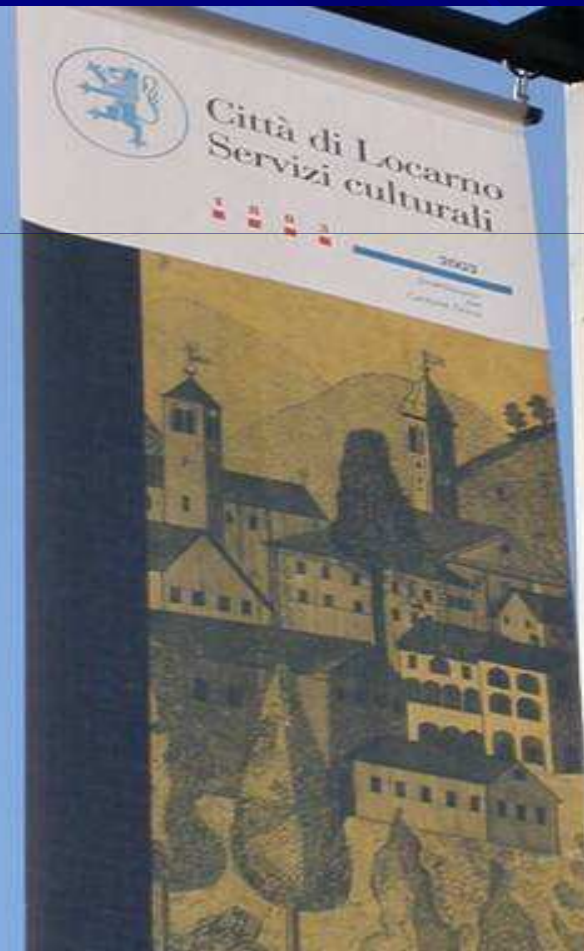


CURRENT SITUATION OF HUMAN DIPHYLLOBOOTHRIOSIS IN EUROPE

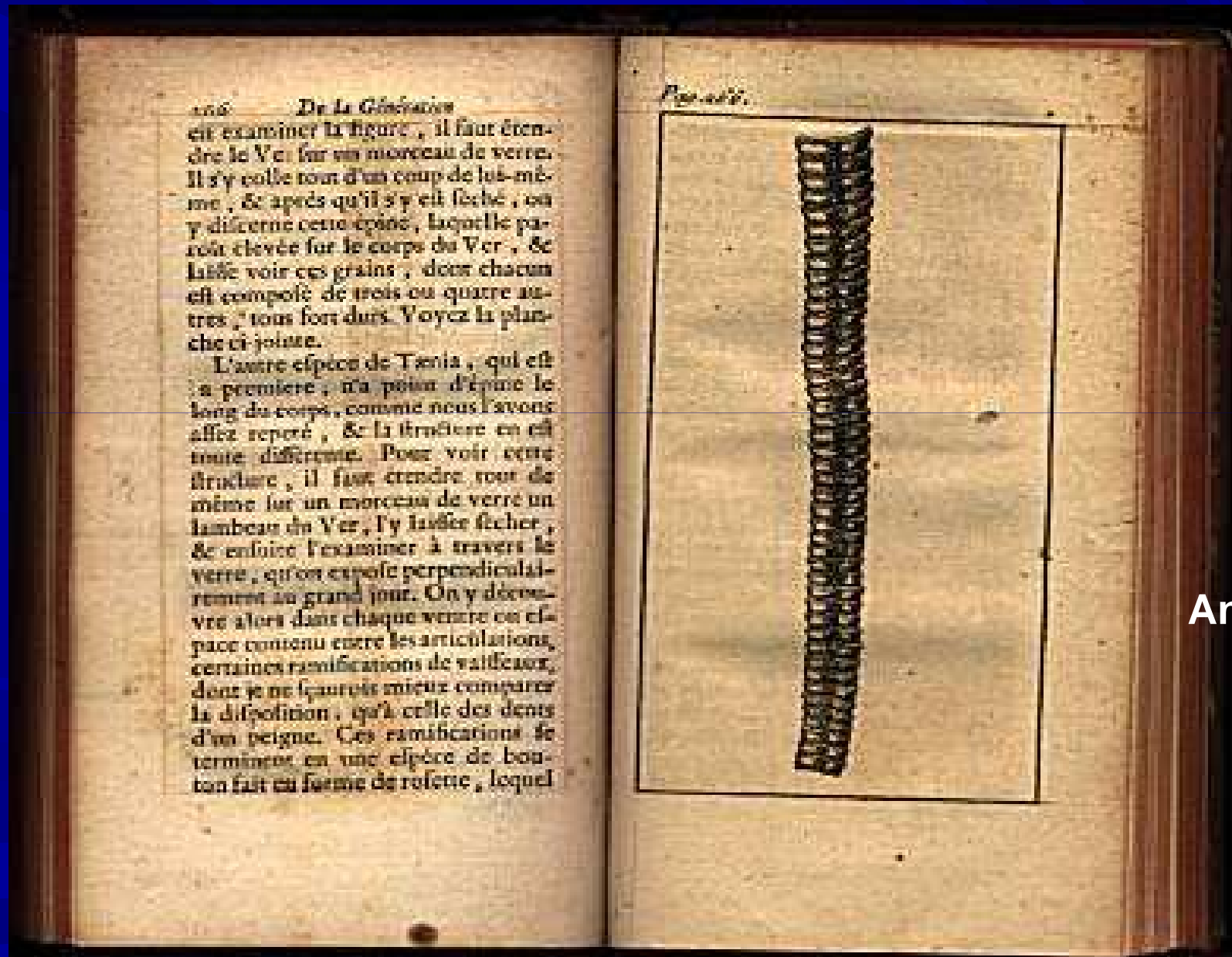
Prof Jean Dupouy-Camet
Parasitology, Paris Descartes
University, Cochin Hospital,
Paris, France



■ First description
by Taddeus Dunus
in Locarno in the
16th century



Reported in Paris in the 18th century



Andry, 1741

Global Distribution of Helminthiasis *Diphyllobothrium Latum*, 1952



Source: "Distribution of Helminthiases: Atlas of Diseases - Plate 4." The American Geographical Society, The Geographic Review, Vol. 42, No. 1, 1952

Helminthiasis distribution

absent
 present

Center for International Development, 2000

Diphyllobothrium latum: the biggest human intestinal parasite (up to 10 meters)



Clinical & biological signs

■ Clinical

- Abdominal pain & discomfort
- Diarrhea
- Worm expelled

■ Biological

- Moderate increase of blood eosinophiles
- No macrocytic anemia
- Eggs at stools examination

"The man ate masu salmon. After a time, a strange object emerged from the anus and was pulled out: it turned out to be 2-3 m long."

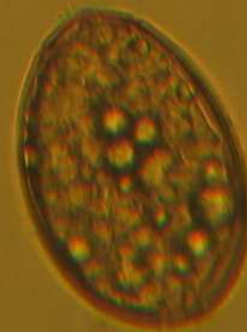


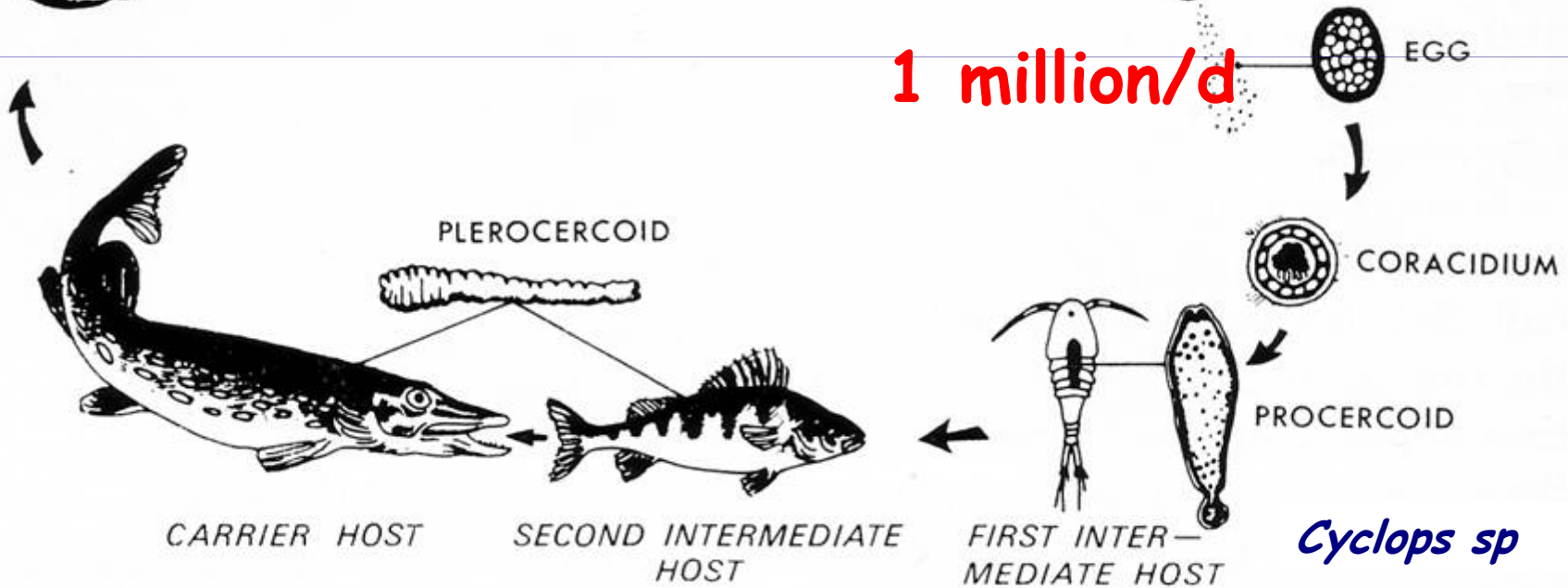
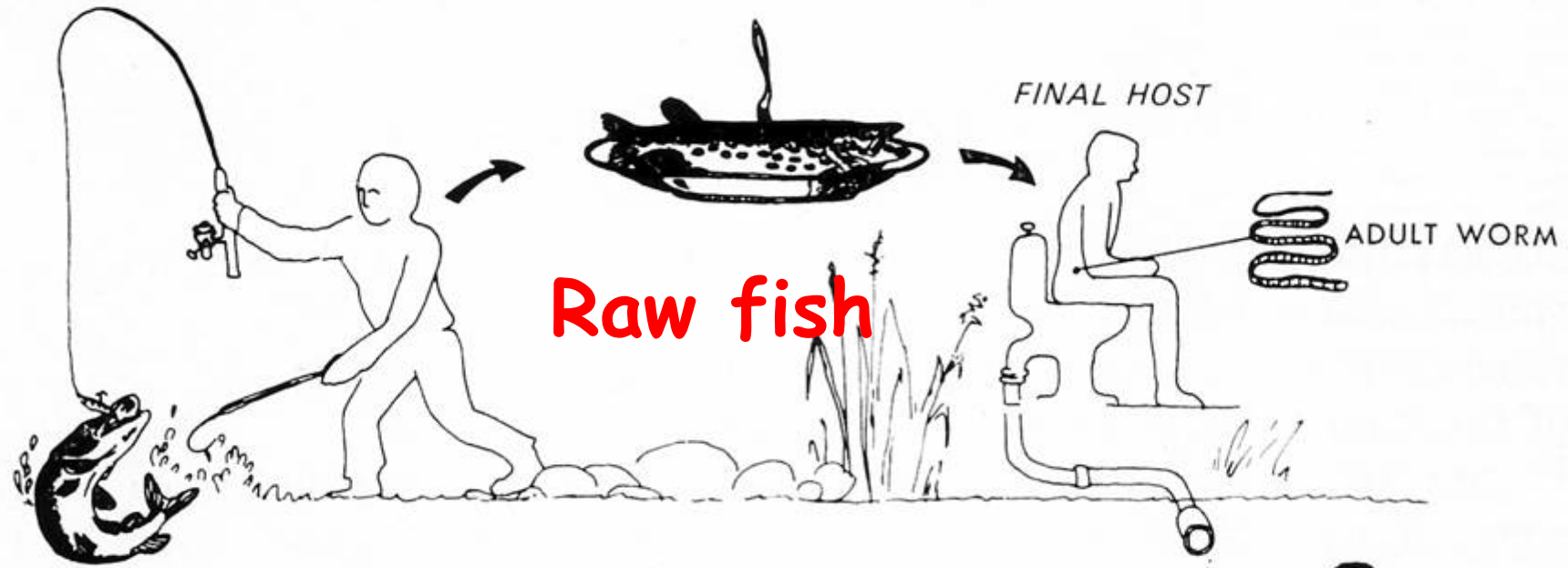
From Shinsen Yamaino Soushi, Woodprint by Daizenosuke Koan (1850).
Courtesy of the Tohoku University Medical Library.

Arizono, EID, 2009

Fasciola hepatica

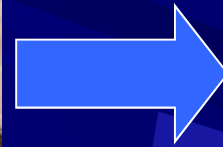
Diphyllobothrium latum



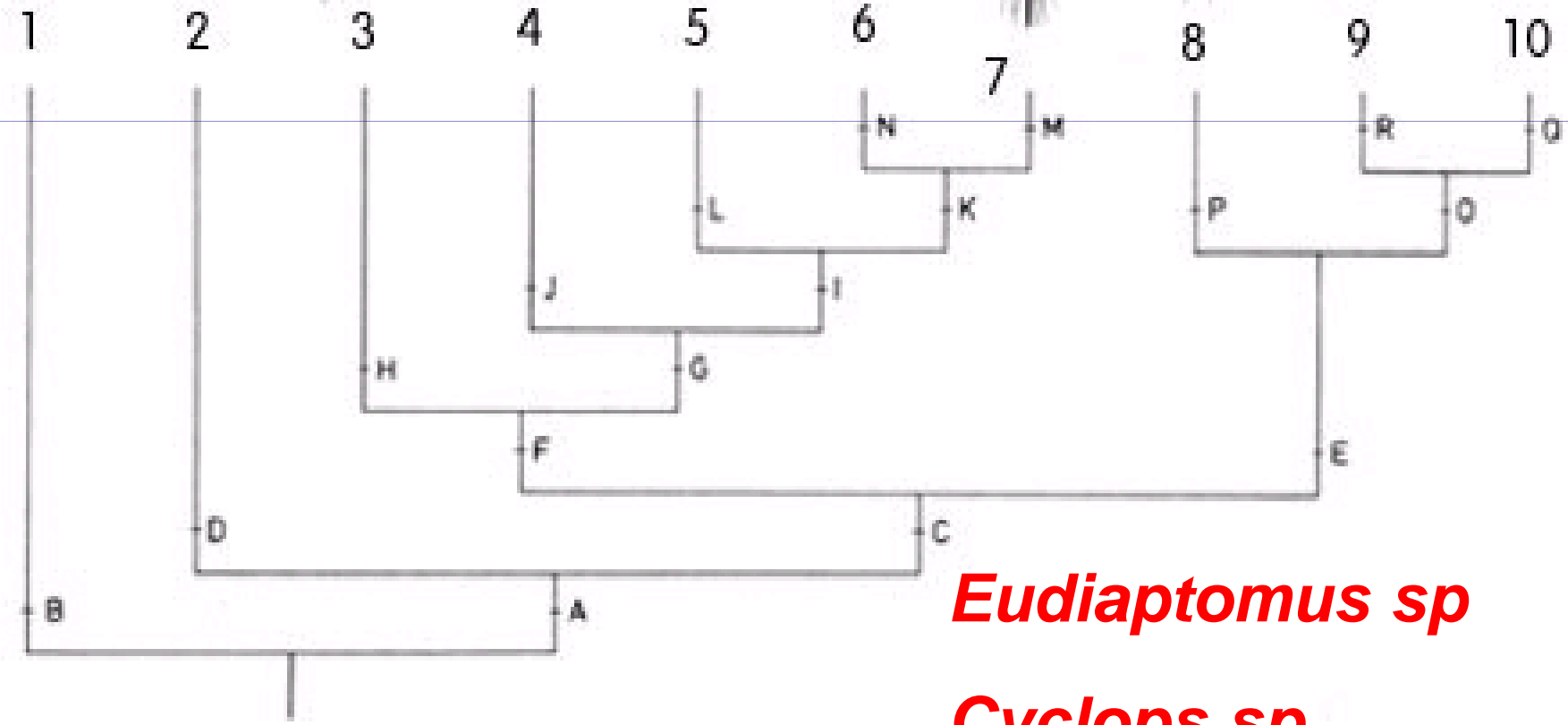
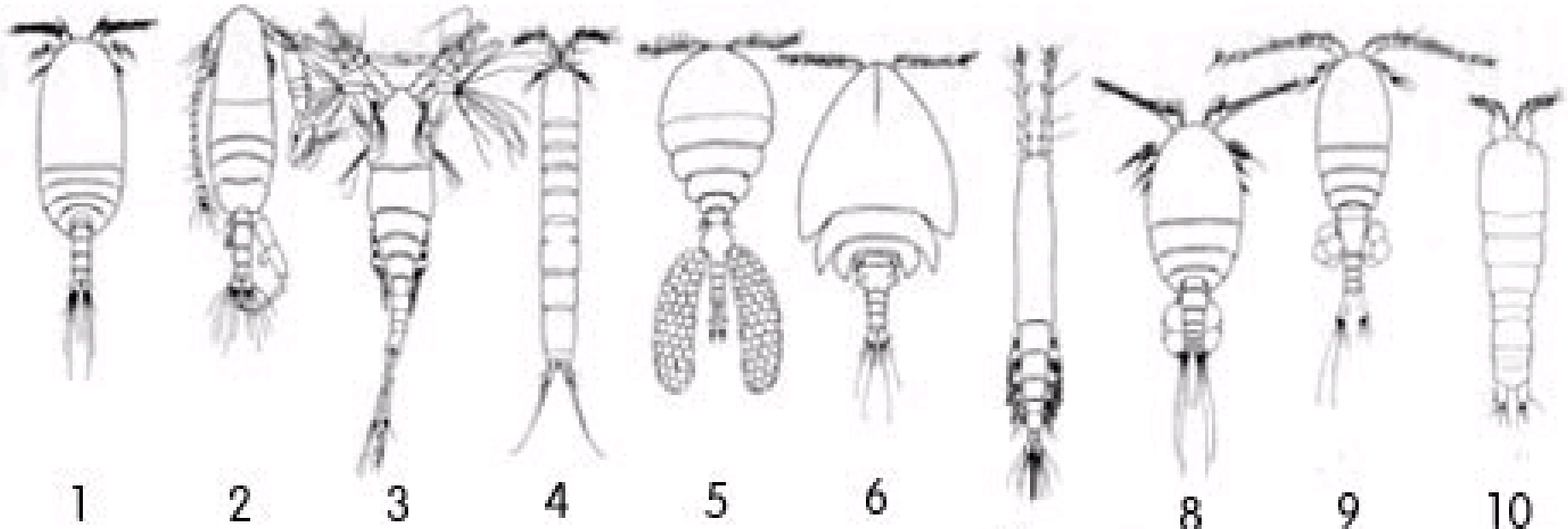


from Almer, 1974 in Von Bonsdorff, 1977

Complex parasitic cycle but high
prolificity of the parasite

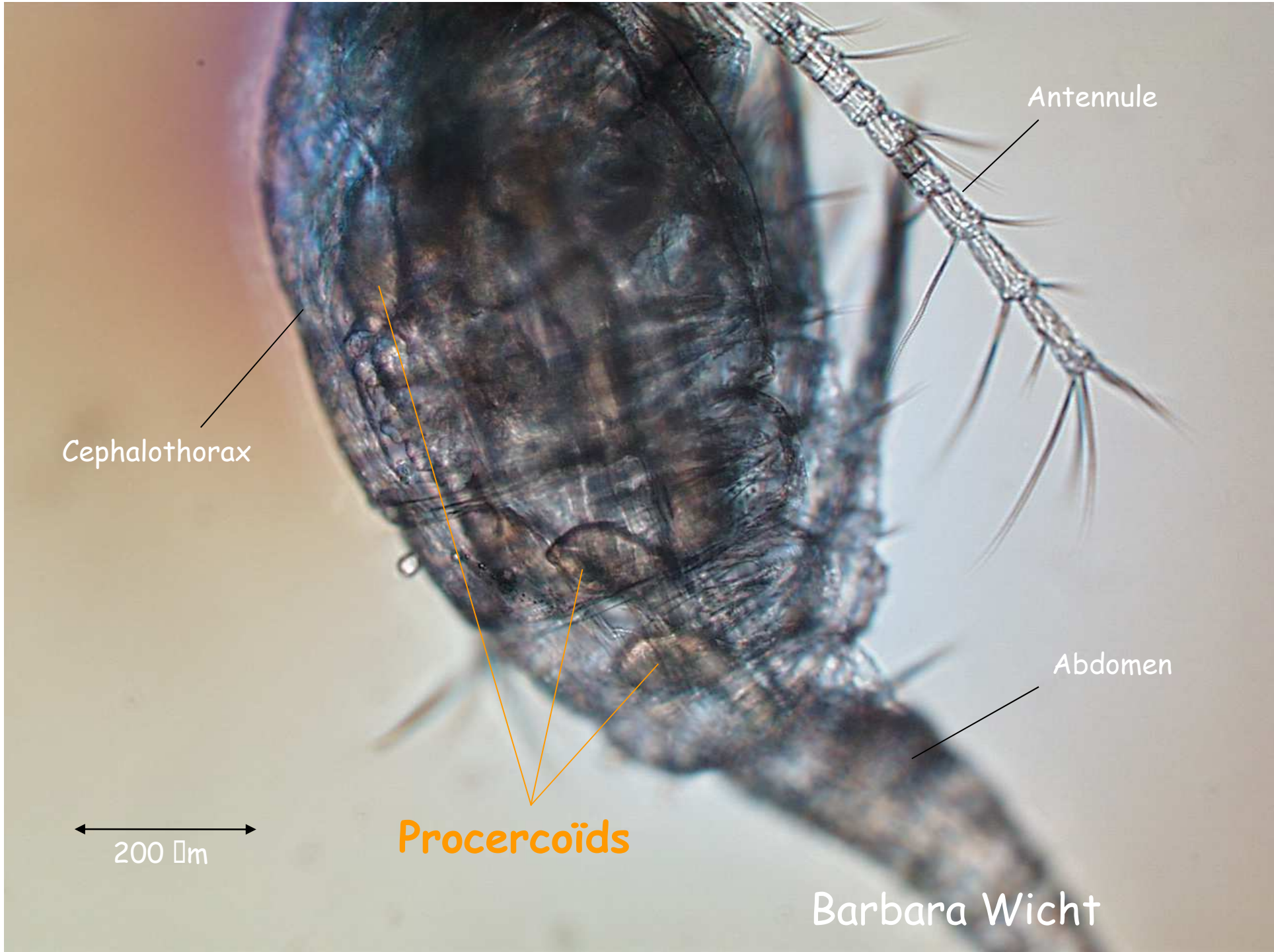


**1 million
eggs/day**



Eudiaptomus sp

Cyclops sp



Antennule

Cephalothorax

Abdomen

Procercoïds

200 μ m

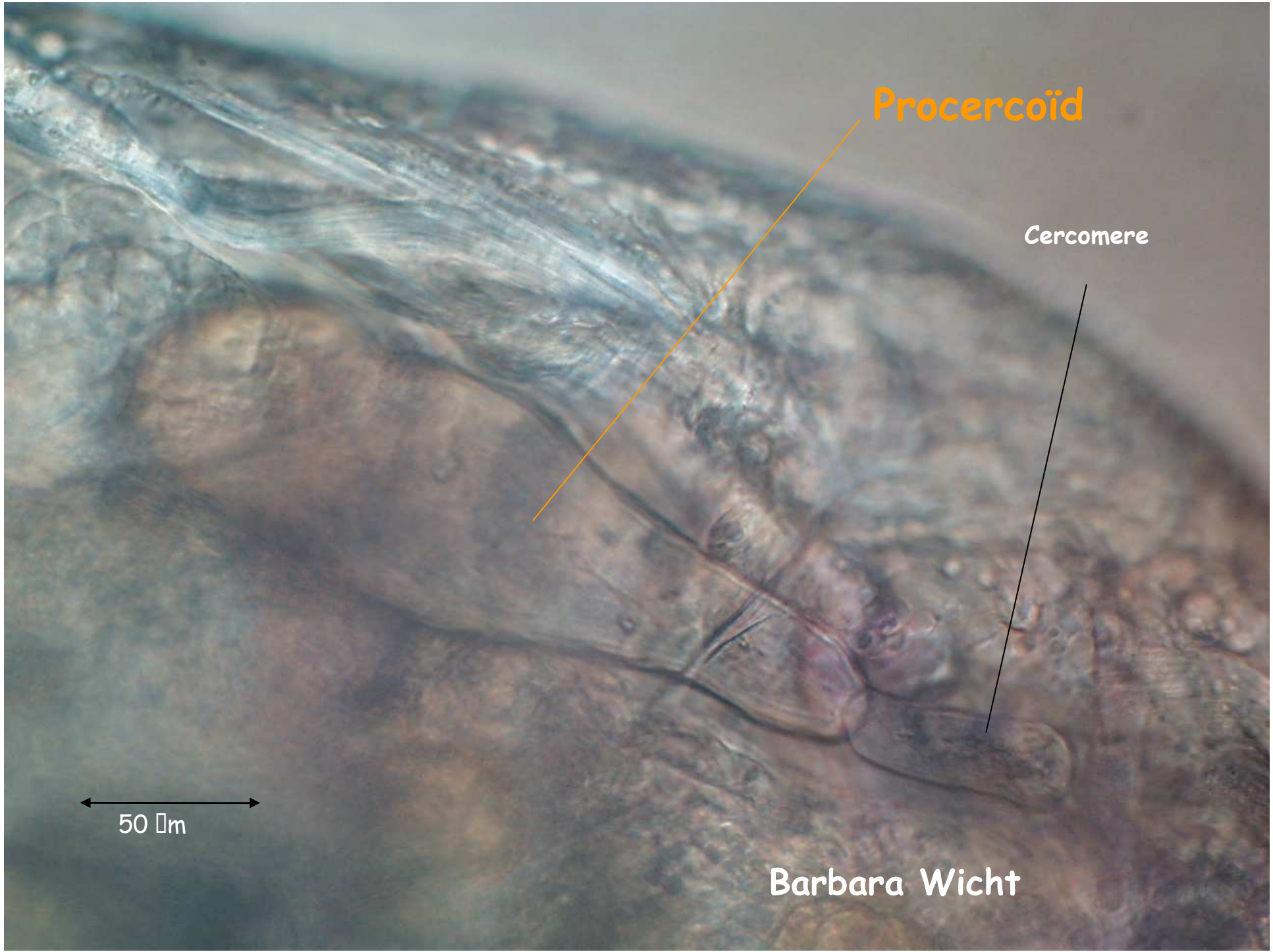
Barbara Wicht

Procercoid

Cercomere

50 μ m

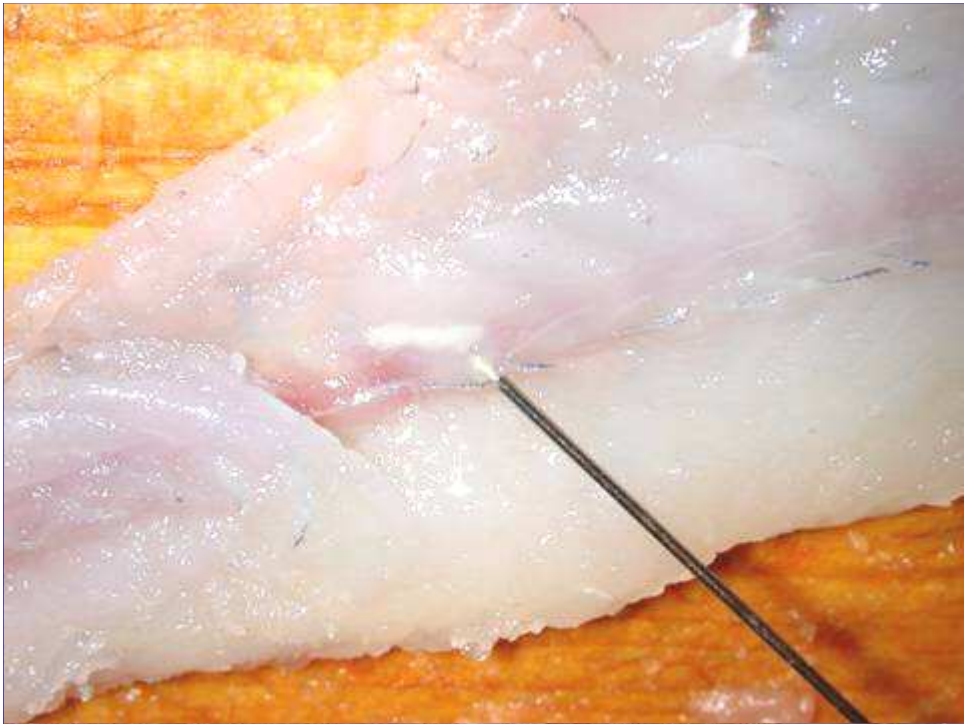
Barbara Wicht



© 2000 Ralf Rebmann



Perca fluviatilis





Prevalence in fish

(Golay 1995, Peduzzi 2001, Nicoulaud 2005)

	Prevalence	Fish
Laggo Maggiore	8 %	<i>Perca fluviatilis</i>
Laggo Orta	33 %	<i>Perca fluviatilis</i>
Lac de Morat	12 % 5 %	<i>Esox lucius</i> , <i>Perca fluviatilis</i>
Lac de Bienne	14% 4 % 0	<i>Esox lucius</i> , <i>Perca fluviatilis</i> <i>Lotta lotta</i>
Lac Léman	4-10 %	<i>Perca fluviatilis</i>

Professional fishermen



Fish mongers



Parasite considered as extinct in the alpine lakes region

- Cases described on the Swiss shores of lago Maggiore (Med Mal Inf, Peduzzi, 1990) and around lakes of Neuchâtel region and Geneva lake (Golay & Mariaux, 1995).

Journals with a low impact factor....

- No data for France and other european countries

bulletin épidémiologique hebdomadaire

RÉPUBLIQUE FRANÇAISE

Ministère de l'Emploi

et de la Solidarité

INSTITUT DE VEILLE SANITAIRE



BEH

Enquête sur l'incidence de la bothriocéphalose
en Haute-Savoie (1993-2000) p. 211

N° 45/2001

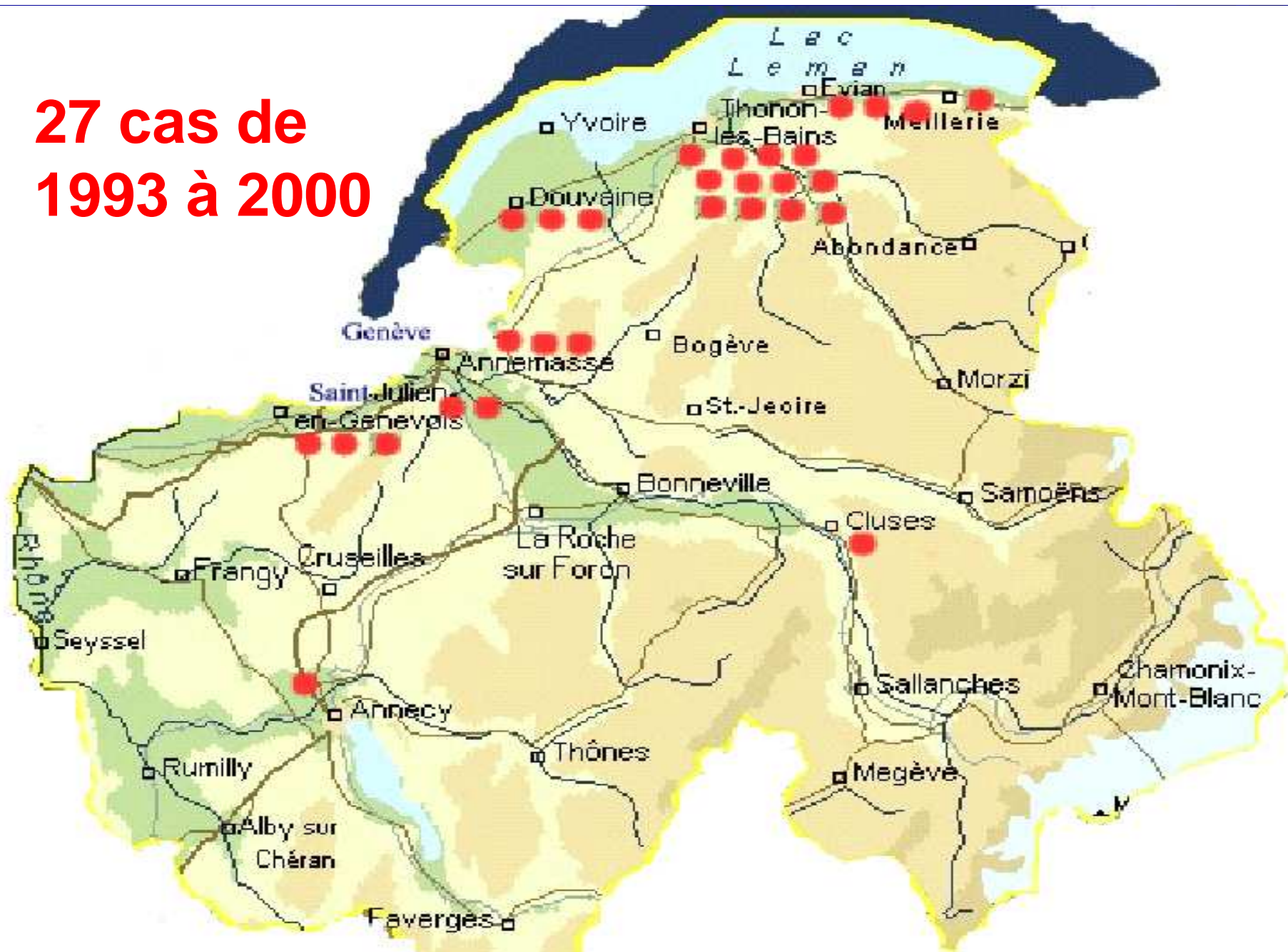
06 novembre 2001

ÉTUDE

ENQUÊTE SUR L'INCIDENCE DE LA BOTHRIOCÉPHALOSE EN HAUTE-SAVOIE (1993-2000)

Laurent Desvois¹, Alain Gregory², Thierry Ancelle¹ et Jean Dupouy-Camet¹

**27 cas de
1993 à 2000**



Cas de bothriocéphalose en Haute-Savoie

APRÈS DES ANNÉES DE CAVALE DANS LES CARAÏBES... **SCHULLER : L'ALLER SIMPLE**



nt de se rendre, Didier Schuller, que l'on voit ici dans l'avion du retour, a fait savoir qu'il ne voulait dénoncer personne. n'al pas de comptes à régler", a-t-il dit...

Photo SIPA

suspense a pris fin hier. Sous le coup d'un mandat d'arrêt international, l'ancien RPR est finalement rentré en France, de lui-même... Arrêté à l'aéroport, immédiatement été présenté au juge chargé de l'enquête sur les HLM des uts-de-Seine, qui l'a interrogé durant quatre heures. Mis en examen pour "trafic influence et recel d'abus de biens sociaux", il devait être incarcéré dans la soirée.

POISSONS DU LAC LÉMAN

Du danger de les manger crus.

Le plus grand des parasites humains, le "diphyllobotrium latum", peut atteindre 10 mètres de long, battant ainsi à plate couture le ténia ! Ce champion-squatter élit domicile dans l'intestin grêle. Pour éviter cette parasitose intestinale, il convient de ne plus manger crus, ou mal cuits, les perches, ombles chevaliers et autres truites sauvages du lac Léman...

● EN PAGE 2, L'ARTICLE DE FRANÇOISE GRUBER



Malgré tout, bon appétit !

YVOIRE

Les tribulations d'un python voyageur

La Douane a remis au vivarium un animal, saisi l'automne dernier au poste de Vallard, en vertu des accords de Washington pour préserver certaines espèces.

European survey

Methods

- **Data base search** : MEDLINE, CABI Helminthological abstract, INIST Pascal, Yahoo & Google.

Key words : *Diphyllobothrium latum*, countries

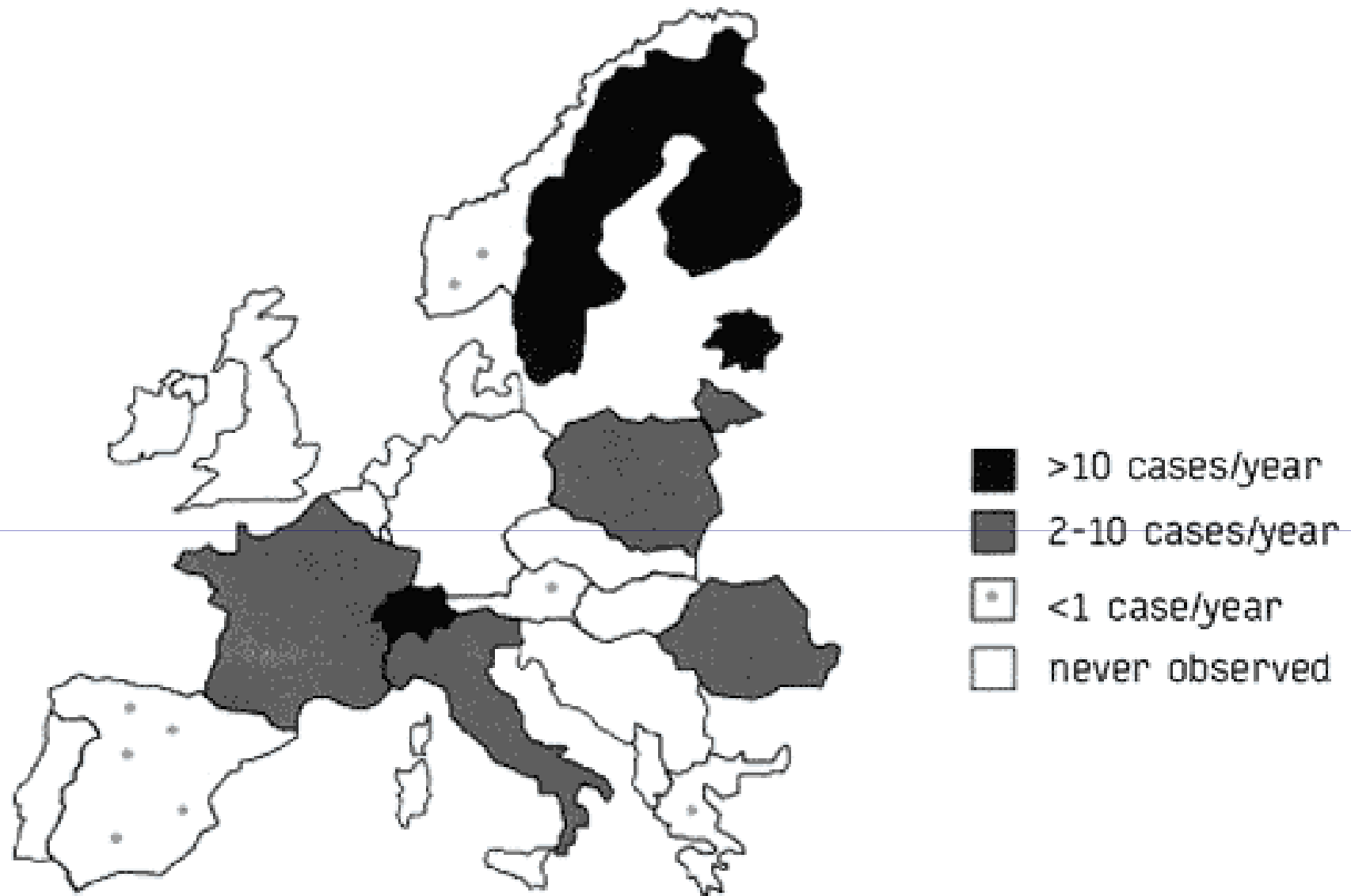
- **Form** :

- Network of European parasitologists
- Medical biology labs on the shores of French and Swiss alpine lakes

- **Limits** : 1980-2002

Results

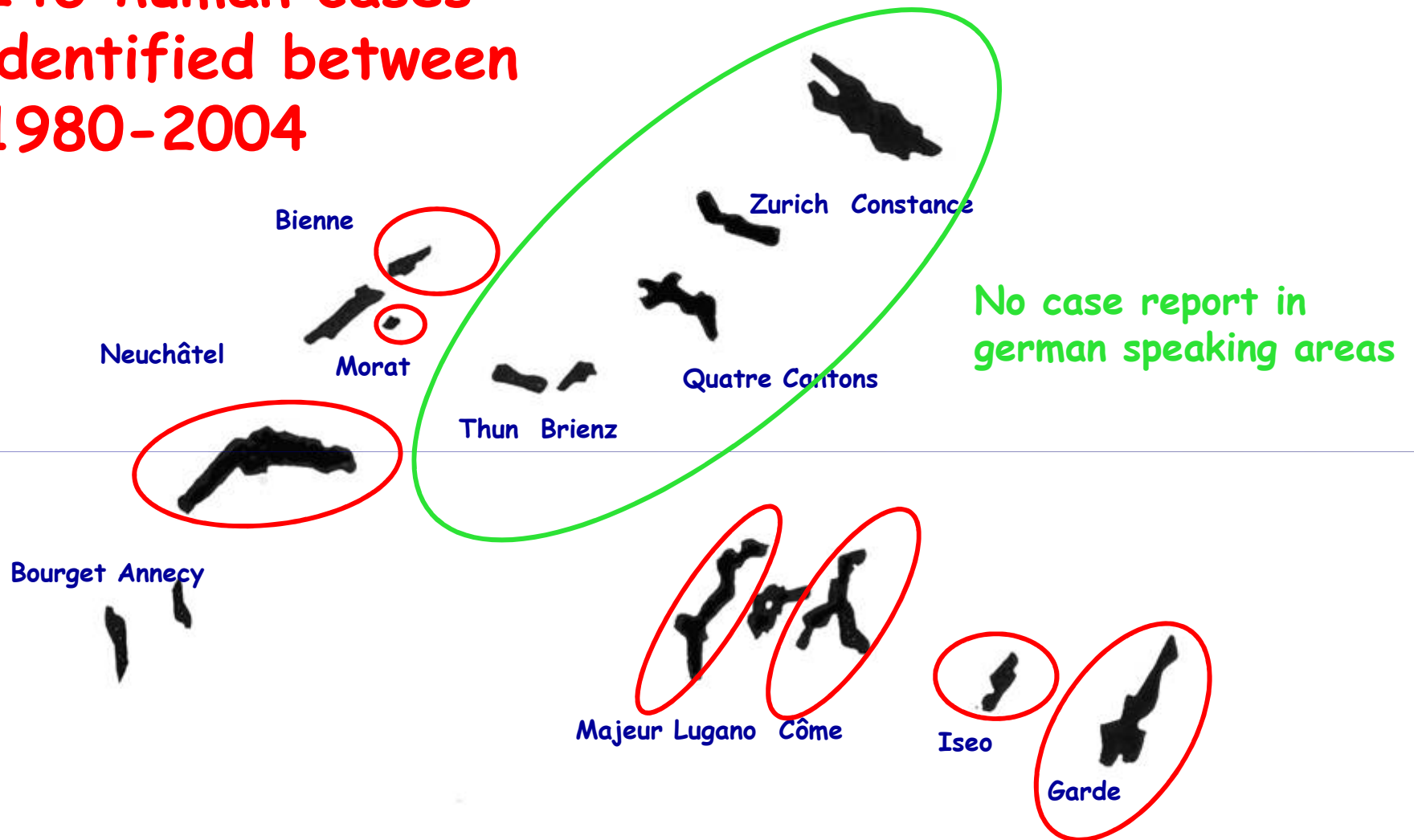
- Information from 24 European countries
- Yearly incidence of locally acquired cases
 - > 10 cases/year : Finland, Sweden, Estonia, Switzerland
 - 2-10 cases/year : France, Italy, Poland, Lithuania & Romania
 - Sporadic cases or no case from the remaining
- Report of sporadic cases acquired abroad or from imported fish



Human diphyllobothriosis in Europe since 1980

Alpine lakes region

246 human cases identified between 1980-2004



Contamination

- Many lake-side restaurants of Italian or French speaking areas offer dishes such as : "carpaccio di persico" (raw perch), sushi , "marinated lake fish fillets in Scandinavian style"
- Not in the restaurants of the German speaking areas

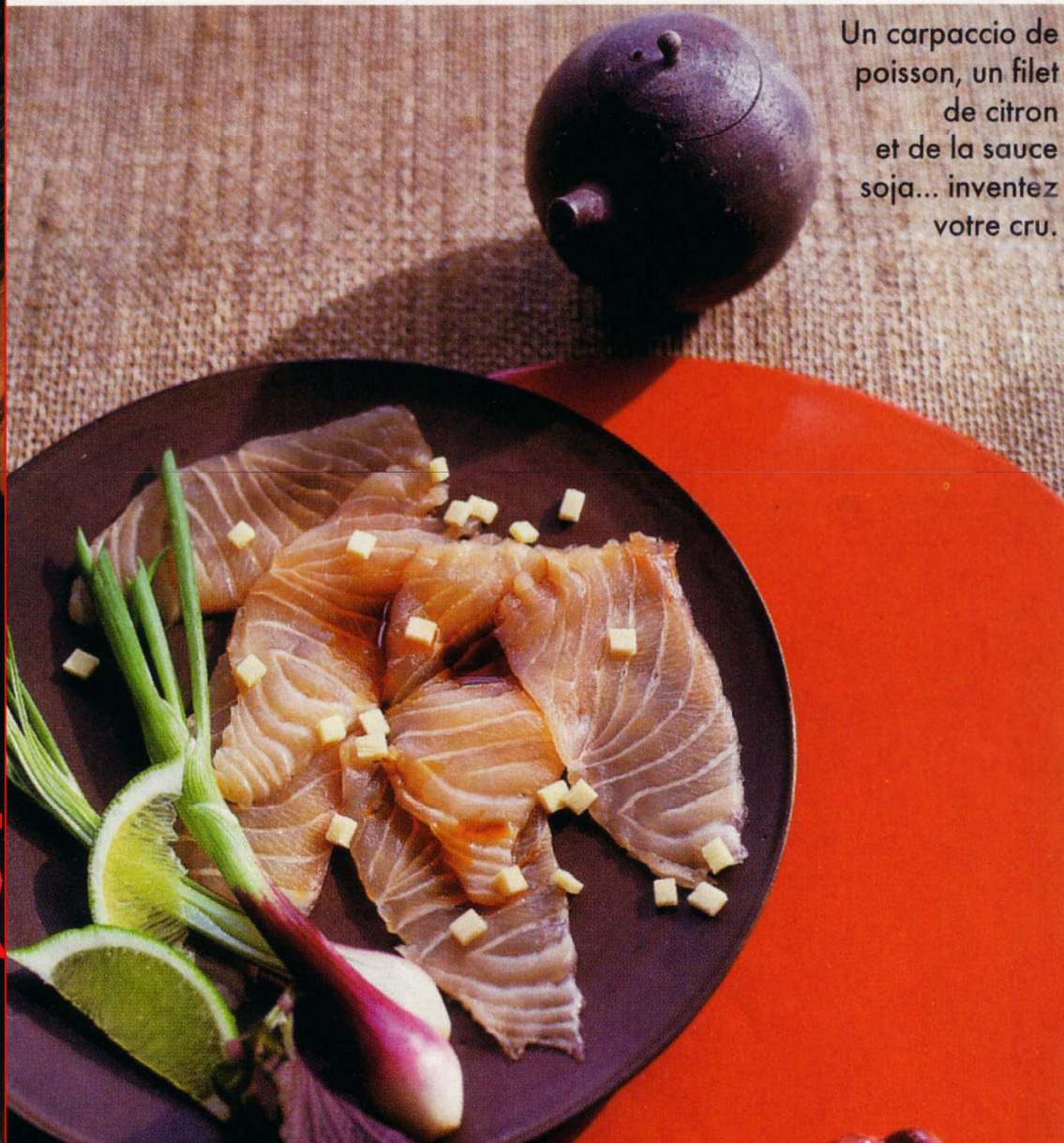


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ATOUT FOR
DE L'ÉTÉ**

**DIÉTÉTIQUE LES NOUVEAUX
GOÛTS DE LA FORME**



Un carpaccio de poisson, un filet de citron et de la sauce soja... inventez votre cru.

A persisting cycle of *D. latum* is a witness of the fecal pollution of the lakes

Waste Water Treatment Plants (WWTP) contribute to fight the parasite

but

- unconnected habitats
- 1-5 % of *D. latum* eggs may escape WWTP treatment
- WWTP may overflow during storms



Animal reservoir ?

- *D. latum* found in small carnivores

Germany : 0.5 % foxes
(Wesbecher, 1994)

Switzerland : 0.4 % dogs
(Sager, 2006) or sporadic cases in foxes (Deplazes, com. pers.)

- *D. latum* does not develop well in these hosts who may only play a minor role



Faecal pollution by boats



- Leisure boats ? new regulations require appropriate sanitary facilities onboard



- Fishermen have small boats without sanitation
- Fishermen are frequently infected!

CURRENT SITUATION OF HUMAN DIPHYLLOBOOTHRIASIS IN EUROPE

J Dupouy-Camet¹, R Peduzzi²

Diphyllobothriasis, a parasitosis caused by the flatworm *Diphyllobothrium latum*, is contracted by consuming raw or undercooked freshwater fish. The aim of this study was to evaluate the situation of this parasitosis during the past 20 years in Europe through the analysis of databases and search engines (Medline, CABI Helminthological abstracts, Yahoo, Google), and through a questionnaire sent to a network of European parasitologists and to microbiological laboratories located on the shores of the large Alpine lakes. This study has shown that several dozen cases have been reported each year in Finland and Sweden, that there have been numerous cases in the French or Italian speaking areas of subalpine lakes, and that sporadic cases only have been observed in Austria, Spain, Greece, Romania, Poland and Norway. Over 30 cases have been identified on the Swiss shores of Lake Maggiore since 1990, and 70 cases

on the Swiss and French shores of Lake Léman between 1993 and 2002. Eight to 12% of perch fillets from Lake Lemman and 7.8 % of perch from Lake Maggiore were infested with larvae. Contamination sources include marinated fish fillets in northern Europe, 'carpaccio di persico' in northern Italy, and perch and charr consumed raw or undercooked around Lake Léman. Factors allowing the continuation of the parasitic cycle include the continued dumping of wastewater into lakes, yachtsmen who also fish, and a possible animal reservoir.

Euro Surveill 2004;9:31-5

Key words : Diphyllobothriasis, parasitosis, lake fish, Europe

Euro Surveill. 2004;9:31-5

Species identification

Meeting French society of Parasitology, dec 2003

**Un risque parasitaire méconnu:
la Bothriocéphalose
par ingestion de saumon sauvage cru**

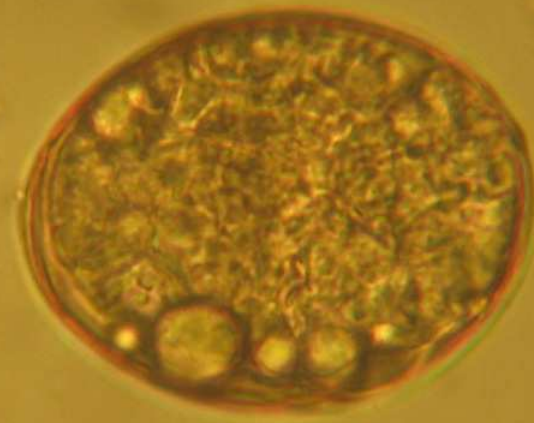
Christelle Estran, Cristel Fissore,
Martine Gari-Toussaint, Pascal Delaunay,
Frédérique Baudinetto, Yves Le Fichoux et Pierre
Marty

Parasitologie-Mycologie
Centre Hospitalier Universitaire de Nice

Bothriocéphalose et saumon sauvage

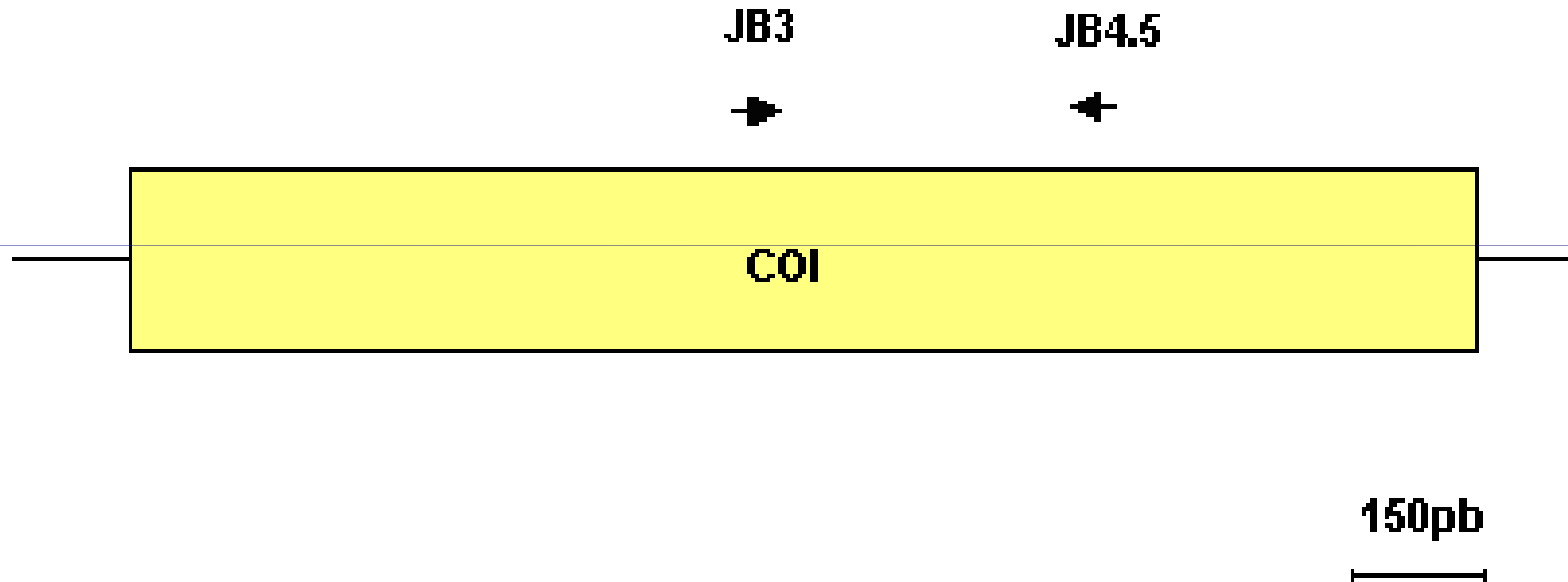


œufs de *Diphylobothrium* sp.



D. latum : 45 x 70 mm

Cytochrome oxydase



Very few sequences in Genebank at that time

T- T+


D. latum

T+

Proteocephalus



Acanthocephalus



10 20 30 40 50 60 70 80

>AF096240 *D. latum* FTTTGGGCATCCTGAGGTTTATGTTATACATTCTCCTGGATTTGGTATTATTAGACATATATGTTTAAAGAAATAGTAT
 >AB015755 *D. nihonkaiense* FTTTGGCCACCCSAAGTATATGTTTTAATTTTACCTGGTTTTGGATGATTAGCCATGTTTGTAGAAATTTAGGTTG
 >*D. specimen proglottid* GTTTTAATTTTACCTGGTTTTGGGATGATTAGCCATGTTTGTAGAAATTTAGGTTG
 >*D. latum* larva FTTTGGGCATCCTGAGGTTTATGTTTTAATTTTACCTGGTTTTGGGATGATTAGCCATGTTTGTAGAAATTTAGGTTG
 >*D. latum* proglottid FTTTGGGCATCCTGAGGTTTATGTTTTAATTTTACCTGGTTTTGGGATGATTAGCCATGTTTGTAGAAATTTAGGTTG

90 100 110 120 130 140 150 160

>AF096240 *D. latum* GTCCTCGGATGTTGTTTGGTTTTTATGGTTTATTGTTTGCTATGTTTCTATAGTTTGTTTAGGAAGAAGGTTGTGAGGTC
 >AB015755 *D. nihonkaiense* TTCATATGACACTTTTGGGTTTACGGGTTGTTATTTGCTATGTTTCTATAGTTTGCCTTAGGTACTGTTGTGTGGGGGC
 >*D. specimen proglottid* TTCATATGACACTTTTGGGTTTACGGGTTGTTATTTGCTATGTTTCTATAGTTTGCCTTAGGTACTGTTGTGTGGGGGC
 >*D. latum* larva TTCATATGACACTTTTGGGTTTACGGGTTGTTATTTGCTATGTTTCTATAGTTTGTTTAGGTAGAGTTGTGTGGGGGC
 >*D. latum* proglottid TTCATATGACACTTTGGGTTTACGGGTTGTTATTTGCTATGTTTCTATAGTTTGTTTAGGTAGAGTTGTGTGGGGGC

170 180 190 200 210 220 230 240

>AF096240 *D. latum* ATCATATGTTTACGTTGGGTTAGATGTAAGACGGCTGTGTTTTTTAGCTCTAATACTATGATTATTGGAGTACCTACA
 >AB015755 *D. nihonkaiense* ATCATATGTTTACAGTGGGTTAGATGTAAGACGGCTGTTTTCTTTAGTTCCGTTACTATGATTATTGGTGTACCGACT
 >*D. specimen proglottid* ATCATATGTTTACAGTGGGTTAGATGTAAGACGGCTGTTTTCTTTAGTTCCGTTACTATGATTATTGGTGTACCGACT
 >*D. latum* larva ATCATATGTTTACGGTGGGTTAGATGTAAGACGGCTGTTTTTTTAGTTCCAGTTACTATGATTATTGGAGTTCTACT
 >*D. latum* proglottid ATCATATGTTTACGTTGGGTTAGATGTAAGACGGCTGTTTTTTTAGTTCCAGTTACTATGATTATTGGAGTTCTACT

250 260 270 280 290 300 310 320


>AF096240 *D. latum* GGTATAAAGGTTTTTACATAATTGTATATGTTGTTCAATGCTCGAGTCAAAAAGASTGATCCTGTTTTATGATGAATTGT
 >AB015755 *D. nihonkaiense* GGTATAAAGGTAATTTTCATGACTATACATGATTTTAAAAAGTCGTGTTTCGTTGCGTGAGCCTGTTTTTTGGTGAGTTTT
 >*D. specimen proglottid* GGTATAAAGGTAATTTTCATGACTATACATGATTTTAAAAAGTCGTGTTTCGTTGCGTGAGCCTGTTTTTTGGTGAGTTTT
 >*D. latum* larva GGTATAAAGGTAATTTCTTTGGTTATATATGATTCTAAAAAGTCGTGTTTCACTGCGTGAGCCTGTTTTTTGGTGAGTTTT
 >*D. latum* proglottid GGTATAAAGGTAATTTCTTTGGTTATATATGATTCTAAAAAGTCGTGTTTCACTGCGTGAGCCTGTTTTTTGGTGAGTTTT

330 340 350 360 370 380 390 400

>AF096240 *D. latum* TTCCTTTATTATCTGTTTACGTTGGTGGGTTACTGGTATAGTATATCGGCTTGTGTTTTASATAAAGTGTAAATG
 >AB015755 *D. nihonkaiense* ATCATTATTGTTGTTATTCACTATCGGGGGTGTACGGGCATTATTCCTTCAGCTTGTGTACTTGATAATATTTGCATG
 >*D. specimen proglottid* ATCATTATTGTTGTTATTCACTATCGGGGGTGTACGGGCATTATTCCTTCAGCTTGTGTACTTGATAATATTTGCATG
 >*D. latum* larva ATCATTATTGTTGTTATTCACTATCGGGGGTGTACGGGTATTATACTCTCAGCTTGTGTACTTGATAATATTTGCATG
 >*D. latum* proglottid ATCATTATTGTTGTTATTCACTATCGGGGGTGTACGGGTATTATACTCTCAGCTTGTGTACTTGATAATATTTGCATG

410 420 430 440 450 460 470

>AF096240 *D. latum* ATACTTGATTTGTTGTAGCTCATTTCATTATGTTCTTTCTTTA
 >AB015755 *D. nihonkaiense* ATACTTGGTTTGTGTAGCTCACTTCCACTATGTAAGTCACTAGGATCTTATATAAGTATAATAGTGT
 >*D. specimen proglottid* ATACTTGGTTTGTGTAGCT
 >*D. latum* larva ATACTTGATTTGTTGTAGCTCATTTCATTATGTTCTTTCTTTA
 >*D. latum* proglottid ATACTTGATTTGTTGTAGCTCATTTCATTATGTTCTTTCTTTA



Putative *Diphyllbothrium nihonkaiense* acquired from a Pacific salmon (*Oncorhynchus keta*) eaten in France; genomic identification and case report[☆]

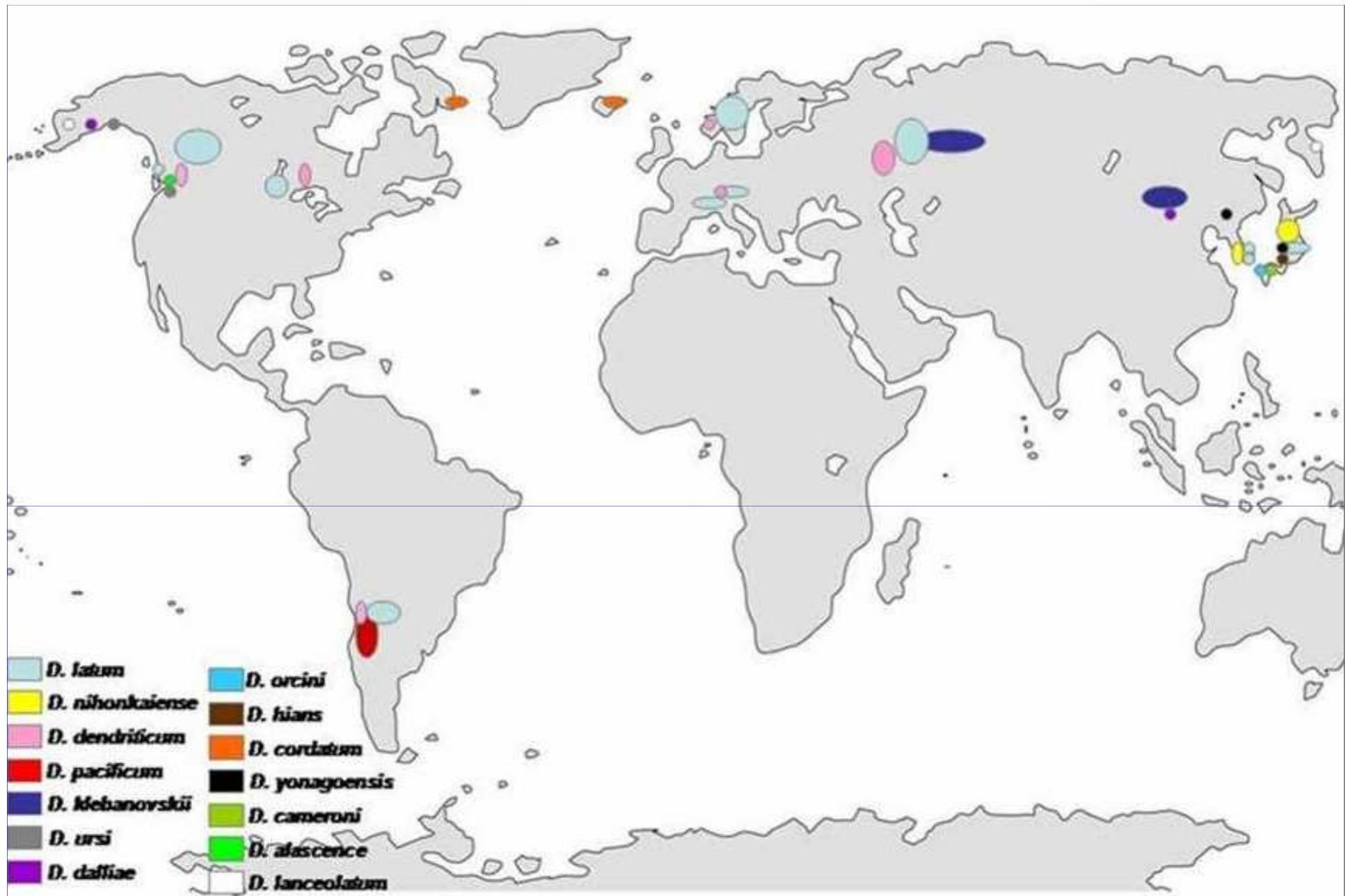
Hélène Yera^a, Christelle Estran^b, Pascal Delaunay^b, Martine Gari-Toussaint^b,
Jean Dupouy-Camet^{a,*}, Pierre Marty^b

Parasitol Int. 2006;55:45-9

Several species of *Diphyllobothrium* infect humans

(Dupouy-Camet & Yera. 2009)

Species	Fish Hosts	Other Hosts	Distribution
<i>D. alascense</i> (Rausch et Williamson, 1958)	Burbot, Smelt	Dogs	Alaska
<i>D. cameroni</i> ^b (Rausch, 1969)	Marine fishes	Monk seal	Japan
<i>D. cordatum</i> (Cobbold, 1869)	Marine fishes	Seals, walrus	North Atlantic
<i>D. dalliae</i> (Rausch, 1956)	<i>Dallia pectoralis</i>	Gulls, dogs	Alaska
<i>D. dendriticum</i> [*] (Nitzsch, 1824)	Salmonids, Coregonus	Fish eating birds	Circumpolar
<i>D. hians</i> ^b (Diesing, 1850)	Marine fishes	Seals	North Atlantic
<i>D. klebanovskii</i> ^c (Muratov and Psokhov, 1988)	Salmonids	Brown bears	Eastern Eurasia,
<i>D. lanceolatum</i> (Krabbe, 1865)	<i>Coregonus</i>	Seals	North Pacific
<i>D. latum</i> [*] (L., 1758)	Pike, Burbot, Percids	Dogs, bears	Cosmopolitan
<i>D. nihonkaiense</i> [*] (Yamane, 1986)	Salmon	?	Japan
<i>D. pacificum</i> [*] (Nybelin, 1931)	Marine fishes	Sea lions, fur seals	Peru, Chile
<i>D. ursi</i> (Rausch, 1956)	Red salmon	Bears	Alaska, British Columbia
<i>D. yonagoense</i> (Yamane, 1981)	Salmon	?	Japan, eastern Siberia
<i>Diplogonoporus grandis</i> ^d (Blanchard, 1889)	Anchovy, sardine	Cetacean	Japan



Distribution of species of *Diphylobothrium* infect humans
(Nicoulaud, 2007)

Genus identification quite easy with conventional methods

Adults:

macroscopic & microscopic examination

- Unarmed scolex with bothria
- Genital apertures on the ventral face of proglottids
- Oval and operculated eggs

Plerocercoid larvae:

macroscopic examination

- a few mm to cm long
- can be mistaken for *Proteocephalus*

Species identification difficult with conventional methods

Adult:

macroscopic & microscopic examination

- *D. latum* (up to 10 m) > others species (3 to 10 m)
- Eggs of *D. latum* (72x57 μm) > *D. nihonkaiense* (57x44 μm)

Plerocercoid larvae:

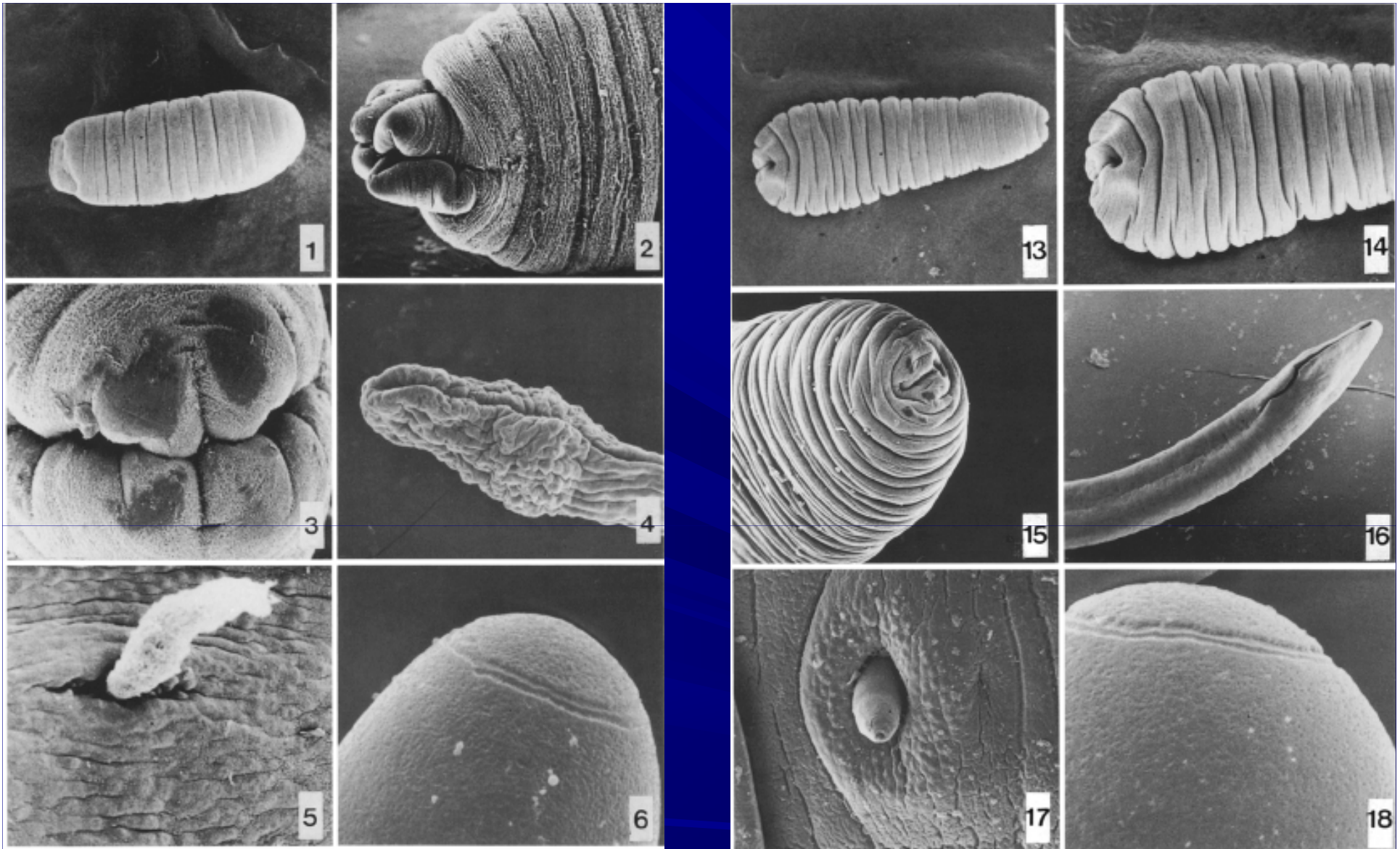
electronic microscopy

- Characteristics differ with the host

Scanning electron microscopic study of four *Diphyllbothrium* species

Y. Yamane¹, G. Bylund², K. Abe¹, Y. Osaki¹, and T. Okamoto¹

Parasitol Res 1989 75:238



Plerocercoids : 1, 2, 13, 14 Scolex of plerocercoid : 3, 15 Adults scolex : 4, 16
 Genital pore : 5, 17 Egg shell : 6, 18

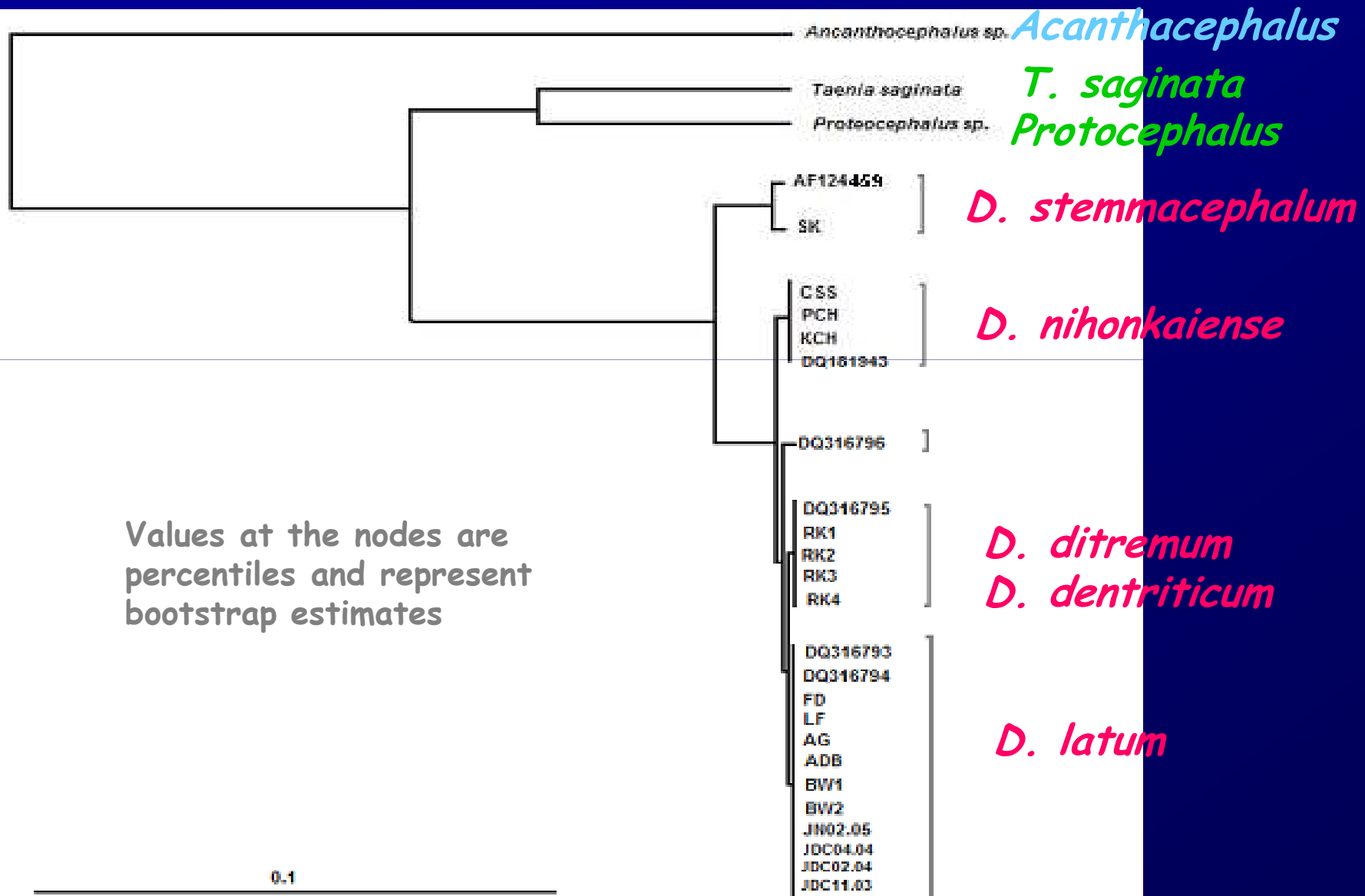
D. dendriticum

D. latum

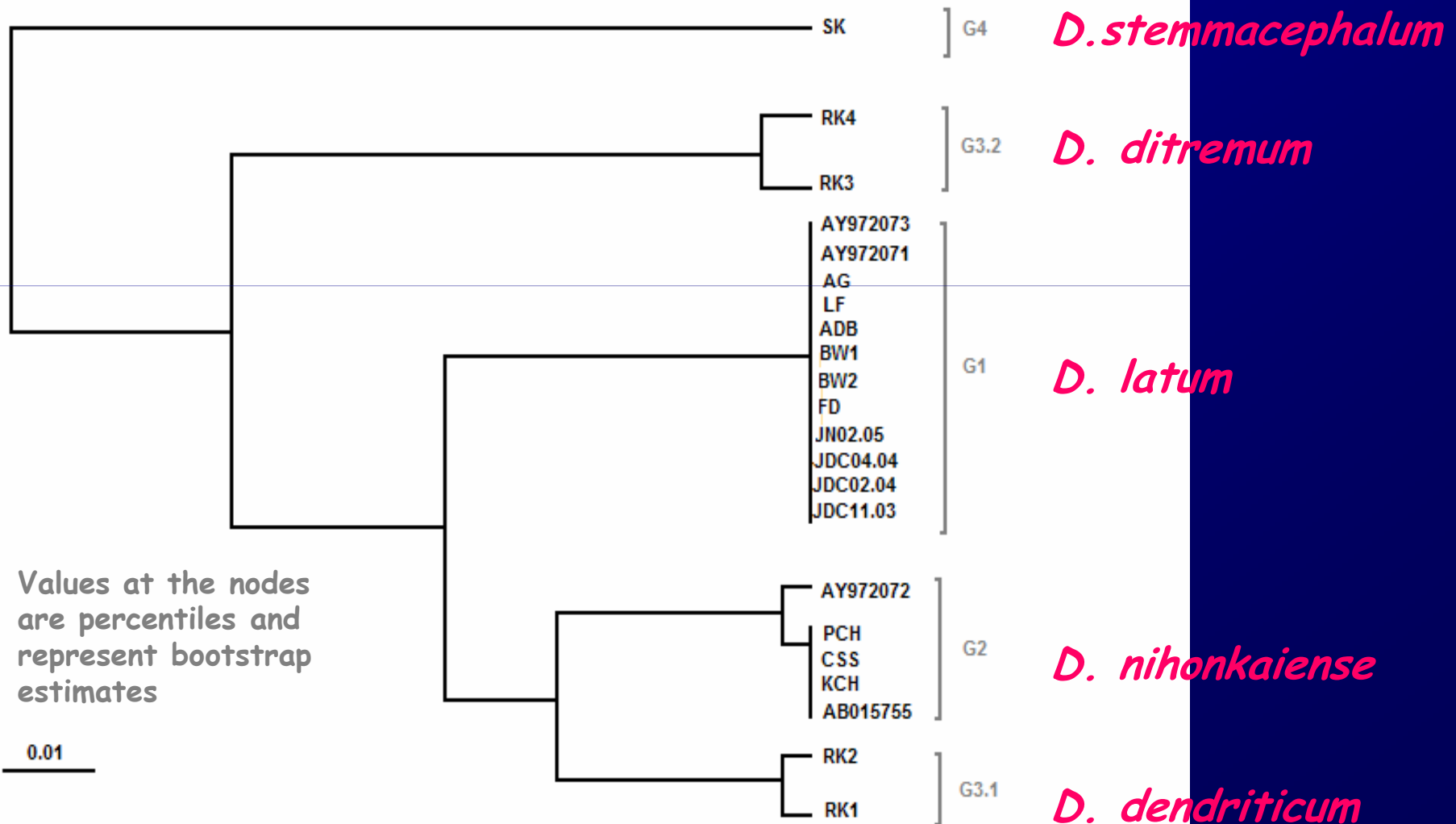
Molecular identification

- DNA amplification & sequencing
- Targets:
 - Partial **18S rDNA**
 - **ITS1-5,8S rDNA-ITS2**
 - Partial **cytochrome c oxidase I : COI**
 - others...
- Specimens: proglottids, eggs & larvae

Tree based on partial 18S rDNA sequences



Tree based on partial COI sequences



56 *Diphyllobothrium*

Jean Dupouy-Camet and H el ene Yera
Universit  Paris Descartes

CONTENTS

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56.1.2	Biology and Pathogenesis	769
56.1.3	Conventional Diagnostic Techniques.....	770
56.1.4	Molecular Diagnostic Techniques	771
56.2	Methods	773
56.2.1	Reagents and Equipments.....	773
56.2.2	Sample Preparation	773
56.2.3	Detection Procedure	773
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	References.....	775

Molecular Detection of Foodborne Pathogens

Edited by Dongyou Liu, CRC Press, Publication Date: 27/07/2009

Conclusions

- Emerging or re-emerging disease

Diphyllobothriasis, Brazil

Jorge Luiz Mello Sampaio,*

Victor Piana de Andrade,*

Maria da Conceição Lucas,* Liang Fung,*

Sandra Maria B. Gagliardi,*

Sandra Rosalem P. Santos,*

Caio Marcio Figueiredo Mendes,*

Maria Bernadete de Paula Eduardo,†

and Terry Dick‡

Cases of human diphyllobothriasis have been reported worldwide. Only 1 case in Brazil was diagnosed by our institution from January 1998 to December 2003. By comparison, 18 cases were diagnosed from March 2004 to January 2005. All patients who became infected ate raw fish in sushi or sashimi.

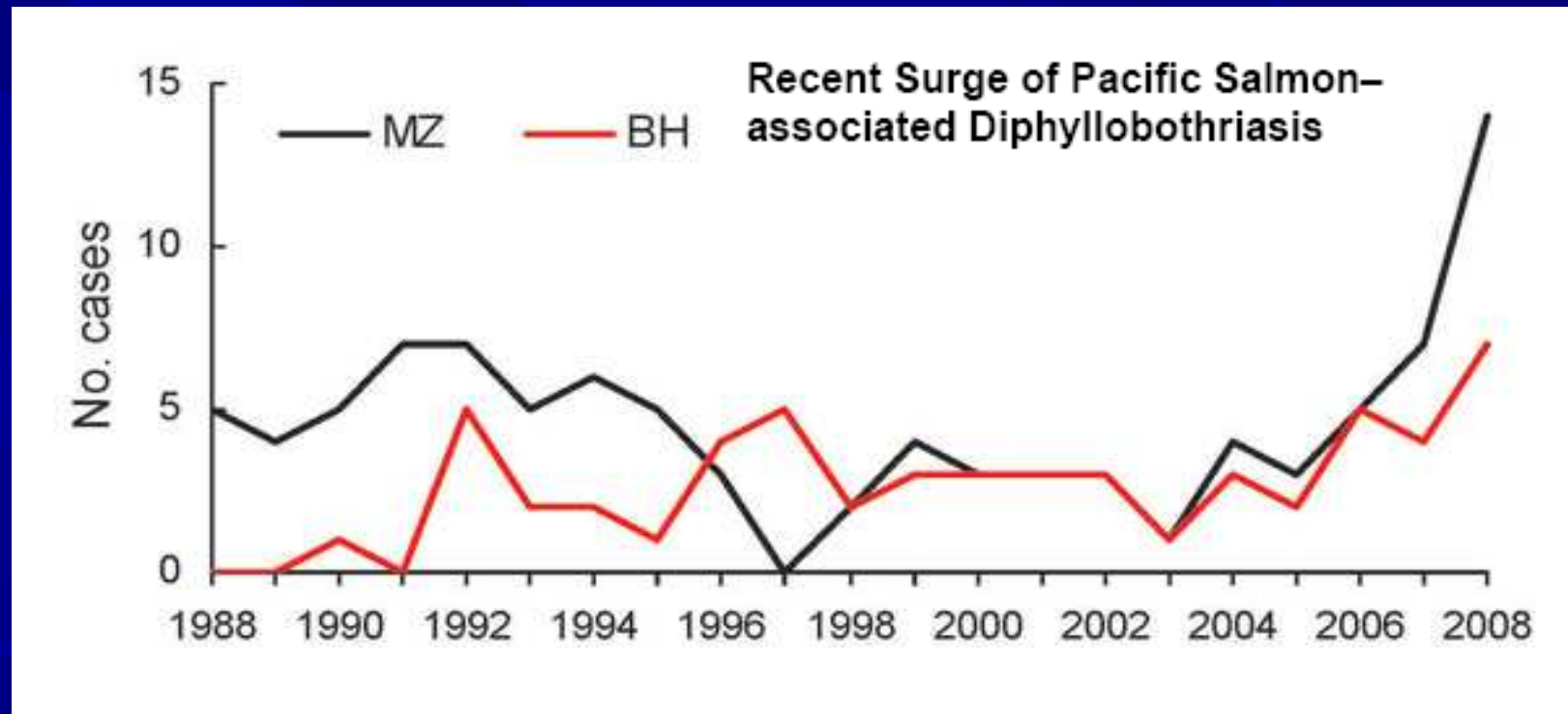
***Diphyllobothrium
latum* Outbreak
from Marinated
Raw Perch,
Lake Geneva,
Switzerland**

Yves Jackson,*
Roberta Pastore, ††
Philippe Sudre, † Louis Loutan,*
and François Chappuis*

Twenty-six wedding guests ate raw marinated perch. Seven confirmed cases and 1 probable case of *D. latum* infection occurred (attack rate 30.8%).

Diphyllobothriasis Associated with Eating Raw Pacific Salmon

Naoki Arizono, Minoru Yamada, Fukumi Nakamura-Uchiyama, and Kenji Ohnishi



Diphyllobothriasis cases, Department of Medical Zoology of the Kyoto Prefectural University of Medicine in Kyoto, [Arizono et al. EID June 2009](#)

Conclusion

- Emerging or re-emerging disease
- Sporadic imported cases caused by other *Diphyllobothrium* species

Putative *Diphyllobothrium nihonkaiense* acquired from a Pacific salmon (*Onchorynchus keta*) eaten in France; genomic identification and case report[☆]

Hélène Yera ^a, Christelle Estran ^b, Pascal Delaunay ^b, Martine Gari-Toussaint ^b, Jean Dupouy-Camet ^{a,*}, Pierre Marty ^b

Parasitol Int. 2006;55

Diphyllobothrium nihonkaiense (Yamane et al., 1986) in Switzerland: First molecular evidence and case reports[☆]

Barbara Wicht ^{a,*}, Floriane de Marval ^b, Raffaele Peduzzi ^a

Parasitol Int. 2007;70

Imported diphyllorbothriasis in Switzerland: molecular evidence of *Diphyllobothrium dendriticum* (Nitsch, 1824)

Barbara Wicht • Floriane de Marval • Bruno Gottstein •
Raffaele Peduzzi

Parasitol Res. 2007;102

First Record of Human Infection with the Tapeworm *Diphyllobothrium nihonkaiense* in
North America

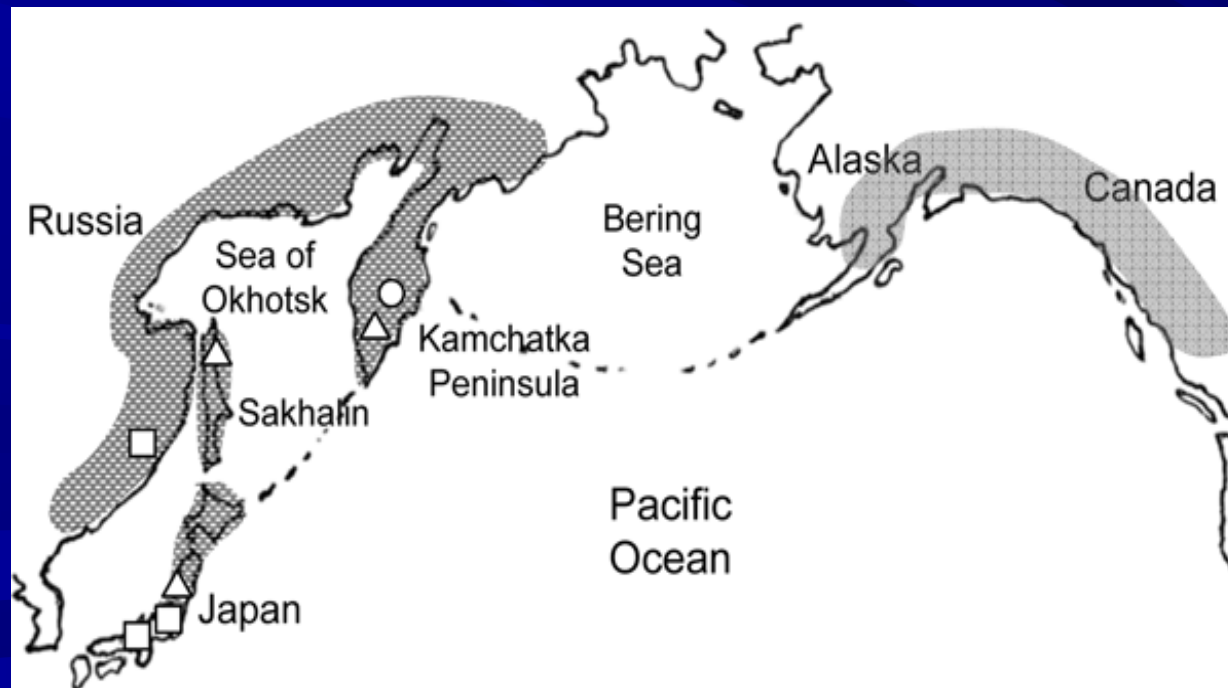
Barbara Wicht, Tomáš Scholz,* Raffaele Peduzzi, and Roman Kuchta

Am J Trop Med Hyg. 2008;78

New risk linked to *Oncorhynchus* sp consumption (Pacific salmon)

- *D. nihonkaiense* in Japan & USA
- *D. klebanovskii* in Russia

D. nihonkaiense = *D. klebanovskii*



Arizono et al.
2009

Conclusion

- Emerging or re-emerging disease
- Imported cases caused by other species
- Inform consumers and fish professionals on prophylactic methods
- Suppress faecal pollution of lakes
- Collaborative studies to type by molecular methods all *Diphyllbothrium* species : eg *D. ursi* specimens required !

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