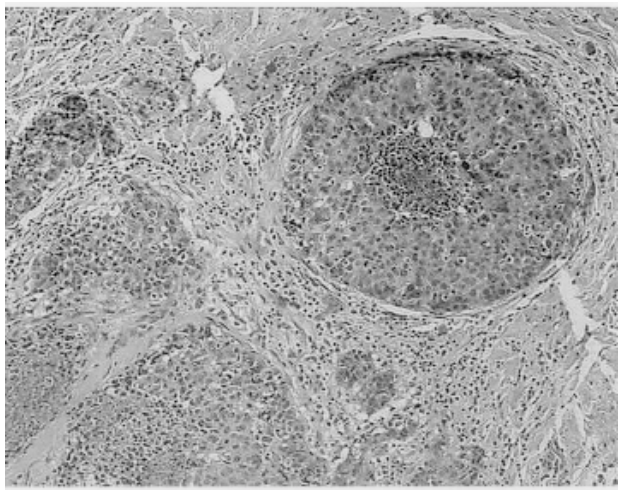

Fig. 3. Poorly differentiated invasive ductal carcinoma: large pleomorphic cells with central necrosis filling the ducts of the glands and infiltrating the stroma

DISCUSSIONS

In 1995 the first group of researchers began to study HNPCC in the Lithuanian population. A retrospective analysis of diagnosed HNPCC cancer case probands and their affected family members shows that a positive family history as a prognostic factor of hereditary cancer has not been appreciated, sought and acted upper in the public health rare sector and has thus not been used as a tool to help in the early diagnosis of these cancers. Results of this study show that a colon cancer (stage I–II) diagnosis in HNPCC probands has an odds ratio OR = = 0.84, which is less than that for the general population. Similar retrospective studies of early diagnosis (stage I–II – Dukes A or B) of HNPCC and sporadic colorectal cancers showed other authors to quote 51% for sporadic cancer and 52.1% for HNPCC (3, 4). Even known confirmed mutations in families do not seem to change the situation for the early diagnosis of cancer in other family members, as seen in the case of family N 092.01, where colon cancer (stage III) was diagnosed. There is no organ screening or family surveillance for those at risk of hereditary cancers, as in this case of HNPCC, which revealed a cancer at its later stage – not a good prognostic factor for successful treatment.

Studies and experience in many Western countries have led to the development of cancer family clinics, genetic-oncology departments for raising the awareness and for discussion about the impact of educational level on the style of association and relationship between doctors and patients. A better understanding of the processes and mechanisms of how healthy people cope with a genetic risk of cancer is important in genetic counselling and decisions-making in prevention or effective screening programs (5–8).

The present study shows alse that the cumulative frequency of each tumour is not the only significant factor in this evaluation. For most cancers the risk increases after the age of 50 years, when the value of surveillance programs becomes less clear. In family N 004.01, breast cancer (stage II) can be interpreted as a sporadic case, because there was no one known relative affected by cancer at this site, and histologically the features were more common for a sporadic cancer type. In family N 095.01 the stomach cancer (diagnosed stage II) was prognostic and needed screening and gastrectomy. Aarnio's (1997) study suggested that no mutation type was associated with a particularly high gastric cancer risk, which would necessitate screening more urgently than in other HNPCC families. In contrast, Vasen (1996) found a study that described an excess of gastric cancer only in families with mutation of the HMSH2 gene, i.e. not in HLMH1-associated families (9, 10). Our observation of HNPCC families suggests that the development of extracolonic cancers, including gastric, is associated more often with mutation of the HMSH2 gene, however in one family with a HLMH1 gene mutation there was an association with gastric cancer in a previous generation.

The question of which extra-colonic tumour sites should be taken into consideration for screening programs is difficult. Furthermore, an effective screening program requires suitable examination methods for detecting early and easily treatable lesions, for example, endoscopy which is readily available for examining stomach lesions. The value of screening for endometrial cancer remains to be proven in practice. Part of the problem can be managed by prophylactic hysterectomy and bilateral oophorectomy in association with the treatment of colorectal cancer or independently of it.

CONCLUSIONS

The results of our study show that gene mutation on HLMH1 or HMSH2 themselves are not markers for cancer development in individuals with HNPCC. From this limited study one can see that identification of the best known HLMH1 and HMSH2 gene mutations in HNPCC families helps improve follow-up for these families depending on gene mutation type and thus to improve early cancer diagnosis. The results of thes study show a need of a national institution that would follow up high risk families and those with hereditary non-polyposis colorectal cancers for diagnosis, genetic testing and further screening and prevention in other family members.

Received 3 June 2003
Accepted 19 September 2003

References

1. Lynch HT, de la Chapella A. Genetic susceptibility to non-polyposis colorectal cancer. J Med Genet 1999; 36: 801–18.
2. Parc YR, Halling KC, Burgart LJ. Microsatellite instability and hMLH1/hMSH2 expression in young endometrial carcinoma patients: Associations with family history and histopathology Int J Canc 2000; 86 (1): 60–6.
3. Elsakov P, Kurtinaitis J, Meškauskas R. Diagnosis of HNPCC in Lithuanian families. Acta medica Lituanica 1998; 5 (4): 339–43.
4. Bertario L, Russo A, Sala P et al. Survival of patients with hereditary colorectal cancer: Comparison of HNPCC and colorectal cancer: cancer in FAP patients with sporadic colorectal cancer. Int J Canc 1999; 80 (2): 183–7.

5. Hoskins IA. Genetic counseling for cancer patients and their families. Oncology 1989; 3: 84–92.
6. Houlston RS, Murday V, Harocopos C et al. Screening and genetic counselling for relatives of patients with colorectal cancer in family cancer clinic. Br Med J 1990; 301: 366–8.
7. Murday V. Genetic counselling in the cancer family clinic. Eur J Canc 1994; 3A: 2012–5.
8. Julian-Reynier C, Eisinger F, Chabal F et al. Cancer genetic consultation and anxiety in healthy consultees" Psychology and Health 1999; 14: 379–90.
9. Aarnio M, Salovaara R, Aaltonen L, Mecklin J-P, Jarvinen H. Features of gastric cancer in hereditary nonpolyposis colorectal cancer syndrome. Int J Canc (Pred. Oncol) 1997; 74: 551–5.
10. Vasen HFA et al. Cancer risk in families with hereditary nonpolyposis colorectal cancer diagnosed by mutation analysis. Gastroenterology 1996; 110: 1020–7.

P. Elsakov, R. Meškauskas

ANKSTYVOJI VĖŽIO DIAGNOSTIKA BEI PAVELDIMO NEPOLIPOZINIO STOROSIOS IR TIESIOSIOS ŽARNOS VĖŽIO PROFILAKTIKA ŠEIMOSE PRIKLAUSOMAI NUO GENŲ MUTACIJŲ IR NUO JŲ SIRGUSIŲ GIMINAIČIŲ VĖŽIO LOKALIZACIJOS

S a n t r a u k a

Paveldimo nepolipozinio storosios ir tiesiosios žarnos vėžio diagnostika remiasi nustatytomis genų mutacijomis. Šeimų, kurioms nustatytas paveldimas nepolipozinis storosios ir tiesiosios žarnos vėžys, nariai gali susirgti bet kurio organo vėžiu, nes tai būdinga šiam sindromui. Dažniausiai susergama gimdos, skrandžio, krūties, inkstų ir kitų lokalizacijų vėžiu. Lietuvoje buvo nustatytos šešios šeimos su paveldimu nepolipoziniu storosios ir tiesiosios žarnos vėžiu. Trims iš jų diagnozė patvirtinta HMLH1 geno mutacija, o kitoms trims šeimoms – HMSH2 geno mutacija. Šeimos buvo stebimos dėl galimo jų šeimos narių bei pačių probandų susirgimo vėžiu. Retrospektyvi analizė bei prospektyviai nustatyto vėžio atvejai šiose šeimose rodo, kad ankstyvoji vėžio diagnostika nepriklauso nuo patvirtintų genų mutacijų. HMLH1 geno mutacija paveldimo nepolipozinio storosios ir tiesiosios žarnos vėžio šeimose negali būti prognozinė tik storosios ir tiesiosios žarnos vėžio žymė. HMSH2 geno mutacija tampa prognozine vėžio žyme už storosios ir tiesiosios žarnos ribų. Gana nedidelis stebėtų šeimų skaičius neleidžia nustatyti optimalios profilaktikos priemonių šiose šeimose, kurios priklausytų nuo genų mutacijų lokalizacijos tarp vėžiu sirgusių giminių. Lietuvoje būtina sukurti tarnybą, kuri nagrinėtų paveldimo vėžio klinikines bei genetines diagnostikos, epidemiologijos bei profilaktikos priemones šioms šeimoms.

Raktažodžiai: paveldimas nepolipozinis storosios ir tiesiosios žarnos vėžys (PNSŽV), HMLH1 ir HMSH2 genų mutacija, ankstyvas paveldimas vėžys