

**DIAGNOSIS AND MULTIDISCIPLINARY TREATMENT OF SPORADICAL
DESMOID TUMORS**

Ph.D. Thesis

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Szeged

2012

PUBLICATIONS RELATED TO THE THESIS

1. **Mátrai Z**, Tóth L, Szentirmay Z, Papp J, Langmár Z, Kásler M Multidisciplinary treatment of intra-thoracic desmoid tumours: case series and narrative review. *Med Sci Monit.* 2012, 18: 17-25. **IF: 1.699***
2. **Mátrai Z**, Tóth L, Szentirmay Z, Papp J, Antal I, Vadász P, Bartal A, Kásler M Surgical challenges of chest wall and intra-thoracic desmoid tumors. *Orvosi Hetilap*, 2011, 152: 3-13.
3. **Mátrai Z**, Tóth L, Gulyás G, Szabó E, Szentirmay Z, Kásler M. A desmoid tumor associated with a ruptured silicone breast implant. *Plast Reconstr Surg.* 2011, 127: 1-4. **IF: 2.635***
4. **Mátrai Z**, Tóth L, Szentirmay Z, Vámos FR, Klepetko W, Vadász P, Kenessey I, Kásler M. Sporadic desmoid tumors of the chest: long-term follow-up of 28 multimodally treated patients. *Eur J Cardiothorac Surg.* 2011, 40: 1170-1176. **IF: 2.293***
5. **Mátrai Z**, Tóth L, Rényi Vámos F, Papp J, Gődény M, Bartal A, Szentirmay Z, Kásler M Giant intra-thoracic desmoid tumour. Case report. *Med. Thor.* 2010, 63: 158-166.
6. **Mátrai Z**, Papp J, Polgár C, Hitre E, Köves I, Oláh E, Andi J, Kiss A, Vámosi Nagy I, Tóth L, Orosz Z Long-term experience with therapy of a Gardner's syndrome female, first presenting with extra-abdominal desmoid tumor, and review of the literature. *Magy Seb.* 2009, 62: 75-82.
7. **Mátrai Z**, Péley G, Rényi Vámos F, Farkas E, Kovács T, Köves I The similarities between the mechanism of wound healing and tumor development--literature review on the occasion of a patient with colonic adenocarcinoma metastasis in a dog-bite wound. *Orv Hetil.* 2005, 146: 99-109.

ΣIF = 6.6272*

* Source: Magyar Tudományos Művek Tárháza

PUBLICATIONS NOT DIRECTLY RELATED TO THE THESIS

1. **Mátrai Z**, Gulyás G, Tizedes G, Tóth L, Langmár Z, Kásler M. Bilateral juvenile fibroadenoma of the breast: Management with subcutaneous mastectomy and silicone implant placement. *European Journal of Gynaecological Oncology*, 2012 (accepted for publication)
2. **Mátrai Z**, Tóth L, Sávolt Á, Típusz A, Palla É, Bartal A, Horti I, Kásler M Breast care nurse. A new specialist in the multidisciplinary care of breast cancer patients. *Magy Onkol.* 2012 (accepted for publication)
3. Bartal A, **Mátrai Z**, Szűcs A, Belinszkaja G, Langmár Z, Rosta A Novel oral anticancer drugs: a review of adverse drug reactions, interactions and patient adherence. *Orv Hetil.* 2012, 153: 66-78.
4. Gulyás G, **Mátrai Z**, Kunos C, Pesthy P, Doleviczényi Z, Kásler M Modern breast surgery. *Orv Hetil.* 2012, 153: 31-38.
5. Bartal A, **Mátrai Z**, Rosta A, Szűcs A Introduction of a clinical protocol for extravasation at the National Institute of Oncology, Budapest, Hungary. *Magy Onkol.* 2011, 55: 14-20.
6. Bartal A, **Mátrai Z**, Szűcs A, Liskay G Main treatment and preventive measures for hand-foot syndrome, a dermatologic side effect of cancer therapy. *Magy Onkol.* 2011, 55: 91-98.

7. Langmár Z, Németh M, Kolozsvári LR, Joó JG, **Mátrai Z** Cancer of the vagina. *Orv Hetil.* 2011, 152: 1974-1976.
8. **Mátrai Z**, Gulyás G, Tóth L, Polgár C, Bidlek M, Szabó E, Láng I, Horváth Z, Udvarhelyi N, Kunos C, Sávolt A, Pesthy P, Kásler M Challenges in oncologic plastic surgery of the breast. *Magy Onkol.* 2011, 55: 40-52.
9. **Mátrai Z**, Gulyás G, Tóth L, Sávolt A, Kunos C, Pesthy P, Bartal A, Kásler M Role of nipple sparing mastectomy in modern breast surgery. *Orv Hetil.* 2011, 152: 1233-1249.
10. **Mátrai Z**, Tóth L, Polgár C, Láng I, Gödény M, Sinkovics I, Horváth Z, Bidlek M, Udvarhelyi N, Bartal A, Sávolt A, Ujhelyi M, Kásler M Sentinel lymph node biopsy after neoadjuvant chemotherapy in breast cancer. *Magy Onkol.* 2011, 55: 73-84.
11. **Mátrai Z**, Gulyás G, Tóth L, Sávolt A, Kunos C, Pesthy P, Bartal A, Szabó E, Kásler M Special considerations in breast cancer treatment of an augmented breast. *Orv Hetil.* 2011, 152: 1679-1691.
12. **Mátrai Z**, Gulyás G, Tóth L, Sávolt A, Kunos C, Pesthy P, Bartal A, Kásler M The place of skin-sparing mastectomy in oncoplastic breast surgery. *Magy Onkol.* 2011, 55: 252-267.
13. **Mátrai Z**, Tóth L, Bidlek M, Szabó E, Farkas E, Sávolt A, Góbor L, Bartal A, Kásler M The potential role of ductoscopy in modern breast diagnostics and therapy. *Orv Hetil.* 2011, 152: 1284-1293.
14. **Mátrai Z**, Tóth L, Saeki T, Sinkovics I, Gödény M, Takeuchi H, Bidlek M, Bartal A, Sávolt A, Dorogi B, Kásler M The potential role of SPECT/CT in the preoperative detection of sentinel lymph nodes in breast cancer. *Orv Hetil.* 2011, 152: 678-688. (*Lajos Markusovszky Price 2011*)
15. **Mátrai Z**, Tóth L, Polgár C, Láng I, Gödény M, Sinkovics I, Horváth Z, Bidlek M, Udvarhelyi N, Bartal A, Sávolt A, Ujhelyi M, Kásler M; Sentinel lymph node biopsy after neoadjuvant chemotherapy in breast cancer. *Magy Onkol.* 2011, 8: 73-84.
16. **Mátrai Z**, Tóth L, Saeki T, Sinkovics I, Gödény M, Takeuchi H, Bidlek M, Bartal A, Sávolt A, Dorogi B, Kásler M The potential role of SPECT/CT in the preoperative detection of sentinel lymph nodes in breast cancer. *Orv Hetil.* 2011, 24: 678-688.
17. **Mátrai Z**, Gulyás G, Tóth L, Sávolt A, Bidlek M, Szabó E, Pesthy P, Kunos C, Bartal A, Kásler M Breast conserving oncoplastic surgery in the treatment of centrally located malignant breast tumors. *Orv Hetil.* 2010, 151: 2105-2112.
18. **Mátrai Z**, Langmár Z, Szabó E, Rényi-Vámos F, Bartal A, Orosz Z, Németh M, Tóth L Granular cell tumour of the breast: case series and review of the literature. *Eur J Gynaecol Oncol.* 2010, 31: 636-640. **IF: 0.633**
19. **Mátrai Z**, Liskay G, Plotár V, Orosz Z, Székely J, Hitre E, Bartal A, Langmár Z, Bocs K, Rényi Vámos F, Sávolt Á, Tóth L Long-term experience with multidisciplinary therapy of twenty-six patients with dermatofibrosarcoma protuberans. *Orv Hetil.* 2009, 150: 1894-1902.
20. **Mátrai Z**, Plotár V, Liskay G, Fejős Z, Vámosi A, Dubóczky Z, Rényi Vámos F, Vámosi Nagy I, Köves I, Bartal A, Schmidt E, Tóth L Recurrent acral lentiginous melanoma successfully treated with Mohs' micrographic surgery. Case report and review of the literature. *Orv Hetil.* 2009, 150: 1071-1082.
21. Góbor L, Czeyda-Pommersheim F, **Mátrai Z**, Köves I Rectum carcinomas treated with Kraske procedure in elderly patients. *Magy Seb.* 2007, 60: 289-292.
22. **Mátrai Z**, Péley G, Kovács T, Rényi VF, Szivós E, Szabó E, Vereczkey I, Török K, Köves I Cytoreductive surgery and intraperitoneal hyperthermic chemoperfusion for peritoneal carcinomatosis caused by recurrent inflammatory myofibroblastic sarcoma: a case report and review of the literature. *Orv Hetil.* 2006, 147: 147-158.

23. **Matrai Z**, Lővey J, Hitre E, Orosz Z, Gödény M, Péley G, Sulyok Z, Rényi Vámos F, Farkas E, Köves I Histologic response after neoadjuvant therapy in rectal adenocarcinoma: own experience and review of the literature. *Orv Hetil.* 2006, 147: 2011-2020.
24. Török K, Péley G, **Mátrai Z**, Bidlek M, Szabó É, Sinkovics I, Polgár C, Farkas E, Orosz Z, Köves I The role of sentinel lymph node biopsy in breast cancer non-invasive-duct. *Magy Seb.* 2006, 59: 173-178.
25. Farkas E, Péley G, **Mátrai Z**, Rényi-Vámos F, Köves I Administration of clexane 4000 U/0.4 ml (40 mg) for preventing venous thromboembolic complications in patients undergoing surgery for malignant tumors. A follow-up study. *Magy Seb.* 2005, 58: 197-200.
26. **Mátrai Z**, Fehér I, Péley G, Rényi Vámos F, Farkas E, Sulyok Z, Kovács T, Köves I Experience with radiofrequency ablation in the treatment of unresectable pelvic recurrence of rectal cancer. *Magy Seb.* 2005, 58: 21-27.
27. Péley G, Farkas E, **Mátrai Z**, Rényi-Vámos F, Kovács T, Köves I The role of surgery in the treatment of malignant melanoma. *Magy Seb.* 2003, 47: 69-77.

QUOTABLE CONGRESS ABSTRACTS

1. **Mátrai Z**, Tóth L., Szentirmay Z., Rényi Vámos F., Klepetko W., Vadász P., Kenessey I., Kásler M. Sporadic desmoid tumors of the chest. Long term follow-up of twenty-eight multimodally treated patients. ECCO 16 – 36th ESMO Multidisciplinary Congress, Stockholm, Sweden *Eur. J. Cancer* 2011, 47: 667 (poster presentation)
2. **Mátrai Z**, Péter I., Tóth L., Csernák E., Antal I., Sávolt Á., Bartal A., Kásler M. High frequency estrogen receptor beta expression in sporadic aggressive fibromatosis. Immunohistochemical analysis of 57 cases. The 4th Conference of Phd Students and the First Conference of Postdoctoral Researchers in Medicine and Pharmacy, Targu Mures, Romania *Acta Medicina Maresiensis* 2011, 57: 4 (oral presentation)
3. **Mátrai Z**. Desmoid tumor associated with a ruptured silicone breast implant. A case report. International Oncoplastic Breast Surgery Symposium, Tokyo, Japan. 2010 Abstract book P25-20 (poster presentation)
4. **Mátrai Z**, Tóth L., Rényi-Vámos F., Szentirmay Z., Papp J., Klepetko W., Szentirmay M., Vadász P., Kenessey I., Sávolt Á., Bartal A., Kásler M. Sporadic desmoid tumors of the chest. Long term follow-up of twenty-eight multimodally treated patients. 4th Scientific Meeting of the Japan-Hungarian Surgical Society, Yokohama, Japan. 2010 Abstract book P94 O10-4 (oral presentation)
5. **Mátrai Z**, Tóth L., Gulyás G., Szabó É., Szentirmay Z., Papp J., Bartal A., Kásler M. Desmoid tumor associated with a ruptured silicone breast implant. The 60th Congress of the Hungarian Surgical Society, Siófok, Hungary. *Magy. Seb.* 2010, 63: 214 (oral presentation)
6. **Mátrai Z**, Orosz Zs., Papp J., Vadász P., Szendrői M., Rahóty P., Pápai Zs., Tóth L. Long-term results of the surgical and combined treatment of sporadic desmoid tumors of the thoracic wall: a multicenter, retrospective trial with 28 patients”, The XXVIII. Congress of the Hungarian Society of Oncology, Budapest, Hungary. *Magy. Onkol.* 2009, 53: 77. (poster presentation) (awarded 3rd place)
7. **Mátrai Z**, Orosz Zs., Oláh E., Papp J., Csekeő A., Vadász P., Szendrői M., Rahóty P., Pápai Zs., Rényi Vámos F., Köves I. Long term follow-up of sporadic desmoid tumors of the thorax. A multicentric retrospective analysis of 25 patients. 59th Congress of the Hungarian Surgical Society, Debrecen, Hungary. *Magy. Seb.* 2008, 61: 176 (oral presentation)

8. **Mátrai Z.**, Papp J., Péter I., Szendrői M., Rahóty P., Pápai Zs., Köves I., Oláh E. Clinical follow-up of desmoid tumors and the impact of APC gene mutations. ECCO 14 The European Cancer Conference, Barcelona, Spain *Eur. J. Cancer* 2007, 5, 404. (poster presentation)
9. **Mátrai Z.**, Papp J., Péter I., Szendrői M., Rahóty P., Pápai Zs., Sáy Z., Oláh E., Köves I. Clinico-pathological follow up of desmoid tumor patients and the impact of APC gene mutations. The XXth Congress of Coloproctological Section of the Hungarian Surgical Society and Joint Meeting of the European Federation for Colorectal Cancer, Debrecen, Hungary. 2007 Abstract book P34 (oral presentation)
10. **Mátrai Z.**, Papp J., Péter I., Szendrői M., Rahóty P., Pápai Zs., Oláh E., Köves I. Clinical follow-up of desmoid tumors and the impact of APC gene mutations. 19th Meeting of European Association for Cancer Research EACR, Budapest, Hungary. 2006 Proceedings of EACR P206 Poster 342 (poster presentation)
11. **Mátrai Z.**, Papp J., Péter I., Szendrői M., Rahóty P., Pápai Zs., Oláh E., Köves I. Clinico-pathological follow up of desmoid tumor patients and the impact of APC gene mutations. 58th Anniversary Congress of the Hungarian Surgical Society, Budapest, Hungary. *Magy. Seb.* 2006, 59: 270. (oral presentation)

1. INTRODUCTION

1.1. Epidemiology

The World Health Organization defines desmoid tumors as „clonal fibroblastic proliferations that arise in the deep soft tissues and are characterized by infiltrative growth and a tendency toward local recurrence but an inability to metastasize”. Regarding the biological background of the tumor, it is classified between benign fibrous tissue proliferation and fibrosarcoma. Desmoids are rarely occurring tumors which account for 0.03% of all neoplasms. The estimated annual incidence in the general population is 2–4 per million. The peak incidence of the tumor is between 25 and 40 years of age. Two different types have been described: most of them belong to the sporadically occurring type (95%) and the rest are associated with hereditary cancer syndromes like the autosomal dominant familial adenomatous polyposis (FAP, Gardner) syndrome. The sporadic types are more common in women than in men with a ratio between 1.8 : 1 and 5 : 1.

1.2. Clinical presentation

Desmoids develop from musculoaponeurotic structures throughout the body and are classified as extra-abdominal, abdominal and intra-abdominal. The clinical manifestation of desmoids is non-specific. Little or no pain accompanies extra-abdominal fibromatoses which typically arise as firm, poorly circumscribed, deep-seated, furtively grown masses. Tumors are fixed to the musculoaponeurotic plane and are usually free in relation to the bone, and joint capsule, only rarely adhering to these. Muscular retractions, deformity, limited or lost joint functions or even the lifethreatening compression of vital organs may be caused when the tumour reaches a large size. When a desmoid is diagnosed, a thorough family history is necessary to be taken, examination, genetic counseling and colonoscopy should be performed in order to outrule Gardner's syndrome.

1.3. Clinical behaviour

The clinical behavior and natural history of desmoids is typically heterogeneous and unpredictable and is characterised not only by tumor growth, proliferation and disease progression but also by stabilization and spontaneous remission. Most desmoids are however slowly growing neoplasms, they aggressively invade surrounding tissues and organs, do not metastasize but bear a high propensity (20% to 85% in 10 years) for local recurrence. Despite their benign nature, the tendency to recur and the infiltrative growth remain significant problems in terms of morbidity and even mortality.

1.4. Etiology

Some authors considered them as non-neoplastic processes and others described desmoids as well-differentiated low grade sarcomas. In fact, the etiology is likely multifactorial and includes genetic, endocrine and physical factors as well.

1.5. The neoplastic nature of desmoids

Multiple studies described the clonal nature of desmoids pointing at malignant capacities that place these tumors into the category of fibroblastic malignancies. The understanding of the molecular etiology of desmoids is mandatory. A common feature detected is deregulated Wnt signaling via β -catenin–dependent activation of latent T-cell factor/lymphoid enhancer factor (Tcf/Lef), a pathway with a critical role for instance in embryogenesis, cell adhesion, carcinogenesis, in adult stem cell survival and self-renewal during wound healing. Desmoids arising in patients with FAP show loss of adenomatous polyposis coli (APC) tumor suppressor function, which leads to high intracellular β -catenin levels and is correlated with constitutive activation of Wnt signaling. In sporadic desmoids most tumors contain specific

point mutations in the catenin beta 1 (CTNNB1) gene, that stabilize β -catenin and achieve a similar result. Several mutations of the APC gene in sporadic desmoids lacking β -catenin gene mutations have been reported and approximately 95% of these lead to expression of a truncated protein. It has been suggested, therefore, that the higher β -catenin protein levels are consequences of the APC gene truncation in the absence of β -catenin mutations. It is proposed that APC mutations cause inadequate regulation of β -catenin activation, leading to proliferation on a cellular level, and the location of the APC or β -catenin mutation dictates the phenotype of desmoid tumor expression.

1.6. The endocrine etiology of desmoids

A possibility of functional interaction between estrogen receptor (ER) and Wnt/ β -catenin signaling has recently been reported in human tumor cell lines and in *Drosophila*, in which estrogen signaling appears to potentiate the effects of nuclear β -catenin, though evidence for interaction of the ER pathway with the APC/ β -catenin pathway is circumstantial.

1.7. Histology

The macroscopical appearance of desmoid tumors is firm and rubbery, and characterised by a relatively homogenous incisional surface of white and greyish network of bundles resembling scar tissue with a relatively poor vascularization. At microscopical presentation a proliferation of elongated, slender, spindle-shaped cells of uniform appearance, arranged in fascicles is seen, sometimes with perivascular oedema. Immunohistochemistry (IHC) is positive for vimentin, alpha smooth muscle actin, muscle actin, and desmin muscle cell markers. Nuclear β -catenin immunoreactivity is staining positive in 67%–80% of cases in the reported series. The IHC studies on hormone receptor status showing uniform negative for ER- α and progesterone receptor, but 7.4% to 100% positive for ER- β .

1.8. Treatment of desmoid tumors

The optimal therapy for desmoids is difficult to be established due to the heterogeneous clinical behavior and anatomical presentation. There is limited or no evidence based medicine for these type of tumors. The general recommendations for the efficacious management of desmoids consist of surgical and nonsurgical modalities and require a multidisciplinary approach. The treatment should be decided on case-by-case basis by the multidisciplinary soft tissue board.

Close observation is an acceptable strategy for asymptomatic patients with small desmoid tumors not infiltrating any nearby structures or for patients of stable disease presenting with few, mild symptoms.

If the preoperative diagnostics finds that a desmoid tumor is technically resectable, aggressive surgery is the first line treatment. To date wide excision with 2 to 4 cm resection margins is generally recognized as the most effective treatment for desmoids. Complete resection of the lesion with negative microscopic surgical margins (R0 resection) is the standard surgical goal, however, it is not always possible due to anatomic boundaries. The overall strategy should be to attempt complete removal using function-preserving approaches to minimize major functional and cosmetic morbidity. Incomplete resection is associated with a higher risk of local recurrence. The importance of a negative surgical margin is thus recently debated. Radiation therapy (RT) is considered an effective option in the therapy of desmoid tumors, that may improve local control. The role of adjuvant RT in achieving local control has not yet been clearly defined and is still controversial. Postoperative RT is considered only in patients with a large tumor and positive margins as suggested in the National Comprehensive Cancer Network (NCCN) guidelines.

For patients with positive resection margins, advanced desmoids that are not amenable to surgery or RT, various medical treatment options including antihormonal therapies, non-steroidal anti-inflammatory drugs (NSAIDs), targeted therapies, and traditional cytotoxic chemotherapies have been investigated.

1.9. Posttreatment surveillance

Given the increased potential for recurrence, regular clinical and possible imaging follow-up have been strongly recommended after therapy. Consensus-based guidelines from the NCCN suggest that a history and physical examination with appropriate imaging (preferably MRI) every 3–6 months for 2–3 years and then annually.

AIMS OF THE THESIS

There is a relative paucity of clinical and pathological data available on sporadic desmoid tumors. As the data found in the English language literature are mostly based on the meta-analysis of case reports or series, or retrospective studies with low number of cases, a global lack of knowledge is recognised regarding risk factors, multidisciplinary treatment modalities, prospective and predictive factors and long-term follow-up results.

Given these facts, the main aims of our investigations were as follows:

1st aim. To establish a data base of sporadic desmoid tumors adequate for analytical purposes and, to collect clinico-pathological data on long term follow-up in order to find scientific answers to the questions of etiology, pathology and clinical behaviour.

2nd aim. On the basis of a detailed clinico-pathological database the first planned step was to confirm the sporadic origin of desmoids by ruling out the FAP associated germ line mutation of the APC gene by clinical and genetic investigations.

3rd aim. To confirm the theory of the etiology of sporadic desmoids, by detecting the β -catenin mutation status with molecular-genetical probes in a large number of tissue samples, quilting as one of the largest single cohort reported in the literature.

4th aim. To analyse and compare the clinical and pathological data of the very rare thoracic as well as the more common abdominal tumors and desmoids located on the extremities.

a. Radicality of first surgery and its influence on local control.

b. The analysis of the surgical techniques and the reconstructive methods after wide soft tissue resections (e.g. long time results with mesh reconstructions after the resection of abdominal desmoids).

c. The correlation of genotyping results with clinical and immunohistochemical features, including time to recurrence and β -catenin protein expression pattern and intensity.

d. To establish possible prospective and predictive factors on the basis of the correlation of genotyping and long-time clinical course.

5th aim. Assessment of the expression of ERs and PR in sporadic desmoid tumors by immunohistochemical analysis using anti-ER- α , anti-ER- β and anti-PR antibodies.

a. The comparison of the clinical tumor status with the adherent ER- β status of patients following endocrine treatment.

MATERIALS AND METHODS

1. The creation of a cohort of sporadic desmoid cases and a data base

With the approval of the Institutional Ethical Board, a cohort of desmoid cases was collected by multicentric way, using the soft tissue tumor database of the National Institute of Oncology (NIO), the Semmelweis University Orthopaedic Clinic and the Thoracic Surgical Clinic of Koranyi National Institute of Pulmonology. The cases were multidisciplinary treated between 1982 and 2011. The medical records and tissue samples were collected by personal research

of the archives of the institutes. All patients were invited for a face to face consultation and clinical investigation. Clinical information including demographic, therapeutic, tumor, and clinical outcome variables were retrieved and tabulated for correlative analyses. Occasional (28 cases) or regular follow-ups (69 cases) were realized by 97 patients in the NIO with the successful completion of the clinical and anamnestic database. Regular follow-ups were performed as recommended in the literature. The tissue samples of all cases were officially acquired from all three pathological departments and histologically revised by an independent reference pathologists in the NIO. Demographic and clinical data were gathered. Therapeutic data acquired consist of the type and number of surgical interventions, surgical margins, reconstructive plastic surgical options, perioperative morbidity, mortality, adjuvant therapy administered and its efficacy using the RECIST 1.1. criteria. Pathological data consist of the pathological tumor size, the circumferential microscopical surgical margin, results of IHC assays. The database is continuously updated enabling long term follow-up and analytical comparison of the data. A limitation of the study was the principally retrospective collection of data, the incidental low-quality of a few decade-old pathological tissue samples, and the low number of cases preventing a reliable statistical comparison of subsets of patients. Although during the nearly 30 years of patient recruiting period, surgical resection and non-surgical therapies as first choice of treatment have not or only minimally (imatinib) changed, a further limitation was proposed by the non-controlled therapies.

2. Confirmation of the sporadical origin of desmoid tumors by ruling out the germ line mutation of APC gene.

All participants were asked for detailed family history, and a three-generation family tree was delineated. Medical records for family members who have developed colon polyps and/or underwent colon surgery were requested, and took a survey in order to evaluate the presence of FAP. Colonoscopy was performed or former medical records were checked for all the patients. Thirty-nine individuals included in this study were referred to genetic counseling and testing to the Department of Molecular Genetics at the NIO. All investigations have been performed in agreement with international guidelines, the study protocols were approved by the Institutional Ethical Board. Written informed consent was signed by each patient. DNA was extracted from peripheral blood samples of all consenting subjects using the classic phenol-chloroform method. The entire coding region and splice junctions of the APC gene were amplified by polymerase chain reaction (PCR). Mutation pre-screening was done on the resulting PCR amplicons using single-strand conformation polymorphism/heteroduplex analysis technique (SSCP/HDA). Bands on gels were visualized by silver staining. Where altered mobility of a sample indicated the presence of a variant, the fragment was subjected to direct bidirectional DNA sequencing. Completed sequencing reactions were electrophoresed. The presence of mutations was confirmed using a different blood sample.

3. Detection of the β -catenin mutation status.

Only sporadic desmoid tumors were evaluated. As tissue samples were only partly suitable in quality for molecular-genetic investigations, an amount of 58 specimens from 51 patients were found to be eligible for the β -catenin mutation analysis. The samples from formalin-fixed, paraffin embedded tissue sections were analysed for CTNNB1 gene mutation status with locked nucleic acid (LNA) probe-based real-time (PCR) followed by melting curve analysis.

4. Three groups were formed following the analysis and comparison of clinical and pathological data: sporadical desmoid tumors of the chest, abdominal wall tumors and fibromatoses originating in the extremities.

Dividing the sporadic desmoid cases into 3 different groups seemed a reasonable option, based on the diversity of the surgical techniques, radicality and methods of tissue reconstruction according to the location of the tumors of the available 94 sporadic cases in the database. The therapeutic protocol for desmoids was invariable during the investigated period. First-line treatment was wide surgical resections. The radicality of the surgery was considered to be R0 if the circumferential surgical margins were microscopically negative, R1 and R2 if microscopic residual tumor and gross residual disease respectively was left behind. The largest diameter of the tumor and the status of the surgical margin were defined by the pathological befund. All operations were performed under antibiotic prophylaxis with amoxicillin or cefazolin (a single intravenous shot of 1000 mg of amoxicillin and 200 mg of clavulanic acid or 1000 mg of cefazolin), or continued until the suction drains were removed over the implanted mesh graft. According to the applied protocol, patients exposed to the potential of major complication (intra-thoracic location or lesion adjacent to joint capsule or great vessels) and those with R1 resections received 20 mg of oral tamoxifen daily, otherwise close surveillance was implemented. Higher dose of tamoxifen (120 mg daily) and sulindac (150 mg/twice daily) were administered to patients with unresectable lesions or R2 resections. In cases of recurrence, a daily dose of 20 to 120 mg tamoxifen and 150 to 300 mg sulindac were administered. The therapy of high dose tamoxifen in combination with sulindac was continued for a maximum of 24 months, followed by maintenance daily doses of 20 mg of tamoxifen and 150 mg of sulindac. Patients who received sulindac were under regular cardiological control. Radiotherapy was indicated in aggressive cases showing rapid progression in an adjuvant setting or in irresectable cases showing tumor progression. The dose of RT for definitive therapy was 50–60 Gy in 5–7 weeks at 1.8–2 Gy per fraction, with wide radiation field margins of at least 5 cm applied in the direction of possible infiltrative growth. Doxorubicin based chemotherapy was administered in a single advanced case locating in the neck with life-threatening morbidity. Operative morbidity was recorded within the first 30 days after surgery or during the same hospitalization. There was no record of malignant disease in the histories of the 94 investigated patients. The soft tissue follow-up protocol of the participating institutes corresponded with the international recommendations. The investigated cases were analyzed for family history, age, gender, previous trauma to the site of the tumor, size and site of the tumor, radicality of surgeries, results of IHC assays, reconstructive plastic surgeries, perioperative morbidity, number of recurrences, time to recurrences, and adjuvant therapeutic modalities. For all patients, analysis of time to recurrence was calculated from the first surgery of primary lesions to the diagnosis of the first recurrence.

5. Assessment of the expression of ERs and PRs in sporadic desmoid tumors by immunohistochemical assays.

Histological samples of sporadically occurring primary desmoids acquired from 67 patients were eligible for the IHC investigations. Each diagnosis was reviewed and confirmed based on the relevant WHO criteria. Normal and neoplastic human breast tissue from the surgical pathology laboratory were used as positive control tests. Normal mouse or rabbit sera were used instead of the first antibodies in negative control tests. Classification as positive was done either according to company guidelines or institutional standard protocols for the positive controls: a minimum of 10% of the tumor cells had to be positive for ER- α , ER- β or progesteron receptor. Only definite nuclear staining was regarded as positive and cases were scored by the percentage of tumor cells staining as 1+ (<10%), 2+ (10-50%), or 3+ (>50%). Normal breast tissue served as control for ER- α , and benign fibroadenoma served as positive control for ER- β , and were run parallel. The staining on the entire array was performed at the clinical IHC facility of NIO on two occasions and each was scored independently.

6. Statistical methods

Categorical data were compared using Fisher's exact probability test and chi-square test. Recurrence free survival analyses were done using the Kaplan—Meier method. Recurrence free survival intervals were determined as the time period from initial diagnosis to the time of first recurrence. Univariate analysis of potential factors affecting recurrence-free survival (gender, age, tumor size, radicality of the first surgery, and impact of the pharmacologic treatment) was performed using Cox-regression hazard model. Differences were considered to be statistically significant when $p < 0.05$. Statistica 8.0 (StatSoft, Tulsa, OK) was used to perform all of the statistical calculations.

RESULTS

1. The successful gathering of a cohort of sporadic desmoid cases and creation of a database

A cohort of altogether 97 desmoid cases was collected by the investigator. Fifty-seven cases were primarily treated in the NIO, 38 cases at the Semmelweis University Orthopaedic Clinic and 2 cases at the Thoracic Surgical Clinic of Koranyi National Institute of Pulmonology. Three cases were excluded from the investigation due to confirmed hereditary origin. In all the cases the personal attendance of the patient, family and personal anamnesis as well physical investigation and completion of the medical records were successfully achieved. The 94 sporadic desmoid cases represented 66 female and 28 male patients. The median age of the patients was 31 years (range, 9—74 years) and fifty-three female patients (80.3%) were premenopausal. Physical, often surgical trauma to the site of the lesion preceded the development of desmoids in 26 patients (28%). Desmoids were located on the region of the chest in 28 patients, in the abdominal wall in 28 patients and in the extremities in 38 patients. As for the surgical radicality of the primary tumor, 52 cases (55.5%) resulted in R0, 39 cases (41.5%) in R1 and 3 cases (3%) in R2 resections. A reconstructive surgical procedure was performed in 54 cases (57%). Perioperative morbidity was detected in 19 cases (20%), while perioperative mortality didn't occur. The median pathological diameter of the primary tumors was 61 mm (range, 20—250 mm). Adjuvant therapy was commenced for 50 patients (53.2%), while 44 patients (46.8%) did not need adjuvant treatment. The non-surgical modalities in the adjuvant setting varied greatly. Tamoxifen monotherapy in 20 cases (40%), tamoxifen in combination with sulindac in 17 cases (34%) including one patient who received imatinib too. Sulindac monotherapy was chosen for 2 patients (4%). Ten patients (20%) received adjuvant RT, 8 cases (16%) in combination with tamoxifen, complemented with sulindac in one case, and with chemotherapy in another one. Tamoxifen was administered to 48 out of the 50 adjuvant cases (96%). Follow-up was completed in all investigated patients and ranged from 8 to 336 months, with a median of 98 months. During the follow-up period a total of 122 local recurrence happened. Taking the 3 cases with R2 resection into consideration, among the 91 cases with R0 or R1 resection 38 cases (41.8%) were recurrence free and local recurrence occurred in 53 cases (58.2%). Among the 91 cases with R0 or R1 resection 38 cases (41.8%) were recurrence free and local recurrence occurred in 53 cases (58.2%). Twenty-two cases developed one, 13 cases two and 18 cases 3 or more local recurrences. Surgical resection due to a recurrent tumor was performed 105 times. The radicality (R0 vs. R1 or R2) of the resections decreased significantly ($P=0.0016$) from the 2nd recurrence on.

2. Confirmation of the sporadic origin by ruling out germ line mutations of the APC gene by clinical and genetic investigations.

Three patients were found to carry a deleterious APC mutation in the screened section of the gene and were subsequently excluded from the further investigations. All of these patients

were diagnosed with both desmoid tumors and prominent colorectal polyposis. The three patients with hereditary desmoids were excluded from the further investigations. At the last follow-up only one of them was alive. The remaining 94 cases were confirmed as sporadic desmoids. None of them had a positive family history, nor developed polyposis controlled by colonoscopy during the follow-up period. In conformity with the literature, our investigation approved the reliability and cost effectivity of genetic testing the APC gene.

3. The detection of the β -catenin mutation status.

3.1. CTNNB1 Mutations Are Highly Prevalent in Sporadic Desmoid Tumors

Fifty-eight tissue samples from 51 patients were eligible for evaluating the prevalence of CTNNB1 mutations. The database provided appropriate clinical information in relation to the 35 female (68.6%) and 16 male (31.4%) patients. Median age was 33 years and ranged from 8 to 66 years. The following sites were involved: chest in 13 cases (25.5%), abdominal wall in 17 cases (33.3%) and extremities in cases 21 (41.2%). Tumors presented with variable size ranging from 20 to 240 mm (median, 83 mm). A mutation was identified in 30 (58.8%) of the 51 primary tumors. The CTNNB1 exon 3 mutation profile in tissue samples taken from recurrent desmoids were 100% identical with the mutation profile of the primary tumor of the same patients. Female patients were more likely than male patients (60% versus 50%) to have a mutation but the difference didn't prove to be significant ($P=0.387$). Next the CTNNB1 mutational spectra was determined. Interestingly, only 3 different point mutations in 2 different codons (41 and 45) could be identified in all mutated samples ($n=30$ of 51 total): ACC to GCC in codon 41 (41A), resulting in the replacement of threonine by alanine, was identified in 15 samples (50%); TCT to TTT in codon 45 (45F), resulting in the replacement of serine by phenylalanine, was identified in 12 cases (40%); TCT to CCT in codon 45 (45P), resulting in the replacement of serine with proline, was identified in 1 sample (3.3%) and a deletion was identified in 2 cases (6.7%). Upon analyzing the incidence of the specific CTNNB1 gene mutations and their correlation to patient and tumor variables, a higher incidence of the 41A and 45F mutation was identified in females versus males (75% versus 25%, and 73.3% versus 26.7% $P=0.922$). In terms of tumor site, lesions involving the extremities showed a significantly higher incidence of the 45F mutation (58.3%, $P=0.018$) and tumors originating in the abdominal wall showed a significantly higher incidence for 41A mutations (60%, $P=0.037$) versus all other sites.

3.2. CTNNB1 45F Mutations Significantly Correlate with Increased Desmoid Recurrence

The investigator assessed the correlation of the mutation of any specific CTNNB1 gene with clinical outcome. The different CTNNB1 gene mutational cohorts were found to present with different recurrence-free survival values, which was significantly inferior for patients with the 45F genotype ($P=0.008$) (Cox-regression). Only one sample was found to harbor a 45P CTNNB1 gene mutation, and so this tumor was excluded from analysis. Younger age (<30) at diagnosis as an increased propensity for local recurrence was further identified by univariate analysis ($P=0.002$). Other parameters just as gender, tumor sites (chest vs. abdominal wall vs. extremities) and tumor size did not show significant difference. Multivariate analysis was not performed due to the low number of cases. The 45F CTNNB1 gene mutation-harboring tumors demonstrated significant difference to WT tumors in comparison with local recurrence rate using the log-rank analysis ($P=0.009$).

4. The analysis and comparison of the clinical and pathological data of the sporadic desmoids in the three subgroups.

4.1. Sporadical desmoid tumors of the chest

There were 19 female and 9 male patients with a median age of 35 years (range, 11—74 years). As many as 64% of the tumors originated in the shoulder girdle. Upon the pathological examination of the primary specimens, an R0 excision was achieved in only 50% of the patients. Of the 27 macroscopically wide surgical excisions, 10 cases (37%) were full-thickness and 17 cases were (63%) partial-thickness chest wall resections. Partial or complete scapula and clavicle resections were performed in six and four cases respectively. The disease of 3 patients necessitated compartment resection. The median pathological diameter of the resected tumors was 50 mm (range, 20—210 mm). Nine patients had their chest walls stabilized by synthetic meshes and no detectable implant complications occurred. Pleural involvement indicated wedge resection of the lung in 3 patients. Soft tissue coverage was achieved with transposed muscles in six patients. The rate of complications was 25%. No operative deaths occurred. In the subsequent follow-ups, first recurrences were noted in 17 (63%) of the 27 resectable (in both R0 and R1) patients while 10 patients (37%) remained recurrence free. The median time to first recurrences was 30 months (range, 10—120 months). Altogether, 35 surgeries were performed on recurrences. One patient died of stroke before the end of the follow-up, another patient was in a weak general condition due to an extensive desmoid in the shoulder girdle, and superior vena cava syndrome was diagnosed in yet another case. One patient needed neurosurgical intervention and stabilization of the spinal. The median duration of anti-estrogen therapy in 18 patients was 68 months (range, 10—171 months) and the median length of tamoxifen and sulindac combination therapy in 12 patients was 48 months (range, 10—118 months). ER- β positivity was observed in 50% of the samples. RT was applied in 2 patients with recurrent tumors. Both lesions were observed to recur and progress 9 and 14 months after treatment, respectively. A total of 16 patients (57%) were found to be recurrence free, whereas residual or recurrent tumors were detected in 12 patients at the time of their last follow-up examination. Statistical analyses of the categorical data such as gender, age, tumor size, ER status, and sulindac treatment using Fisher's exact probability test and the chi-square test established that these factors did not correlate with surgical radicality. Since tamoxifen was not an independent factor, it could not be used in the multivariate analysis. However, local recurrence rate was significantly affected by the surgical radicality of the primary tumor ($P=0.00065$) and sulindac treatment ($P=0.029$) in the univariate analysis by log-rank statistics. Factors that were not significant included gender, age, tumor size, and ER positivity. The significant difference on the factors affected by the recurrence-free survival group was demonstrated by Kaplan—Meier curves. The multivariate Cox regression model confirmed, that the local recurrence rate was only significantly affected by the surgical radicality of the primary tumor ($P=0.001$).

4.2. Sporadical desmoid tumors of the abdominal wall.

Twenty-three female and 5 male patients belonged to this subgroup with a median age of 29 (range, 19—59 years). Antedescendent surgical or physical trauma to the site of the tumor was identified in 7 patients (25%). Primary tumors were located in the anterior abdominal wall in 100%, including the inguinal region in 4 cases. Pathological results of the primary specimens showed that an R0 excision was achieved in 18 cases (64%). Of the 28 macroscopically wide surgical excisions, 18 cases (64%) were full-thickness and 10 cases were (36%) partial-thickness abdominal wall resections. The median pathological diameter of the resected tumors was 63 mm (range, 20—180 mm). Synthetic meshes were utilised for the stabilisation of the abdominal wall in all the 28 cases. The applied antibiotic prophylaxis ensured the lack of detectable implant complications, only a superficial wound infection localised to the subcutis was detected. Soft tissue coverage with transposed musculocutaneous flaps was not necessary to perform. No perioperative deaths occurred. The rate of complications was 10.5%. First

recurrences were recorded in 10 (35.7%) of the 28 resectable (R0 and R1) patients during the subsequent follow-ups. The follow-ups were completed in all of the investigated patients. Eighteen patients (64%) were found to be recurrence free. The median time to first recurrences was 26 months (range, 6—110 months). Altogether, 17 surgical interventions were performed due to recurrences. All patients were alive and tumor free at the last follow-up. Abdominal wall hernias developed in 3 patients, and in one case mesh bulging was confirmed. The median duration of tamoxifen therapy in 11 patients was 25 months (range, 8—36 months). Four patients received tamoxifen and sulindac combination therapy for the median length 20 months (range, 8—25 months). ER- β positivity was observed in 13 cases (46.4%) by IHC analysis. All the patients were found to be recurrence free at the time of last checkup. Statistical analyses of the categorical data (gender, age, tumor size, ER status) using Fisher's exact probability test and the chi-square test determined that these categories did not correlate with surgical radicality. Because of the low number of sulindac use, the cases using tamoxifen and sulindac were summed up. The significance of tamoxifen and sulindac ($p=7.96*10^{-4}$) is explained by the adjustment for R1-resected cases and for recurrent tumors, corresponding to our adjuvant protocol. As a result, tamoxifen was not an independent factor and, it could not be used in the multivariate analysis. In the univariate analysis by Cox-regression model, the local recurrence rate was significantly affected by the surgical radicality of the primary tumor ($P=0.006$). Factors that were not significant included gender, age, tumor size, and ER positivity. The significant difference on the factor affected by the recurrence-free survival group was demonstrated by Kaplan—Meier curves.

4.3. Sporadical desmoid tumors of the extremities.

The subgroup included 24 female and 14 male patients with a median age of 30 years (range, 9—59 years). Eleven tumors (29%) were located in the upper extremity, while 27 (71%) tumors in the lower extremities. Regarding the pathological results of the primary specimens, an R0 excision was achieved in only 19 cases (50%). Macroscopically wide surgical resection was performed in 32 cases (84%) whereas 4 patients (10.5%) underwent compartment resection of the buttock resection, adductor compartment, quadriceps and the entire brachioradialis muscle. The tumor of one patient originated in the popliteal fossa, thus the popliteal arteria had to be grafted. Although en block resection with additional bone resection was performed in 1 case, amputations were not performed. The median pathological diameter of the resected tumors was 70 mm (range, 25—250 mm). Soft tissue reconstruction was performed with transposed myocutaneous flaps in 6 patients (15%), and additional 4 patients needed full- or partial thickness skin graft coverage. No operative deaths occurred. The rate of perioperative complications was 23.7%. First recurrences were noted in 28 (77%) of the 36 radically resectable patients. The median time to first recurrences was 32 months (range, 6—108 months). Ten patients (26.3%) proved to be recurrence free. Altogether 52 surgeries were carried out on recurrences and the follow-ups were completed in all of the investigated patients. At the last follow-up all the patients were alive. Twenty-seven patients (71%) were disease free, and 4 patients had stable disease, while 7 patients had recurrent tumors with slow progression. Five patients complained about limitation of joint motion, 4 patients experienced neurological symptoms like loss of motoric functions, and 7 patients suffered from hypesthesia of the affected region or the distal part of the extremity was experienced. The median duration of anti-estrogen therapy in 19 patients was 43.2 months (range, 11—78 months). The median length of tamoxifen and sulindac combination therapy in 4 patients was 32 months (range, 9—68 months). ER- β positivity was observed in 12 cases (42.8%). RT was administered for 7 patients (18.2%). Five of them were observed to be tumor free while recurrence was found in 2 cases with a median of 21 months following treatment. A total of 27 patients (71%) were found to be recurrence free, whereas residual or recurrent tumors were

detected in 11 patients (29%) at the time of their last checkup. As many as 36.3% of the recurrences exhibited stable disease according to RECIST 1.1 criteria during the last 6 months of follow-up. However 63.7% of the tumors were characterized by a slow progression despite the ongoing multimodal therapy. Categorical data such as gender, age, tumor size, ER status did not correlate with surgical radicality determined by Fisher's exact probability test and the chi-square test. Because of the low number of cases treated with sulindac statistical analysis was unable to perform. The significance of tamoxifen ($P=0.007$) and RT ($P=0.001$) is explained by the adjustment for R1 or R2 resected cases, corresponding to our adjuvant protocol. Neither of these can be considered as an independent factor and thus, could not be used in the multivariate analysis. In the univariate analysis by Cox-regression model, the local recurrence rate was significantly affected by the surgical radicality of the primary tumor ($P=0.013$). Factors that were not significant included gender, age, tumor size and ER positivity. The significant difference on the factor affected by the recurrence-free survival group was demonstrated by Kaplan—Meier curves.

4.4 . Comparisons of the clinical results among the three cohorts of different tumor locations.

Significant differences were not detected among the three cohorts in terms of surgical radicality of the primary tumor (R0 versus R1 and R2) ($P=0.281$), gender ($P=0.236$), age ($P=0.201$), ER- β status ($P=0.122$) and tumor size ($P=0.435$).

The recurrence-free survival showed a decreasing trend among tumor locations as follows: abdominal, chest and extremities. Significance was confirmed using log-rank analysis between tumor locations 'abdominal' and 'extremity' ($P=0.0061$), while no significance was found between 'abdominal' versus 'chest' ($P=0.086$) and 'chest' versus 'extremities' ($P=0.253$).

5. Assessment of the expression of ERs and PRs in sporadic desmoid tumors by immunohistochemical assays.

The assessment included the tissue samples of 67 sporadic desmoid patients: 51 female (76.1%) and 16 (23.9%) male patients with a median age of 32 years (range 9-65 years). The tumor was located in the chest in 19 patients (28.35%), abdominally in 20 patients (29.85%) and in the extremities in 28 patients (41.8%). Stains for ER- α and PR were uniformly negative, whereas 36 cases (53.7%) displayed expression of ER- β : 3+ expression in 31 cases (86.1%), 2+ expression in 4 cases (11.1%) and 1+ expression in one case (2.8%). There was no significant correlation between the ER- β status and gender ($P=0.732$), age ($P=0.281$) or location ($P=0.289$) and size ($P=0.924$) of the tumor. R0 resection was achieved in 38 cases (56.7%) only. The median pathological tumor diameter of the investigated cases was 80 mm (range 25-210 mm). Thirty-two patients (47.7%) in the IHC screened group received adjuvant tamoxifen treatment. Fourteen of the 32 patients (43.75%) treated with tamoxifen and subjected to clinical assessment had ER- β positive tumors. At a median follow-up time of 110 months (range, 5 to 336) 43 cases (64.2%) had local recurrences, and no difference between the ER- β positive (12/14) and ER- β negative (18/18) patients treated with tamoxifen were detected ($P=0.184$).

DISCUSSION

1. Successful creation of a cohort of sporadic desmoid cases and a database

The relatively large number of patients with sporadic primary desmoids, the constant therapeutic modalities, and the long term clinical follow-up (median 98 months) together proved to be eligible for molecular-genetic and IHC investigations and scientific analysis of

the data. The database was registered to the national and the European databases of rare diseases and soft tissue tumors, to support additional scientific research of sporadic desmoids.

2. Confirmation of the sporadic origin of the desmoid tumors.

Testing for APC germline mutations in FAP families has become an efficient tool for predictive testing of subjects at possible risk and is now commercially available and has led to changes in management guidelines, particularly for those whose tests indicate they are not mutation carriers. If the germline mutation causing FAP is detected in a given family, carriers are selected who need to undergo surveillance by regular endoscopies. Presymptomatic genetic diagnosis of supposed FAP patients has been feasible with linkage and direct detection of APC mutations. Beside the desmoids in all 3 germline mutated cases other manifestations of FAP syndrome appeared such as multiple colon polyps or polyposis. After the exclusion of the FAP-associated cases, the remaining 94 patients with negative clinical and genetic tests and family anamnesis did not show any signs of FAP during the median follow-up time of 98 months. Colonoscopy was performed in every 5 years in the investigated population. Sporadically, polypectomies were performed, but multiple polyps or polyposis were not detected. On the bases of the above detailed experiences and results the author agrees that genetic testing together with accurate family anamnesis, physical investigation and colonoscopy is a reliable and non-invasive method to confirm FAP even without positive family anamnesis. It is imperative that FAP families be referred for genetic counselling, genetic testing, early diagnosis, and clinical management to centres that are aware of the complexity of both surgical intervention and possible implications of molecular findings for individual patients.

3. The detection of the β -catenin mutation status.

The present study identified CTNNB1 gene mutations in 58.8% of the investigated 51 cases. This finding further confirms the results of a recently published series, strongly suggesting that CTNNB1 mutations are highly prevalent in sporadic desmoids. In conformity with other publications we have also observed that desmoid CTNNB1 mutational spectra are rather limited. Interestingly, only three specific mutations are noted: 2 involving codon 45 and 1 involving codon 41, with all mutations clustered at Threonine 41 (41A) and Serine 45 (45F and 45P) in all topographies, suggesting that these residues have crucial functions in the β -catenin/Wnt signaling pathway. In our study we identified that risk of recurrence in desmoids is significantly associated with the specific CTNNB1 mutation of 45F ($P=0.022$). A larger than threefold increased risk of recurrence is found in tumors with this mutation, proposed to begin to be manifest at a significantly earlier time point in the desmoid tumor-host encounter. Our findings suggest that the biological consequences of the 3 different CTNNB1 mutations in exon 3 are not equivalent. Our study supports a role for 45F mutation as a prognostic factor strongly associated with recurrence. Due to the high propensity for recurrence of desmoids, the use of nonsurgical approaches as adjuvant strategies may be useful in preventing recurrence, especially in recurrence-prone cases such as the 45F mutated lesions. The causal relationship between the 45F mutation, β -catenin expression, and desmoid recurrence are still to be confirmed in further investigations. Our study suggests that genotyping of CTNNB1 exon 3 may provide information regarding risk of recurrence and personalised adjuvant therapies and as a diagnostic test is could be useful in situations such as insufficient core needle biopsies or differentiating a post-surgical scar from desmoid recurrence. Prospective trials are needed to test such possibilities.

4. Comparisons of the clinical results among the three cohorts of different tumor locations

According to our results we can conclude that the surgical radicality of the primary tumor served as a significant prognostic factor on local control rate. Another important conclusion is that the available surgical radicality significantly declines following the second local recurrence. Possible reasons for the phenomenon are proposed to be local anatomy, the surrounding postoperative scarry tissues and employed radiotherapy.

Among the three cohorts of different tumor locations, recurrence-free survival showed a decreasing trend as follows: 'abdominal', 'chest' and 'extremities', with a significantly better local control in 'abdominal' versus 'extremities' group. Significant difference was not detectable among the three cohorts in terms of the surgical radicality of the primary tumor, so the aforementioned can be explained by the significant occurrence of 45F and 41A CTNNB1 mutations in the groups of 'extremity' and 'abdominal'.

5. Assessment of the expression of ERs and PRs in sporadical desmoid tumors.

In general, it has been suggested that desmoids respond to the manipulation of ER signalling. ERs are members of the steroid/thyroid/retinoid hormone receptor superfamily of nuclear receptors, binding estrogen with high affinity. Binding to its ligand causes a change in receptor conformation that results in dimerisation and binding to specific promoter sequences of the DNA. The activated receptor/DNA complex then recruits other cofactors from the nucleus, which results in transcription of a protein, causing a change in cell function. Two main isoforms of the human ERs (ER- α and ER- β) have been identified. Recently IHC studies have found uniform ER- α negativity but high rate of expression of ER- β . In the present IHC study of 67 primary desmoids, 36 patients (53.7%) were found positive for ER- β . At a median follow-up time of 110 months no statistical difference between the ER- β positive and ER- β negative patients treated with tamoxifen were detected.

NEW STATEMENTS

- I. The multicentric creation of a cohort of primary sporadic desmoid tumors was succesfully carried out, and proved to be one of the ten largest in the English language literature, and the largest in Hungary and suitable for clinico-pathological investigations and to join international databases.
- II. Genetic testing of the APC gene is a reliable investigation together with family tree analysis and clinical investigations, and so sporadic cases were succesfully separated and followed-up; mutation of the CTNNB1 gene was detectable with high prevalence in sporadic desmoid tumors; mutation of the 45F gene is characteristic for higher recurrence rate, and serving as prognostic factor, the optimal choice of individualized adjuvant therapies is enabled.
- III. The cohort of sporadic desmoids of the chest is the second largest of its kind reported in the literature. Surgical radicality of the primary tumor was confirmed to significantly (P=0.001) determine the recurrence rate.
- IV. The cohort of the abdominal wall sporadic desmoids revealed that surgical radicality of the primary tumor as significant prognostic factor (P=0.006) is crucial for the disease-free survival.
- V. The recurrence rate in sporadic desmoids of the extremities was confirmed to be significantly (P=0.013) determined by the surgical radicality of the primary tumor as well.
- VI. Significant difference was not detectable among the three cohorts in terms of the surgical radicality of the primary tumor, but significance (P=0.0061) was confirmed between tumor locations 'abdominal' and 'extremity' in terms of the recurrence-free survival, which can be explained by the significant occurrence of 45F (P=0.018) and 41A (P=0.037) CTNNB1 mutations in the two groups.

- VII. The achieved surgical radicality of the re-resections is decreasing significantly ($P=0.0016$) from the 2nd recurrence, which must be taken into consideration by the management of the recurrences.
- VIII. Although a former universal statement existed that sporadic desmoid tumors are ER negative, our investigations confirmed a high rate of ER- β positivity.

ACKNOWLEDGEMENT

First of all I wish to thank my promoter, Professor Dr. Miklós Kásler, Ph.D., D.Sc., Dr.h.c. for his continuous support and encouragement throughout my Ph.D. studies. Furthermore, I wish to thank for his valuable inspiration and for introducing me to the oncological world. Besides, I would like to thank him for providing me with the professional environment and facilities of the National Institute of Oncology to complete my thesis.

I greatly acknowledge to Professor Dr. Zoltán Szentirmay, Ph.D. and Erzsébet Csernák, M.Sc. for the molecular-genetic investigations of β -catenine, and all the colleagues from the Department of Surgical and Molecular Oncopathology who supported my research.

I wish to express my sincere gratitude to Dr. Ilona Péter, Ph.D for the immunohistochemical analysis of estrogen receptors.

I am grateful to Professor Dr. Edit Oláh Ph.D, D.Sc. academician and Dr. János Papp, Ph.D. for performing the genetic testing of APC gene.

I thank Dr. István Kenessey, Ph.D. for the egregiuos help in the statistical analysis.

I would like to show my great appreciation to Professor Dr. Miklós Szendrői, Ph.D., D.Sc. and Professor Dr. Pál Vadász, Ph.D. for their assistance in the creation of the database.

I thank Professor Dr. László Tóth, Ph.D. and Dr. István Köves, Ph.D. for their support during my studies.

I wish to thank Dr. Rényi Vámos Ferenc, Ph.D. for the help by professional analysis of chest desmoids.

I wish to thank Dr. Emil Farkas, Dr. Ákos Sávolt and all my co-workers, colleagues and staff members in the National Institute of Oncology for assisting my studies and research activities and for creating a supportive and pleasant working environment.

Finally, an honorable mention goes to my wife, parents, family and friends for their understanding and support on me in completing this thesis.