EUROPEAN JOURNAL OF CANCER 47 (2011) 1305–1311

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Mutational spectrum and therapy response of metastasized GIST in Central Switzerland – A population-based study

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ARTICLE INFO

Article history:
Received 25 October 2010
Received in revised form 31 December 2010
Accepted 20 January 2011
Available online 18 February 2011

Keywords:
Gastrointestinal stromal tumour
GIST
Mutations
Imatinib
Sunitinib
c-kit
PDGFRα

ABSTRACT

Background: Until now, no population-based studies investigated the mutational status of primary GIST (PT) and corresponding metastases and correlated these data with response to Imatinib or Sunitinib therapy.

Patients and methods: In a retrospective observation study, all metastatic GISTs of the last 15 years of our institution were investigated for mutations in *c-kit* and in *PDGFRα* gene in each PT and corresponding metastasis. Correlation with clinical outcome and response to Imatinib or Sunitinib therapy was performed.

Results: In 13 PT *c-kit* mutations in exon 9 (3), exon 11 (7) and exon 13 (1), 2 wild type genotypes, and no *PDGFRα* mutation were detected. In three metastases a switch from heterozygosity to homozygosity and one additional exon 13 mutation was observed. All 10 persons with available follow-up received Imatinib as first-line chemotherapy. Five of them (3 exon 9 mutations, 1 wild type, 1 additional exon 13 mutation) stopped Imatinib due to tumour progression. In three cases, Sunitinib as second-line chemotherapy was ended due to the same reasons.

Conclusions: Our data support previous observations, that *PDGFRα* mutations play no important role in metastasized GISTs. The influence of Imatinib and Sunitinib therapy in metastasized GISTs with wild type genotype and *c-kit* exon 9 mutations needs further investigation.

1. Introduction

Gastrointestinal stromal tumours (GIST) are the most common mesenchymal neoplasms of the gastrointestinal tract with an incidence of approximately 0.65–1.5 cases per 100,000 inhabitants.[1–6] The most frequently affected sites are the stomach and the small intestine, but GISTs may also develop in any other part of the GI tract, peritoneum and retroperitoneum. The finding, that approximately 95% of these tumours express CD117 (*c-kit* tyrosine kinase receptor),[7,8] has dramatically increased the diagnostic accuracy of these tumours. On a molecular level most GISTs harbour hyperactivating somatic mutations either in *c-kit* or *PDGFRα* genes, occurring mostly in the juxtamembrane domains or in the extracellular region.[9,10] This is the rationale for the treatment option with tyrosine kinase inhibitors like Imatinib (Gleevec®, Novartis, Switzerland) or Sunitinib (Sutent®, Pfizer, USA). The localisation of these mutations determines the efficiency of these substances, to inhibit the stimulatory effect of c-kit and PDGFRα proteins on cell growth.[11,12]

In the last few years some unselected population-based studies from various European regions have been published,

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0959-8049/$ - see front matter
doi:10.1016/j.ejca.2011.01.012

in which the mutational status of primary GISTs was investigated.[2–6] In one of these studies, the genomic status of metastases was reported,[2] but no correlation to the genomic status of corresponding primary tumour on the one hand and to the response after treatment with Imatinib and Sunitinib on the other hand was given.

The aim of our study was to evaluate the status of *c-kit* and *PDGFRα* mutations in metastasized GISTs of an unselected population in Central Switzerland, to describe the mutational spectrum of primary and corresponding metastatic tumours. In addition we correlated these results with the response to treatment with tyrosine kinase inhibitors Imatinib and Sunitinib.

2. Materials and methods

2.1. Ethical considerations

This was a retrospective study with no study-driven clinical intervention. No material was sent to external institutions. Approval of the local ethics committee was obtained (Reference No. EK 915).

2.2. Retrieval of cases

The record files of the Institute of Pathology of the Cantonal Hospital Lucerne were searched for all patients with GIST diagnosed in the period from 1995 to 2009. The search included all sites of the gastrointestinal tract as well as the intra-abdominal, mesenteric, omental, pelvic and retroperitoneal regions. In addition to GIST the search included all benign and malignant mesenchymal neoplasm as well as tumour-like conditions including fibromatosis, desmoids and inflammatory pseudotumour. The pathology reports and histological slides of patients with metastasized disease were re-evaluated (M.R.) and, in questionable cases, GIST diagnosis was confirmed by CD117 immunohistochemistry (see below).

Since only metastasized tumours were investigated no risk stratification of malignancy or pathological staging of the GIST was performed.

Clinical data including the treatment procedures (e.g. chemotherapy with Imatinib or Sunitinib, radiation, other therapies) after diagnosis of the primary and metastatic tumour, the response to therapy, and the state of disease at the end of the observation period were obtained from the treating oncologists.

2.3. Immunohistochemical and molecular analysis

Tissue probes of primary and metastatic tumours were fixed in 4% buffered formalin, embedded in paraffin and, after histopathological diagnosis, archived at the Institute of Pathology, Cantonal Hospital, Lucerne, Switzerland. All formalin-fixed paraffin-embedded tumour blocks were reviewed (M.R.) for quality and tumour content, and a single representative tumour block from primary tumour and every available metastases of each case was selected for immunohistochemical and molecular analyses.

Immunohistochemical reactions were performed with a Benchmark automatic immunostaining device (Ventana Medical System, Tucson, AZ, USA) according to the manufacturer's instructions. Three-μm thick tissue sections were incubated after heat induced antigen retrieval with antibodies against CD117 [clone A4502, DakoCytomation (Glostrup, Denmark), 1:50 dilution]. Positive and negative controls were included in each slide run.

For molecular analysis, micro-dissection of selected areas of the representative tumour tissue blocks was performed.

For molecular analysis of *c-kit* and *PDGFRα* gene, DNA was extracted using the QIAamp DNA FFPE Tissue Kit (Qiagen, Hilden, Germany). PCR for *c-kit* exons 8, 9, 11–18 and *PDGFRα* exons 10, 12, 14, 18 was performed in 20 μl volumes using 50 ng of template DNA, 0.2 U HotstarTaq polymerase (Qiagen, Hilden, Germany), 2.5 mM $MgCl_2$, 0.2 mM of dNTP (peqlab, Erlangen, Germany) and 400 nM of each primer (see Table 1). The cycling conditions were as follows: one cycle of 95 °C 15 min; 50 cycles of 95 °C 30 s, 60 °C 30 s and 72 °C 45 s; one cycle of 72 °C 5 min. The PCR products were purified with ExoSapIT (usb, Cleveland, Ohio). The sequencing reaction was carried out with BigDye Terminator Mix v 1.1 (Applied Biosystems, Foster City, CA) according to the manufacture's protocol. After cleaning the products with DyeEx (Qiagen) the probes were analysed using the abi 3130 sequencing system (Applied Biosystems) and the Chromas software. Each sequencing reaction was performed at least twice starting from independent PCR reactions. Detected mutations were confirmed in the sequence as sense and antisense strands. Our results were compared with the normal sequence available on gene ID 3815 (KIT) and gene ID 5156 (PDGFRα).

3. Results

Of the 87 GIST (crude incidence rate: 0.97 per 100,000 inhabitants of Central Switzerland per year), diagnosed from 1995 to 2009 in our institution, 13 patients (14.9%) had metastatic disease, which equals a crude incidence rate of 0.14 per 100,000 inhabitants of Central Switzerland per year. All primary and all metastatic tumours showed a strong CD117 expression. In three of the 13 metastatic cases, the metastases were diagnosed at the same time as the primary tumour.

Patients and tumour characteristics are summarised in Table 2. The patients collective included eight women (61.5%) and five men (38.5%), with a median age of 68 years (range 31–80) at the first GIST diagnosis. Primary tumours (PT) arose in the stomach (six cases; 46.2%), small intestine (four cases; 30.8%) and one case each in the pancreas, gastrocolic ligament and rectovaginal septum. Metastases were found in the liver (six cases, 33.3%), peritoneum, not otherwise specified (four cases, 22.2%), omentum (three cases, 16.7%), and retroperitoneum, mesenterium, rectovaginal septum, lymph nodes and bone (one case each, 5.6%). Five of the 13 patients (38.5%) had metastases in different organs (see Table 2). In two other patients with peritoneal metastases (patients A and C), tissue samples of two different, not otherwise specified peritoneal tumour sites were examined.

Histologically 10 of 13 PT showed a spindle cell type (76.9%), two PT a mixed spindle cell/epitheloid type (15.4%), and one tumour of the small intestine an epitheloid type (7.7%).

Table 1 – Used PCR-primer for c-kit and pdgfr-α.

Exon	Primer sequence 5′ → 3′
c-kit Exon 8 forward	TTCTGCCCTTTGAACTTGCT
c-kit Exon 8 reverse	AATTGCAGTCCTTCCCCTCT
c-kit Exon 9 forward	TCCTAGAGTAAGCCAGGGCTT
c-kit Exon 9 reverse	CAGAGCCTAAACATCCCCTTA
c-kit Exon 11 forward	CCAGAGTGCTCTAATGACTGA
c-kit Exon 11 reverse	CCTTAAAGTCACTGTT ATGTGTACC
c-kit Exon 12 forward	GGTTTGCCATAGAGAACATCG
c-kit Exon 12 reverse	CAAAAAGCACAACTGGCAAA
c-kit Exon 13 forward	TCCTGTATGGTACTGCATGC
c-kit Exon 13 reverse	AAAGGCAGCTTGGACACGGCT
c-kit Exon 14 forward	TGGGAGGCAGAATTAATCT
c-kit Exon 14 reverse	AACCCTTATGACCCCATGAA
c-kit Exon 15 forward	GACCCATGAGTGCCCTTCTA
c-kit Exon 15 reverse	TTGACCATTGGCACTGCTAC
c-kit Exon 16 forward	GCCACTGTCTTTTCCTTTCC
c-kit Exon 16 reverse	TGGCTCTAAAATGCTCTGTTCTC
c-kit Exon 17 forward	TGTATTCACAGAGACTTGGC
c-kit Exon 17 reverse	GCAGGACTGTCAAGCAGAGA
c-kit Exon 18 forward	CATTTCAGCAACAGCAGCAT
c-kit Exon 18 reverse	CAAGGAAGCAGGACACCAAT
pdgfr-α Exon 10 forward	TCCACTCATTGCCATGACTC
pdgfr-α Exon 10 reverse	AGATGCGGCTCAGCTGATGA
pdgfr-α Exon 12 forward	GGTGCACTGGGACTTTG GTAATTCAC
pdgfr-α Exon 12 reverse	CAAGGGAAAAGGGAGTCTTGGAG
pdgfr-α Exon 14 forward	GAGAACAGGAAGATGGTAGCTCA
pdgfr-α Exon 14 reverse	TTCACAACCACATGTGTCCA
pdgfr-α Exon 18 forward	CCAGTCTTGCAGGGGTGATGCTAT
pdgfr-α Exon 18 reverse	AACAGGCACCGAATCTCTAGAAGC

The median of the greatest diameter of PT was 9.5 cm (range 4–25 cm). Only one PT was less than 5 cm (7.7%), six PT ranged between 5 and 10 cm (46.2%), and five PT were greater than 10 cm (38.5%). For one PT, confirmed by biopsy, no reliable tumour size was available.

The mitotic count (per 50 high power fields (hpf)) ranged from 1 to 100 with a median of 8. The highest counts were observed in PT of the stomach (up to 100 mitoses per 50 hpf) and the small intestine (up to 40 mitoses per 50 hpf), whereas the extragastrointestinal PT showed a low mitotic count (range 1–6 mitoses per 50 hpf).

Molecular analyses could be performed in all PT and all above described corresponding metastases (see also Table 2). In patients with more than one metastatic site, the different metastases of the respective patient always showed the same genetic alterations. In two cases with PT in the stomach (patients C and H; 15.4%) no genetic alterations in the *c-kit* or in the *PDGFR-α* genes could be detected either in PT or in metastases. The other 11 cases (84.6%) showed changes in the *c-kit* gene, but none in the *PDGFR-α* gene. *C-kit* exon 9 alterations could be detected in two PT of the small intestine and in the one PT of the pancreas. In all three cases the same well known duplication of codon 502–503 (p.502_503dup AY) was found in PT as well as in metastatic tumours in heterozygous form. Both PT and metastasis of the GIST of the septum rectovaginale showed a heterozygous exon 13 point mutation (p.K642E). The vast majority of PT (7 patients with PT in stomach, small intestine and lig. gastrocolicum) showed heterozygous exon 11 alterations: One point mutation (p.W557G; case I), five deletions (p.W557_K558del in case D and J, p.K558_E562del in case G, p.E554_N567del in case K, and p.V559_G565del in case M), and one duplication (p.P577_R588dup12 in case L). In two of the cases (patients J and L) metastatic tumours showed homozygosity for the same alteration as in the PT. In the metastatic tumour of patient D, two changes compared to the PT could be seen: Homozygosity of the exon 11 deletion and an additional heterozygous point mutation in exon 13 (p.V654A). Of the above mentioned genetic alterations, all but two have been previously described. The two exceptions are the exon 11 deletion (p.E554_N567del) of patient K and the exon 11 duplication (p.P577_R588dup12) of patient L.

Full clinical data were available in 10 of 13 cases. In 2 patients metastatic GIST was initially diagnosed within the last 3 months of the study period, so no clinical data of the disease progression exist. One patient did not return for a follow-up after diagnosis of metastatic disease (25 months after PT diagnosis). The median observation period was 47 months (range 1–180). The progression free survival after diagnosis of PT ranged from 6 to 114 months (median 22 months). At the end of the follow-up period (15th January 2010) 4 patients had died of disease, 3 patients had a stable disease and 3 patients showed no evidence of disease. The range of overall survival after diagnosis of PT was 6–180 months (median 47 months).

All 10 patients with appropriate clinical data received Imatinib as first-line tyrosine kinase inhibitor therapy during the observation period. Among these, the therapeutic regime varied considerably (see also Table 2): One patient (case C) received Imatinib as preoperative therapy (400 mg/day for 1 month) of PT and then again after resection of metastatic tumour (800 mg/day). Another patient (case J) received Imatinib as postoperative therapy (400 mg/day) of PT for 12 months and again after relapse of disease (400 mg/day). The other 8 patients received Imatinib after the radiological and/or histological diagnosis of metastases or local recurrency with an initial dose of 400 mg/day. So in 4 patients (C, D, H; J) the tissue probes of metastases used in this study were taken after the start of Imatinib therapy and in 6 patients (A, B, E, F, G and I) before.

In 1 case (patient A) a dose elevation to 600 mg/day was performed after 38 months for another 25 months. In two other patients (E and F) the Imatinib dosage was increased to 800 mg/day after 20 months for 1 month and after 38 months for 3 months, respectively. The therapy was switched to Sunitinib in 5 patients (37.5 mg in four cases, 50 mg in one case, see also Table 2) due to tumour progression (3 patients), intolerance (1 patient) or a combination of both (1 patient) during Imatinib application. The Sunitinib therapy in 3 of these 5 patients was stopped due to the same reasons (2 patients with intolerance, 1 patient with tumour progression). In patient E, Imatinib therapy was replaced by radiotherapy (30 Gy for 2 weeks). In 2 patients, Imatinib therapy was terminated without substitution because of tumour progression (patient B) and side-effects (patient H), respectively.

In all 3 patients with alterations of the *c-kit* exon 9 a tumour progression was observed during Imatinib therapy, which was, therefore, stopped. Whereas 1 of the 2 remaining

Table 2 – Patients' and tumour characteristics.

Patient ID	Sex	Age (years)	Site primary tumour (PT)	PT diameter (cm)	PT mitoses (per 50 hpf)	PT histology	Mutation PT	Site of metastasis (M)	Mutation M	Observation period (months)	Progression free survival (months)	Time spread PT – M (months)	State of disease	Duration (months) and dosage Imatinib-therapy	Withdrawel Imatinib therapy (reason)	Duration (months) and dosage Sunitinib therapy	Withdrawel Sunitinib therapy (reason)
A	m	71	Small intestine	9	22	S	c-kit Exon 9 (p.502_503dupAY) heterozygous	Peritoneum	c-kit Exon 9 (p.502_503dupAY) heterozygous	93	23	23	DOD	38 (400 mg/day) 25 (600 mg/day)	Yes (TP)	1 (50 mg/day)	Yes (SE)
B	f	68	Jejunum	8	4	S	c-kit Exon 9 (p.502_503dupAY) heterozygous	Mesenterium	c-kit Exon 9 (p.502_503dupAY) heterozygous	47	19	19	DOD	3 (400 mg/day)	Yes (TP, SE)	No	
C	f	31	Stomach	21	21	S	None	Peritoneum	None	52	20	20	DOD	1 (400 mg/day before PT surgery) 4 (800 mg/day after metastasis)	Yes (TP)	1 (37.5 mg/day)	Yes (SE)
D	m	64	Stomach	23	8	S	c-kit Exon 11 (p.W557_K558del) heterozygous	Retroperitoneum	c-kit Exon 11 (p.W557_K558del) homozygous; c-kit exon 13 (p.V654A) heterozygous	79	17	78	DOD	28 (400 mg/day)	Yes (TP)	30 (37.5 mg/day)	Yes (TP)
E	f	72	Septum rectovaginale	n/a (biopsy)	1	S	c-kit Exon 13 (p.K642E) heterozygous	Septum rectovaginale	c-kit Exon 13 (p.K642E) heterozygous	180	114	114	SD	20 (400 mg/day) 1 (800 mg/day)	Yes (SE)	No	
F	f	66	Pancreas	4	2	S	c-kit Exon 9 (p.502_503dupAY) heterozygous	Liver, omentum	c-kit Exon 9 (p.502_503dupAY) heterozygous	168	104	104	SD	38 (400 mg/day) 3 (800 mg/day)	Yes (TP, SE)	12 (37.5 mg/day)	No
G	f	65	Stomach	8	24	S	c-kit Exon 11 (p.K558_E562del) heterozygous	Liver	c-kit Exon 11 (p.K558_E562del) heterozygous	41	11	11	SD	18 (400 mg/day)	No	No	
H	f	68	Stomach	11	4	S	None	Liver, lymph node	None	6	6	0	NED	4 (400 mg/day)	Yes (SE)	No	
I	f	68	Jejunum	8	40	S	c-kit Exon 11 (p.W557G) heterozygous	Omentum, peritoneum	c-kit Exon 11 (p.W557G) heterozygous	35	22	22	NED	1 (400 mg/day)	Yes (SE)	9 (50 mg/day)	No
J	m	65	Stomach	10	40	S	c-kit Exon 11 (p.W557_K558del) heterozygous	Liver	c-kit Exon 11 (p.W557_K558del) homozygous	26	24	24	NED	12 (400 mg/day after PT) 1 (400 mg/day after metastasis)	No	No	
K	m	55	Stomach	7.5	100	S/E	c-kit Exon 11 (p.E554_N567del) heterozygous	Liver	c-kit Exon 11 (p.E554_N567del) heterozygous	n/a	n/a	0	n/a	n/a	n/a	n/a	n/a
L	f	74	Lig. gastrocolicum	25	6	S/E	c-kit Exon 11 (p.P577_R588dup12) heterozygous	Liver, bone	c-kit Exon 11 (p.P577_R588dup12) homozygous	n/a	n/a	25	n/a	n/a	n/a	n/a	n/a
M	m	80	Small intestine	15	7	E	c-kit Exon 11 (p.V559_G565del) heterozygous	Omentum, peritoneum	c-kit Exon 11 (p.V559_G565del) Heterozygous	n/a	n/a	0	n/a	n/a	n/a	n/a	n/a

Sex (m = male; f = female); PT histology (S = spindle cellular; E = epitheloid); state of disease (DOD = death of disease; SD = stable disease; NED = no evidence of disease); n/a = not available; TP = tumour progression; SE = side-effects.

patients, in which the Imatinib therapy had to be ended due to tumour progression, showed no detectable genetic alteration, the other one showed more genetic alterations in the metastasis compared to the PT: The heterozygous deletion in exon 11 of the *c-kit* gene of the PT changed to a homozygous deletion in metastasis and an additional point mutation in exon 13 of the *c-kit* gene was detected.

Unfortunately, no clinical follow-up data exist for the two cases (patients K and L) that harbour the novel complex genetic alterations in exon 11 of the *c-kit* gene.

4. Discussion

The present study is, to our knowledge, the first, which focuses on metastatic GIST in a population-based collective, analysing both the mutational status and response to tyrosine kinase inhibitor therapy.

Our population data with a GIST crude incidence of 0.97 per 100,000 inhabitants per year and a proportion of 14% metastasized GIST is comparable to data reported by other groups.[2–6]

Compared to the study of Braconi and colleagues,[2] which provides data of primary metastasized tumours of their whole GIST collective, the mutational status of our collective, included primary and secondary metastasized tumours, shows a slightly higher amount of wild type GIST (15.4% versus 5%) and a similar proportion of *c-kit* mutations (84.6% versus 90%). In contrast to their study (5%) we could not detect any *PDGFRα* mutation.

Also the site distribution of tumours (46.2% stomach, 30.8% small intestine, and 24% other sites) is comparable to the data presented by Braconi et al. A difference could be observed in the histological type of the tumours: While the vast majority of tumours in the present series showed a spindle cell type (76.9%) and only 15.4% and 7.7% a mixed or epithelioid type, respectively, more epitheloid (26%) and mixed type (31%) GISTs were reported in the metastatic group in their study.

The genetic alterations of the investigated metastatic GIST's are comparable to those, reported in literature for recurrent and metastasized GIST. The most frequently altered exon in the primary tumours of our study group was *c-kit* exon 11 (54%) with five deletions, one duplication, and one missense point mutation. Other observed genetic alterations occurred in the *c-kit* exon 9 (23%) and *c-kit* exon 13 (8%), while two PT (15%) showed a *c-kit* and *PDGFRα* wild type status. Similar results have also been described in previous publications.[2,9,13,14]

The fact that we could not detect any *PDGFRα* mutations in our group of malignant GIST supports the assumption, that GISTs with this genetic alteration are associated with a lower malignant potential and, therefore, have a better prognosis.[10,14]

The exon 11 alterations, which mostly occur in gastric GIST,[5,14] were also mostly detected in our PT of the stomach (4 of 7 cases). In 3 of these 4 cases, for which a clinical follow-up was available, a stable disease or no evidence of disease was observed, which is congruent with published data.[12] No correlation between outcome and type of exon 11 alterations could be seen. So the PT of patient D, who died of disease, had the same heterozygous exon 11 deletion (p.W557_K558) as patient J, who showed no evidence of disease. Interestingly, both showed a switch to homozygosity in their metastatic tumours, which was described as a prognostically unfavourable factor.[15] In addition the metastasis of patient D harboured a point mutation in exon 13. Previous studies showed that exon 13 alterations result in poor response to tyrosine kinase inhibitor therapy.[16] This could be responsible for the more aggressive course of the disease of this patient, while receiving Imatinib and Sunitinib without any clinical or radiological response. It remains debatable, whether the genetic changes form PT to metastasis in these two cases, in which the here investigated metastatic tissue probes were taken after the start of the drug therapy, were a consequence of the administration of Imatinib and/or sunitinib. The other 3 patients with exon 11 alterations, who showed a stable disease or no evidence of disease, initially received Imatinib therapy. In 1 patient the therapy was stopped after the end of the adjuvant cycle (1 year) with no evidence of residual disease. In another patient a change to Sunitinib was necessary due to liver toxicity.

Exon 9 alterations generally seem to be associated with a worse course of disease. In our study all three observed cases showed the frequent p.502_503AY duplication[12,17–19] heterozygously in PT and in metastases. The 2 patients with exon 9 mutations and PT in the small intestine showed a tumour progression under tyrosine kinase inhibitor therapy (Imatinib and Sunitinib) and died of disease. One of these two patients (A) showed a tumour progression despite a dosage elevation of Imatinib up to 600 mg/day. The third patient with exon 9 alteration, who's PT was located in the pancreas, shows stable disease under Sunitinib treatment after Imatinib therapy was stopped due to tumour progression and side-effects.

One of the two patients with wild type GIST was diagnosed only 6 months before the end of our observation period, so no reliable follow-up data is available. The other wild type patient died of disease, showing a rapid tumour progression during tyrosine kinase inhibitor treatment (Imatinib, Sunitinib). This poor response of wild type GIST patients was also repeatedly reported in the literature.[9,11,12]

In two patients (K and L) we detected novel exon 11 alterations so far not described: A 14 amino acids deletion (p.E554_N567) in case K with heterozygosity in both PT and metastasis and a 12 amino acids duplication (p.P577_R588) in case L, being heterozygous in PT and homozygous in the metastasis. Unfortunately, no clinical follow-up was available.

Until now only a few cases of GIST in the rectovaginal septum have been described in the English literature.[20–29] In all these reports, similarly to our case, this localisation seems to be associated with a favourable prognosis. In the few cases with reported genotyping,[24,25] no *c-kit* exon 13 alteration – like in our case – was described.

Taken together, our study, which is based on a population-based collective of metastatic GIST, confirms the observations of other researchers: GISTs with a malignant behaviour are mostly associated with genetic alterations in exons 9 and 11

of the *c-kit* gene or harbour wild-type *c-kit*. *PDGFRα* alterations on the other hand only rarely result in metastatic disease.

In the herein described setting of a population-based observational study with a small number of cases with different, not standardised treatment modalities, it remains difficult, to draw certain conclusions for a possible relation between the genetic alterations of *c-kit* and *PDGFRα* in PT and metastases, respectively, and the response to tyrosine kinase inhibitor therapy. In tendency, our observations support the results of previous studies, in that GISTs with a wild type genotype or a *c-kit* exon 9 alteration in primary tumour or a secondary mutation in metastasis show a worse response to Imatinib or Sunitinib treatment than other *c-kit* alterations. But, especially in patients with exon 9 mutations and treatment with elevated doses of Imatinib according to the results of the Meta-GIST group,[30] further studies with a larger number of patients with metastasized GIST are needed to confirm these findings.

Funding

This work was supported by an unrestricted research grant of Novartis Pharma Schweiz AG.

Conflict of interest statement

None declared.

Acknowledgements

The authors thank the treating oncologists A. Gschwend, R. Sperb, R. Winterhalder and T. Froesch for providing clinical data. We are very grateful to A. Vogetseder for linguistic revision of the manuscript.

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