



Subtype Distribution of *Blastocystis* in Humans



Supaluk Popruk

Department of Protozoology
Faculty of Tropical Medicine
Mahidol University

The Joint International Tropical Medicine Meeting (JITMM, 13 December 2013)

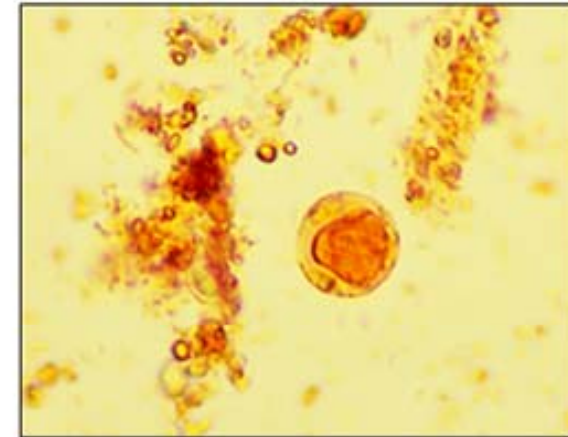
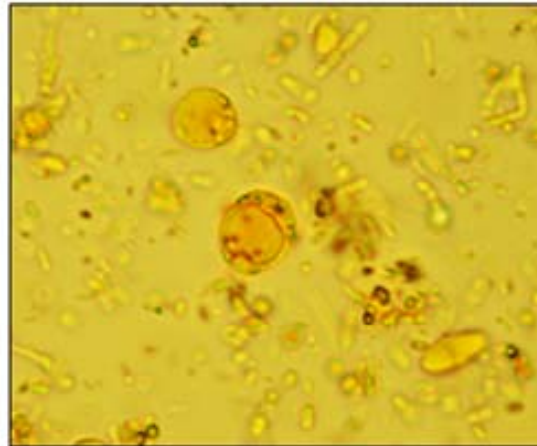


***Blastocystis* spp.**

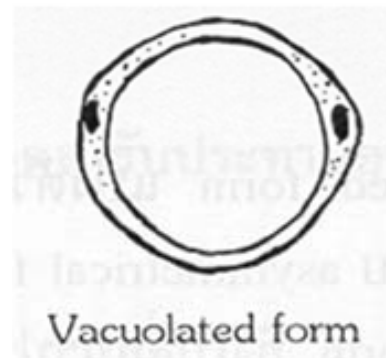
- Inhabits in intestinal tracts of humans and was named *Blastocystis hominis*
- But the name was later changed to ***Blastocystis* spp.** due to an indistinguishable difference between those found in humans and in other animals
- Worldwide distribution
- Mode of transmission: Fecal-oral route

Morphology of *Blastocystis* spp.

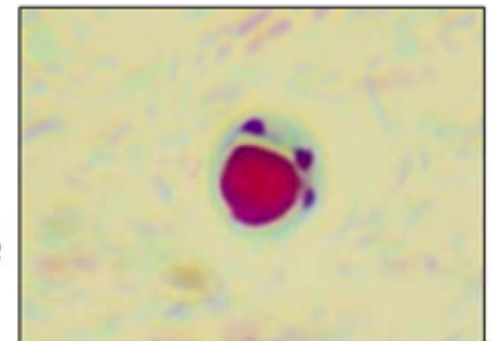
1. Vacuolar form
2. Granular form
3. Amoeboid form
4. Cyst form



Blastocystis : vacuolated
form in iodine



Blastocystis :
with trichrome





Blastocystis spp.

- 17 subtypes were found in humans and animals
- ST1-ST4 are more common in humans
- pathogenicity is inconclusive
- microscopy, culture & molecular techniques

Blastocystis isolated from humans belong to the same subtypes seen in animals, suggesting that animals may act as reservoirs for *Blastocystis* and may be linked to **zoonotic transmission**





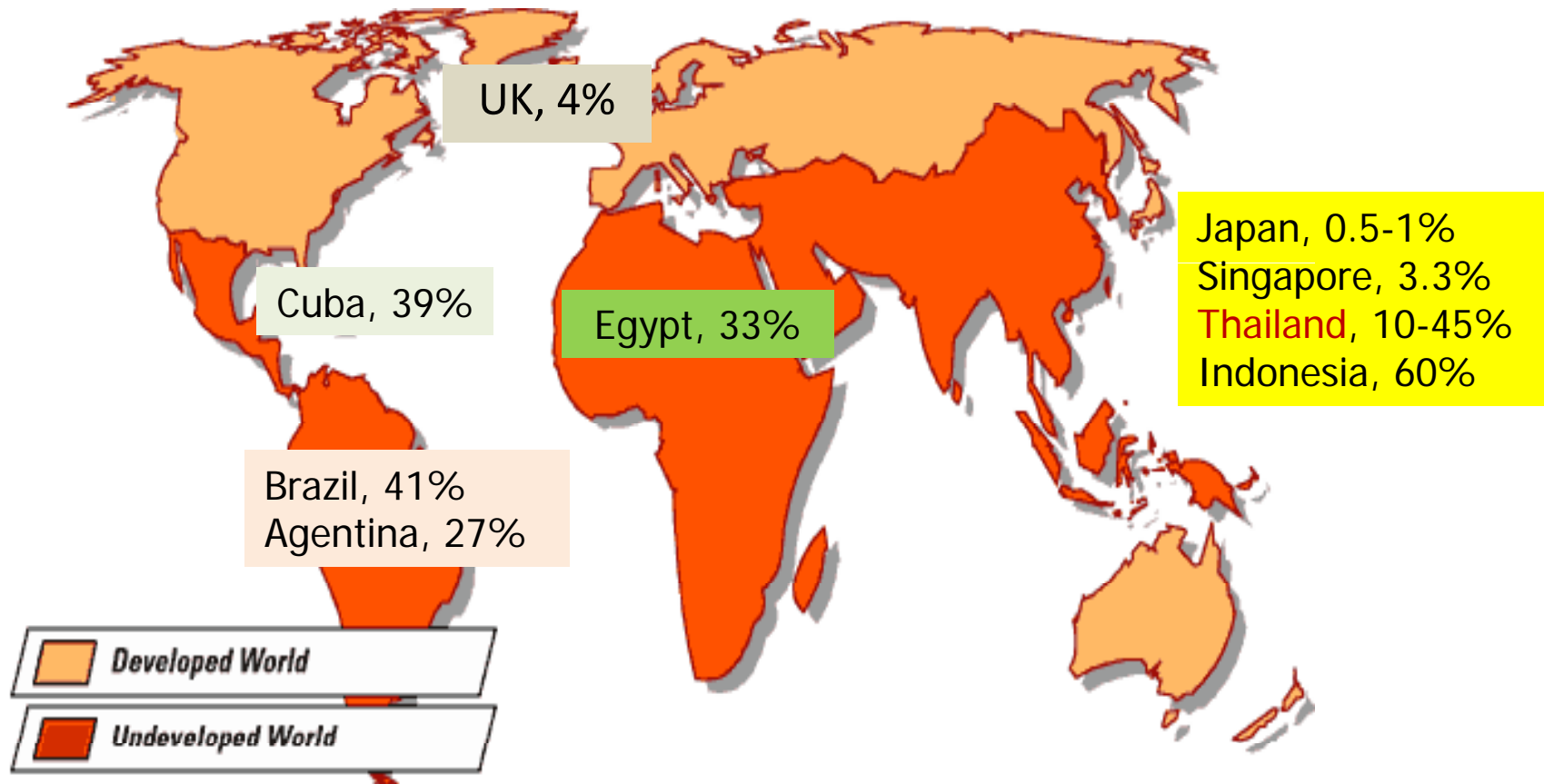
Blastocystis subtypes in various hosts

Subtype	Hosts
1	Human, vervet monkey, pig
2	Human, macaque, pig
3	Human, cattle, pig, baboon
4	Human, rat, pig
5	Human, pig, cattle
6	Human, turkey, chicken
7	Human, duck
8	Human, lemur, pheasant
9	Human





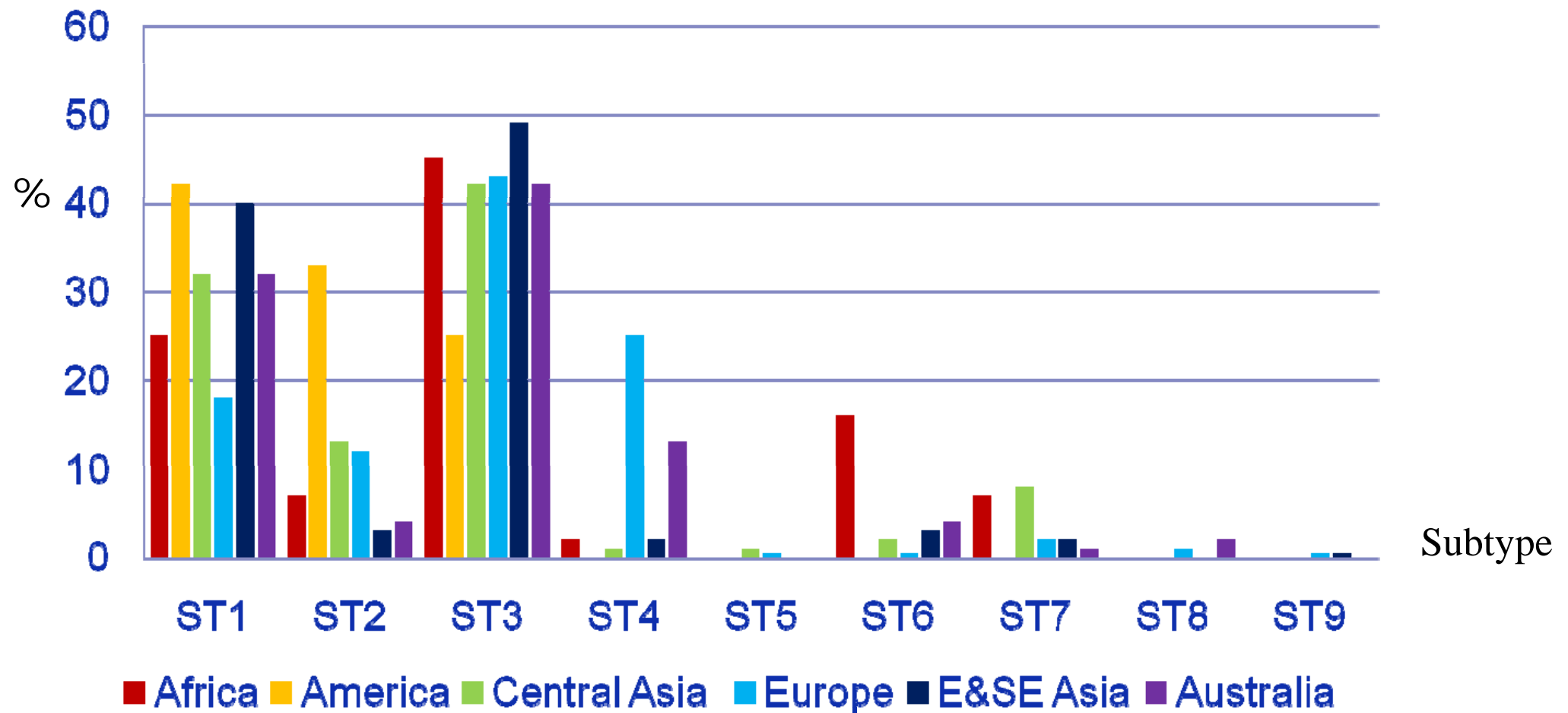
Prevalence of *Blastocystis*



Rayan *et al.*, 2010



Prevalence of *Blastocystis* subtypes by geographic regions





No. of subtype observation

Americas total: 93

ST1=39

ST2=31

ST3=23

100% Sequencing

Brazil

Colombia

USA





No. of subtype observation

Americas total: 93

ST1=39

ST2=31 **100% Sequencing**

ST3=23



No. of subtype observation

Europe total: 1149

ST1=186

ST2=146

ST3=506

ST4=278

ST5-ST9=33

Mixed ST=51

90% Sequencing

10% STS



- Denmark
- France
- Germany
- Ireland
- Italy
- Spain
- Sweden
- UK



No. of subtype observation

Americas total: 93

ST1=39

ST2=31 100% Sequencing

ST3=23

No. of subtype observation

Europe total: 1149

ST1=186

ST2=146

ST3=506

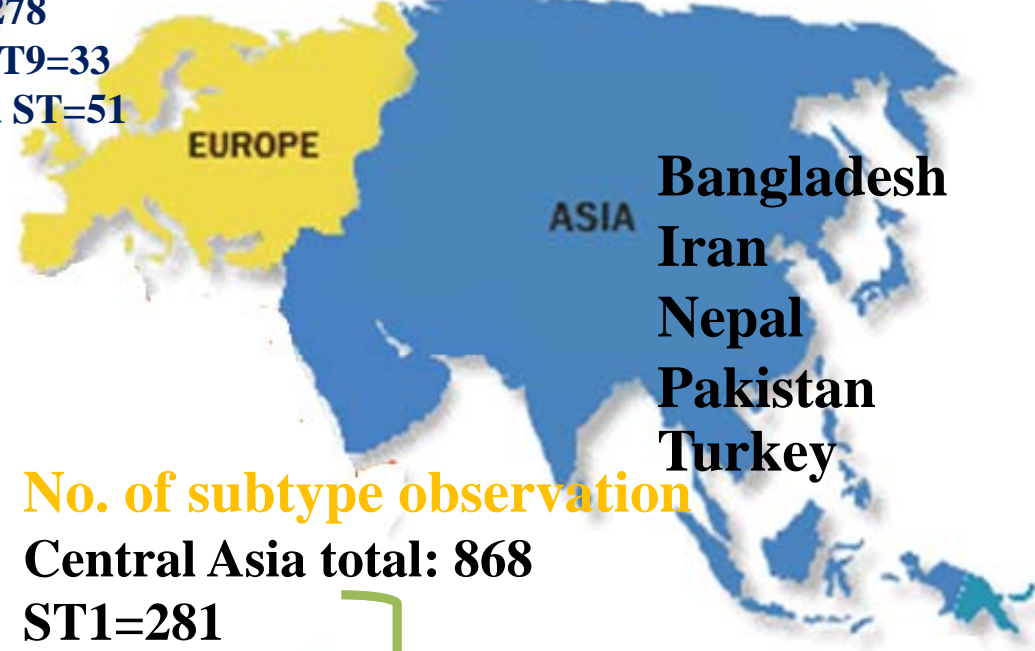
ST4=278

ST5-ST9=33

Mixed ST=51

90% Sequencing

10% STS



No. of subtype observation

Central Asia total: 868

ST1=281

ST2=118

ST3=368

ST4=9

ST5=15

ST6=70

Mixed ST=90

95% STS

5% Sequencing



No. of subtype observation

Americas total: 93

ST1=39

ST2=31

ST3=23

100% Sequencing

NORTH AMERICA

LATIN AMERICA

No. of subtype observation

Europe total: 1149

ST1=186

ST2=146

ST3=506

ST4=278

ST5-ST9=33

Mixed ST=51

90% Sequencing

10% STS

EUROPE

No. of subtype observation

Central Asia total: 868

ST1=281

ST2=118

ST3=368

ST4=9

ST5=15

ST6=70

Mixed ST=90

95% STS

5% Sequencing

ASIA

No. of subtype observation

E and SE-Asia total: 692

ST1=279

ST2=26

ST3=338

ST4=11

ST6=25

ST7=11

ST9=2

Mixed ST=24

100% STS

China

Japan

Malaysia

Singapore

Thailand



No. of subtype observation

Americas total: 93

ST1=39

ST2=31

ST3=23

100% Sequencing

NORTH AMERICA

LATIN AMERICA

No. of subtype observation

Europe total: 1149

ST1=186

ST2=146

ST3=506

ST4=278

ST5-ST9=33

Mixed ST=51

90% Sequencing

10% STS

EUROPE

No. of subtype observation

Africa total: 266

ST1=64

ST2=17

ST3=120

ST4=6

ST6=41

ST7=18

Mixed ST=17

95% Sequencing

5% STS

AFRICA

Egypt

Liberia

Libya

Nigeria

Tanzania

No. of subtype observation

Central Asia total: 868

ST1=281

ST2=118

ST3=368

ST4=9

ST5=15

ST6=70

Mixed ST=90

95% STS

5% Sequencing

ASIA

No. of subtype observation

E and SE-Asia total: 692

ST1=279 100% STS

ST2=26

ST3=338

ST4=11

ST6=25

ST7=11

ST9=2

Mixed ST=24

No. of subtype observation

Americas total: 93

ST1=39

ST2=31 100% Sequencing

ST3=23

NORTH AMERICA

LATIN AMERICA

No. of subtype observation

Europe total: 1149

ST1=186

ST2=146 90% Sequencing

ST3=506 10% STS

ST4=278

ST5-ST9=33

Mixed ST=51

EUROPE

No. of subtype observation

E and SE-Asia total: 692

ST1=279 100% STS

ST2=26

ST3=338

ST4=11

ST6=25

ST7=11

ST9=2

Mixed ST=24

AFRICA

No. of subtype observation

Africa total: 266

ST1=64

ST2=17

ST3=120

ST4=6

ST6=41

ST7=18

Mixed ST=17

95% Sequencing

5% STS

No. of subtype observation

Central Asia total: 868

ST1=281

ST2=118 95% STS

ST3=368 5% Sequencing

ST4=9

ST5=15

ST6=70

Mixed ST=90

ASIA

No. of subtype observation

Australia total: 103

ST1=33

ST2=5

ST3=44

ST4=14

ST6=4

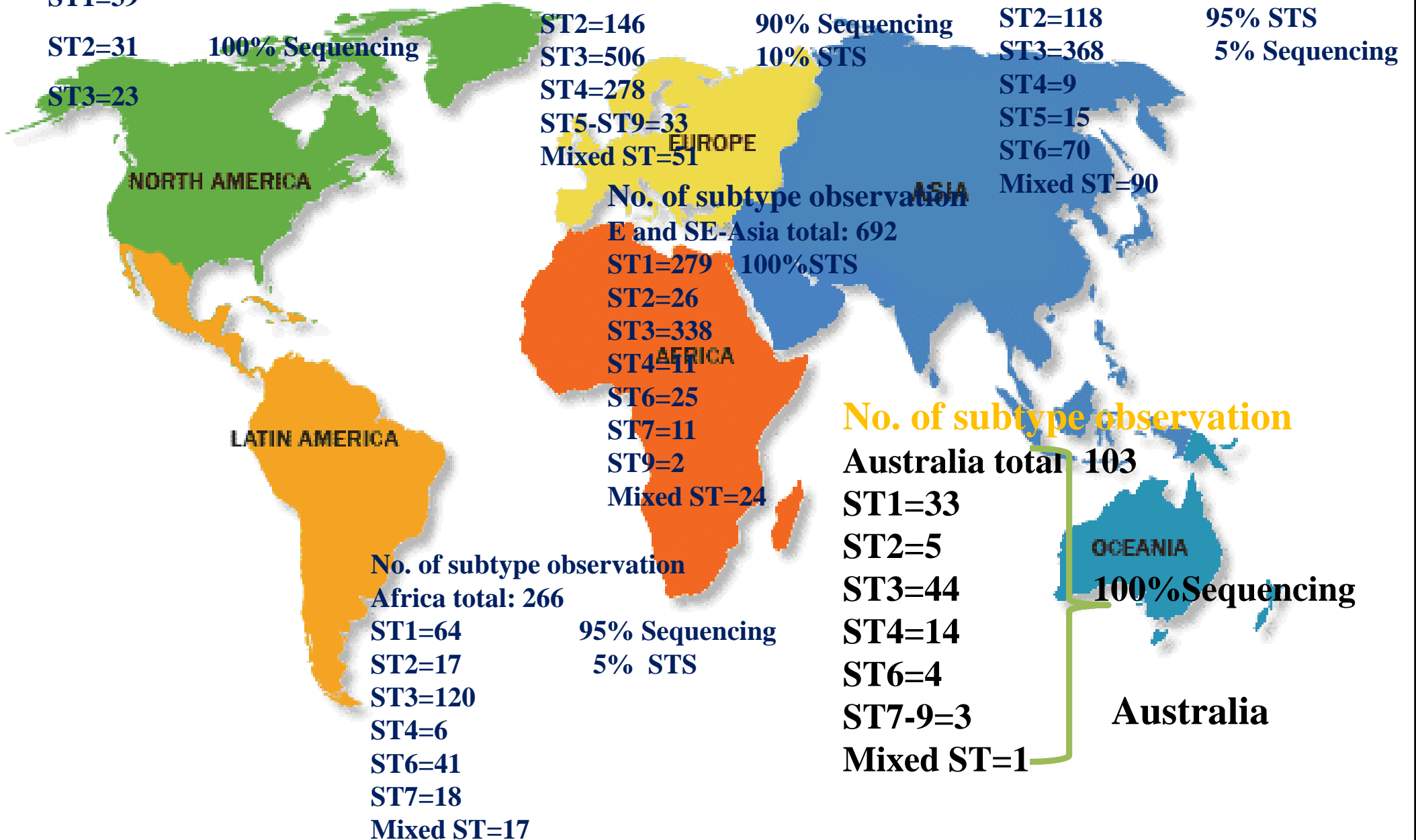
ST7-9=3

Mixed ST=1

OCEANIA

100% Sequencing

Australia





No. of *Blastocystis* subtype observation

Earth total: 3171

ST1=882

ST2=343

ST3=1399

ST4=318

ST5=9

ST6=89

ST7=118

ST8=10

ST9=3

Mixed ST=191





Subtyping has been approached in mainly 2 ways:

- **STS primers method (sequence-tagged sites primers)**
- **Genus-specific primers with subsequent sequencing for ST identification**

STS primers method

- **Available for ST1-ST7 (7 pairs of primers, separately perform in each primer pairs)**
- **Human carriage due to other STs remains undetected.**
- **detect mixed infection**
- **Some of the primer pairs appear rather insensitive, especially **ST4****



What did you observe in subtype distribution of *Blastocystis* spp. ?

- Geographic areas
- Methodologies





**MAHIDOL
UNIVERSITY**

Wisdom of the Land

MOLECULAR DETECTION AND SUBTYPING OF *BLASTOCYSTIS* SPP. IN THAI CHILDREN AND ADOLESCENT ORPHANS

Ms. AI-RADA PINTONG

5436335 TMTM/M



Research questions



- What is the prevalence and subtype of *Blastocystis* in this orphanage?
- What is the potential source of *Blastocystis* in this orphanage?

Objectives

- To investigate the prevalence and subtype of *Blastocystis* in an orphanage, Pathum Thani Province, Thailand
- To identify the potential source of *Blastocystis* contamination by subtyping and phylogenetic analysis



Process of this study

STEP1 Prevalence study

STEP2 Evaluation of the potential source of transmission based on the subtyping study

1) Human specific subtype

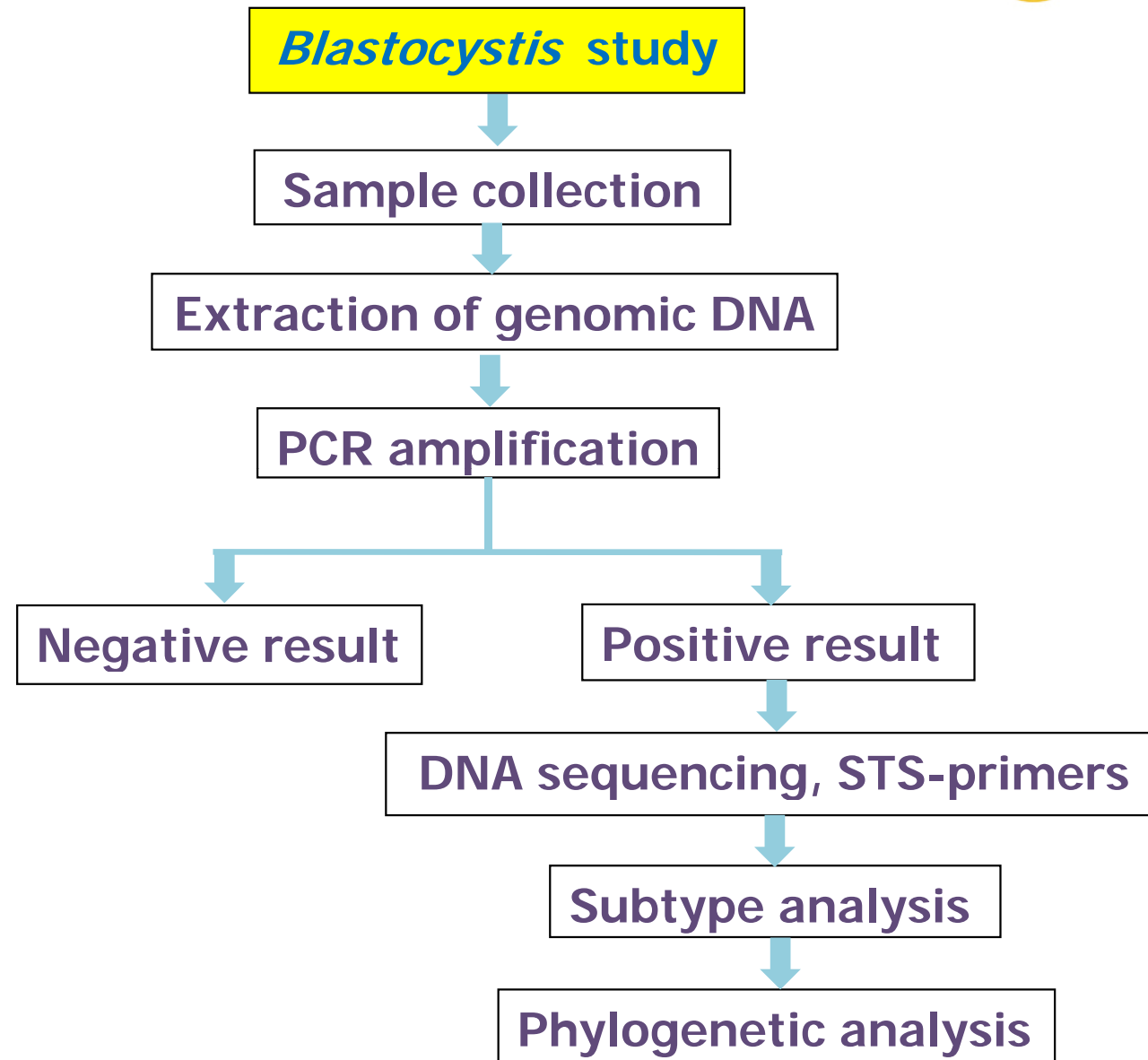
Evaluation of water supply, human stool samples

2) Zoonotic subtype

Zoonotic transmission

Animal contact

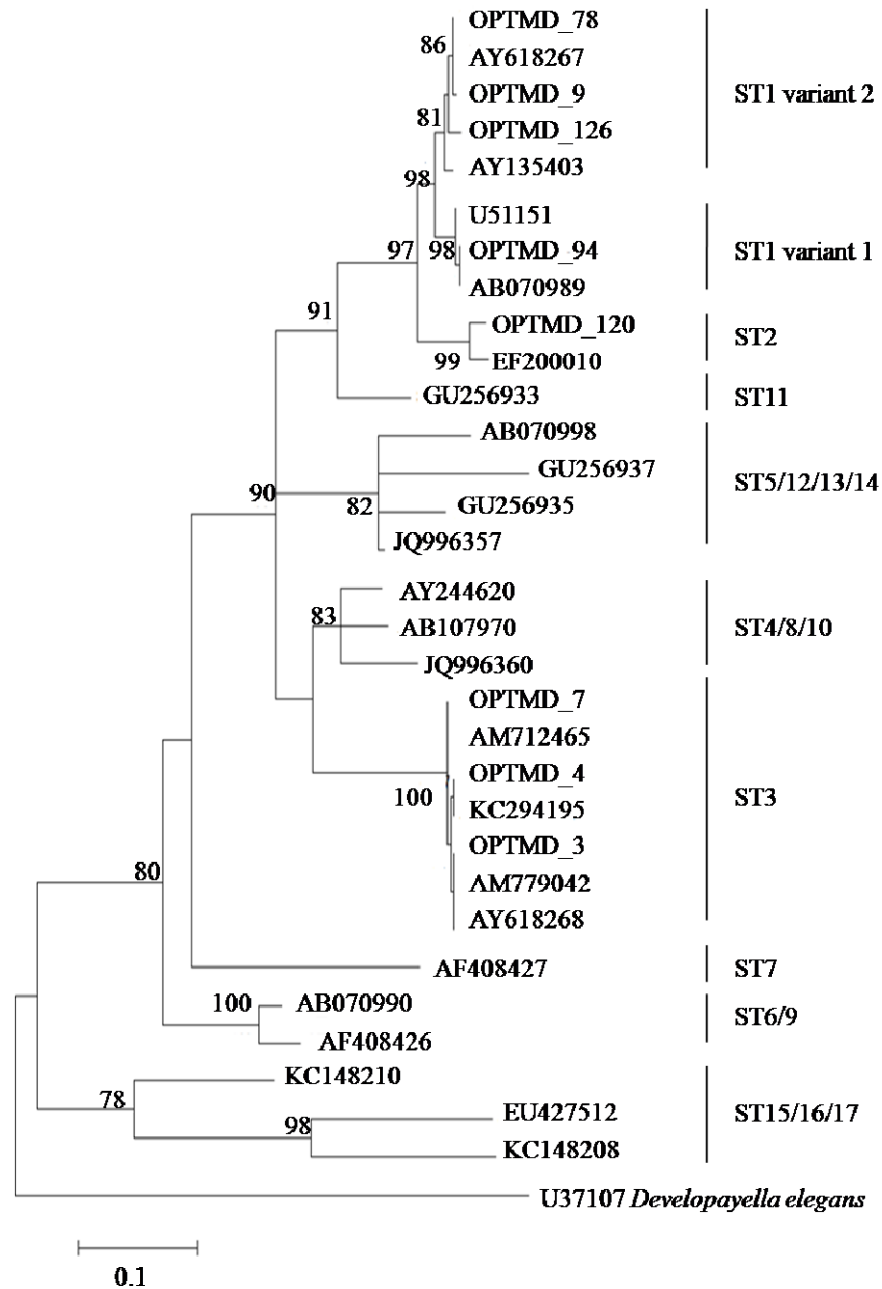
Study design



Results

Samples	Prevalence	Subtype
Human stool	51.2% (64/125)	ST3 with 51/64 (79.69%), followed by ST1 with 12/64 (18.75%), and ST2 with 1/64 (1.56%)
Animal stool	Negative	-
Drinking water	Negative	-

Results



Phylogenetic

- ST1 subgroups: variants 1 and 2
- ST3 showed no distinct subgroup
- OPTMD isolates 3, 4, and 7 (KF285448-KF285450) showed highly similar sequences

Discussion

- 51.2% the highest so far reported in Thailand
- Predominant subtype was ST3 in this study
- Animal stool and drinking water samples were negative
- The potential source of *Blastocystis* was human via human to human
- Orphans live together and share facilities within the household

Thank You

