SHORT REPORT

Increased risk of cancer in patients with fumarate hydratase germline mutation

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Hereditary leiomyomatosis and renal cell cancer (HLRCC) is a tumour predisposition syndrome caused by heterozygous germline mutations in the fumarate hydratase (*FH*) gene. The condition is characterised by predisposition to benign leiomyomas of the skin and the uterus, renal cell carcinoma (RCC), and uterine leiomyosarcoma (ULMS). To comprehensively examine the cancer risk and tumour spectrum in Finnish *FH* mutation positive families, genealogical and cancer data were obtained from 868 individuals. The cohort analysis of the standardised incidence ratios (SIR) was analysed from 256 individuals. *FH* mutation status was analysed from all available individuals (n = 98). To study tumour spectrum in *FH* mutation carriers, loss of the wild type allele was analysed from all available tumours (n = 22). The SIR was 6.5 for RCC and 71 for ULMS. The overall cancer risk was statistically significantly increased in the age group of 15–29 years, consistent with features of cancer predisposition families in general. *FH* germline mutation was found in 55% of studied individuals. Most RCC and ULMS tumours displayed biallelic inactivation of *FH*, as did breast and bladder cancers. In addition, several benign tumours including atypical uterine leiomyomas, kidney cysts, and adrenal gland adenomas were observed. The present study confirms with calculated risk ratios the association of early onset RCC and ULMS with *FH* germline mutations in Finns. Some evidence for association of breast and bladder carcinoma with HLRCC was obtained. The data enlighten the organ specific malignant potential of HLRCC.

Hereditary leiomyomatosis and renal cell cancer (HLRCC) is a recently recognised tumour predisposition syndrome caused by heterozygous germline mutations in the fumarate hydratase (*FH*; fumarase) gene (HLRCC, MIM 605839; MCUL, MIM 150800).[1 2] To date, 114 mutation positive families have been reported, mainly in Europe and North America.[1 3–10] Leiomyomas of the skin and uterus are the most common feature of HLRCC. RCC has been reported in families from North America, the UK, Finland, and Poland.[1 3 5 8–11] Predisposition to uterine leiomyosarcoma has been observed only in Finnish HLRCC families. In HLRCC patients from other populations, one case of leiomyosarcoma of the skin, a single case of bladder and brain cancer, and one malignant and two benign breast tumours have been reported.[8 10 12] Of note, no clear correlation between the type of *FH* mutation and phenotype has been found.[10 11] Previously, homozygous *FH* mutations have been shown to cause a recessive condition with progressive encephalomyopathy, referred to as fumarase deficiency (MIM 606812). Phenotypic similarities between HLRCC and FH deficiency have been observed in one family with FH deficiency, in which a patient's heterozygous parent had cutaneous leiomyomas.[2] To date, no cancer has been reported in these patients or their first degree relatives.

MATERIALS AND METHODS

Studies on high risk populations provide a unique opportunity to examine *FH* related cancer predisposition. To gain comprehensive insight into potential cancer phenotype in *FH* mutation positive families, the risk of cancer and the tumour spectrum was examined in seven Finnish families with HLRCC (FAM1-7) and one kindred with FH deficiency (FAM8) (table 1). The characteristic leiomyomatosis phenotype was not further scrutinised in this study. The study was approved by the Helsinki University Central Hospital authorised ethics review committee and all samples were derived after obtaining the individual's informed consent or authorisation from National Authority for Medicolegal Affairs.

RESULTS AND DISCUSSION

Genealogical and cancer data from church parish registries, death certificates, the Population Register Centre, and the Finnish Cancer Registry were obtained from 868 family members. To avoid selection bias, only individuals of the completely traced generations (n = 256) were included in the cohort analysis of the standardised incidence ratios (SIR). No selection according to mutation status was performed. Furthermore, all cancers in the index patients (five cancers in eight patients) and other patients with RCC or ULMS used for identification of the families (seven cancers in six patients) were excluded. The risk analysis provided statistically significant results, with a 6.5 fold risk (95% confidence interval (CI) 2.1 to 15.0) for RCC and a 71 fold risk (95% CI 8.6 to 260) for ULMS compared with the general population (table 2). The excess was concentrated in the age groups 15–29 and 30–44 years. Moreover, the overall risk of cancer was statistically significantly increased in the age group of 15–29 years (SIR 6.60, 95% CI 1.40 to 19.0) (table 2), although of note, this group included only three cancers (RCC, uterine leiomyosarcoma, and Hodgkin's lymphoma). The risk for other tumour types did not reach statistical significance (data not shown).

We next investigated the tumour spectrum in *FH* mutation positive individuals. Of 98 patients tested, 54 harboured a germline *FH* mutation, and approximately half of these patients had been diagnosed with cancer (table 3). Comparison of the age at cancer diagnosis of mutation positive and negative patients showed that mutation positive individuals are prone to cancer at a younger age (p = 0.009)

Abbreviations: CT, computed tomography; FH, fumarate hydratase; HLRCC, hereditary leiomyomatosis and renal cell cancer; PGL, paraganglioma syndrome; RCC, renal cell cancer; SIR, standardised incidence ratio; ULMS, uterine leiomyosarcoma

Table 1 Family data

Family	No. of family members	Phenotype	Germline mutation
FAM1*	137	RCC	541delAG
FAM2*	97	RCC	541delAG
FAM3†	59	Leiomyomas	R300X
FAM4	208	ULMS	H153R
FAM5	37	RCC	H153R
FAM6‡	42	Leiomyomas	H153R
FAM7‡	6	ULMS	541delAG
FAM8§	282	FHD	Q333P

Family identification previously reported by: *Launonen *et al*[1], ††Kiuru *et al*[3 4], and §Remes *et al*.[13 14] RCC, renal cell cancer; ULMS, uterine leiomyosarcoma; FHD, FH deficiency.

(fig 1), although this difference did not remain significant after exclusion of the probands. In *FH* mutation positive patients, the most prominent cancer types were RCC (n = 12) and uterine ULMS (n = 5), affecting 22% of all mutation positive individuals and 15% of mutation positive women, respectively. In addition, four patients had atypical leiomyomas, a variant of leiomyoma sometimes difficult to discern from leiomyosarcoma. As both atypical leiomyomas and leiomyosarcomas were frequent in *FH* mutation positive individuals, it is possible that the underlying *FH* mutation promotes malignant transformation of leiomyomas. Several cases of breast carcinoma and hematopoietic or lymphoid malignancies were observed. Cancer cases detected in *FH* mutation positive individuals are summarised in table 3.

To get insight into which tumours might have arisen due to the *FH* germline mutation, we examined somatic (biallelic) inactivation of the gene in 22 tumours (table 3, fig 2). Almost all analysed RCC and ULMS tumours as well as the one bladder and all three studied breast carcinomas had lost the wild type *FH* allele. One of the breast carcinomas and the bladder carcinoma were detected in the family with FH deficiency. These results suggest possible association of these two tumour types with *FH* germline mutations. Two of the patients with breast cancer had also been diagnosed with ULMS, supporting this concept. Strikingly, one patient with ULMS and breast cancer was also affected with multiple myeloma and non-Hodgkin's lymphoma.

In addition to the malignant tumours, data on benign tumours other than skin and uterine leiomyomas were obtained. Radiology reports available from 33 mutation positive individuals revealed benign kidney cysts in 14 individuals (42%). Scanning was mainly performed by ultrasound, but in 10 cases alternatively or in addition by computed tomography (CT). Reported prevalence of kidney cysts in general population varies, probably due to detection method used. Ultrasound and CT screenings have indicated prevalence to vary between 11.9–17.6% and 24–41%, respectively.[15–19] Thus, cystic lesions in the present study were observed somewhat more frequently than usual, especially at a young age. For comparison, in individuals younger than 40 years, prevalence was 36% compared with 4.6–8.2% (ultrasound and CT, respectively) in the general population.[18 19] Five of the 14 individuals with cysts were diagnosed with RCC. These included four multiple cyst cases, of which one was bilateral. Seven RCC patients had no cysts or the presence of them was not scrutinised. Thus, correlation between RCC and cysts is not probable. The remaining nine individuals with cysts displayed both single (n = 6) and multiple lesions (n = 3), of which two were bilateral. Diameter of these lesions was 20–60 mm in the cases where the information was available in the radiology report.

Table 2 Risk of cancer in families positive for *FH* mutation

	Observed	Expected	SIR	95% CI
RCC				
All	5	0.78	6.50	2.10 to 15.0
0–14	–	0.01	0.00	0.00 to 260
15–29	1	0.00	230	5.90 to 1300
30–44	2	0.04	45.0	5.50 to 160
45–59	–	0.22	0.00	0.00 to 17.0
60–74	1	0.35	2.80	0.07 to 16.0
75+	1	0.14	7.00	0.18 to 39.0
ULMS				
All	2	0.02	71.0	8.60 to 260
0–14	–	0.00	–	–
15–29	1	0.00	2100	52.0 to 11 000
30–44	1	0.00	180	4.60 to 1000
45–59	–	0.01	0.00	0.00 to 540
60–74	–	0.00	0.00	0.00 to 840
75+	–	0.00	0.00	0.00 to 2100
Total cancer				
All	34	24.73	1.40	0.95 to 1.90
0–14	–	0.23	0.00	0.00 to 16.0
15–29	3	0.45	6.60	1.40 to 19.0
30–44	5	1.97	2.50	0.82 to 5.90
45–59	6	6.16	0.97	0.36 to 2.10
60–74	15	10.40	1.40	0.81 to 2.40
75+	5	5.51	0.91	0.29 to 2.10

Members were divided into six age groups. SIR, standardised incidence ratio.

In addition, one liver haemangioma and adrenal gland adenomas including one bilateral tumour in four individuals (12%) were reported. Of note, the frequency of adenomas was higher than in the general population (0.5–2%).[20] Interestingly, RCC, kidney cysts, adrenal gland pheochromocytomas, and liver haemangiomas are associated with von Hippel-Lindau syndrome, and RCC and adrenal gland pheochromocytomas in hereditary paraganglioma syndrome (PGL). The molecular background in these syndromes also overlaps. The predisposing genes in the three syndromes have been implicated in the hypoxia pathway, and the genes predisposing to HLRCC and PGL operate in the mitochondrial Krebs cycle.[21–23] These similarities might provide clues to further understand the pathways of tumorigenesis in these diseases.

The present study has for the first time, using the exceptional databases and resources available in Finland, thoroughly evaluated the spectrum and relative risk of different cancers in patients with *FH* mutation. The results of the study confirm with calculated risk values the association of early onset RCC and ULMS with *FH* germline mutations. In addition to Finland, several cases of RCC have also been reported in North American HLRCC families; In a

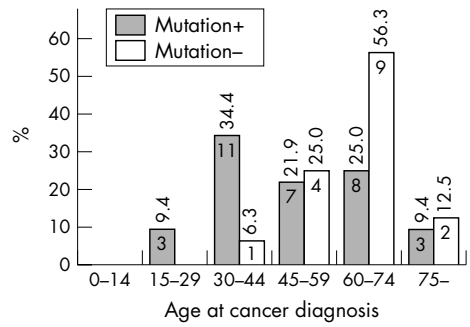

Figure 1 Age at cancer diagnosis in mutation positive and negative patients. The age distribution of cancer cases by percentage value is calculated within groups with different mutation status. The value inside the bar indicates number of cancer cases in the age group.

Table 3 Cancer cases in *FH* mutation-positive individuals

Tumour	n	Age(s) at diagnosis, years (median)	Somatic second hit detected/ analysed (%)
RCC	12*	26, 32, 33, 35, 36, 39, 42, 48, 49, 68, 71, 90 (40.5)	10/12 (83)
ULMS	5*	27, 30, 32, 35, 39 (32)	3/3 (100)
Breast cancer	4	50, 53, 55, 61 (52.5)	3/3 (100)
Bladder	1	71	1/1 (100)
Non-Hodgkin's lymphoma	1	63	0/1
Hodgkin's lymphoma	1	24	0/1
Chronic lymphatic leukaemia	1	48	0/1
Oesophageal cancer	1	53	0/1
Basal cell cancer	2	70, 83	0/1
Multiple myeloma	1	61	Not analysed
Prostate cancer	1	63	Not analysed
Liver/bile duct cancer (obligatory carrier)	1	82	Not analysed
Unknown origin	1	42	Not analysed

*Seven RCCs and two ULMSs have previously been published and have been reviewed by Kiuru and Launonen 2004.[11] RCC, renal cell cancer; ULMS, uterine leiomyosarcoma.

recent study, the frequency of HLRCC families with RCC was as high as 62%.[10] The average frequency of North American HLRCC kindreds with RCC is 35%, indicating high cancer risk in populations also other than Finns,[8 10] although criteria for family recruitment obviously affects the results of the different studies. However, no cases of ULMS have been identified in HLRCC families in North America. Data suggesting correlation of certain mutations to RCC have been reported,[10 24] but no such evidence concerning ULMS has been obtained. Difference in incidence of ULMS, however, may be due to the high frequency of myomectomy and hysterectomy at a young age carried out in affected women in North America; for example, in one cohort, 57% of women with cutaneous or uterine leiomyomas had undergone hysterectomy before 30 years of age.[8] Interestingly, as in Finnish families, those women included cases with uterine atypia (n = 2). The present study also provides evidence that other cancer types, including breast and bladder carcinoma, may be promoted by loss of *FH*. Of note, *FH* mutation carriers with bladder and breast tumours have been reported recently in HLRCC families in other populations.[10 12] Although the observations of the present study have been derived from a high risk population, it is likely to reflect that of other populations. This study provides insight into organ specific malignant potential associated HLRCC, and should promote further studies in families with *FH* germline mutations and awareness in patient management.

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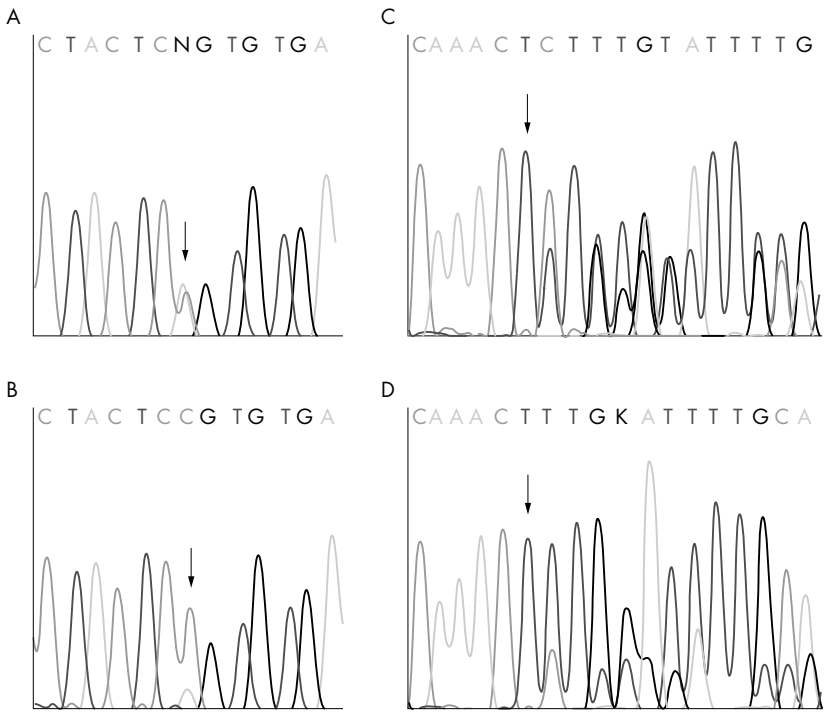

Figure 2 Examples of LOH detected in breast cancer. Chromatograms A and C illustrates normal breast tissues displaying germline mutations Q333P and 541delAG, respectively. LOH in breast tumours (B, D) is observable by the lower wild type allele signal in the presence of normal tissue contamination.

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