ORIGINAL PAPER

Genetic variability of *Taenia saginata* inferred from mitochondrial DNA sequences

Sima Rostami • Reza Salavati • Robin N. Beech • Zahra Babaei • Mitra Sharbatkhori • Majid Fasihi Harandi

Received: 1 December 2014 / Accepted: 7 January 2015 / Published online: 17 February 2015 © Springer-Verlag Berlin Heidelberg 2015

Abstract Taenia saginata is an important tapeworm, infecting humans in many parts of the world. The present study was undertaken to identify inter- and intraspecific variation of T. saginata isolated from cattle in different parts of Iran using two mitochondrial CO1 and 12S rRNA genes. Up to 105 bovine specimens of T. saginata were collected from 20 slaughterhouses in three provinces of Iran. DNA were extracted from the metacestode Cysticercus bovis. After PCR amplification, sequencing of CO1 and 12S rRNA genes were carried out and two phylogenetic analyses of the sequence data were generated by Bayesian inference on CO1 and 12S rRNA sequences. Sequence analyses of CO1 and 12S rRNA genes showed 11 and 29 representative profiles respectively. The level of pairwise nucleotide variation between individual haplotypes of CO1 gene was 0.3-2.4 % while the overall nucleotide variation among all 11 haplotypes was 4.6 %. For 12S rRNA sequence data, level of pairwise nucleotide variation was 0.2-2.5 % and the overall

S. Rostami · Z. Babaei Department of Medical Parasitology, School of Medicine, Kerman University of Medical Sciences, Kerman 76169-14111, Iran

R. Salavati · R. N. Beech Institute of Parasitology, McGill University, Ste. Anne de Bellevue, QC, Canada

M. Sharbatkhori

Department of Medical Parasitology and Mycology, School of Medicine, Golestan University of Medical Sciences, Gorgan, Iran

S. Rostami · M. F. Harandi (🖂)

Research Center for Hydatid Disease in Iran, Kerman University of Medical Sciences, Kerman 76169-14111, Iran e-mail: fasihi@kmu.ac.ir nucleotide variation was determined as 5.8 % among 29 haplotypes of 12S rRNA gene. Considerable genetic diversity was found in both mitochondrial genes particularly in 12S rRNA gene.

Keywords Taenia saginata · Mitochondrial cox1 · 12S rRNA · Iran

Introduction

The beef tapeworm, Taenia saginata is an important taeniid species infecting humans in many parts of the globe. Taeniasis/cysticercosis are recognized by the World Health Organization (WHO) as neglected tropical diseases. Taeniasis caused by T. saginata is a cyclozoonosis in which the tapeworm uses human and cattle as definitive and intermediate hosts, respectively. The prevalence of T. saginata/ Taenia solium infection in man ranged from 0.3 to 64.2 %. Forty five to 77 million people have been estimated to be infected with T. saginata in the world (Abunna et al. 2008; Megersa et al. 2010; Roberts et al. 1994). In Iran, recent information indicates 0.5 % prevalence of T. saginata in northern province of Mazandaran (Kia et al. 2005). Prevalence of taeniasis depends on the level of sanitation and food habits like consuming raw or undercooked infected beef. Low prevalence rates have been observed in developed countries and higher rates of infection occur in less-developed countries especially in African states (Cabaret et al. 2002). Furthermore, bovine cysticercosis causes considerable economic losses mainly due to the condemnation of infected organs and/or carcasses. Results of several studies in different parts of the

	10	20	30	40	50	60	70	80																																																															
				.		.		I																																																															
T.saginata.NC_009938	TTTGTTTTTGTGTAGA	TTG-TGTGTG:	TTATTTATC	TTTAAAAATAG	GTTAAATTI	TTTTTATTTAA	GTTTTAAGTA	TTCA																																																															
IRTSSR2																																																																							
IRTSSR3	• • • • • • • • • • • • • • • • • •			· · · · · · · · · · · ·	•••••																																																																		
IRTSSR4	•••••	•••-•••	• • • • • • • • • • •	•••••	•••••	•••••	•••••	••••																																																															
IRTSSR5 IRTSSR6	A																																																																						
IRTSSR7																																																																							
IRTSSR8	•••••	••••	.A	•••••	•••••		• • • • • • • • • •	• • • • •																																																															
IRTSSR9	•••••	•••	•••••	Δ	•••••	•••••	•••••	••••																																																															
IRTSSR11				· · · · · · · · · · · · ·																																																																			
IRTSSR12					•••••																																																																		
IRTSSR13	C	••••	• • • • • • • • • • •	•••••	•••••	•••••	•••••	• • • • •																																																															
IRTSSR15	CA																																																																						
IRTSSR16	•••••	–		c																																																																			
IRTSSR17	••••••	••••		e	•••••	•••••	•••••	• • • • •																																																															
IRTSSR10																																																																							
IRTSSR20																																																																							
IRTSSR21	•••••	•••-•••	• • • • • • • • • •	•••••	•••••	•••••	• • • • • • • • • • •	••••																																																															
IRTSSR22 IRTSSR23	A A		•••••	•••••	•••••	•••••	•••••	• • • • •																																																															
IRTSSR24				•••••																																																																			
IRTSSR25	A	••••		•••••	•••••																																																																		
IKTSSK20 IRTSSR27				• • • • • • • • • • • • • •	•••••			• • • • •																																																															
IRTSSR28																																																																							
IRTSSR29					•••••			• • • • •																																																															
T.hydatigena.GQ228819	C T .A	CGA.T.A	.A	AC T	•••••	TA TC	G.AGA.	т.																																																															
T.pisiformis.GU569096	CT.A		CA	AA		AA.T	TA.A.C.T	.A.T.																																																															
T.crassiceps.NC002547	T .A	T .A	C.A	AA.CTGT	•••••	G	TAAT	.A. T .																																																															
T.solium.NC004022	CA.C	· · · · · · · · · · · · · · · · · · ·	CZ	¥A		•••••	.AA.	A																																																															
T.multiceps.GQ228818 T.asiatica.AF445798				•••••																																																																			
T.asiatica.NC_004826	CACA						A.																																																																
E.granulosus.NC008075	GG	ACA		G.GCTGT	•••••	G.GAC	G.G <mark>T</mark> G	. AAT .																																																															
	90	100	110	120	130	140	150	160																																																															
	.			.		•••••		••••																																																															
T.saginata.NC_009938 IRTSSR1	TTTTTATATAAAATTT	ATATTTGTGA	CAGGGATTAC	JATACCCCATT	AACGTAT-T	TTGTAAT-AT	TATCTTAGTT	-A-GT																																																															
INIODINI																																																																							
IRTSSR2			· · · · · · · · · · · ·			· · · · · · · · · ⁻ · · ·		C																																																															
IRTSSR2 IRTSSR3					· · · · · · · · · - ·	· · · · · · · · - · · ·		C																																																															
IRTSSR2 IRTSSR3 IRTSSR4 IRTSSR5				A	· · · · · · · · · · · · · · · · · · ·	· · · · · · · · · · · · · · · · · · ·	A.	ľ ľ ľC.																																																															
IRTSSR2 IRTSSR3 IRTSSR4 IRTSSR5 IRTSSR6				.	· · · · · · · · · · · · · · · · · · ·	· · · · · · · · · · · · · · · · · · ·		P P PC. P P																																																															
IRTSSR2 IRTSSR3 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7				A	· · · · · · · · · · · · · · · · · · ·			P P C. P C. P P P C.																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR8				۸ ۸	· · · · · · · · · · · · · · · · · · ·	· · · · · · · · · · · · · · · · · · ·	A 	 																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR8 IRTSSR9 IRTSSR10				A			A 	I - . I - . I - . I - . I - . I - . I - . I - . I - . I - . I - . I - . I - .																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR8 IRTSSR9 IRTSSR10 IRTSSR11				A			. A . A	C C.																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR8 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR12							. A . A	 																																																															
IRTSSR2 IRTSSR3 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR8 IRTSSR9 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14			λ. 					C C C. C C C. C																																																															
IRTSSR2 IRTSSR3 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR8 IRTSSR9 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR15			A	A				C																																																															
IRTSSR2 IRTSSR3 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR6 IRTSSR8 IRTSSR8 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR15 IRTSSR15 IRTSSR16			A				. A.	C																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR8 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR18			A				. A.	C - C -																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR8 IRTSSR9 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR16 IRTSSR16 IRTSSR17 IRTSSR19				Δ				2 - . 3 - .																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR9 IRTSSR10 IRTSSR10 IRTSSR12 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR16 IRTSSR16 IRTSSR18 IRTSSR19 IRTSSR19 IRTSSR20				Δ				C - C - <td< th=""></td<>																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR8 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR14 IRTSSR16 IRTSSR16 IRTSSR19 IRTSSR19 IRTSSR21 IRTSSR21				λ			A 	2 - . 2 - .																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR12 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR17 IRTSSR18 IRTSSR19 IRTSSR20 IRTSSR21 IRTSSR23				Δ			A A	2 - . 2 - .																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR17 IRTSSR18 IRTSSR20 IRTSSR20 IRTSSR21 IRTSSR23 IRTSSR23 IRTSSR23 IRTSSR23				λ			A A	C																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR9 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR17 IRTSSR18 IRTSSR19 IRTSSR20 IRTSSR21 IRTSSR22 IRTSSR23 IRTSSR23 IRTSSR24 IRTSSR24 IRTSSR26			Z	A			A A																																																																
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR9 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR15 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR18 IRTSSR20 IRTSSR21 IRTSSR21 IRTSSR23 IRTSSR24 IRTSSR24 IRTSSR26 IRTSSR27				A			A 	2 - 2 -																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR14 IRTSSR16 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR21 IRTSSR21 IRTSSR22 IRTSSR23 IRTSSR23 IRTSSR24 IRTSSR26 IRTSSR27 IRTSSR27 IRTSSR27				X																																																																			
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR8 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR14 IRTSSR14 IRTSSR16 IRTSSR16 IRTSSR20 IRTSSR20 IRTSSR21 IRTSSR21 IRTSSR22 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR24 IRTSSR25 IRTSSR26 IRTSSR26 IRTSSR27 IRTSSR28 IRTSSR38 I IRTSSR38 I IRTSSR38 I I I I I I I I I I I I I I I I I I I				Δ			A																																																																
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR8 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR14 IRTSSR16 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR20 IRTSSR20 IRTSSR21 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR25 IRTSSR26 IRTSSR26 IRTSSR27 IRTSSR27 IRTSSR28 IRTSSR38 I IRTSSR38 I IRTSSR38 I I I I I I I I I I I I I I I I I I I	A A A			Δ			.G	C - C - <td< th=""></td<>																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR14 IRTSSR16 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR21 IRTSSR22 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR29 T. hydatigena.GQ228819 T. saginata.AY684274 T. pisiformis.GD569096	A. A			λ				2 - 2 - <tr td=""></tr>																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR14 IRTSSR16 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR21 IRTSSR21 IRTSSR22 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR24 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR29 T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium NC004022	A A			Δ				C																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR14 IRTSSR16 IRTSSR16 IRTSSR26 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR29 IRTSSR29 IRTSSR29 IRTSSR29 IRTSSR29 IRTSSR29 IRTSSR29 IRTSSR29 IRTSSR29 IRTSSR29 IRTSSR29 IRTSSR29 IRTSSR29 IRTSSR29 IRTSSR29 IRTSSR29 IRTSSR29 I.saginata.AY684274 I.s	A			Δ.				C																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR14 IRTSSR15 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR20 IRTSSR20 IRTSSR21 IRTSSR21 IRTSSR23 IRTSSR24 IRTSSR24 IRTSSR25 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR29 T. hydatigena.GQ228818 T. crassiceps.NC002547 T. solium.NC004022 T. multiceps.GQ228818 T. asiatica.AF445798	A			А. 			.G	C - A TA C - C - C - C - C - C - C - C - <tr td=""> </tr> <tr><th>IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR4 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR1 IRTSSR1 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR17 IRTSSR16 IRTSSR21 IRTSSR21 IRTSSR21 IRTSSR21 IRTSSR23 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR26 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR29 T.hydatigena.GQ228818 T.casiatica.AF445798 T.asiatica.NC.004826 E granucess W0002617</th><th>A. A. A</th><th></th><th></th><th>А.</th><th></th><th></th><th>.G</th><th>C C</th></tr> <tr><th>IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR20 IRTSSR20 IRTSSR21 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR24 IRTSSR26 IRTSSR27 IRTSSR27 IRTSSR28 IRTSSR29 T.bydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU560966 T.crassiceps.Nc002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.NC004025 E.granulosus.NC008075</th><th>A. A. A</th><th></th><th></th><th>A.</th><th></th><th></th><th>.G.TC.</th><th>C - <td< th=""></td<></th></tr> <tr><th>IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR20 IRTSSR21 IRTSSR22 IRTSSR22 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR26 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR29 T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.Nc002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.NC_004826 E.granulosus.NC008075</th><th>A. A. A</th><th></th><th>190</th><th>A</th><th></th><th></th><th>.G. TC. 230</th><th>C C C C C</th></tr> <tr><th>IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR20 IRTSSR21 IRTSSR21 IRTSSR22 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IR</th><th>A. A. A</th><th></th><th>190</th><th>A</th><th>T. C-A T</th><th></th><th>.G.TC. 230</th><th>C C</th></tr> <tr><th>IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR4 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR14 IRTSSR16 IRTSSR16 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR29 I.hydatigena.GQ228819 I.saginata.AY684274 T.pisiformis.GU569096 I.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 I.asiatica.NC45798 I.asiatica.NC009938 IRTSSR2 I.saginata.NC009938 IRTSSR1</th><th>A. A. A</th><th></th><th>190 </th><th>A</th><th></th><th></th><th>.G.TC. 230</th><th>C C C C C - C C C [240]</th></tr> <tr><th>IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR14 IRTSSR16 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR20 IRTSSR21 IRTSSR22 IRTSSR23 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 I. hydatigena.GQ228819 T. sginata.AY684274 T. pisiformis.GQ569096 T. crassicgens.CQ228818 T. asiatica.AF445798 T. asiatica.AF445798 T. asiatica.AF445798 T. asiatica.NC008075 I. saginata.NC009938 IRTSSR1 IRTSSR1 IRTSSR1 IRTSSR1</th><th>A. A. A</th><th></th><th>190 </th><th>A.</th><th></th><th></th><th>.G.TC. 230</th><th>C C</th></tr> <tr><th>IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR21 IRTSSR22 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR26 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 I.sajiata.AY484274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.sojiata.AF445798 T.asiatica.NC004928 I.asiatica.NC004928 I.asiatica.NC004938 IRTSSR1 IRTSSR1 IRTSSR1 IRTSSR1 IRTSSR1 IRTSSR3 IRTSSR1 IRTSSR2 IRTSSR3 IRTSSR3 IRTSSR3 IRTSSR3 IRTSSR4</th><th>A. A. A</th><th></th><th>190</th><th>A</th><th></th><th></th><th>.G.TC</th><th>C C C C C C C C C C C</th></tr>	IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR4 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR1 IRTSSR1 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR17 IRTSSR16 IRTSSR21 IRTSSR21 IRTSSR21 IRTSSR21 IRTSSR23 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR26 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR29 T.hydatigena.GQ228818 T.casiatica.AF445798 T.asiatica.NC.004826 E granucess W0002617	A. A			А.			.G	C C	IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR20 IRTSSR20 IRTSSR21 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR24 IRTSSR26 IRTSSR27 IRTSSR27 IRTSSR28 IRTSSR29 T.bydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU560966 T.crassiceps.Nc002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.NC004025 E.granulosus.NC008075	A. A			A.			.G.TC.	C - C - <td< th=""></td<>	IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR20 IRTSSR21 IRTSSR22 IRTSSR22 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR26 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR29 T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.Nc002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.NC_004826 E.granulosus.NC008075	A. A		190	A			.G. TC. 230	C C C	IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR20 IRTSSR21 IRTSSR21 IRTSSR22 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IR	A. A		190	A	T. C-A T		.G.TC. 230	C C	IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR4 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR14 IRTSSR16 IRTSSR16 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR29 I.hydatigena.GQ228819 I.saginata.AY684274 T.pisiformis.GU569096 I.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 I.asiatica.NC45798 I.asiatica.NC009938 IRTSSR2 I.saginata.NC009938 IRTSSR1	A. A		190 	A			.G.TC. 230	C C C C C - C C C [240]	IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR14 IRTSSR16 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR20 IRTSSR21 IRTSSR22 IRTSSR23 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 I. hydatigena.GQ228819 T. sginata.AY684274 T. pisiformis.GQ569096 T. crassicgens.CQ228818 T. asiatica.AF445798 T. asiatica.AF445798 T. asiatica.AF445798 T. asiatica.NC008075 I. saginata.NC009938 IRTSSR1 IRTSSR1 IRTSSR1 IRTSSR1	A. A		190 	A.			.G.TC. 230	C C	IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR21 IRTSSR22 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR26 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 I.sajiata.AY484274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.sojiata.AF445798 T.asiatica.NC004928 I.asiatica.NC004928 I.asiatica.NC004938 IRTSSR1 IRTSSR1 IRTSSR1 IRTSSR1 IRTSSR1 IRTSSR3 IRTSSR1 IRTSSR2 IRTSSR3 IRTSSR3 IRTSSR3 IRTSSR3 IRTSSR4	A. A		190	A			.G.TC	C C C C C C C C C C C
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR4 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR1 IRTSSR1 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR17 IRTSSR16 IRTSSR21 IRTSSR21 IRTSSR21 IRTSSR21 IRTSSR23 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR26 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR29 T.hydatigena.GQ228818 T.casiatica.AF445798 T.asiatica.NC.004826 E granucess W0002617	A. A			А.			.G	C C																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR20 IRTSSR20 IRTSSR21 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR24 IRTSSR26 IRTSSR27 IRTSSR27 IRTSSR28 IRTSSR29 T.bydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU560966 T.crassiceps.Nc002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.NC004025 E.granulosus.NC008075	A. A			A.			.G.TC.	C - C - <td< th=""></td<>																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR20 IRTSSR21 IRTSSR22 IRTSSR22 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR26 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR29 T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.Nc002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.NC_004826 E.granulosus.NC008075	A. A		190	A			.G. TC. 230	C C C																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR14 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR20 IRTSSR21 IRTSSR21 IRTSSR22 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IR	A. A		190	A	T. C-A T		.G.TC. 230	C C																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR4 IRTSSR5 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR14 IRTSSR16 IRTSSR16 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR29 I.hydatigena.GQ228819 I.saginata.AY684274 T.pisiformis.GU569096 I.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 I.asiatica.NC45798 I.asiatica.NC009938 IRTSSR2 I.saginata.NC009938 IRTSSR1	A. A		190 	A			.G.TC. 230	C C C C C - C C C [240]																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR14 IRTSSR16 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR20 IRTSSR21 IRTSSR22 IRTSSR23 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 I. hydatigena.GQ228819 T. sginata.AY684274 T. pisiformis.GQ569096 T. crassicgens.CQ228818 T. asiatica.AF445798 T. asiatica.AF445798 T. asiatica.AF445798 T. asiatica.NC008075 I. saginata.NC009938 IRTSSR1 IRTSSR1 IRTSSR1 IRTSSR1	A. A		190 	A.			.G.TC. 230	C C																																																															
IRTSSR2 IRTSSR4 IRTSSR4 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR7 IRTSSR7 IRTSSR10 IRTSSR10 IRTSSR11 IRTSSR12 IRTSSR13 IRTSSR13 IRTSSR14 IRTSSR15 IRTSSR16 IRTSSR16 IRTSSR21 IRTSSR21 IRTSSR22 IRTSSR22 IRTSSR23 IRTSSR24 IRTSSR25 IRTSSR25 IRTSSR26 IRTSSR27 IRTSSR26 IRTSSR27 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR28 IRTSSR27 I.sajiata.AY484274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.sojiata.AF445798 T.asiatica.NC004928 I.asiatica.NC004928 I.asiatica.NC004938 IRTSSR1 IRTSSR1 IRTSSR1 IRTSSR1 IRTSSR1 IRTSSR3 IRTSSR1 IRTSSR2 IRTSSR3 IRTSSR3 IRTSSR3 IRTSSR3 IRTSSR4	A. A		190	A			.G.TC	C C C C C C C C C C C																																																															

Fig. 1 Alignments of the 11 representative profiles of CO1 sequences and 29 representative profiles of 12S rRNA sequences, among *Taenia saginata* isolates from Iran with key reference sequences (for other

taeniid species) from previous studies. *Echinococcus granulosus* sensu stricto was used as outgroup

TRUSSRA					Δ			
TDMCCD7					· · · · · · · · · · · · · · · · · · ·			
INISSN/	• • • • • • • • • • • • • • • •	•••••	•••••	•••••	•••••	••••••		
INISSKO	• • • • • • • • • • • • • • •	•••••	••••••	•••••	• • • • • • • • • • • • •	••••••	•••••	• • • • •
IRTSSR9	• • • • • • • • • • • • • • • • • • • •	•••••	•••••	•••••	• • • • • • • • • • • • •	•••••	•••••	• • • • •
IRTSSR10		•••••						
IRTSSR11								
IRTSSR12								
IRTSSR13								
TRTSSR14				C				
INISSNI IDMOOD1E		••••••	•••••			••••••		
IRTSSRI5	• • • • • • • • • • • • • • • • • • • •	••••••	•••••	•••••	A	•••••	•••••	• • • • •
IRTSSR16	• • • • • • • • • • • • • •	•••••	•••••	•••••	• • • • • • • • • • •	• • • • • • • • • • •	•••••	• • • •
IRTSSR17								
IRTSSR18								
IRTSSR19								
TRTSSR20								
TDWCCD21		••••••	••••••			•••••		
IRISSR21	• • • • • • • • • • • • • • •		••••••	•••••		•••••	•••••	• • • • •
IRTSSR22	• • • • • • • • • • • • • • • •	· · · · · · · · · · · · A	•••••	•••••	· · · · · · · A · · · ·	••••••	•••••	• • • • •
IRTSSR23	• • • • • • • • • • • • • • • •	• • • • • • • • • • • •	• • • • • • • • • •	• • • • • • • • • •	A	• • • • • • • • • • •	• • • • • • • • • • • •	• • • •
IRTSSR24					.			
IRTSSR25								
IRTSSR26								
TRTSSR27		Δ			Δ			
TDWCCD20			••••••			•••••		
INISSN28	• • • • • • • • • • • • • • • •	••••••	•••••	•••••	• • • • • • • • • • • • •	••••••		
IRTSSR29	• • • • • • • • • • • • • • • •	••••••••	• • • • • • • • • •	•••••	•••••	•••••	•••••	• • • • •
T.hydatigena.GQ228819	AA	C.	.C				A	
T.saginata.AY684274								
T.pisiformis.GU569096					A.T			
T.crassiceps.NC002547							T	
T. solium NC004022					. A.			
T multiconc (0020010	•••••							• • • • •
1.multiceps.GQ228818	•••••	••••••	•••••	•••••	•••••	••••••	•••••	• • • • •
T.asiatica.AF445798	•••••••••••	••••••	• • • • • • • • • •	• • • • • • • • • •	• • • • • • • • • • • •	• • • • • • • • • • •	• • • • • • • • • • •	• • • • •
T.asiatica.NC_004826								
E.granulosus.NC008075		c.	A	T A	.CATA.G.	GA.	G	c.
	250	260	270	200	200	200	21.0	220
	230	260	270	200	290		310	520
		1	••••			• • • • • • • •		• • • •
T.saginata.NC009938	TATTATGTTGGTGT	ATATCTGGTTT/	AATATTATCO	GTTGAGTGAT	GTATAAGTTT	GTGTAGGTTT-	TAGTTAAGCTA	AGTC
IRTSSR1							••••••	
IRTSSR2							•••••••	
IRTSSR3							т	r
TRTSSR4						с -		
TPTSSP5								
TDWCCD6		••••••	••••••	•••••				
IRISSRO	•••••	• • • • • • • • • • • • •	• • • • • • • • • •	•••••	• • • • • • • • • • • • •	• • • • • • • • • • • • •		
IRTSSR7	• • • • • • • • • • • • • • • •	• • • • • • • • • • • •	• • • • • • • • • •	• • • • • • • • • •	• • • • • • • • • • •	• • • • • • • • • • •	· · · · · · · · · · · · · · · · · · ·	
IRTSSR8								
IRTSSR9							·	r
IRTSSR10 IRTSSR11			•••••	•••••	• • • • • • • • • • • • •		т.	•••
IRTSSRIZ		••••••	•••••	•••••	• • • • • • • • • • • •	••••	•••••	•••
IRTSSR13	.C	•••••	• • • • • • • • •	• • • • • • • • • •	• • • • • • • • • • • •	c	•••••	• • •
IRTSSR14	.C							
IRTSSR15							T .	
IRTSSR16								
TRTSSR17	Ψ							
TDWCCD10					• • • • • • • • • • • • •			•••
INISSNIC	~				• • • • • • • • • • • • •	••••••		• • •
INISSNIP			• • • • • • • • • •	• • • • • • • • • •	• • • • • • • • • • • • •	• • • • • • • • • • • • • • • •	· · · · · · · · · · · · · · · · · · ·	• • •
IRTSSR20		• • • • • • • • • • • •	• • • • • • • • •	• • • • • • • • • •	• • • • • • • • • • • •	•••••	r .	• • •
IRTSSR21	· · · · T · · · · · · · · · ·	• • • • • • • • • • • •	• • • • • • • • •	• • • • • • • • •	• • • • • • • • • • • •	• • • • • • • • • • •	• • • • • • • • • • • •	•••
IRTSSR22								• • •
IRTSSR23								
IRTSSR24						– .		
IRTSSR25						– .	.A	
IRTSSR26							.A	• • •
IRTSSR27								
IRTSSR28							.A	
IRTSSR29								
	CG			Δ		Ψ	т с	
T hydatigers CO228910		•••••		· · · · · · · · · · · · · · · ·	5.5			•••
T.hydatigena.GQ228819					• • • • • • • • • • • •			•••
T.hydatigena.GQ228819 T.saginata.AY684274			-	m z z			C	
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096		· · · · · · · · · · · · · · · · · · ·	T .	T.A.A				• • •
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547			т. т.	T.A.A C.T.AG.		TA	.TTC	
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022		AG	T.	T.A.A C.T.AG. A.AG.	TA	TA	.TTC	••••
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818		A			TA	TA	.TTC 	•••
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798		.A			TA	TA	.TTC C 	••••
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.NC004826		.AG			TA	TA	.TC.	••••
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.NC_004826 E.granulosus.NC008075		.A				TA	.T. TC .C .C	· · · · · · · · · · · · · · · · · · ·
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.NC_004826 E.granulosus.NC008075		.GG			-TA		.TTCCC	· · · · · · · · · · · · · · · · · · ·
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.NC_004826 E.granulosus.NC008075					TA		.T	
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.NC_004826 E.granulosus.NC008075	. T	. A			TA		.TTC .C 	400
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.NC_004826 E.granulosus.NC008075	.T			T.A.A. C.T.A.G. A.AG A.AG A.A. A.C A.A. 	TA		.TTCCC	400
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.NC_004826 E.granulosus.NC008075				T.A.A C.T.A.G. A.AG. A.AG. A.A. A.C. A.A. A.A. 			.TTC	400
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.NC04826 E.granulosus.NC008075 T.saginata.NC009938 IRTSSR1	.T			T.A.A C.T.AG. A.AG. A.A. A.A AC A.A 360 			.TTC	400
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.NC_004826 E.granulosus.NC008075 T.saginata.NC009938 IRTSSR1 IRTSSR1	.T	.A					.TTCCCC	400
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.AF445798 T.asiatica.NC_004826 E.granulosus.NC008075 T.saginata.NC009938 IRTSSR1 IRTSSR1 IRTSSR3	.T	.GGG					.T. TC. .C. C. .C 	400 AAA
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.NC004826 E.granulosus.NC008075 T.saginata.NC009938 IRTSSR1 IRTSSR1 IRTSSR3 IRTSSR4	.T				TA T		.TTC	400 AAA
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.AF445798 T.asiatica.NC_004826 E.granulosus.NC008075 T.saginata.NC009938 IRTSSR1 IRTSSR2 IRTSSR3 IRTSSR4 IRTSSR4 IRTSSR5	.T						.T	400 AAA
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.NC004826 E.granulosus.NC008075 T.saginata.NC009938 IRTSSR1 IRTSSR2 IRTSSR3 IRTSSR4 IRTSSR5 IBTSSR4 IRTSSR5 IBTSSR6	.T						.TTC. 	400 AAA
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.NC_004826 E.granulosus.NC008075 T.saginata.NC009938 IRTSSR1 IRTSSR2 IRTSSR3 IRTSSR5 IRTSSR5 IRTSSR5 IRTSSR5 IRTSSR5 IRTSSR5 IRTSSR5	.T						.T	400 AAA
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.NC004026 E.granulosus.NC008075 T.saginata.NC009938 IRTSSR1 IRTSSR1 IRTSSR3 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR6 IRTSSR7 IRTSSR7	.T	.GGGG					.TTCAGGACTTA	400 AAA
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.NC004826 E.granulosus.NC008075 T.saginata.NC009938 IRT5SR1 IRT5SR1 IRT5SR3 IRT5SR4 IRT5SR4 IRT5SR5 IRT5SR4 IRT5SR5 IRT5SR5 IRT5SR7 IRT5SR7	T. 330 TATGTGCTGCTTATA				TA TA T		.TC. 	400 AAA
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.AF445798 T.asiatica.NC_004826 E.granulosus.NC008075 T.saginata.NC009938 IRTSSR1 IRTSSR2 IRTSSR3 IRTSSR4 IRTSSR5 IRTSSR5 IRTSSR6 IRTSSR6 IRTSSR8 IRTSSR8 IRTSSR8	.T						.T.C. 390 	400 AAA
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.NC004826 E.granulosus.NC008075 T.saginata.NC008075 T.saginata.NC009938 IRTSSR1 IRTSSR1 IRTSSR4 IRTSSR5 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR7 IRTSSR8 IRTSSR7 IRTSSR8 IRTSSR9 IRTSSR10	.T						.TC	400 AAA
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.NC_004826 E.granulosus.NC008075 T.saginata.NC009938 IRTSSR1 IRTSSR2 IRTSSR3 IRTSSR4 IRTSSR5 IRTSSR6 IRTSSR6 IRTSSR7 IRTSSR8 IRTSSR8 IRTSSR8 IRTSSR9 IRTSSR10 IRTSSR11	.T						.TC. 	400
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.NC004826 E.granulosus.NC008075 T.saginata.NC009938 IRTSSR1 IRTSSR1 IRTSSR4 IRTSSR4 IRTSSR6 IRTSSR6 IRTSSR7 IRTSSR8 IRTSSR8 IRTSSR9 IRTSSR10 IRTSSR11 IRTSSR12	.T						.TTCAGGACTTA	400 AAA
T.hydatigena.GQ228819 T.saginata.AY684274 T.pisiformis.GU569096 T.crassiceps.NC002547 T.solium.NC004022 T.multiceps.GQ228818 T.asiatica.NC00422 E.granulosus.NC008075 T.saginata.NC009938 IRT5SR1 IRT5SR3 IRT5SR4 IRT5SR4 IRT5SR5 IRT5SR4 IRT5SR6 IRT5SR7 IRT5SR8 IRT5SR9 IRT5SR10 IRT5SR10 IRT5SR11 IRT5SR13	.T		T. T. T. T. T. 350				.TC. 	400 AAA

IRTSSR14	C
IRTSSR15	
TRTSSR10	Δ
IRTSSR18	
IRTSSR19	
IRTSSR20	
IRTSSR21	
IRTSSR22	······
IRTSSR24	A
IRTSSR25	
IRTSSR26	
IRTSSR27	
IRTSSR28	
IRTSSR29	
T.nydatigena.GQ228819	······································
T.pisiformis.GU569096	AT T TA A A -AT GTA T T
T.crassiceps.NC002547	
T.solium.NC004022	
T.multiceps.GQ228818	
T.asiatica.AF445798	
T.asiatica.NC_004826	
E. granuiosus. Neuvou / 5	
	410 420 430
T.saginata.NC009938	GTAACGTTAAATTAGTTTGTTAATGTGAAGTAAGTTT
IRTSSR1	·····T·······
LKISSKZ TRTSSRZ	π
IRTSSR4	T
IRTSSR5	TTT
IRTSSR6	· · · · T · · · · · · · · · · · · · · · · · · ·
IRTSSR7	·····T································
IRTSSR8	····· <u>T</u> ·······························
IRTSSR9	····· T
IRISSRIU IRTSSRI1	·····
IRTSSR12	
IRTSSR13	· · · · T · · · · · · · · · · · · · · · · · · ·
IRTSSR14	·····T·······T·······
IRTSSR15	·····T········T·······················
IRTSSRI6	· · · · · · · · · · · · · · · · · · ·
TRTSSRIB TRTSSRI9	т т
IRTSSR20	· · · · · · · · · · · · · · · · · · ·
IRTSSR21	· · · · T · · · · · · · T · · · · · · · · · · · · · · · · · · ·
IRTSSR22	···· T ································
IRTSSR23	·····
IRTSSR24	······································
IRTSSR26	· · · · · · · · · · · · · · · · · · ·
IRTSSR27	···· T ······
IRTSSR28	···· T ································
IRTSSR29	·····
T.saginata AV684274	······································
T.pisiformis.GU569096	TTA.AAGAA
T.crassiceps.NC002547	T
T.solium.NC004022	TA.C
T.multiceps.GQ228818	T
T.asiatica.NC 004826	G
E.granulosus.NC008075	T
2	
	±0 20 30 40 30 60 70 80
T.saginata.NC009938	TTTGGTATGATTAGTCATATATGTTTAAGAATAAGTATGTGTCCAGATGCTTTTGGTTTTTATGGTTTGTTGTTGTTGCTAT
IRTSCO1	
IRTSCO2	
IRTSCO3	
IRTSC05	······································
IRTSCO6	
IRTSCO7	
IRTSCO8	T
IRTSCO9	
IRTSCOLU IRTSCOLL	C
T.saginata.AB465242	·····
T.saginata.AB533168	
T.saginata.AB465247	
T.saginata.AB533169	
T.asiatica.AB533170	G
I.Saginata.AB533172 T asiatica AB533175	<i>C</i> . *
T.asiatica AB608739	
T asiatica NC004826	G A

T solium NC004022	Δ		ст	т. т.		c	G A	
T.multiceps.GO228818				c			A A	
T.hydatigena.GQ228819	AT		.G	AT			AAA	
T.pisiformis.GU569096	A			т	G		A	A
T.crassiceps.NC002547	A T A	T	.G.A	AA. TGT	T		AA	••••
E.granulosus.NC008075	A	T	.GTT.	. GCTAA . TTT	G	G <mark>C</mark>	G	• • • • •
	0.0	1.0.0	110	100	100	1.40	1 5 0	1.00
	90	100	110	120	130	140	120	100
T saginata NC009938	GTTTTCAATACTCTC		AGTGTGTGG	GTCATCATAT	CTTTACCCT		TTAAGACTGC	TGTGT
IRTSCO1								
IRTSCO2								
IRTSCO3								
IRTSCO4		•••••		•••••			• • • • • • • • • • •	
IRTSC05	•••••	••••	••••••	•••••	• • • • • • • • •	••••••	•••••	• • • • •
IRTSCO6	•••••	•••••	•••••	•••••	• • • • • • • • • •	••••••	•••••	
IRTSCO7	•••••	•••••	••••••	•••••	•••••	••••••		c
IRTSCO0		•••••		Δ	•••••	••••••	•••••	• • • • •
IRTSCO10							A	
IRTSCO11								
T.saginata.AB465242						G		
T.saginata.AB533168		••••				• • • • • • • • • • •		
T.saginata.AB465247	•••••	•••••		•••••	• • • • • • • • • •	• • • • • • • • • • • •	• • • • • • • • • • •	
T.saginata.AB533169	· · · · · · · · · · · · · · · · · · ·	•••••	••••••	•••••	•••••	••••	•••••••••••••••••••••••••••••••••••••••	• • • • •
T.asiatica.AB533170	A	•••••	••••••	•••••	•••••	A	A.	• • • • •
T aciatica AB533175	Δ				•••••	Δ		
T.asiatica.AB608739	A						A	
T.asiatica.NC004826				G			A.	
T.solium.NC004022.	A	AA	A	.A			G	A.
T.multiceps.GQ228818		A	A.	.c	A	• • • • • • • • • • • •		A.
T.hydatigena.GQ228819	C	T	· · · · · · · · · · ·	•••••	T	••••••	• • • • • • • • • • •	т.
T.pisiformis.GU569096	TT	AT	AA.	•••••	T	A	.AC	· · · <u>·</u> ·
T.crassiceps.NC002547	T	ATC	· · · · · · T · · · ·	•••••		T		T.
E.granulosus.NC008075	· · · · · · · · · · · · · · · · · · ·	· · · · · · · · · · · · ·	GT	•••••	· · · · · · · · · · ·	· · · · · · · · · · · · · · · ·		r .
	170	180	190	200	210	220	230	240
T.saginata.NC009938	TTTTTAGTTCGGTTA	CTATGATAAT	AGGAGTACC	ACAGGAATAA	AGGTTTTTA	CTTGACTTTAT	ATGCTTTTAA	ATTCT
IRTSCO1	•••••	•••••	•••••	•••••	• • • • • • • • • •	• • • • • • • • • • • •	• • • • • • • • • •	• • • • •
IRTSCO2	•••••	•••••	•••••	•••••	•••••	•••••	•••••	• • • • •
IRTSCO3	•••••	••••••	•••••	•••••	•••••	•••••	•••••	• • • • •
IRTSCO4				G				
IRTSCO6								
TPTSCO7								
IRTSCOR	••••••	•••••	•••••••••	•••••••••	•••••		•••••••	••••
IRTSCO9								
IRTSCO10	T			c				
IRTSCO11		•••••		.				
T.saginata.AB465242	•••••••••••••	•••••		· · · · · · · · · · ·		•••••	• • • • • • • • • • • •	• • • • •
T.saginata.AB533168	••••••••••••	•••••	• • • • • • • • • • •	•••••••••	•••••	•••••	•••••••	• • • • •
T.saginata.AB465247	••••••	•••••	•••••••	•••••••••	•••••	•••••	•••••	•••••
T asiatica AB533170								
T.saginata.AB533172				.				
T.asiatica.AB533175				.				
T.asiatica.AB608739				.				
T.asiatica.NC004826	•••••	•••••	•••••••	•••••••••	• • • • • • • • •	• • • • • • • • • • •	• • • • • • • • • • •	••••
T.solium.NC004022.	T	•••••	.TG	ГGGТ.	•••••	G	•••••	.A
T hydatigena GO228819	т. с.	тт	т с	ст г т т	c	GT A		
T.pisiformis.GU569096		.A	т	г. т т.	C	.A		
T.crassiceps.NC002547	T	т.		гт	G	T .G		G
E.granulosus.NC008075	CT	т.	G <mark>T</mark> '	F T T	G	GT.A	T .GG.	G
	250	260	270	280	290	300	310	320
						1		
T.saginata.NC009938	CGTGTAAATAAGAGT	GATCCTATA	TGTGGTGAA	FAGTTTCTTTT	ATAGTGTTG	TTTACTTTTGO	TGGTGTGACT	GGTAT
IRTSCO1	· · · · · · · · · · · · · · · · · · ·		• • • • • • • • • •					
IRTSCO2	•••••	••••••	•••••	• • • • • • • • • • • •	•••••	• • • • • • • • • • •	•••••	••••
IRTSCO3	•••••	•••••	•••••	••••••	•••••	•••••	•••••	• • • • •
IRTSCO4	•••••	•••••	•••••	• • • • • • • • • • • •	•••••	••••••	•••••	••••
IRISCOS	•••••	•••••	•••••	••••••	•••••	••••••	•••••	• • • • •
IRTSCO7								
IRTSCO8	G							
IRTSCO9	T							
IRTSCO10	•••••	•••••						
IRTSCO11	•••••	•••••	•••••		•••••	•••••	•••••	••••
T.saginata.AB465242	•••••	•••••	•••••	••••••	• • • • • • • • •	• • • • • • • • • • • •	•••••	• • • • •
T.saginata.AB533168	•••••	•••••	•••••	•••••	•••••	•••••	•••••	••••
I.SAYINATA.AB40524/ T saginata AB533160	•••••	•••••			•••••			••••
T.asiatica.AB533170			с. С			C		
T.saginata.AB533172	· · · · · · · · · · · · · · · · · · ·							
T.asiatica.AB533175			G.			C		
T.asiatica.AB608739	G	••••••	G.			c		
T.asiatica.NC004826	G		G.					
m asliem MC004000					•••••			
1.S011um.NC004022.	T	GG.T		AG	A	A	AC	· · · · ·

T.hydatigena.GQ228819	.AG	G. T	G.TA	.T	T		.GT
T.pisiformis.GU569096	CA	G.G			CA.	A.	A
T.crassiceps.NC002547	G <mark>C</mark>			.T	T A.	G	TA .
E.granulosus.NC008075	A T G TT .	GG.T	AGG	. T		GG.	.ATG
	330	340	350	360	370	380	390
	1 1	1 1	1 1	1 1	1 1 1	500	550
T caginata NC009939			· · · · · · · · · · · · · · · · ·	·····	╎・・・・│・・・・│ フ ヘ ᲚᲚ©ᲚᲚ©ᲚᲚ		••
TPTSCO1	1919119101001	IGCGIAIIGGA	IMAAGIIIIG	CAIGAIACII	SALLIGIIGII	Getektittekt	141
IRESCO2	••••••	•••••	•••••		• • • • • • • • • • • • •	•••••	•••
TRESCO2	•••••		•••••		• • • • • • • • • • • • •	•••••	•••
IRISCO3	• • • • • • • • • • • • • • • •	•••••••••	•••••	•••••	• • • • • • • • • • • • •	•••••	•••
TDECOF	•••••	•••••	• • • • • • • • • • •	•••••	• • • • • • • • • • • • •	•••••	••••
INTSCOS	••••••	•••••	• • • • • • • • • • •	•••••	•••••••••••	•••••	• • •
IRTSCO6	•••••	•••••	•••••		· · · · · · · · · · · · · · · · · · ·	•••••	• • •
IRTSCO7	••••••	•••••	••••••		•••••	•••••	• • •
IRTSCO8	••••••	••••••	• • • • • • • • • •	•••••	• • • • • • • • • • • •	•••••	• • •
IRTSCO9	••••••	•••••	• • • • • • • • • •	•••••	• • • • • • • • • • • •	•••••	• • •
IRTSCO10	••••••••••	••••••••••	• • • • • • • • • • •	•••••	• • • • • • • • • • • •	•••••	• • •
IRTSCO11	••••••	T	• • • • • • • • • •	•••••	• • • • • • • • • • •	• • • • • • • • • • • •	• • •
T.saginata.AB465242	• • • • • • • • • • • • • •	•••••		•••••		• • • • • • • • • • • • •	• • •
T.saginata.AB533168	• • • • • • • • • • • • • •	•••••		•••••		• • • • • • • • • • • • •	• • •
T.saginata.AB465247	• • • • • • • • • • • • •	•••••		•••••			• • •
T.saginata.AB533169							
T.asiatica.AB533170		T				c	
T.saginata.AB533172							
T.asiatica.AB533175		T		· · · · · · - · · · ·		c	
T.asiatica.AB608739		T				c	
T.asiatica.NC004826		T				c	
T.solium.NC004022.	.A.TC.A	T		•••••	. G		
T.multiceps.GQ228818	A	TA	A				
T.hydatigena.GQ228819	AA	TA	с .т				
T.pisiformis.GU569096	AAA	TTA	A		A	G	
T.crassiceps.NC002547	A. A		С.т				
E granulosus NC008075	Δ Ψ	TCA	π λ		G G G		

world indicate 0.007-26.3 % of cattle infected with *Cysticercus bovis* (Abunna et al. 2008; Dorny and Praet 2007). Annual monetary cost of human taeniasis is estimated at US\$ 263,000 in the USA mainly due to medical costs and wage losses. In England, annual losses due to bovine cysticercosis are estimated about £4 million (Roberts et al. 1994; Silva and Costa-Cruz 2010). Understanding the genetic identity of the parasite as well as the extent of genetic variation within *T. saginata* is essential for diagnosis and control of taeniasis.

Several studies have investigated genetic variation of human taeniid tapeworms, however, most of the research activity has been focused on T. solium and human cysticercosis. Relatively limited information is available on genetic variation within T. saginata from different parts of the world. The complete mitochondrial genome of T. saginata has been determined in 2007 (Jeon et al. 2007). Genetic characterization of T. saginata has been carried out in Ethiopia, Indonesia, and Thailand (Hailemariam et al. 2013; Okamoto et al. 2010). Molecular characterization in Iranian isolates of T. saginata has not been demonstrated so far. Comprehensive molecular studies in this area are required to increase our knowledge on the genetic diversity of this species and to provide effective vaccine against the parasite. Mitochondrial genes especially cytochrome c oxidase subunit 1 (CO1) are universally accepted markers for molecular identification of helminth parasites (Gasser et al. 1999). The purpose of the present study was to identify inter- and intraspecific variation of *T. saginata* in a large number of cattle isolates from different parts of Iran using PCR sequencing of two mitochondrial genes, i.e., CO1 and 12S rRNA.

Materials and methods

One hundred and five specimens of *T. saginata* metacestode were collected from cattle in 20 slaughterhouses from Tehran, Alborz, and Kerman provinces of Iran from August 2010 to July 2011. The scolex was detached from the infected organs and each isolate was transferred to the Parasitology Lab of the School of Medicine, Kerman University of Medical Sciences. After three times rinsing with normal saline, the specimens were stored at -20 °C until used.

Genomic DNA of the isolates was extracted from each scolex by using High Pure PCR Template Preparation Kit (Roche, Mannheim, Germany) as described elsewhere (Rostami et al. 2013). The DNA was used for PCR amplification of cytochrome c oxidase subunit I (CO1) and 12S ribosomal DNA (12S rRNA) genes. A 400-bp fragment of CO1 was amplified by PCR with the forward JB3 (5'-TTTTTTGGGCATCCTGAGGTTTAT-3') and reverse

CO1	No. of isolates	Accession no.	12S rRNA	No. of isolates	Accession no.
IRTSCO1	70	JQ756969	IRTSSR1	56	KF362126
IRTSCO2	16	JQ756970	IRTSSR2	10	KC344674
IRTSCO3	8	JQ756971	IRTSSR3	6	KC344676
IRTSCO4	2	JQ756972	IRTSSR4	3	KC344677
Other haplotypes	9	JQ756973–JQ756979	Other haplotypes	30	KC344678-KC344701

 Table 1
 Frequency distribution of *Taenia saginata* haplotypes from Iranian cattle for two mitochondrial CO1 and 12S rRNA genes with the corresponding GenBank accession numbers

JB4.5 (5'-TAAAGAAAGAACATAATGAAAATG-3') primer (Bowles et al. 1992).

Polymerase chain reactions (50 μ L) were performed using 3.5 mM MgCl₂, 250 mM each of dNTPs, 25 pmol of each primer and 2 U Taq polymerase, and 4 μ l (50–100 ng/ml) of DNA template, under the following thermal profile: 5 min at 94 °C as an initial denaturation step, followed by 35 cycles of

30 s at 94 °C, 45 s at 50 °C, 35 s at 72 °C, and a final extension step of 10 min at 72 °C.

A taeniid-specific primer pair was designed for amplification of 12S rRNA gene using Primer-BLAST (http://www.ncbi.nlm.nih.gov/tools/primer-blast/). PCR was carried out using 12SRF (5'-AGGGGATAGGAC ACAGTGCCAGC-3') as the forward and 12SRR (5'-



Fig. 2 Genetic relationships of *T. saginata* isolated from cattle and other published for taeniid and *E. granulosus* as outgroup. The relationships were inferred based on phylogenetic analysis of cox1 and 12S rRNA data

using Bayesian inference. The accession numbers and sources of sequences are shown in Table 1. Nodal support is given as a pp value

E.granulosus.NC008075



Fig. 2 (continued)

CGGTGTGTACAT GAGCTAAAC-3') as reverse primers under the following thermal conditions: 5 min at 94 °C as a primary denaturation step, followed by 35 cycles of 30s at 94 °C, 45 s at 57 °C, 35 s at 72 °C, and a final extension of 10 min at 72 °C. The primers amplified an approximately 450-bp fragment of 12S rRNA gene. Negative control (no DNA) was included in each experiment. The PCR products were electrophoresed on 1 % (w/v) agarose gel containing ethidium bromide.

All amplicons were sequenced by an ABI-3730XL capillary machine (Macrogen Inc., South Korea). Sequence data were analyzed with Blast program while alignments were conducted using the softwares ClustalX and Bioedit (Fig. 1). CO1 and 12S rRNA nucleotide sequences of the representative isolates were submitted to NCBI GenBank (Table 1). Two phylogenetic analyses of the sequence data were inferred by Bayesian inference on CO1 and 12S rDNA sequences (Fig. 2) using the program MrBayes v.3.1.2 (http://mrbayes.csit.fsu.edu/index.php). Posterior probabilities (pp) were designed for 2,000,000 generations (ngen: 2,000,000). The TreeviewX v.0.5.0 program (Page 1996) was used to depict the resulting trees. Two dendrograms were drawn by using the sequences obtained in this study as well as reference sequences available for T. saginata and representative Taenia species in GenBank. Echinococcus granulosus sensu stricto G1 genotype (Accession No. NC008075) was applied in the model as outgroup. Pairwise comparisons were made for the two mitochondrial gene sequences of T. saginata isolates of the present study as well as the other published studies.

	2	3	4	5	9	7	∞	10	11	12	13	14	15 1	6 17	18	19	20	21	22	23	24 2	5 2	5 2'	
1. T. saginata.NC009938																								i i
2. IRTSC01 0	.000																							
3. IRTSC02 0	.003 0.0	03																						
4. IRTSC03 0	.003 0.0	03 0.00	05																					
5. IRTSC04 0	.003 0.0	03 0.00	0.0 20	-05																				
6. IRTSC05 0	.003 0.0	03 0.00	05 0.0	05 0.00)5																			
7. IRTSC06 0	.003 0.0	03 0.00	0.0 20	05 0.00	0.00:	\$																		
8. IRTSC07 0	.010 0.0	10 0.00	0.0 80	13 0.0	3 0.01	3 0.013																		
9. IRTSC08 0	.005 0.0	05 0.00	0.0 80	0.0 80	8 0.00	8 0.008	0.016																	
10. IRTSC09 0	.008 0.0	0.0 80	10 0.0	10 0.0	0 0.01	0.010	0.018	0.013																
11. IRTSC010 0	.013 0.0	13 0.0	16 0.0	16 0.0	6 0.01	5 0.016	0.024	0.018 0.	018															
12. IRTSC011 0	.005 0.0	05 0.00	0.0 80	03 0.00	8 0.00	8 0.008	0.016	0.010 0.	013 0.01	8														
13. T. saginata.AB465242 0	.003 0.0	03 0.00	0.0 20	05 0.00	5 0.00	5 0.005	0.013	0.008 0.	010 0.01	6 0.008														
14. T. saginata.AB533168 0	.000 0.0	0.0 0.00	33 0.0	03 0.00	3 0.00	3 0.003	0.010	0.005 0.	0.0 800	3 0.005	0.003													
15. T. saginata.AB465247 0	.000 0.0	0.0 0.00	0.0	03 0.0(3 0.00	3 0.003	0.010	0.005 0.	0.0 800	3 0.005	0.003	0.000												
16. T. saginata.AB533169 0	.000 0.0	0.0 0.00	0.0	03 0.0(3 0.00	3 0.003	0.010	0.005 0.	0.0 800	3 0.005	0.003	0.000	0.000											
17. T. saginata.AB533170 0	.026 0.0	126 0.02	29 0.0	24 0.02	9 0.02	9 0.029	0.037	0.032 0.	034 0.03	37 0.026	0.029	0.026	0.026 0	.026										
18. T. saginata.AB533172 0	.000 0.0	0.0 0.00	0.0	03 0.00	3 0.00	3 0.003	0.010	0.005 0.	008 0.01	3 0.005	0.003	0.000	0.000 0	0.0 0.0	26									
19. T. saginata.AB533175 0	.026 0.0	126 0.02	29 0.0	24 0.02	0.02	9 0.029	0.037	0.032 0.	034 0.03	37 0.026	0.029	0.026	0.026 0	0.026 0.0	00 0.0	56								
20. T. asiatica.AB608739 0	.026 0.0	126 0.02	29 0.0	24 0.02	20 0.02	9 0.029	0.037	0.032 0.	034 0.03	37 0.026	0.029	0.026	0.026 0	0.0 900.	00 0.0	26 0.00	0							
21. T. asiatica.NC004826 0	.032 0.0	32 0.05	34 0.0	29 0.05	34 0.03	4 0.034	0.043	0.037 0.	040 0.04	12 0.032	0.034	0.032	0.032 0	0.032 0.0	05 0.0	32 0.00	5 0.005							
22. T. solium.NC004022 0	.127 0.1	27 0.13	30 0.1	24 0.15	30 0.12	4 0.130	0.139	0.130 0.	136 0.13	9 0.120	0.130	0.127	0.127 0	0.127 0.1	39 0.1	27 0.13	9 0.139	0.145						
23. T. multiceps.GQ228818 0	.066 0.0	166 0.0t	58 0.0	63 0.06	8 0.06	8 0.068	0.077	0.071 0.	074 0.08	30 0.066	0.068	0.066	0.066 0	0.0 990.0	83 0.0	90.0 99	3 0.083	0.089	0.108					
24. T. hydatigena.GQ2288190	.129 0.1	29 0.15	32 0.1	26 0.15	32 0.12	9 0.0132	0.141	0.129 0.	138 0.14	1 0.129	0.132	0.129	0.129 0	0.129 0.1	41 0.1	29 0.14	1 0.141	0.147	0.141	0.132				
25. T. pisiformis.GU569096 0	.117 0.1	17 0.12	20 0.1	14 0.12	20 0.120	0.117	0.129	0.120 0.	126 0.13	32 0.111	0.120	0.117	0.117 0	.117 0.1	29 0.1	17 0.12	9 0.129	0.135	0.138	0.123	0.179			
26. T. crassiceps.NC0025470	.120 0.1	20 0.12	23 0.1	17 0.12	23 0.120	0 0.123	0.132	0.120 0.	129 0.13	10.114	0.123	0.120	0.120 0	.120 0.1	26 0.1	20 0.12	6 0.126	0.132	0.163	0.135	0.114 0	0.157		
27. E. granulosus.NC0080730	.206 0.2	06 0.20	99 0.2	02 0.20	9 0.20	5 0.206	2.220	0.209 0.	209 0.22	20 0.199	0.202	0.206	0.206 0	.206 0.2	20 0.2	0.22	0 0.22(0.226	0.199	0.234	0.176 (0.227 0	.179	
T. saginata: NC009938, Al T. serialis: AM503319, T	B465242 crassice	2, AB53 ps: NC	33168, 00254	, AB46. 7, T. so	5247, A <i>lium</i> : N	B533169 C004022), AB53. 2, <i>T.asia</i>	170, Al tica: NC	3533172 004826,	, <i>T. mult</i> AB533	<i>iceps</i> : G 175, AE	Q2288 860873	18, <i>T. sc</i> 9, and <i>E</i>	olium: N 3. granu	C00402 losus: N	12, T. pi UC008(siformi. 175	s: GU5	69096 a	nd T. hy	datiger	ıa: GQ2	228819	

Pairwise comparison of nucleotide sequence differences (%) in CO1 gene among T saginata isolates and some other taeniids Table 2

Table 3 Pairwise con	nparisc	nn of nu	cleotid	le sequ	ence d	ifferenc	es (%)	in 12S	rRNA	gene a	mong	T. sagi	<i>nata</i> iso	lates ar	nd som	ie othe	er taenii	ls								
1 2	3	4	5	9	٢	8	6	10	=	2	3 1	4 1	5 16	17	18	19	20	21	22	23	24	25	26	27 2	8 29	6
1. T. saginata.N																										
2. IRTSSR1 0.005																										
3. IRTSSR2 0.007 0.00	12																									
4. IRTSSR3 0.005 0.00	5 0.00	2																								
5. IRTSSR4 0.014 0.01	4 0.01	6 0.014																								
6. IRTSSR5 0.007 0.00	12 0.00	5 0.007	7 0.012																							
7. IRTSSR6 0.007 0.00	0.00	9 0.007	7 0.016	500.0	_																					
8. IRTSSR7 0.012 0.01	2 0.01	4 0.004	1 0.004	0.014	0.014																					
9. IRTSSR8 0.005 0.00	15 0.00	7 0.005	5 0.014	1 0.007	0.007	0.012																				
10. IRTSSR9 0.005 0.00	15 0.00	7 0.000	0.014	1 0.007	0.007	0.007	0.005																			
11. IRTSSR10 0.009 0.00	9 0.01	2 0.005	0.006	0.007	0.012	0.016	0.009	0.009																		
12. IRTSSR11 0.006 0.00	15 0.00	7 0.005	5 0.019	0.007	0.012	0.012	0.009	0.005	0.014																	
13. IRTSSR12 0.009 0.00	15 0.00	7 0.005) 0.014	0.002	0.012	0.016	0.009	0.009	0.009 (.005																
14. IRTSSR13 0.012 0.01	2 0.01	4 0.012	2 0.012	0.005	0.014	0.019	0.012	0.012	0.007 0	0.016 0	.012															
15. IRTSSR14 0.016 0.01	2 0.01	4 0.016	5 0.021	0.00	0.019	0.024	0.016	0.016	0.016 (0.012 0	.007 0	600.														
16. IRTSSR15 0.016 0.01	2 0.01	4 0.012	2 0.026	0.014	0.009	0.019	0.016	0.012	0.021 0	007 0	.012 0	019 0	014													
17. IRTSSR16 0.005 0.00	15 0.00	7 0.005	5 0.014	1 0.007	0.007	0.012	0.005	0.005	0.009	0 600.0	0 600	012 0	016 0.0	16												
18. IRTSSR17 0.009 0.00	0.06	7 0.005	0.019	0.012	0.012	0.016	0.009	0.009	0.014 0	0.014 0	.014 0	0.016 0	021 0.0	121 0.00	05											
19. IRTSSR18 0.014 0.01	2 0.00	9 0.014	1 0.024	0.014	0.016	0.021	0.009	0.014	0.019 (0.016 0	.016 0	.021 0	024 0.0	24 0.0	14 0.0	14										
20. IRTSSR19 0.012 0.00	7 0.00	9 0.007	7 0.021	0.005	0.014	0.014	0.012	0.007	0.016 0	007 0	.012 0	0.014 0	014 0.0	14 0.0	12 0.0	16 0.0	119									
21. IRTSSR20 0.007 0.00	12 0.00	5 0.002	2 0.016	0.005	0.009	0.009	0.007	0.002	0.012 0	0.002 0	.007 0	0.014 0	014 0.0	0.0 0.00	07 0.0	12 0.0	14 0.00	5								
22. IRTSSR21 0.007 0.00	12 0.00	5 0.007	7 0.016	0.005	0.00	0.014	0.007	0.007	0.012 0	007 0	.007 0	0.014 0	014 0.0	14 0.00	07 0.00	0.0	14 0.00	9 0.00	10							
23. IRTSSR22 0.012 0.01	2 0.01	4 0.012	0.021	0.014	0.005	0.019	0.012	0.012	0.016 0	0.016 0	.016 0	.019 0	024 0.0	14 0.0	12 0.0	16 0.0	21 0.01	9 0.01	4 0.014							
24. IRTSSR23 0.009 0.00	9 0.01	2 0.005	0.019	0.012	0.002	0.016	0.009	0.009	0.014 0	0.014 0	.014 0	016 0	021 0.0	12 0.00	0.0 60	14 0.0	19 0.01	6 0.02	0.012	0.002						
25. IRTSSR24 0.009 0.00	00.0 6	7 0.005	0.019	0.012	0.002	0.016	0.009	0.009	0.014 0	0.014 0	.014 0	016 0	021 0.0	12 0.00	0.0 60	0.0 60	14 0.01	6 0.01	2 0.012	0.007	0.005					
26. IRTSSR25 0.012 0.00	0.00	9 0.012	0.021	0.00	0.005	0.019	0.012	0.012	0.016 0	0.012 0	.012 0	0.019 0	019 0.0	0.0 0.0	12 0.0	16 0.0	19 0.01	4 0.00	600.0 6	0.009	0.007	0.007				
27. IRTSSR26 0.007 0.00	12 0.00	5 0.007	7 0.016	0.005	0.009	0.014	0.007	0.007	0.012 0	007 0	.007 0	014 0	014 0.0	14 0.00	07 0.0	12 0.0	14 0.00	9 0.00	5 0.005	0.014	0.012	0.012	0.005			
28. IRTSSR27 0.007 0.00	0.00	9 0.007	7 0.016	0.009	0.005	0.014	0.007	0.007	0.012 0	012 0	.012 0	0.014 0	019 0.0	14 0.00	07 0.0	12 0.0	16 0.01	4 0.00	00.0	0.009	0.007	0.007	0.009	0.009		
29. IRTSSR28 0.005 0.00	15 0.0C	7 0.005	5 0.014	1 0.007	0.007	0.012	0.005	0.005	0.009 (0 600'	0 600.	012 0	016 0.0	16 0.00	05 0.0	0.0 60	14 0.01	2 0.00′	7 0.007	0.012	0.009	0.009	0.007	0.002 0	.007	
30. IRTSSR29 0.002 0.00	12 0.00	5 0.002	0.012	0.005	0.005	0.009	0.002	0.002	0.007 (007 0	.007 0	0 600'	014 0.0	14 0.04	02 0.0	0.0	0.00	9 0.00	5 0.005	0.009	0.007	0.007	0.009	0.005 0	.005 0.0	002
T. multiceps: GQ228818 NC008075	3, FJ74	4755; 1	r. sagin	iata: N	C0099	38, T. s	olium:	NC004	022, T.	pisifor	mis: G	U5690	96; T. h	ydatige	ena: G(22288	19; Т. с	rassice ₁	ps: NC	002547	; T. sol	ium: N	C0040	22; E. g	ranulos	:sns

Results

All 105 isolates were successfully amplified on both CO1 and 12S rRNA genes. Sequence analyses of CO1 and 12S rRNA genes showed 11 and 29 representative profiles respectively, designated as IRTSCO1 to IRTSCO11 for CO1 and IRTSSR1 to IRTSSR29 for 12S rRNA (Table 1). The level of pairwise nucleotide variation between individual haplotypes of CO1 gene were determined to be 0.3-2.4 % while the overall nucleotide variation among all 11 haplotypes was 4.6 % (Table 2). For 12S rRNA sequence data, the level of pairwise nucleotide variation was found 0.2-2.5 % and the overall nucleotide variation was determined as 5.8 % among 29 haplotypes (Table 3). Total mutations were 19 and 26 in CO1 and 12S rRNA that occurred in 18 and 25 segregation sites respectively. Figure 2 showed dendrograms based on COI and 12S rRNA gene sequences using the Bayesian inference method.

Discussion

Molecular characterization of parasites of medical and veterinary importance is a crucial factor in understanding epidemiology and control of parasitic infections. *T. saginata* is a zoonotic parasite of human and cattle with a worldwide distribution. It constitutes considerable medical and economical losses in endemic countries including Iran. Annual monetary burden of bovine cysticercosis in Iran was estimated at about US\$ 410,000 (Jahed Khaniki et al. 2010).

Several genomic regions have been used for phylogenetic studies of different *Taenia* species including 18S and 28S ribosomal RNA as well as mitochondrial genes (Yan et al. 2013; Hoberg 2006). In the present study, we investigated the mitochondrial CO1 and 12S rRNA genes diversity of 105 cattle isolates of *T. saginata* from Iran. CO1 is a universally accepted marker for the study of genetic variation and evolutionary biology of helminth parasites (Le et al. 2000). Intraspecific variation in *T. saginata* has been investigated in several studies (Hailemariam et al. 2013; Okamoto et al. 2010), however, more studies using larger sample size are required to improve our understanding on the significance of genetic variation within this important taeniid species.

Level of nucleotide variation in CO1 and 12S rRNA between *T. saginata* isolates from the present study and six other *Taenia* species was found to be 2.6–14.1 % and 2.8–19.5 %, respectively. This is in agreement with the expected level of nucleotide variations in CO1 in the genus *Taenia* that has been estimated to be 2.5–15.8 % by McManus and Bowles (1994).

As shown in Table 2, the level of pairwise nucleotide difference among the isolates of the present study and other T. saginata isolates is 2.4 % whereas pairwise comparison of CO1 sequences between isolates of the present study and T. asiatica is 2.6 %. Much higher pairwise differences have been observed between T. saginata and other Taenia species other than T. asiatica (6.3-15.8 %). Taenia asiatica formerly known as Taiwan taenia or Asian taenia; had been recognized in southeast Asia in 1980s (Fan 1988). This tapeworm was classified as a subspecies of T. saginata namely, T. saginata asiatica. Eom and Rim (1993) described it as a new species; Taenia asiatica. However, recent findings indicate probable hybridization between T. saginata and T. asiatica (Okamoto et al. 2010; Yamane et al. 2012). Regarding Mayr's definition of species, a species is defined as a group of organisms that are capable of interbreeding (Mayr 1996). In light of the fact that the two Taenia species are not reproductively isolated and the relatively low level of nucleotide difference between the two species shown in this study, it may be speculated that T. saginata and T. asiatica are either very closely related species or basically they are two subspecies of a single species.

Further studies on the molecular characterization of the parasite are clearly required from other geographical localities in different parts of the world. The present study provided mitochondrial data on the cattle isolates of *T. saginata* in Iran. In-depth studies on nuclear genes are essential to provide a comprehensive picture on the extent and significance of genetic variation within different *T. saginata* populations.

Acknowledgments The authors wish to thank all veterinary staff of different abattoirs that helped in collecting parasite specimen for this study. This work is done as a part of a PhD thesis done by S.R. and was financially supported by the Vice-Chancellor for Research, Kerman University of Medical Sciences, grant No. 90-128.

References

- Abunna F, Tilahun G, Megersa B, Regassa A, Kumsa B (2008) Bovine cysticercosis in cattle slaughtered at Awassa municipal abattoir, Ethiopia: prevalence, cyst viability, distribution and its public health implication. Zoonoses Public Health 55(2):82–88
- Bowles J, Blair D, McManus DP (1992) Genetic variants within the genus *Echinococcus* identified by mitochondrial DNA sequencing. Mol Biochem Parasitol 54(2):165–173
- Cabaret J, Geerts S, Madeline M, Cl B, Barbier D (2002) The use of urban sewage sludge on pastures: the cysticercosis threat. Vet Res 33(5): 575–597
- Dorny P, Praet N (2007) *Taenia saginata* in Europe. Vet Parasitol 149(1): 22–24
- Eom KS, Rim HJ (1993) Morphologic descriptions of *Taenia asiatica* sp. n. Korean J Parasitol 31(1):1–6
- Fan PC (1988) Taiwan Taenia and taeniasis. Parasitol Today 4(3):86-88
- Gasser RB, Zhu X, McManus DP (1999) NADH dehydrogenase subunit 1 and cytochrome c oxidase subunit I sequences compared for members of the genus *Taenia* (Cestoda). Int J Parasitol 29(12):1965–1970
- Hailemariam Z et al (2013) Molecular identification of species of *Taenia* causing bovine cysticercosis in Ethiopia. J Helminthol 88(3):376–380

- Hoberg EP (2006) Phylogeny of *Taenia*: species definitions and origins of human parasites. Parasitol Int 55:S23–S30
- Jahed Khaniki GR, Raei M, Kia EB, Motevalli Haghi A, Selseleh M (2010) Prevalence of bovine cysticercosis in slaughtered cattle in Iran. Trop Anim Health Prod 42(2):141–143
- Jeon H-K, Kim K-H, Eom KS (2007) Complete sequence of the mitochondrial genome of *Taenia saginata*: comparison with *T. solium* and *T. asiatica*. Parasitol Int 56(3):243–246
- Kia EB, Masoud J, Yalda A, Mahmoudi M, Farahani H (2005) Study on human taeniasis by administring anti-taenia drug. Iran J Public Health 34(4)
- Le TH, Blair D, McManus DP (2000) Mitochondrial genomes of human helminths and their use as markers in population genetics and phylogeny. Acta Trop 77(3):243–256
- Mayr E (1996) What is a species, and what is not? Philos Sci: 262-277
- McManus DP, Bowles J (1994) Asian (Taiwan) Taenia: species or strain? Parasitol Today 10(7):273–275
- Megersa B, Tesfaye E, Regassa A, Abebe R, Abunna F (2010) Bovine cysticercosis in cattle slaughtered at Jimma Municipal Abattoir, South western Ethiopia: prevalence, cyst viability and its socioeconomic importance. Vet World 3(6):257–262

- Okamoto M, Nakao M, Blair D, Anantaphruti MT, Waikagul J, Ito A (2010) Evidence of hybridization between *Taenia saginata* and *Taenia asiatica*. Parasitol Int 59(1):70–74
- Page RDM (1996) Tree view: an application to display phylogenetic trees on personal computers. Comput Appl Biosci 12:357–358
- Roberts T, Murrell KD, Marks S (1994) Economic losses caused by foodborne parasitic diseases. Parasitol Today 10(11):419– 423
- Rostami S, Talebi S, Babaei Z, Sharbatkhori M, Ziaali N, Rostami H, Harandi MF (2013) High resolution melting technique for molecular epidemiological studies of cystic echinococcosis: differentiating G1, G3, and G6 genotypes of *Echinococcus granulosus* sensu lato. Parasitol Res 112:3441–3447
- Silva CV, Costa-Cruz JM (2010) A glance at *Taenia saginata* infection, diagnosis, vaccine, biological control and treatment. Infect Disord Drug Targets 10:313–321
- Yamane K et al (2012) Recent hybridization between *Taenia asiatica* and *Taenia saginata*. Parasitol Int 61(2):351–355
- Yan H, Lou Z, Li L, Ni X, Guo A, Li H, Zheng Y, Dyachenko V, Jia W (2013) The nuclear 18S ribosomal RNA gene as a source of phylogenetic information in the genus Taenia. Parasitol Res 112:1343–1347