Strong family history of uterine leiomyomatosis warrants fumarate hydratase mutation screening

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Abstract: Hereditary leiomyomatosis and renal cell cancer (HLRCC) is a tumor predisposition syndrome characterized by cutaneous and uterine leiomyomas and renal cell cancer. HLRCC is caused by heterozygous germline mutations in the fumarate hydratase (FH) gene. A Finnish family with nine closely related women with uterine leiomyomas was detected by an alert gynecologist. No cutaneous or renal cell tumors were reported in the family when it was referred to genetic analyses. Samples were available from seven patients, and a novel germline FH mutation was detected in five of them. Mutation carriers were symptomatic, had multiple tumors and were diagnosed at an early age. This study emphasizes the importance of considering FH mutation screening when gynecologists encounter families with multiple severe uterine leiomyoma cases. Due to possibility of phenocopies more than one patient should be tested. Early mutation detection allows regular screening of the mutation carriers and enables early detection of possible highly aggressive renal tumors. It may also affect family planning as multiple myomas at early age may significantly reduce fertility.

Key words: hereditary leiomyomatosis and renal cell cancer (HLRCC) / uterine leiomyoma / fumarate hydratase (FH) / mutation screening / tumor predisposition syndrome

Introduction

Uterine leiomyomas (ULM, also called myomas and uterine fibroids) are benign tumors which arise from uterine smooth muscle cells. They are the most common tumors of the female reproductive tract. Clinically significant lesions have been observed in ~25% of Caucasian women, but thorough pathological examinations imply the prevalence to be as high as 77% (Stewart, 2001). Uterine leiomyomas can have a major impact on women’s health and quality of life by causing numerous symptoms such as abdominal pain, abnormal menstrual bleeding and infertility problems. Furthermore, these lesions are the most frequent indication for hysterectomy and thus generate considerable economic costs for the health-care system (Stewart, 2001).

Many lines of evidence suggest that genetic factors play a role in the development of myomas. Twin studies have shown that there is a strong element of heritability in women undergoing hysterectomy for leiomyomata (Snieders et al., 1998). Familial predisposition to uterine leiomyomas has been suggested in Japanese and Caucasian populations (Vikhlyaeva et al., 1995; Sato et al., 2002). Moreover, first degree relatives of women with myomas were discovered to have a 2.2 times increased risk of developing myomas (Vikhlyaeva et al., 1995). The clinical features of familial uterine leiomyomas also seem to be more severe than in sporadic cases (Okolo et al., 2005). Although the existence of a heritability component of uterine leiomyomata has been confirmed, still surprisingly little is known about their molecular background, particularly when considering the prevalence and socio-economic impact of uterine leiomyomas. Thus far, the only known genetic factor conferring a high risk for developing uterine leiomyomas are the germline mutations in the fumarate hydratase (FH) gene (Tomlinson et al., 2002). These mutations underlie hereditary leiomyomatosis and renal cell cancer (HLRCC, OMIM 605839, also known as multiple cutaneous and uterine leiomyomatosis, MCUL) syndrome. HLRCC is a rare autosomal dominant tumor predisposition syndrome characterized by cutaneous and uterine leiomyomas and renal cell cancer (RCC; Launonen et al., 2001). FH gene encodes fumarate hydratase, an enzyme of the tricarboxylic acid cycle (Tomlinson et al., 2002). Biallelic inactivation of the FH gene is frequently observed in the tumors, and thus FH is classified as a tumor-suppressor gene according to Knudson’s two-hit hypothesis (Tomlinson et al., 2002). Despite careful investigative efforts, no
clear correlation between the type of FH mutation or FH enzyme activity and HLRCC phenotype has been established. In contrast to the benign nature of leiomyomas, the renal cell tumors in the context of HLRCC are exceptionally aggressive and have a poor prognosis.

HLRCC has typically been diagnosed by detection of cutaneous leiomyomas or both cutaneous and uterine leiomyomas. Uterine leiomyomas alone as a sign of HLRCC are under-recognized. Uterine leiomyomas in HLRCC patients are severely symptomatic and have an early age of onset, which differentiates them from sporadic lesions (Alam et al., 2005; Lehtonen, 2011). Early diagnosis and regular screening of mutation carriers is especially important in the view of RCC risk. It is also relevant regarding family planning, as multiple uterine leiomyomas at a young age may significantly reduce fertility. Here we report a Finnish HLRCC family, in which nine closely related women with uterine leiomyomas only awoke the suspicion of a possible genetic component behind the development of the lesions. A novel FH mutation was detected in this family.

**Case Report**

The Finnish family reported here was identified by a gynecologist. It includes nine women with uterine leiomyomas in three generations (Fig. 1). Four patients have been diagnosed before the age of 30 years, three before 50 years and for two the age at diagnosis is not known. In addition, one woman in the family has been diagnosed with ovarian clear cell carcinoma at the age of 33 years. Her uterus has been removed, with no detection of myomas. Some women in the family have been diagnosed also with other gynecological tumors (Table I). No cutaneous leiomyomas were reported in any of the patients when the family was referred to genetic testing, and none of the family members has been diagnosed with RCC.

Germline mutations were then searched in the coding regions and exon–intron boundaries of the FH gene. Genomic DNA was extracted from blood using a standard procedure. Primer pairs flanking intronic splice sites for all 10 exons of the FH gene were used to amplify the genomic DNA. The PCR products were purified with ExoSAP-IT purification kit (USB Corporation, Cleveland, OH, USA) and sequenced using Big Dye Terminator v.3.1 kit (Applied Biosystems, Foster City, CA, USA) and ABI3730 Automatic DNA Sequencer (Applied Biosystems, Foster City, CA, USA). Sequences were analyzed manually and with Mutation Surveyor software (v.3.30, Soft Genetics, State College, PA, USA). Sequencing primers and PCR conditions are available upon request. All patients signed an informed consent before entering the study. The study has been approved by the ethics committees of the hospital districts of Helsinki and Uusimaa, and the Northern Ostrobothnia.

Sequencing revealed a novel heterozygous missense mutation (c.583A>AG, p.M195V) in exon five in five women with myomas (Fig. 1). The mutation is not a known polymorphism, resides in a highly conserved chromosomal region and was not detected among 94 healthy Finnish control individuals from the same geographical area obtained from Finnish Red Cross Blood Transfusion Service. Interestingly, another missense mutation affecting the same codon (c.584T>C, p.M195T; formerly named as M152T) has been identified in a North American HLRCC family, further supporting the pathogenic nature of the change (Toro et al., 2003). Also a mutation in the next

![Figure 1](https://academic.oup.com/humrep/article-abstract/27/6/1865/621558)
Table 1 Clinical features of uterine leiomyoma patients.

<table>
<thead>
<tr>
<th>ID</th>
<th>FH mutation</th>
<th>Age at diagnosis</th>
<th>Symptoms</th>
<th>Number of myomas</th>
<th>Age at hysterectomy</th>
<th>Other tumors</th>
</tr>
</thead>
<tbody>
<tr>
<td>II-1</td>
<td>M195V</td>
<td>36</td>
<td>Hypermenorrhea</td>
<td>Multiple</td>
<td>37</td>
<td>No</td>
</tr>
<tr>
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<td>M195V</td>
<td>28</td>
<td>Hypermenorrhea</td>
<td>1</td>
<td>28</td>
<td>Thyroid cyst</td>
</tr>
<tr>
<td>II-5</td>
<td>M195V</td>
<td></td>
<td>No</td>
<td>1</td>
<td>30</td>
<td>No</td>
</tr>
<tr>
<td>II-7</td>
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<td>49</td>
<td>No</td>
<td>1</td>
<td>61</td>
<td>Endometrial adenocarcinoma, adrenal adenoma</td>
</tr>
<tr>
<td>III-1</td>
<td>M195V</td>
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<td>Multiple</td>
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<td>III-2</td>
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<td>2</td>
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<td>No</td>
</tr>
<tr>
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<td>Multiple</td>
<td>27</td>
<td>Severe endometriosis, thyroid follicular adenoma</td>
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<tr>
<td>III-6</td>
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<td>35</td>
<td>No</td>
<td>2</td>
<td></td>
<td>No</td>
</tr>
</tbody>
</table>

Discussion

HLRCC is a highly penetrant tumor predisposition syndrome characterized by leiomyomas of the skin and uterus and in some families also RCC. Thus far, 180 HLRCC families have been identified worldwide (Lehtonen, 2011). The primary indication for HLRCC syndrome has been the detection of cutaneous leiomyomas or both cutaneous and uterine leiomyomas.

We here report a Finnish HLRCC family, in which a novel tumor predisposing mutation in the FH gene was detected in five women with uterine leiomyomas. These women were symptomatic, had multiple tumors and were diagnosed at an early age. This emphasizes the importance of FH mutation screening in families with multiple severe uterine leiomyoma cases.

Patients with uterine leiomyomas alone are seldom recognized as cases of HLRCC. Depending on the age of the studied individuals, 79–100% of women with FH mutation have uterine leiomyomas (Lehtonen, 2011). The myomas in HLRCC patients are often severely symptomatic, numerous, large and occur typically at the age of 20–35 years compared with around 40 years in sporadic cases (Toro et al., 2003; Alam et al., 2005; Lehtonen, 2011). The presence of a large and prominent eosinophilic nucleolus surrounded by a halo has also been described in HLRCC-associated uterine leiomyomas (Garg et al., 2011), but this observation needs to be validated in a larger sample series.

RCC in HLRCC is an exceptionally aggressive disease with poor prognosis. The tumors display mostly papillary type II histology and
occur at a younger age compared with sporadic patients, many even in patients younger than 30 years (Launonen et al, 2001; Tomlinson et al., 2002; Lehtonen, 2011). The lesions are typically solitary and unilateral (Tomlinson et al., 2002; Vahteristo et al., 2010; Lehtonen, 2011). These highly malignant renal tumors have been detected in ~20% of the HLRCC families worldwide (Launonen et al., 2001; Tomlinson et al., 2002; Toro et al., 2003; Vahteristo et al., 2010). Of note, up to 32% and 50% of the US and Finnish HLRCC families, respectively, have at least one RCC patient, and in many of these families there is more than one RCC case (Launonen et al., 2001; Tomlinson et al., 2002; Toro et al., 2003; Lehtonen, 2011). The increased RCC risk in the Finnish HLRCC families has been estimated to be 6.5-fold compared with the general population, and as high as 230-fold and 45-fold in the age groups of 15–29 and 30–44 years, respectively (Lehtonen et al., 2003). This strategy has proven successful, and for example in a Finnish HLRCC family a regular screening of a mutation carrier led to a diagnosis of an aggressive but early stage RCC and successful surgical removal of the tumor (Vahteristo et al., 2010). The incomplete penetrance of skin leiomyomas in women with FH mutation—only 55% of the female mutation carriers are affected by the age of 35 years when all males are affected—and the fact that the majority of the patients find these lesions tolerable and never seek for medical advice should be better acknowledged (Alam et al., 2005). Thus, severely symptomatic uterine leiomyomas could be the first detectable symptom of HLRCC. Indeed, the family reported here was identified by an alert gynecologist with the diagnosis of myomas in several family members at an early age. A highly penetrant tumor predisposing mutation in the FH gene was found in five women with myomas. After gene identification, family members have been offered genetic counseling and mutation screening, and also skin lesions have been observed subsequently in all mutation carriers who have attended for counseling thus far. Therefore, as the penetrance of skin lesions is incomplete in women, or they can be subtle and easily missed as happened here, the severe familial uterine leiomyomatosis alone warrants FH mutation screening. Of note, two patients with myomas did not harbor the mutation. These sporadic tumors were diagnosed at a later age and were clinically less severe (Table I). The presence of such phenocopies is not unexpected considering the high prevalence of myomas, and emphasizes the importance of genetic testing of more than one patient in the family when HLRCC is suspected.

The striking pedigree of the Finnish family greatly implied a genetic component behind the development of myomas, and indeed a novel pathogenic germline FH mutation was identified. The possibility of the HLRCC syndrome should be taken into account also in less dramatic families, and the diagnosis of a patient with symptomatic uterine leiomyomas, early age of onset and a family history of myomas should arouse the suspicion of the possibility of hereditary predisposition. When the clinical features are suggestive of HLRCC, even in the absence of cutaneous leiomyomas or RCC in the family, counseling and genetic testing should be offered to confirm the diagnosis. When a germline FH mutation is identified, all members of the family should be offered appropriate counseling and genetic testing, and clinical follow-up should be organized for the mutation carriers. Counseling should include information on the possible effects on reproduction, as multiple myomas at an early age may significantly reduce fertility. Also the significantly increased risk for very aggressive RCC should be recognized.

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### Authors’ roles

J.T. performed the molecular analyses and wrote the first draft of the manuscript. O.U. collected the clinical information and was the contact person towards the patients. M.R. identified the family and first suggested hereditary predisposition. L.A.A. participated in study design and drafting of the manuscript. P.V. designed and supervised the study and wrote the final draft of the manuscript.

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### Conflict of interest

None declared.

### References


