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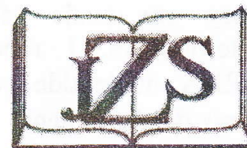
Identification and Molecular Characterization of *β-Tubulin* Gene from Dermatophyte Pathogen *Microsporum Canis.*



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Abstract

Microsporum canis is the most common pathogen causing dermatophytosis in dogs and cats, which can transmit the pathogen to humans. The disease is increasing in many European countries. Studies regarding the structure, expression, and organization of *M. canis* genes have been relatively limited because of its non-aggressive and non-life threatening nature. Microtubules are essential cytoskeleton polymers in all eukaryotes. They are made of α and *β-tubulin* heterodimers. In this study the DNA and the structural organization of *β-tubulin* gene of *M. canis* is described. The DNA of 2487 bp has been identified and sequenced. The ORF of 1341 bp encoding a protein of 447 amino acids with a molecular weight of 49.8 kD is interrupted by eight introns (56-187bp). The gene is highly homologous to *β-tubulin* of *Trichophyton rubrum* (98%) and *Aspergillus parasiticus* (91%). This data may form one of the bases for the development of molecular investigation of dermatophyte fungi *M. canis*.

Keywords: *Microsporum canis*, Dermatophytes, *β-tubulin*, gene expression.

Introduction:

Microsporum canis is a member of the family of fungal dermatophytes that has the capacity to invade keratinized tissue of humans and animals. They establish infection within the cornified layers of the skin, hair and nails [1]. *M. canis* infection is most common in cats and dogs, which can transmit the pathogen to humans [2]. *Microsporum canis* is a worldwide-distributed zoophilic and zoonotic dermatophyte, which is responsible for most cases of feline ringworm. Its prevalence is on the rise in developed countries as well as in Europe [3,4]. Cats are acting mainly as reservoir for *M.canis*. Dermatophytosis in dogs caused by *M. canis* is mostly a companied by bacterial infection. The disease is transmissible to human causing a variety of skin lesions called ringworm [5].

Very little is known about the structural organization of *M. canis*. Genes identified in *M.canis* are *actin*: a structural gene essential for assessing the viability of dermatophytes in skin [6]; *ubiquitin*: a gene necessary for cell cycle regulation [7]; MEP1, MEP2, MEP3: they are secreted Metalloprotease [8]; *Metallothionein*: is a low molecular weight protein, known for their affinity for binding heavy metals[9]; Sub1, Sub2, Sub3:are subtilisin like protease [10]; MEP4, MEP5: they are secreted metalloprotease [11]; *heat shock protein* (accession: AY521222) and *sconC*: a sulphur metabolism negative regulator [12]. These genes are the milestone in the build up information about the structural organization of dermatophytes fungus *M. canis*. In this study, I identified and characterized the structural organization of *β-tubulin* gene.

Microtubules are essential cytoskeleton polymers in all eukaryotes.

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They are made of α and *β-tubulin* heterodimers. I report the DNA and mRNA nucleotide sequence of the *M. canis β-tubulin* gene.
Its structure is compared with those of *T. rubrum*, *A. oryzae*, *N. crassa*, *C. albicans*, and *S. cerevisiae*.

Materials and Methods

Samples: *M. canis* was obtained from clinical specimens collected at the Department of Dermatology, Medical University of Vienna, and cultured in Sabouraud's glucose broth in 75 cm culture flasks at 27°C for one week.
The mycelia mass was collected by centrifugation, extra fluid removed by filtration through 0.45 μM filter and washed twice with PBS. The mycelia mass was flash-frozen in liquid nitrogen and ground to fine powder in a porcelain mortar. The powder was used for DNA and RNA preparation.
Nucleic acid extraction: High molecular weight DNA from *M. canis* was extracted with phenol/chloroform and precipitated with alcohol as described previously [9]. Briefly, the mycelia powder was digested with 300 μg/ml proteinase K incubated at 60°C for 1 hr. The DNA was extracted by Phenol-chloroform and chlorophorm-isoamyl alcohol. The extracted DNA was precipitated by ethanol, DNA pellet is dried and dissolved in bi-distillated water and stored at -20°C. Total RNA from *M. canis* was extracted as described previously [13].
Briefly, the mycelia powder was homogenized with tissue homogenizer after the addition of Guanidine isothiocyanide. The homogenized tissue loaded on caesium chloride and centrifuged over night at 40.000 rpm. RNA pellet was washed once with Absolute and once with 70% ethanol, air dried, dissolved in bi-distillated water and stored at -70°C.
cDNA synthesis and PCR: First strand cDNA synthesis was performed with 1 μg total RNA, 60U MuLV reverse transcriptase (Roche Applied Science, Vienna, Austria), and 2.5 μM oligo (dT) primers in a 20 μl reaction volume. 1μl cDNA was subjected to PCR amplification on thermal cycler according to standard protocol.
The nucleotide sequence of the cDNA was compared to the sequences in gene databanks of the National Center for Biotechnology Information (NCBI, NIH).
5'end sequence of the cDNA was obtained by using SMART™ RACE cDNA amplification Kit (Clontech, Palo Alto, CA) as indicated by the manufacturer.
For analysis of the structural organization of *M. canis β-tubulin*, PCR analysis was performed for genomic DNA according to a standard protocol [14] using synthetic oligonucleotides (figure 1).

s1: 5'-GGG ACC CCG ATA TAC ACA ACA-3'
s2: 5'-GGT AAC CAA ATT GGT GCC-3'
s3: 5'-TGT TCG ACC CCA AGA ACA TG-3'
s4: 5'-TCC AGG AGC TCT TCA AGC GTG T-3'
s5: 5'-GCA GAT GTA CCA TCC TTC GAG TGT GA-3'
as1: 5'-GTA TCA GAG GTG AAG CTC CAT TC-3'
as2: 5'-GAC GGC CAA CTT GCG GAG ATC A--3'
as3: 5'-TTG GAG GTC AGA AGA TCC GGT-3'
as4: 5'-TAC CAG AAC AAG AAA GCC TTC-3'

Figure 1: Sequences of (s) forward and (as) reverse primers used for the amplification of *M.canis β-tubulin* DNA and cDNA.

Results and Discussion

Using s2/as4 primers homologues to the conserved region of *ß-Tubulin* in *T.rubrum* (accession: AAV33733) a fragment of *ß-tubulin* gene from *M.canis* cDNA was amplified. 5′and 3′ end sequence of the cDNA was obtained by primer extension as indicated in materials and methods.

The sequences of primers used for the amplification of DNA and cDNA were illustrated in figure 1. The DNA of 2487 bp has been sequenced. The ORF of 1341 bp is interrupted by eight introns (187, 66, 70, 61,56,58,57 and 61 bp) they are situated mainly towered the 5′end of the gene (figure 2 and 3). Analysis of 13 genes from *M.canis* reported so far (*actin*, *ubiquitin*, MEP1, MEP2, MEP3, MEP4, MEP5, *Metallothionein*, Sub1, Sub2, Sub3, *sconC* and *ß-tubulin* the gene reported in this work) suggest that introns may interrupt the gene once, e.g. MEP2 [8] or twice, e.g. *Metallothionein* [8] or as many as 5 times, e.g. sconC [12]. The total number of introns found in the 13 reported genes are 46 (average of 3.5).

This work shows that *ß-tubulin* gene in *M. canis* is interrupted eight times by introns of 56-187bp. Filamentous fungi introns are short, on average less than 100bp [15]. The smallest *M.canis* intron reported so far is 45 bp from Sub2 [10] and the largest is 187 bP from *ß-Tubulin* (accession no. **DQ449623**).

As in introns of other filamentous fungi the introns of *M.canis ß-tubulin* are started with GTXXG were as the majority of XX are AA, and ended either with TAG or CAG. All the eight introns are located within the ORF. There is just one Known *M.canis* gene, *sconC* [12] which has an intron upstrem of the ATG. Such an intron may play a role in the transcriptional activation of the gene [16]. In the sequence surrounding the ATG start codon a purine (adinine) is present at position -3; this has been suggested to be important for translational activity in filamentous fungi [15].

The deduced amino acid sequence of *M. canis ß-tubulin* is 447 (figure 3) with molecular weight of 49.8 kD. Using database NCBI/Blast and ExPASy proteomics and sequence analysis tools, I found that the amino acid sequence of *M.canis ß-tubulin* was 98% identical to that of *ß-tubulin* protein of *Trichophyton rubrum* (accession: AAV33733) and 91% identical to that of *ß-tubulin* of *Aspergillus oryzae* (accession: BAE64122) and 90% identical to that of *Neurospora crassa* (accession: CAE85615).

The phylogenic tree of *ß-tubulin* gene in different dermatophyte funi indicates the close similarity between *T. rubrum* and *M. canis* (figure 5).

GGGACCCCGATATACACAACAATCTCCAGCTCGACCTCGAACACTCCAGTCCACTCACCCTC
AAGACCTCCGTCAACCACATCAAT**ATGCGTGAAATC***GTAAGTCTAGTTCTCTTCCACTCCTCGCTTCC*
CCTCCAACTGTGCCAAAACACGGTCTGCACAGGCCAAGAAAGGGGGGGGAGGGCCACCACACGACCATGTCCCA
GCGGGAAAAAAACGAGCGTCGAGTTTCATAACCCCAATGTAGAGCTTCCAGCATCAAACTAACGTGTATTGCTTG
*CATAG***GTCCATCTCCAAACTGGCCAATGT***GTAAGCTTTGATCGTTCCCTGGTTCGTTGACAGGAACCCGT*
*TGAGTTAACAGCTATTGACACCCAG***GGTAACCAAATTGGTGCCGCTTTCTG***GTAAGCATTCAACACGCCA*
*ATCATGCTTGTATATAGTCGTGTACGATCGTTACTGACTGAAATTGTATAG***GCAAACCATTGCTGGTGAGC**
ATGGTCTCGATGGATCCGGCCAGTGAGTAAATCCGCAGGAGTCGAAGTAGGGCTCGAGGACTCGGTTATTG
ACGTGATAATAG**CTATACCGGATCTTCTGACCTCCAATTGGAGCGCATGAACGTCTACTTCAAC**
GAG*GTTGGCACAACCAAAGCCCTTCTCTTCAGCAGAATACTAACTCATTGGAGGCACAG***GCCTCTAGCAAAA**
AATACGTTCCTCGTGCTGTGCTTGTTGATCTCGAGCCCGCTGCGCTCGATGCTGTCCGTGCC
GGTCCTTTCGGCCAACTTTTCCGCCCCGACAACGTCGTCTTCGGTCAGTCTGGTGCCGGAAA
CAACTGGGCCAAGGGTCACTACACTGAGGGTGCCAACTTGGTCGACCAGGTCATTGATGTCG
TTCGTCGTGAGGCCGAAGGATGTGACTGCCTTCAGGGTTTCCAGATCACCCACTCTCTCGGT
GGTGGTACCGGTGCCGGTATGGGTACCCTCTTGATCTCTAAGATCCGTGAAGAGTTCCCAGA
CCGTATGATGGCCACCTTCTCCGTTGTCCCATCCCCAATGGTCTCTGACACCGTTGTCGAAC
CATACAACGCCACTCTCTCCATCCACCAGCTCGTTGAGCACTCCGACGAGACCTTCTGTATC
GACAATGAG*GTATGCTTTACTCCCGTGCTCATTTATATGTGACGTGGGCATATCTAACAGCGTGTAG***GCCTT**
GTACAACATTTGCATGAGAACCCTCAAGCTCACCAACCCATCTTATGGTGACCTCAACCACC
TCGTCTCTGCCGTCATGTCCGGTGTCAGCACCAGTCTTCGTTTCCCCGGTCAGCTCAACTCT
GATCTCCGCAAGTTGGCCGTCAACATGGTTCCATTCCCTCGTCTCCACTTTTTCATGGTTGG
ATTCGCTCCTCTCACCAGCCGCAACGCCTACTCTTTCCGTGCCGTCTCCGTACCAGAGTTGA
CCCAGCAGATGTTCGACCCCAAGAACATGATGGCTGCCACTGACTTCCGCAGCGGCCGCTAC
CTTACCTGCTCTGCCATCTT*GTAAGTCTCGCTCGTCCTTACATCGTAATCTTTCTTAATCACTAACACAAT*
*CAATAG***CCGTGGTAAGGTTTCCATGAAGGAGGTTGAGGACCAGATGCGCAACATCCAGAACAA**
GAACTCTGCCTACTTCGTTGAATGGATTCCCAACAACGTCCAGACTGCTCTCTGCTCCATTC
CTCCACGTGGTCTCCAGATGTCCTCCACCTTCGTCGGAAACTCCACCTCCATCCAGGAGCTC
TTCAAGCGTGTCGGTGACCAGTTCACTGCTATGTTCCGCAAGAAGGCTTTCTTGTTCTGGTA
CACTGGCGAGGGTATGGACGAGATGGAGTTCACTGAGGCTGAGAACAACATGAATGACTTGG
TCAGCGAATACCAACAGTACCAGGACGCCTCCGTCTCTGACGGTGAGGAGGAATA*GTGAGTGTT*
*CACCTATCCAACTACTAATCATGATTATTTGCTAACTTACCCTACCAAACAG***CCTTGAGGAGGACCAGCTC**
GAGGCCGAAGAGTAAATGCCACGTACCCGTATAATTGAGACTTAAATGTCTCTCTTATCCTC
CACCTTGCATTCGCTTTGCAAAATATCCCTTTCTCCCGCACAGCAGATGTACCATCCTTCGA
GTGTGAGCCTTGCGTTTAACCAGCCTATGCTTACACCTACGCTCCCCTTATCTTGTCTGCTT
AATAATTCCCCCAACTAGCGTGTCCGTTTTGAGTCGAATATTAGTCACTAGGTGATAACAAA
AACAGGATCAATTGGCTGTTTGATATCCCTCCATCCCAAATATCTTTTAATTTCATCATTTT
TCCTTTGTTTTCGGAGAACCAATTATTAAAAAAATTCCCCTTCAATTTTCAAGGTATTCACT
CCCCTTACCTTCTCTTTGTCTCATTAGATACTGGCATACAGTATTGAAGACAAAAAAGAGCG
AATGGAGCTTCACCTCTGATAC

Figure 2.
Nucleotide sequence of M.canis β- tubulin gene. The genomic DNA sequence of M.canis β-tubulin contains eight introns (indicated in italic-red letters) of 187,66,70,61,56,58,57 and 61 bp. respectively. The coding region is written in bold uppercase. The primers are underlined. The start and stop codon are double-underlined.

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                Exon1 >< Exon2>< Exon3>2< Exon4      >2< Exon5          >< Exon6
M.canis         MREIVHLQTGQCGNQIGAAFWQTIAGEHGLDGSGHYTGSSDLQLERMNVYFNEASSKKYV  60
T.rubrum        MREIVHLQTGQCGNQIGAAFWQTIAGEHGLDGSGHYTGSSDLQLERMNVYFNEASSKKYV  60
A.oryzae        MREIVHLQTGQCGNQIGAAFWQTISGEHGLDGSGVYNGSSDLQLERMNVYFNEANGDKYV  60
N.crassa        MREIVHLQTGQCGNQIGAAFWYTISGEHGLDASGVYNGTSELQLERMNVYFNEASGNKYV  60
C.albicans      MREIIHLSTGQCGNQIGAAFWETICGEHGLDNNGTYVGNNELQKSKLDVYFNEATSGKYV  60
S.cerevisiae    MREIIHISTGQCGNQIGAAFWETICGEHGLDFNGTYHGHDDIQKERLNVYFNEASSGKWV  60
                ****:*:.************** **.****** .*  *  .::* .:::******.. *:*
                                          Exon6
M.canis         PRAVLVDLEPAALDAVRAGPFGQLFRPDNVVFGQSGAGNNWAKGHYTEGANLVDQVIDVV 120
T.rubrum        PRAVLVDLEPAALDAVRAGPFGQLFRPDNVVFGQSGAGNNWAKGHYTEGANLVDQVIDVV 120
A.oryzae        PRAVLVDLEPGTMDAVRAGPFGELFRPDNFVFGQSGAGNNWAKGHYTEGAELVDQVIDVV 120
N.crassa        PRAVLVDLEPGTMDAVRAGPFGQLFRPDNFVFGQSGAGNNWAKGHYTEGAELVDQVLDVV 120
C.albicans      PRAVLVDLEPGTIDNVKTSQIGNLFRPDNFIFGQSSAGNVWAKGHYTEGAELVDSVLDVV 120
S.cerevisiae    PRSINVDLEPGTIDAVRNSAIGNLFRPDNYIFGQSSAGNVWAKGHYTEGAELVDSVMDVI 120
                **:: *****.::* *: . :*:****** :****.*** **********:***.*:**:
                                          Exon6
M.canis         RREAEGCDCLQGFQITHSLGGGTGAGMGTLLISKIREEFPDRMMATFSVVPSPMVSDTVV 180
T.rubrum        RREAEGCDCLQGFQITHSLGGGTGAGMGTLLISKIREEFPAGMMATFSVVPSPMVSDTVV 180
A.oryzae        RREAEGCDCLQGFQVTHSLGGGTGAGMGTLLISKIREEFPDRMMATFSVVPSPKVSDTVV 180
N.crassa        RREAEGCDCLQGFQITHSLGGGTGAGMGTLLISKIREEFPDRMMATFSVVPSPKVSDTVV 180
C.albicans      RREAEGCDSLQGFQITHSLGGGTGSGMGTLLISKIREEFPDTMMATFSVVPSPKVSDTVI 180
S.cerevisiae    RREAEGCDSLQGFQITHSLGGGTGSGMGTLLISKIREEFPDRMMATFSVLPSPKTSDTVV 180
                ********.*****:*********:****************  *******:*** .****:
                     Exon6          ><          Exon7
M.canis         EPYNATLSIHQLVEHSDETFCIDNEALYNICMRTLKLTNPSYGDLNHLVSAVMSGVSTSL 240
T.rubrum        EPYNATLSIHQLVEHSDETFCIDNEALYNICMRTLKLTNPSYGDLNHLVSAVMSGVSTSL 240
A.oryzae        EPYNATLSVHQLVEHSDETFCIDNEALYDICMRTLKLSNPSYGDLNHLVSAVMSGVTTCL 240
N.crassa        EPYNATLSVHQLVENSDETFCIDNEALYDICMRTLKLSNPSYGDLNHLVSAVMSGVTVSL 240
C.albicans      EPYNATLSVHQLVENSDETFCIDNEALYNICQNTLKLPQPSYAELNNLVSSVMSGVTTSL 240
S.cerevisiae    EPYNATLSVHQLVEHSDETFCIDNEALYDICQRTLKLNQPSYGDLNNLVSSVMSGVTTSL 240
                ********:*****:***************:** .**** :***.:**:***:*****:..*
                                          Exon7
M.canis         RFPGQLNSDLRKLAVNMVPFPRLHFFMVGFAPLTSRNAYSFRAVSVPELTQQMFDPKNMM 300
T.rubrum        RFPGQLNSDLRKLAVNMVPFPRLHFFMVGFAPLTSRNAYSFRAVSVPELTQQMFDPKNMM 300
A.oryzae        RFPGQLNSDLRKLAVNMVPFPRLHFFMVGFAPLTSRGAHSFRAVSVPELTQQMFDPKNMM 300
N.crassa        RFPGQLNSDLRKLAVNMVPFPRLHFFMVGFAPLTSRGAHHFRAVSVPELTQQMFDPKNMM 300
C.albicans      RYPGQLNSDLRKLAVNLVPFPRLHFFMVGYAPLTSMGSKSFRSVTVPELTQQMFDAKNMM 300
S.cerevisiae    RYPGQLNSDLRKLAVNLVPFPRLHFFMVGYAPLTAIGSQSFRSLTVPELTQQMFDAKNMM 300
                *:***************:**************:*****:  .:  **::************.****
                         >2<                  Exon8
M.canis         AATDFRSGRYLTCSAIFRGKVSMKEVEDQMRNIQNKNSAYFVEWIPNNVQTALCSIPPRG 360
T.rubrum        AATDFRSGRYLTCSAIFRGKVSMKEVEDQMRNIQNKNSAYFVEWIPNNVQTALCSIPPRG 360
A.oryzae        AASDFRNGRYLTCSAIFRGKVSMKEVEDQMRNIQSKNQSYFVEWIPNNIQTALCSIPPRG 360
N.crassa        AASDFRNGRYLTCSAIFRGKVSMKEVEDQMRNVQNKNSSYFVEWIPNNVQTALCSIPPRG 360
C.albicans      AASDPRNGRYLTVAAFFRGKVSVKEVDDEMHKIQTRNSSYFVDWIPNNVQTAVCSVPPKD 360
S.cerevisiae    AAADPRNGRYLTVAAFFRGKVSVKEVEDEMHKVQSKNSDYFVEWIPNNVQTAVCSVAPQG 360
                **:* *.***** :*:******:***:*:*::*.:*. ***:*****:****:**:.*:.
                                          Exon8
M.canis         LQMSSTFVGNSTSIQELFKRVGDQFTAMFRKKAFLFWYTGEGMDEMEFTEAESNMNDLVS 420
T.rubrum        LQMSSTFVGNSTSIQELFKRVGDQFTAMFRKKAFLFWYTGEGMDEMEFTEAESNMNDLVS 420
A.oryzae        LKMSSTFIGNSTSIQELFKRVGDQFTAMFRRKAFLHWYTGEGMDEMEFTEAESNMNDLVS 420
N.crassa        LKMSSTFVGNSTAIQELFKRIGEQFTAMFRRKAFLHWYTGEGMDEMEFTEAESNMNDLVS 420
C.albicans      LDMSATFIGNSTSIQELFKRVGDQFSAMFRRKAFLHWYTSEGMDEMEFTEAESNMNDLVS 420
S.cerevisiae    LDMAATFIANSTSIQELFKRVGDQFSAMFKRKAFLHWYTSEGMDELEFSEAESNMNDLVS 420
                *.*::**:.***:*********:*:**:*:****.***.*****:**:***:***.*******
                   Exon8           >2<  Exon9
M.canis         EYQQYQDASVSDGEE------EYLEEDQLEAEE------ 447
T.rubrum        EYQQYQDASVSDGEE------EYLEDEQLEGEE------ 447
A.oryzae        EYQQYQDASISEGEE------EYGEEEPLEAEE------ 447
N.crassa        EYQQYQDAGVDEEEE------EYEEEAPLEGEE------ 447
C.albicans      EYQQYQEASIDEEE------LEYADEIPLEDAAME--- 449
S.cerevisiae    EYQQYQEATVEDDEEVDENGDFGAPQNQDEPITENFE 457
                ******:*  :.: *       ::       :
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Figure 3. Multiple alignment of the deduced amino acid sequence of *M. canis ß-tubulin* with that of *T.rubrum* (accession no.AAV33733), *A.oryzae* (accession no.BAE64122), *N.crassa* (accession no.CAE85615), *C. albicans* (accession no. AAA34375) and *S. cerervisiae* (accession no.V01296) using ExPASY ClustalW-EBI Tool. The borders of amino acid regions encoded by different exons are indicated by arrows above the sequences. Shaded numbers between arrows indicate the phase (position within the triplet) of the exon-exon border unless the phase is 0. Amino acid conservation is marked by asterisks, colons, and periods for identical, highly similar, and weakly similar residues, respectively.

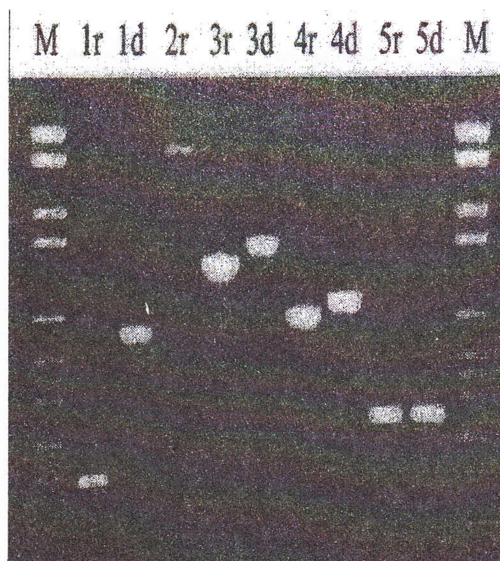

Figure 4.PCR amplification of *M.canis β-tubulin* cDNA (lines 1r,2r,3r,4r,5r) and DNA (lines 1d,3d,4d,5d). The primer pairs used for the amplification of the samples in line 1r,1d is s1-as3; in line 2r is s1-as1; in lines 3r,3d is s3-as1; in lines 4r,4d is s4-as1 and in lines 5r,5d is s5-as1. (M) is DNA molecular marker VI. The PCR products were loaded on 1,2% agarose gel and visualized by ethidium bromide.

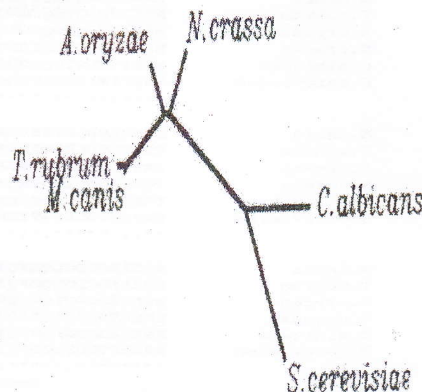

Figure 5. The phylogenic tree of β-tubulin in *M.canis* , *T.rubrum, C. albicans, S.cerevisiae, N.crassa* and *A. Oryzae* indicate the close association between different types of dermatophytes.

In summary, I report the complete genomic sequence and organization of *β-tubulin* gene in *M. canis* and its comparison with other related fungus. This data may form one of the bases for the development of molecular investigation of dermatophyte pathogen *M.canis*.The nucleotide sequence of *M.canis β-tubulin* is deposited in the GenBank database and assigned accession number: **DQ44962.**

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ناسینەوە و روونکردنەوەی رەفتاری *β-tubulin* جینی کەڕووی پێست لە جۆری *Microsporum canis*

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پوختە

كەڕووی پێست *M.canis* هۆی سەرەكی نەخوشی كەڕووی پێستە لە سەگ و پشیلە ولەوانەوە ئەگوازرێتەوە بۆ مروڤ . نەخۆشیەكە لە زۆر ولاّتی ئەوروپی لەزیادبووندایە . لێكۆڵینەوە دەربارەی شێوەی پێكهاتن و دەربڕین و رێكخستنی جینەكانی *M.canis* زۆر سنور دارە , لەبەر كەمی مەترسی بۆ سەرژیان . لەم لێكۆڵینەوەدا DNA وشێوەی پێكهاتن و رێكخستنی جینی *β-tubulin* ی *M.canis* روون دەكرێتەوە . ئەم جینەلەگەڵ *α-tubulin* مایكرۆتبیوول دروست ئەكەن كەبەشێكی گرنگی چوار چێوەی شانەی Eukaryote پێك دێنێ. *β-tubulin* پێكهاتوە لە ٢٤٨٧ نیوكلیۆتاید , ١٣٤١ ی پرۆتین دروست ئەكەن , پرۆتینەكەی ٤٤٧ ترشی ئامۆنیك و كێشی گەردیلەیی ٤٩,٨ KD ە . جینەكەلەلایەن هەشت intron وە بڕدراوە كەلە ٥٨ تا ١٨٧ نیوكلیۆتید پێكهاتون. زۆر لە *β-tubulin* ی كەڕووەكانی كەئەجی و ٩٨٪ وەك یەكن لەگەل *T.rubrum* و ٩١٪ وەك *A.parasiticus* .ئەم زانیاریانە لەوانەیە بنەمایەكبن بۆ گەشەكردنی لێكۆڵینەوەی جینی لە كەڕووی پێست لە جۆری *M-canis* .

التشخيص والتعريف الجزيئي ل *β-tubulin* جين للفطر الجلدي المرضي *Canis Microsporum*

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الخلاصة

الفطر الجلدي المرضي *Microsporum canis* (*M.Canis*) هو احد المسببات المرضية الجلدية للكلاب والقطط الذي ينتقل منهم الى الانسان .المرض في ازدياد مستمر في عدد من الدول الاوربية . الدراسات المتعلقة في التركيب الجيني لمسبب هذا المرض (*M.canis*) محدودة جدا وهذا يعود الى كون المرض هادئ ولا يسبب تهديداً مباشراً للحياة.احد المكونات المهمة لهيكل الخلايا بشكل عام هو Microtubules والذي يتكون من *β-tubulin* و α. في هذه الدراسة تم التعرف وتشخيص الحامض النووي والتركيب الجيني لاحد مكونات Microtubules وهو *β-tubulin* جين .فوجدنا بان الجين يتكون من ٢٤٨٧ نيوكليوتايد منها ١٣٤١ تحمل المعلومات الوراثية لتكوين بروتين مكون من ٤٤٧ حامض اميني بوزن نوعي مقداره KD ٤٩,٨ .يقاطع الجين ثمانية Intron باحجام ٥٦-١٨٦ نيوكليوتايد . ان لهذا الجين تشابة كبير مع *β-tubulin* في *Trichophyton rubrum* (٩٨٪) و *Aspergillus parasiticus* (٩١٪) . ان هذه المعلومات سوف تساعد في وضع الحجر الاساس للدراسات الجينية للفطريات الجلدية وخاصة *M.canis*.

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