

# Genetic diversity and population structure analysis of various *Taenia multiceps*

## isolates from definitive and intermediate hosts worldwide



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## Introduction

- *Taenia multiceps* is a ubiquitous taeniid cestode that circulates in a two-host life cycle.
- Various canids are definitive hosts and carry the adult worms in intestine.
- A broad range of domestic and wild ungulates can serve as intermediate hosts and carry the larval stage (coenuri).
- The dog-sheep/goat cycles are the most common.
- In sheep and goats, the coenuri typically inhabit CNS, and are often fatal.



Source: <https://www.istockphoto.com/photo/border-collie-dog-herding-a-flock-of-sheep-gm511644479-46598650>

- Few surveys from some East Mediterranean and African countries observed extra-CNS coenuri mostly in subcutaneous tissues of sheep and goats.

## Background

- Great debate whether CNS and non-CNS coenuri belong to the same parasite species/strain.
- Verster (1969) considered both types of coenuri to belong to the same parasite species based on morphology.
- Several cross-transmission attempts and results were controversy.
- Studies conducted on isolates from Iran found no morphological or molecular differences
- Christodoulopoulos et al. (2016) have observed distinct morphological differences, but not enough to typify 2 separate species, and proposed 3 genetic variants, one can produce both cerebral and non-cerebral coenuri, the other 2 variants can produce only cerebral coenuri.
- Varcasia et al. (2012): new genetic variant for non-cerebral coenuri from goats in UAE.

## Shortcomings

- For comparison, studies utilized a few isolates, either cerebral or non-cerebral, from limited regions.
- Even in Christodoulopoulos et al. (2016) investigation, non-cerebral coenuri have been collected from several countries, but have been compared to the cerebral isolates from Greece only.
- This could influence results impartiality.

**Therefore**, a study that analyzes all of the worldwide sequenced *T. multiceps* isolates would provide a clear picture about the molecular situation of both types of coenuri.

**Aim** The present study provides the first comprehensive genetic analysis for all published *T. multiceps* nucleotide sequences from various definitive and intermediate hosts

# Data collection

- Systematic search for website of the National Center for Biotechnology (<http://www.ncbi.nlm.nih.gov>) to collect all *T. multiceps* partial cox1 nucleotide sections (the most used).
- Combinations of various terms: *Taenia multiceps*, *Coenurus cerebralis*, *Coenurus gaigeri*, *Coenurus skrjabini*, sheep, goat, cattle, dogs, and cox1.
- 233 partial cox1 sequences were collected.
- Sequences represented 442 isolates from sheep (n = 297), goat (n = 109), cattle (n = 6), dog (n = 26), fox (n = 1), mouflon (n = 1) and yak (n = 1).
- Isolates belonged to various developmental stages of *T. multiceps* either adult worms (n = 27) or coenuri (cerebral = 342; non-cerebral = 72).

# Genetic analysis

- Alignment and trimming of the collected sequences using ClustalW module of MEGA 6.
- Maximum Likelihood phylogenetic analysis using MEGA6.
- Transformation of the aligned sequences to Nexus format using MEGA6.
- Haplotype networks in relation to country of origin, developmental stage and host species using PopArt1.7.
- Population structure analysis using DnaSP6.
- Various indices were estimated: e.g., haplotype and nucleotide diversity, pairwise genetic difference, gene flow

# Results

- The aligned cox1 sections = 337bp in length.
- 59-point mutations, of which 28 were parsimony informative.
- Nucleotide diversity ( $\pi$ ) =  $0.00915 \pm 0.00032$ .
- 51 haplotypes.
- Haplotype diversity =  $0.859 \pm 0.00009$  SD.



Low nucleotide + high haplotype  
**rapid population expansion**  
from small effective population size

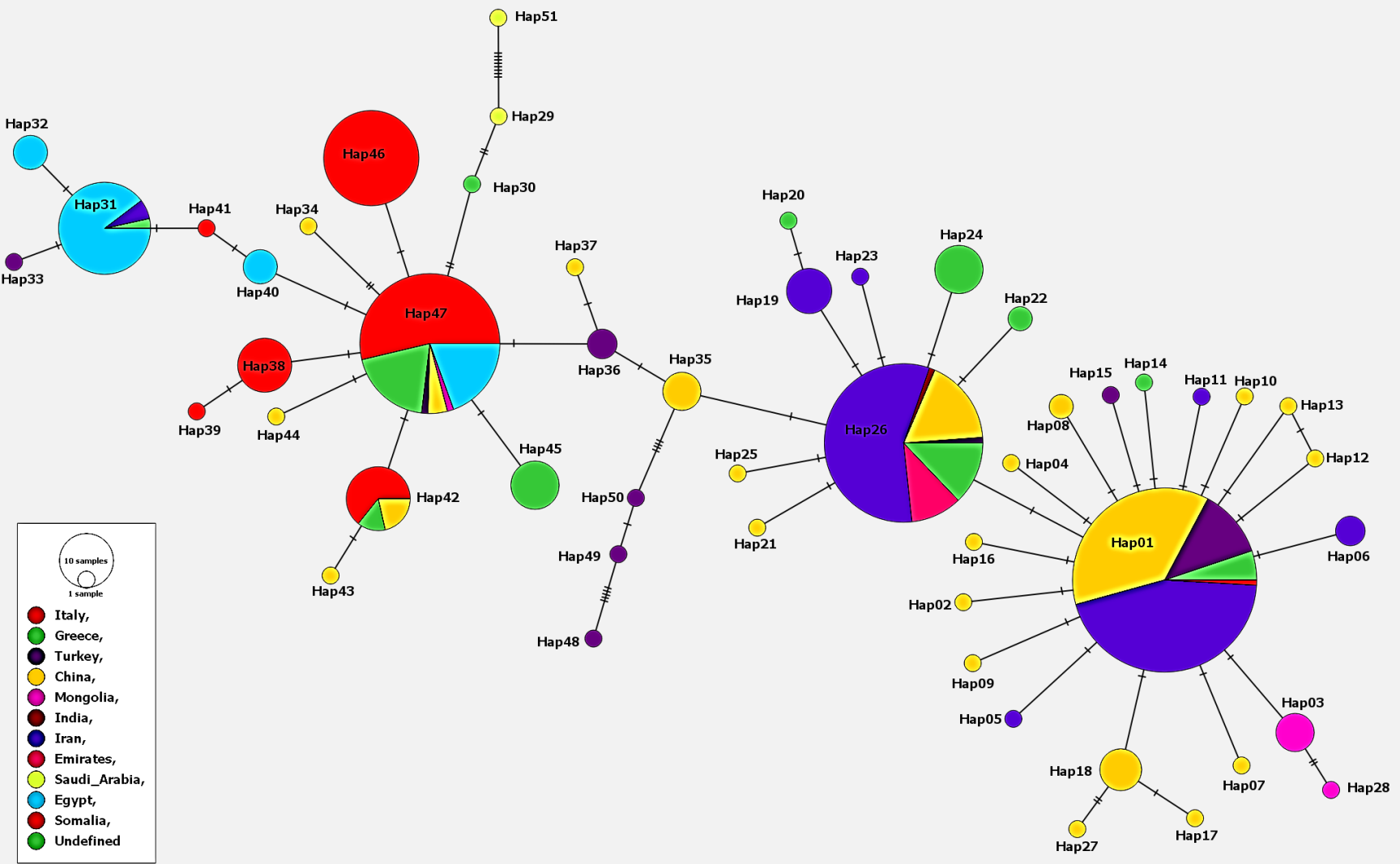


**Confirmed by**

**negative value of Tajima's D (-1.90106) and Fu's Fs (-35.055) tests.**

- Sequences came from 12 countries
- **6 populations** identified acc. To geography, due to limited number of isolates
- China, Turkey, Iran, Egypt, Italy, Greece

- 5 major haplotypes.
- 2 in China, Turkey, Iran.
- 3 in Egypt, Italy.
- Haplotype distribution could be related to geographical neighborship (e.g., Turkey, Iran).
- Greece circulate worldwide.

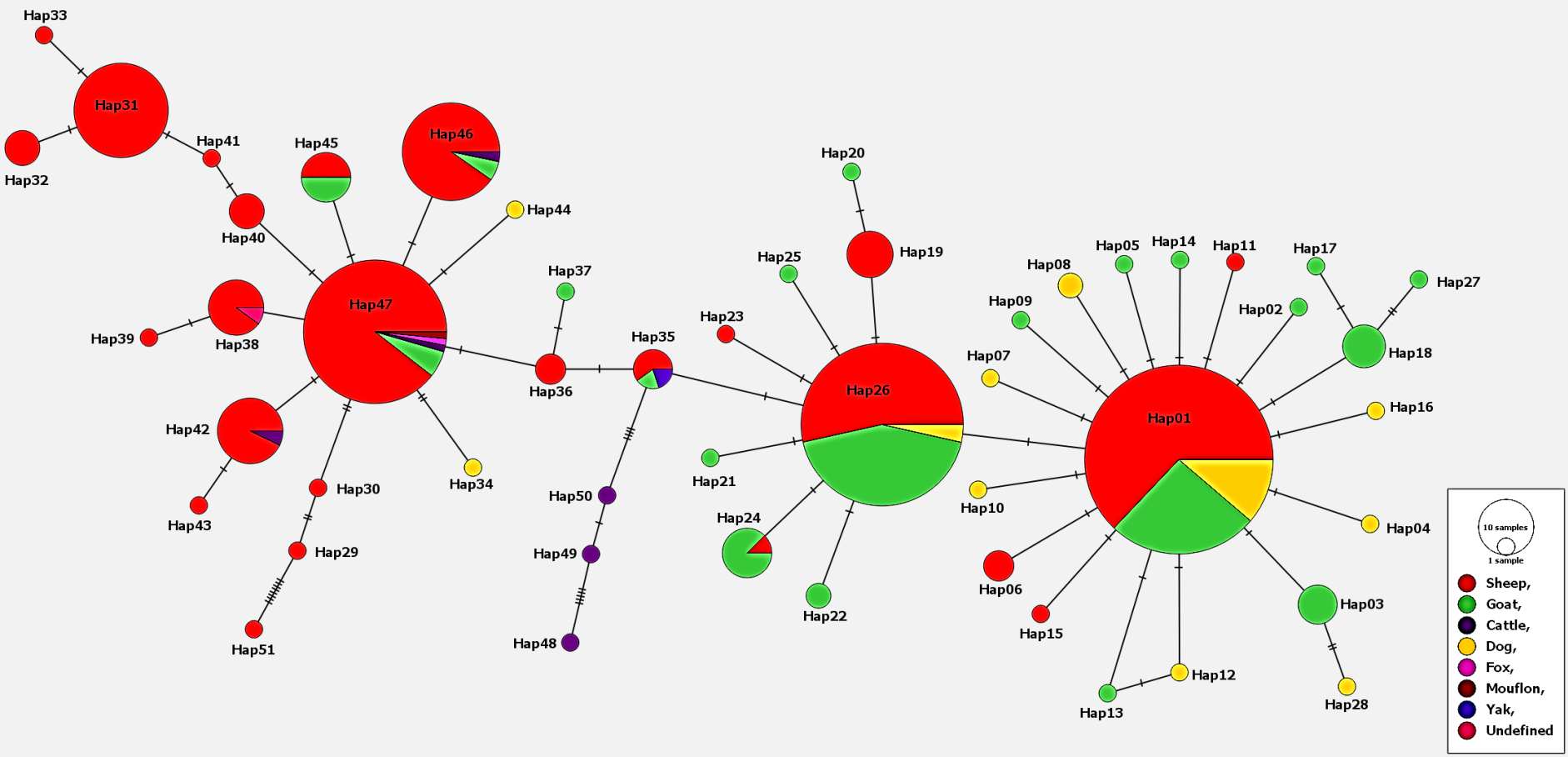


**Table 9. Genetic indices between different populations of *Taenia multiceps* calculated from cox1 partial nucleotide sequences**

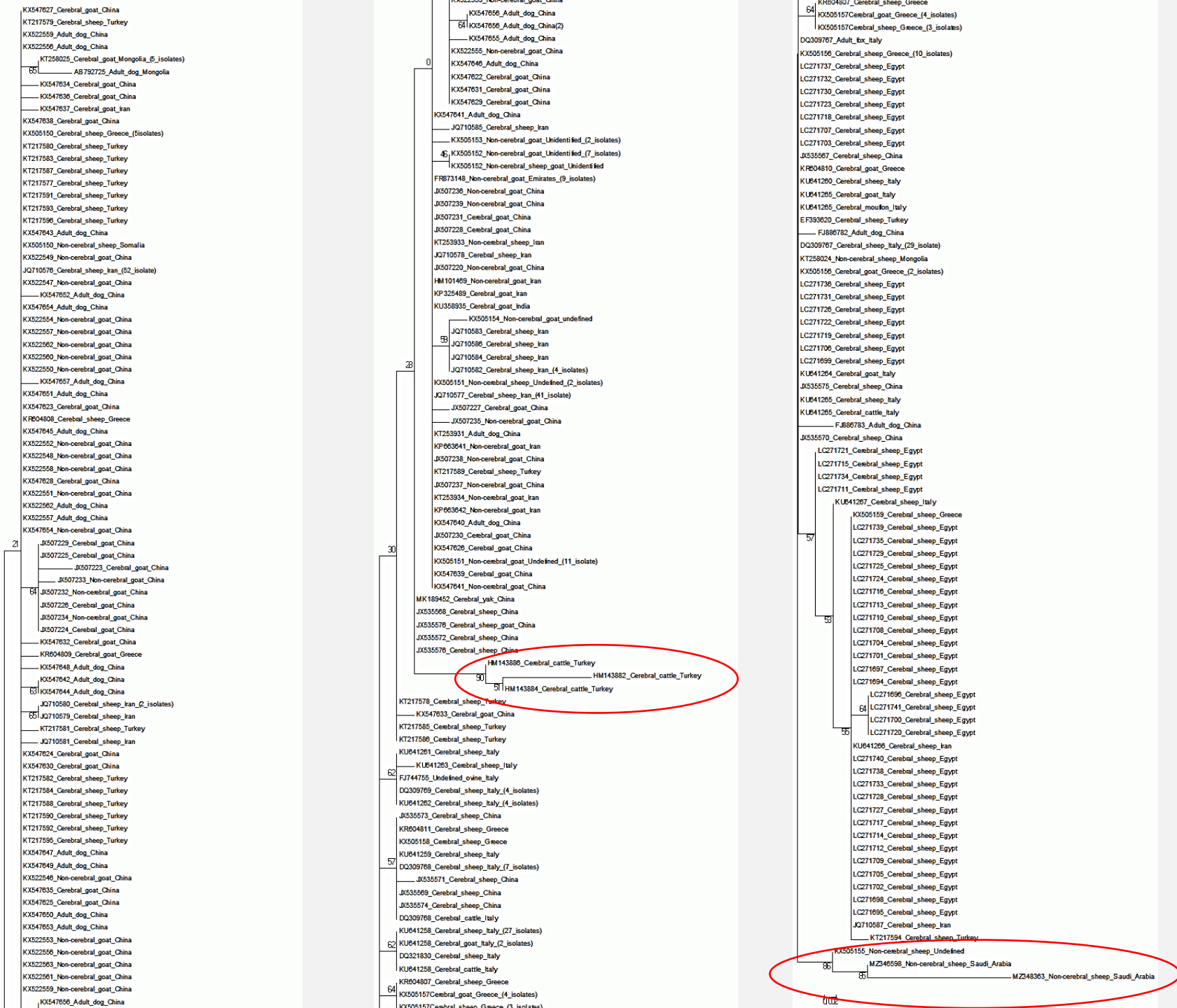
<b>Population 1</b>	<b>Population 2</b>	<b>Fst</b>	<b>Gst</b>	<b>Nm</b>	<b>Kxy</b>	<b>Dxy</b>	<b>Da</b>
<b>China</b>	Greece	0.43113	0.06325	3.73	3.55989	0.01059	0.00457
<b>China</b>	Iran	0.05237	0.02875	8.52	1.45347	0.00433	0.00023
<b>China</b>	Turkey	0.02359	0.01072	22.32	2.51609	0.00749	0.00018
<b>China</b>	Egypt	0.70753	0.14898	1.44	5.74753	0.01711	0.0121
<b>China</b>	Italy	0.68005	0.14225	1.53	4.3183	0.01285	0.00874
<b>Greece</b>	Iran	0.53163	0.10996	2.02	3.36466	0.01001	0.00532
<b>Greece</b>	Turkey	0.3285	0.10709	2.08	3.9543	0.01177	0.00387
<b>Greece</b>	Egypt	0.42524	0.11519	1.92	3.27016	0.00973	0.00414
<b>Greece</b>	Italy	0.19121	0.05733	4.11	1.95381	0.00581	0.00111
<b>Iran</b>	Turkey	0.06688	0.03935	6.10	2.15145	0.0064	0.00043
<b>Iran</b>	Egypt	0.77725	0.19887	1.01	5.53007	0.01646	0.01279
<b>Iran</b>	Italy	0.77635	0.2051	0.97	4.16937	0.01241	0.00963
<b>Turkey</b>	Egypt	0.60684	0.19729	1.02	5.87847	0.0175	0.01062
<b>Turkey</b>	Italy	0.56375	0.13422	1.61	4.61174	0.01373	0.00774
<b>Egypt</b>	Italy	0.5291	0.13346	1.62	2.625	0.00781	0.00413

Fst, Wright's F-statistics for pairwise genetic distance; Gst, genetic differentiation index based on the frequency of haplotypes; Nm, gene flow value; Kxy, average proportion of nucleotide differences between populations; Dxy, average number of nucleotide substitutions per site between populations; Da number of net nucleotide substitutions per site between populations

- No specific pattern for distribution among various hosts.
- Dogs, sheep, goat intermingled.
- Wildlife (e.g., fox, mouflon, yak) share the same haplotypes with domestic animals.
- Cattle from Italy share the same haplotypes with sheep.
- Genetic variant in cattle from Turkey and sheep from Saudi Arabia (confirmed in ML tree).

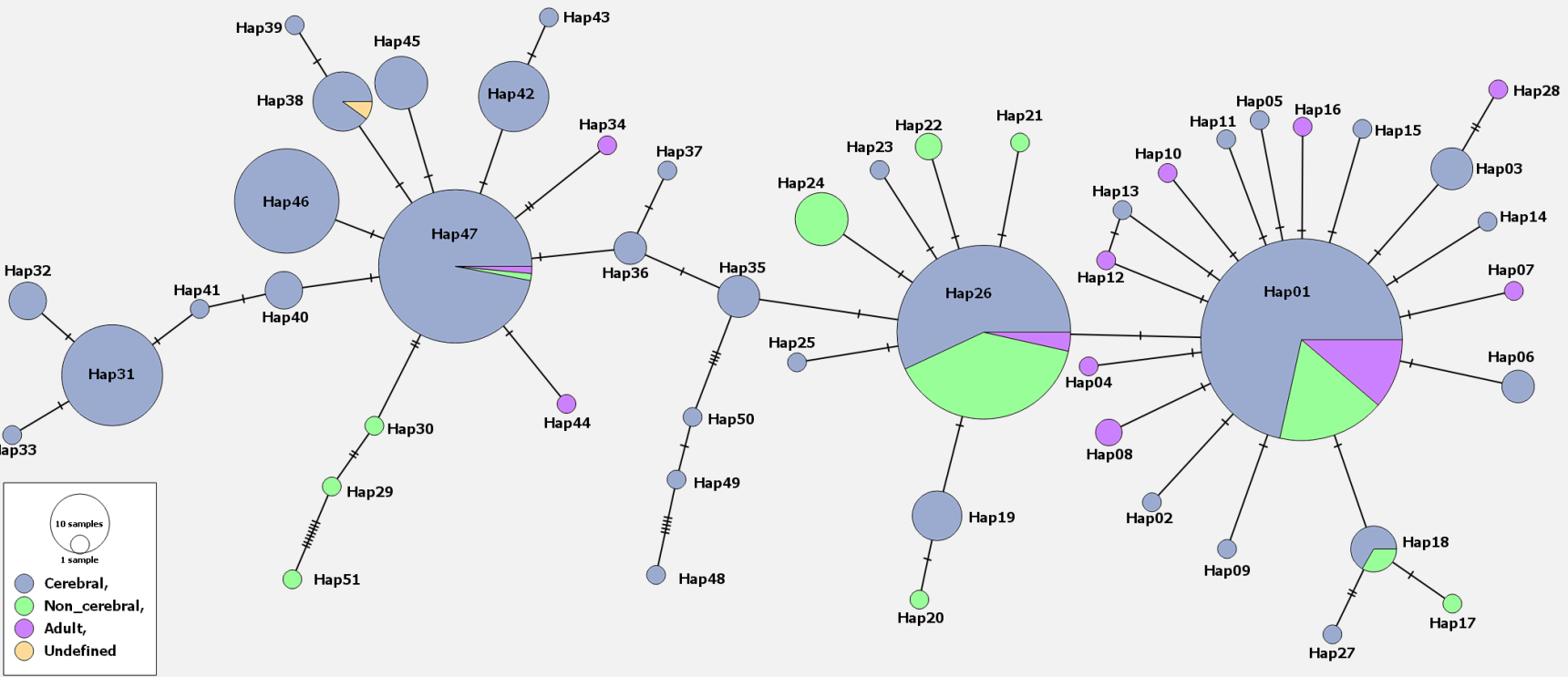








- Genetic analogy between cerebral and non-cerebral isolates.
- Haplotypes from Egypt, Italy are mostly cerebral due to lack of sequenced goat isolates in the 2 countries.
- Low pairwise distance and high gene flow values for cerebral and non-cerebral populations.
- Dogs, sheep, goat intermingled.



## Conclusion

- High genetic differentiation among *Taenia multiceps* sequenced isolates
- Geographical based haplotype distribution is partly suggested.
- No specific patterns for haplotype distribution acc. to host species or coenuri locations.
- *Taenia multiceps* genetic variants are suggested but can not be confirmed due to limited number of sequenced isolates from these variants.

Many thanks