



Working Group Black Yeasts
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The use of introns in population dynamics

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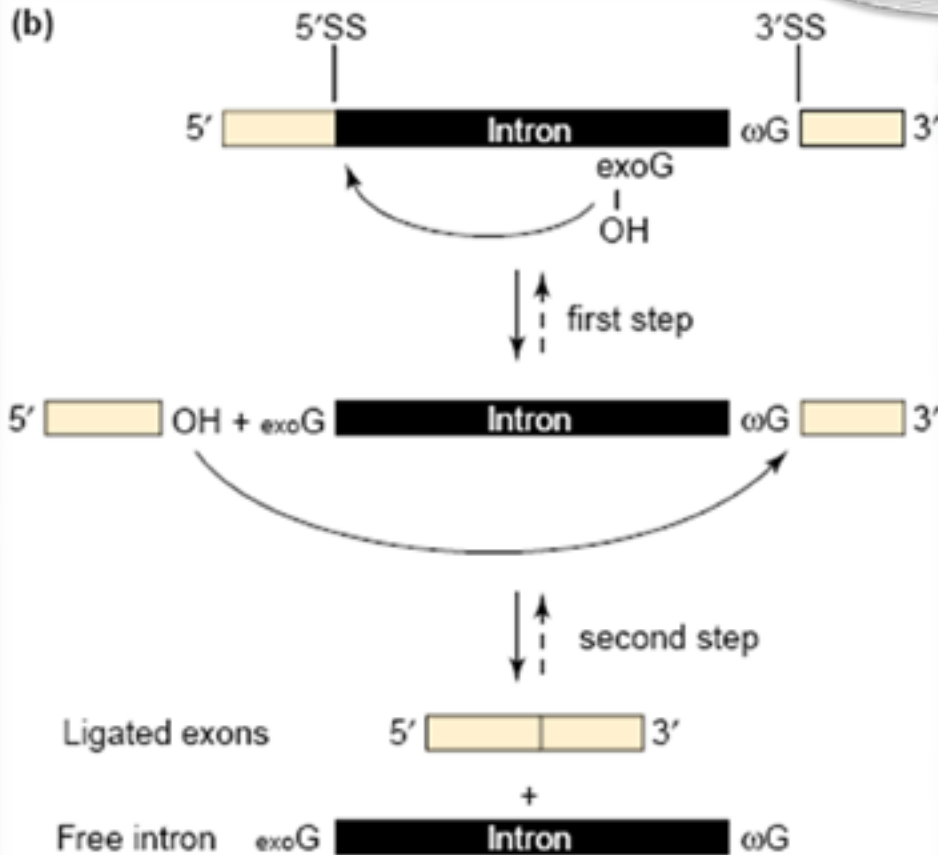
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What are group I introns ?



TRENDS in Genetics

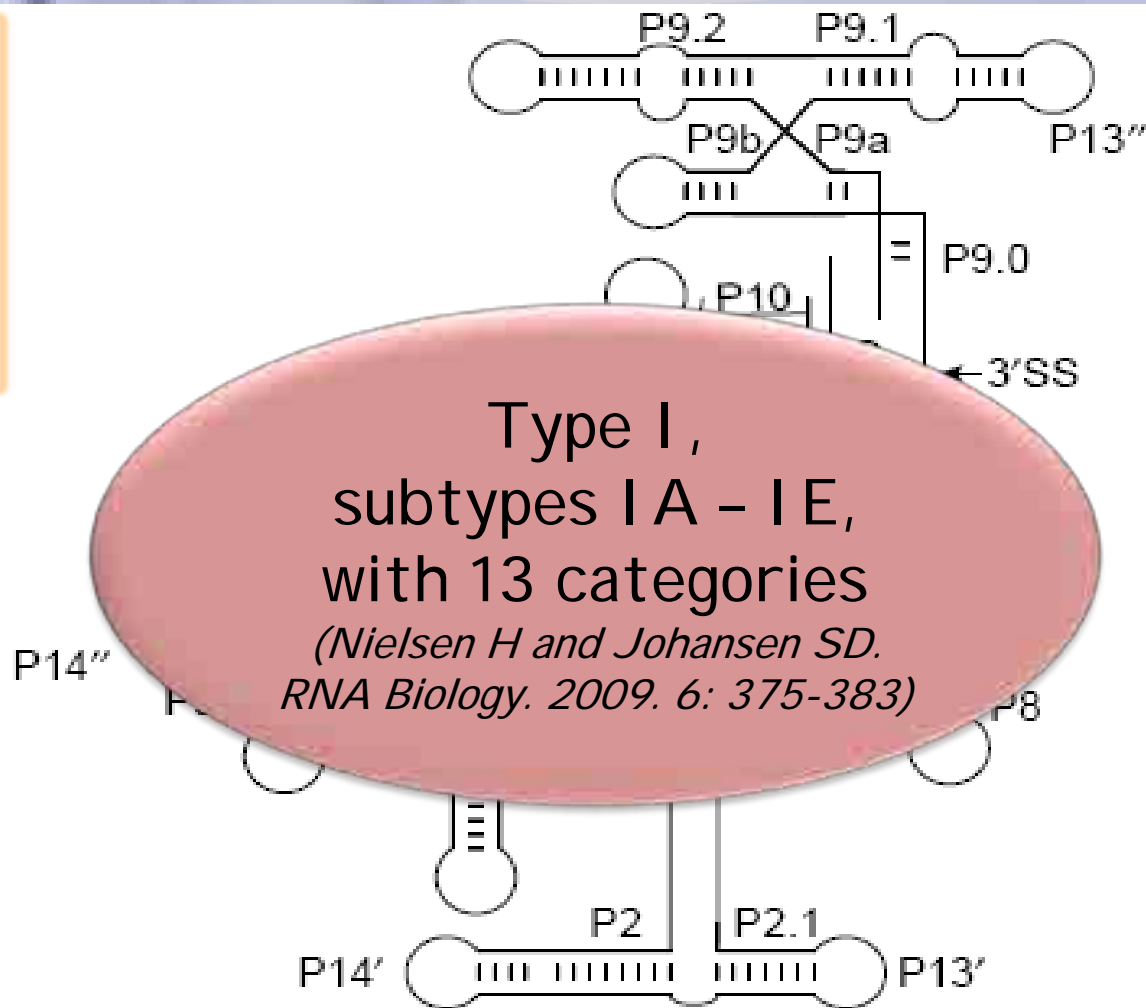
ü A distinct class of non coding DNA interrupting genes

ü Able to remove by themselves from RNA precursors

Widely distributed :
plants, bacteria, and fungi
in nuclear or mitochondrial genomes

What are they looking like ?

specific
primary and
secondary
structure
essential for
self - splicing



Michel and Weshof. J. Mol. Biol. 1990. 216: 585-610 ;
Nikoh et al. Mol. Biol. Evol. 2001. 18 : 1631-1642 ;
Holst Jensen et al. Mol. Biol. Evol. 1999. 16: 114-26

28S rDNA group-I introns: a powerful tool for identifying strains of *Beauveria brongniartii*

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New type of molecular marker used to differentiate species and able to resolve relationships among closely related strains of a single species

Obligatory group I introns with unusual features at positions 1949 and 2449 in nuclear LSU rDNA of Didymiaceae myxomycetes

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Evolutionary Dynamics of Multiple Group I Introns in Nuclear Ribosomal RNA Genes of Endoparasitic Fungi of the Genus *Cordyceps*

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Vertical transmission throughout speciation → Loss / acquisition via horizontal transfer = Evolutionary history

Exophiala dermatitidis



Isolation of Fungi, Especially *Exophiala dermatitidis*, in Patients Suffering from Cystic Fibrosis

A Prospective Study

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N. Schnitzler^{b,c}



Molecular diversity of oligotrophic and neurotropic members of the black yeast genus *Exophiala*, with accent on *E. dermatitidis*

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Table 1. Strains investigated, listed according to populations based on ITS sequencing, M-13 fingerprint, RAPD and SSU-introns.

Isolate	Alternative identification	Source	Origin	No. of nucl. diff.	M-13 group	RAPD group ¹	SSU length
CBS 578.76	CDC B-3567 NCMH 1224 KMU 1405	Brain , pancreas, liver, ileum, lymph nodes, meninges	Taiwan	0	A	1	3000
CBS 207.35	UTMB 3827 NCMH 252 ATCC 28869 WD 252	Skin	Japan	0	A	1	3000
CBS 581.76		Brain?	Japan	0	A	-	3000
CBS 153.94	CDC B-3554 NCMH 1211 KMU 1308	Liver	Japan	0	A	-	3000
NCMH 1213	CDC B-3556 KMU 1166	Disseminated	Japan	-	A	-	3000
NCMH 1214	CDC B-3557 KMU 1948	Skin	Japan	-	A	-	3000
CBS 709.95		Subcutaneous	Korea	0	A	-	3000
NCMH 1388		Subcutaneous cyst	USA	-	A	-	3000
NCMH 1218	CDC B-3561 KMU 1616	Brain , skin, lung, lymph nodes	Japan	-	A	-	3000
CBS 109154	DH 11215	Brain	Korea	-	A	-	-
NCMH 1210	CDC B-3553 KMU 1309 IFM 4837	Disseminated	Japan	-	A	-	3000
CBS 292.49		Faeces	Brazil	0	A	1	3000
DH 11380		Sputum, CF	Finland	0	A	-	3000
CBS 971.87		Unknown	Iraq	-	B	5	1800
CBS 100341	RKI 4669/92 BIschVa300/8	Blood	Germany	3	B	6	1800
CBS 100340	IFM 4848 INPA 109	Liver of bat	Brazil	3	B	-	1800
CBS 106.92	DBVPG 4256	Grape	Italy	3	B	7	1800
CBS 149.90		Sputum, CF	Germany	1	B	4	1800

To screen
the ribosomal
genes
SSU + LSU

Strains
genotype A
and B

To type the
different
introns

Group I
introns but
IC ? IB ?
Or...?

Evolutionary
hypothesis
?

Distribution of
introns
i.e A/B/types

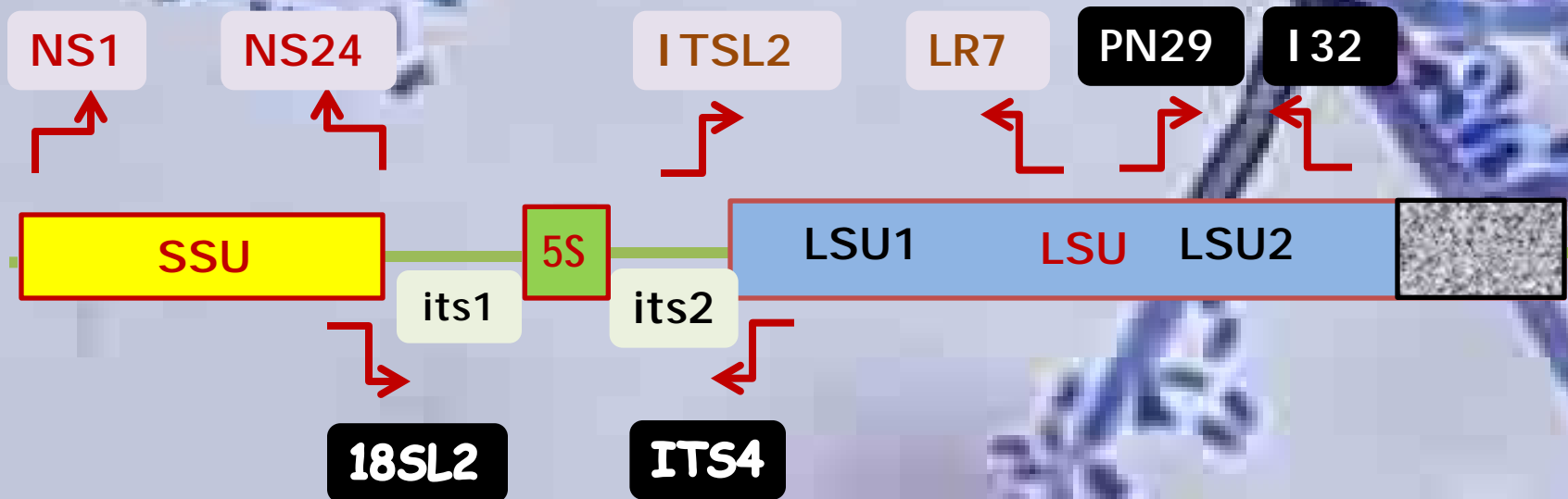
Materials and methods

- 92 strains of *Exophiala dermatitidis*

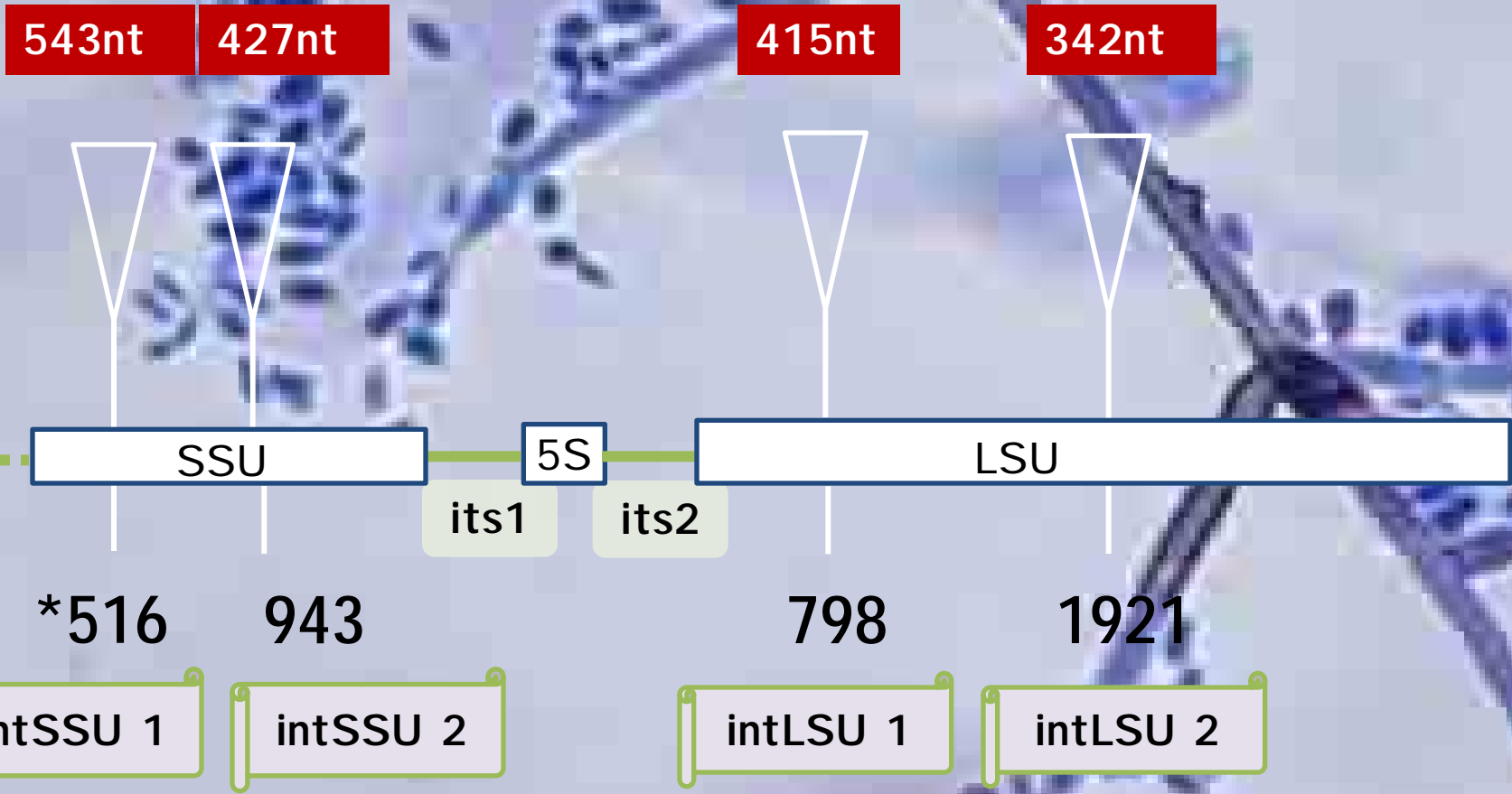
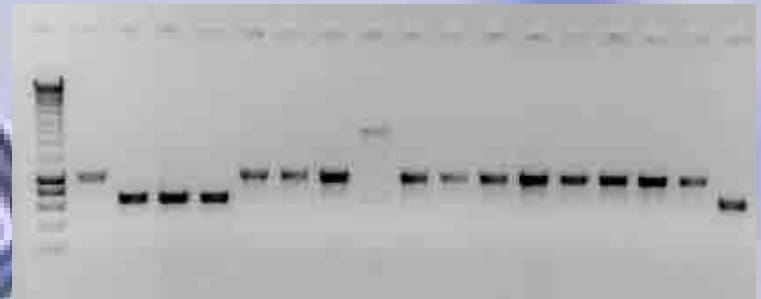
57 of genotype A / 35 of genotype B

- DNA extraction

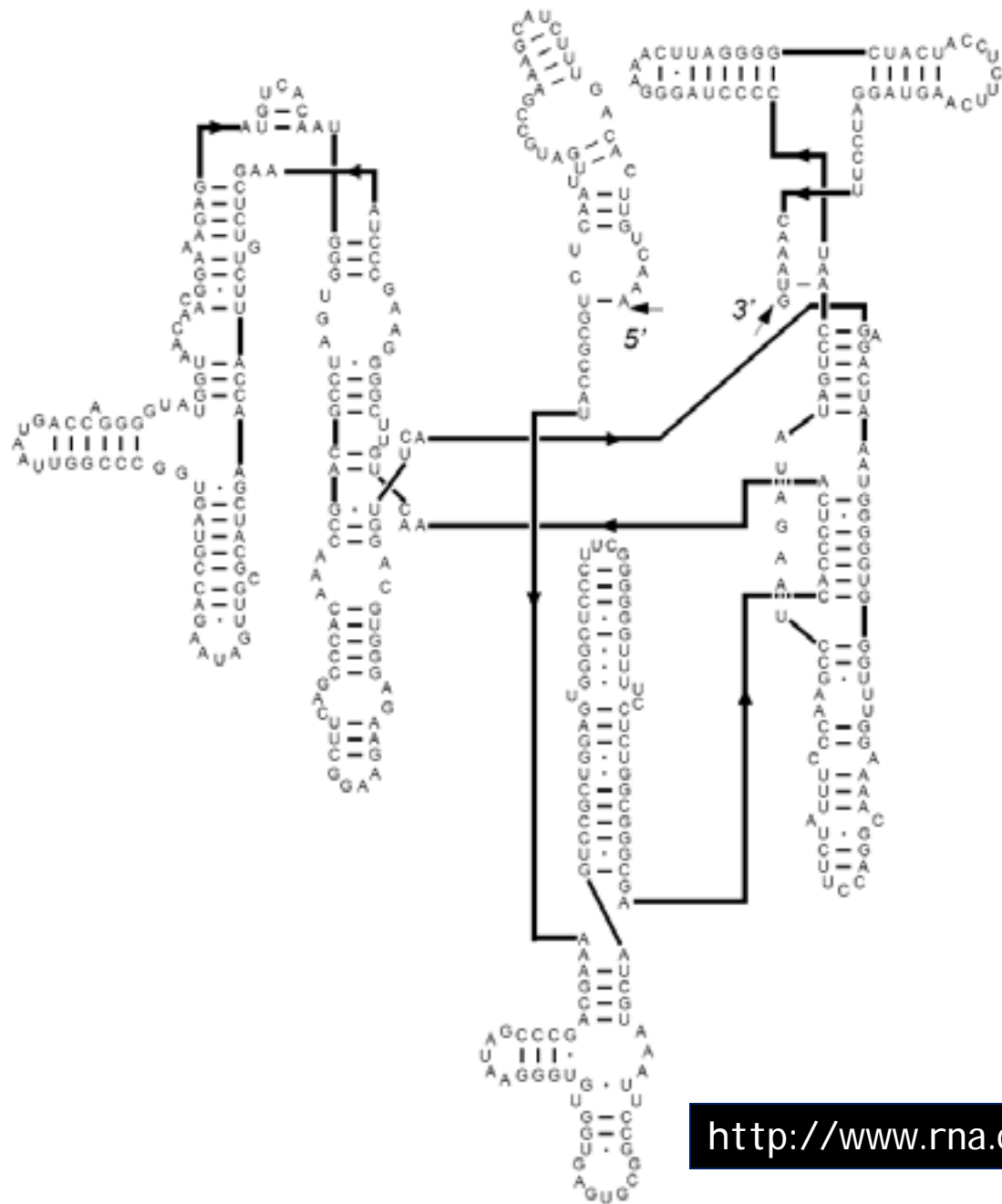
- PCR / sequencing



Results



* *E. coli* numbering



	Group A	Group B
Clinical strains	39 (69%)	7 (20%)
symptomatic	20	4
colonisation	19	3
Environment	1	10 (72%)
Unknown	1	3
Prevalence of introns		
SSU	42 (87,5%)	1 (3%)
LSU 1	45 (93%)	29 (80%)
LSU 2	44 (92%)	5 (14%)
Total of analysed strains	48*	35

SSU and LSU2 introns are mostly associated to genotype A strains

* 9 incomplete pattern

Genotype A WITHOUT introns

6 without intron SSU

2 eucalyptus, 1 pectin, 1 faeces flying fox, 2 railway

3 without intron LSU1

1 eucalyptus, 1 berry, 1 faeces Bonobo

4 without intron LSU2

1 faeces flying fox, 1 eucalyptus, 1 pectin, 1 faeces Bonobo

è Environment or human made environment


Genotype B WITH introns

1 with intron SSU : steam bath

5 with intron LSU2 : 1 sauna, 1 steam bath, 1 treated wood,
1 sputum, 1 flying fox liver

è Human made environment or colonisation

Conclusions



Four type I C1 introns with different
insertion sites
sequences
evolutionary histories



intLSU1 : the most stable within genotype A and B

è vertical inheritance ? Ancestral origin ?



intSSU 1, SSU2, LSU2 :

è horizontal transmission ?



For *Exophiala dermatitidis*
Introns = new evolutionary marker

Molecular diversity and distribution of populations with AFLP.
Sudhadham et al. (PhD - thesis)



Scytalidium
sp.



Scytalidium
dimidiatum



Scytalidium
hyalinum



	domain	Nb analyzed	WITH INTRONS	WITHOUT INTRON
<i>S. hyalinum</i>	LSU1	13	13	0
	LSU2	16	16	0
<i>S. dimidiatum</i>	LSU1	28	23	5
	LSU2	27	6	21
- Clinical	LSU1	23	20	3
	LSU2	22	6	16
- Environment	LSU1	5	3	2
	LSU2	5	0	5
<i>Scytalidium sp.</i>	LSU1	17	1	16
	LSU2	15	3	12

58 LSU1 and LSU2 pattern

INTRONS

High resolution markers at various taxonomic levels from closely related strains of a single species to separating species

What are
their role? ...

Where are
they coming
from? ...

To be continued ...

Thank you

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