Culturable diversity and Biochemical Features of thraustochytrids from Coastal Waters of Southern China

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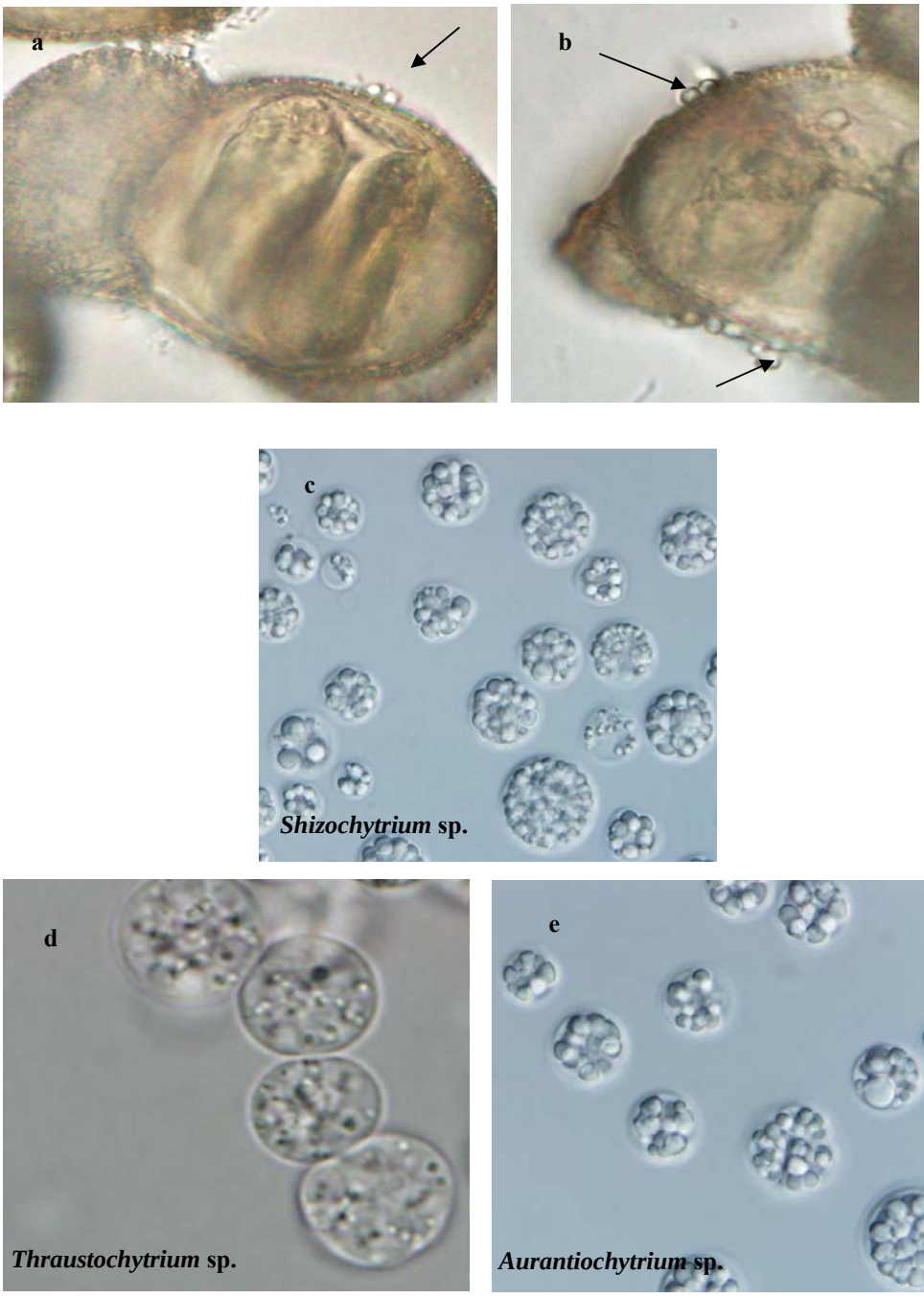

Fig. S1 Light micrographs of the thraustochytrid cells; a) attached to pine pollen surface (marked with arrow) and different species isolated in the present study; b) *Shizochytrium* sp.; c) *Aurantiochytrium* sp.; d) *Thraustochytrium* sp.

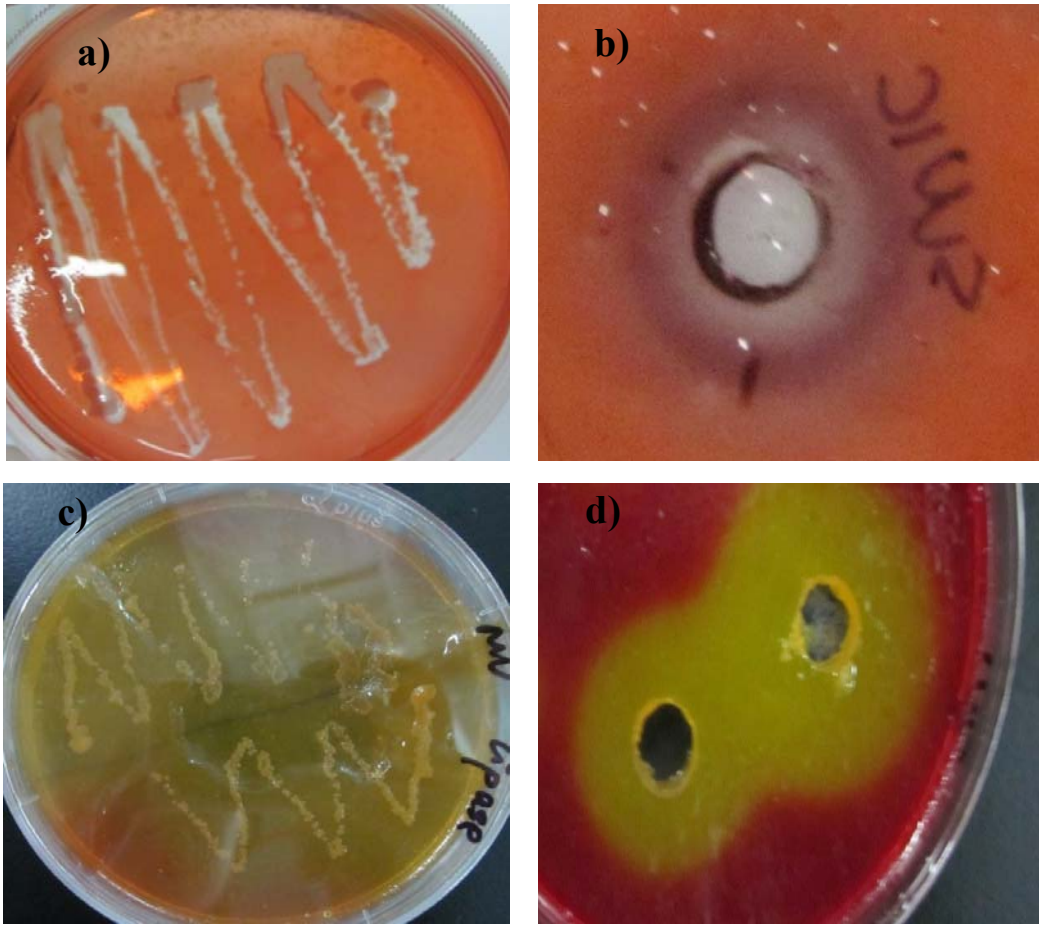

Fig. S2 Qualitative screening of CMCase and lipase enzyme activity of the thraustochytrid isolates. (a) Thraustochytrid isolate, PKU#SW1, grown on CMC-MV plate; (b) Cell free culture supernatant of the thraustochytrid isolate, PKU#SW1; **(c)** Thraustochytrid isolate, PKU#Sed1, grown on chromogenic MV plate containing olive oil and phenol red; and (d) Cell free culture supernatant of the thraustochytrid isolate, PKU#Sed1

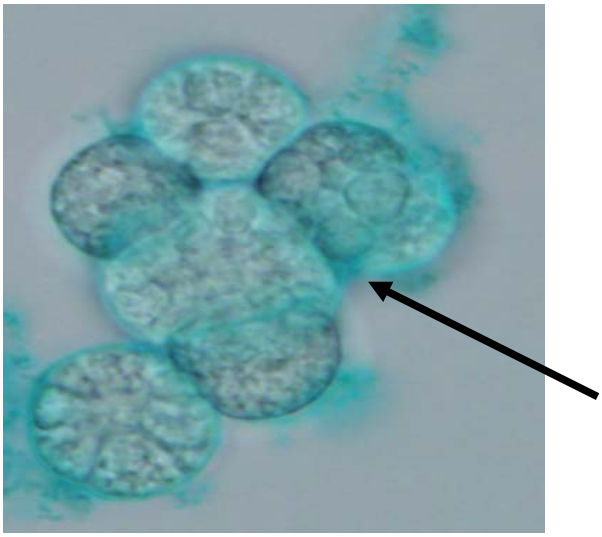

Fig. S3 Phase contrast micrograph of Alcian Blue stained (EPS sheath marked with arrow) cell of the thraustochytrids isolate, PKU#SW1.

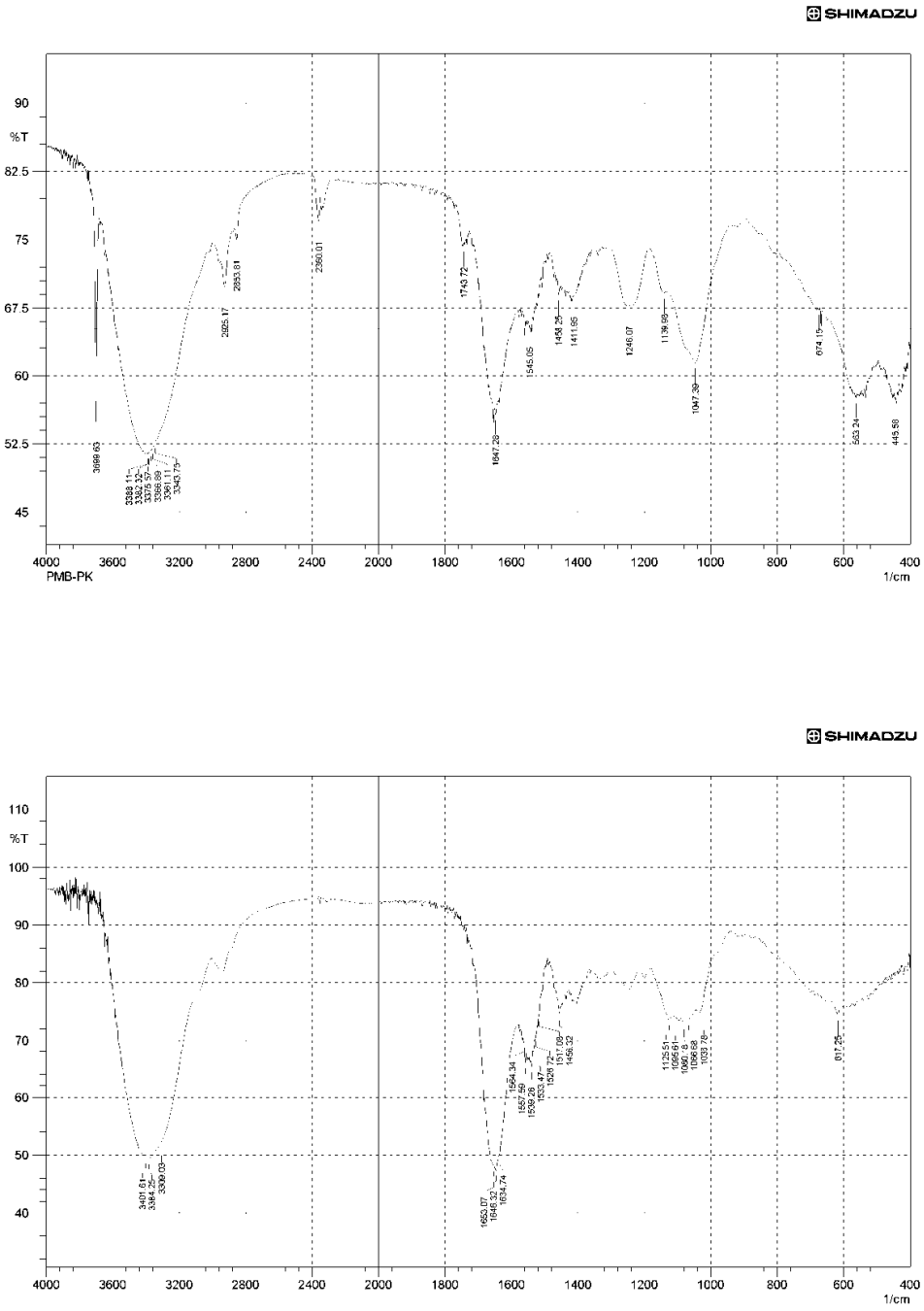

Fig. S4 FTIR spectra of the EPS samples of the thraustochytrids. PKU#Sed1 (Top Panel) and PKU#SW1 (Bottom Panel)

Table S1 Qualitative plate screening results of extracellular enzymes by 10 representative thraustochytrid isolates; Estimation of lipase produced by four strains of thraustochytrids; Biomass, Total fatty acid (TFA) and docosahexaenoic acid (DHA) yield of four thraustochytrid isolates at 4th day of growth

Isolate no.	Thraustochytrid genera	Protease	Cellulase	Lipase	Growth media with olive oil (2nd day)		Growth media without olive oil (1st day)		Biomass (g l^{-1})	TFA (% biomass)	DHA (% TFA)	DHA (g l^{-1})
					OD660	Lipase activity (U ml^{-1})	OD660	Lipase activity (U ml^{-1})				
PKU#Mn4	*Shizochytrium* sp.	-	+	++	6.914±0.570	4.630±1.517	6.914±0.570	4.630±1.517	7.1 ± 0.1	51.5 ± 5.6	44.3 ± 1.3	1.6
PKU#Mn15		-	+	+								
PKU#Mn35		-	+	+								
PKU#Mn11	*Aurantiochytrium* sp.	-	+	+								
PKU#Sed1		-	+	+++	6.607±0.289	11.874±12.110	6.607±0.289	11.874±12.110	7.5 ± 1.2	37.9 ± 21.5	41.2 ± 0.2	1.2
PKU#SW7		-	+	+								
PKU#SW1	*Thraustochytrium* sp.	-	++	++	2.737±0.233	2.635±1.004	2.737±0.233	2.635±1.004	3.8 ± 0.9	23 ± 15.6	30.3 ± 4.4	0.3
PKU#SW2		-	+	++	1.876±0.202	3.202±1.769	1.876±0.202	3.202±1.769	3.3 ± 1.1	8.0 ± 0.9	27.3 ± 1.4	0.07
PKU#SW8	*Thraustochytriidae* sp.	-	+	+								
PKU#Mn16		-	+	+								

The result of enzyme activities (intensity of clearance zone), (-): negligible; (+): moderate; (++): high; (+++): very high activity

Table S2 Effect of glucose concentration on EPS production (± SD) of the isolate PKU#Sed1 (5th day of growth)

% Glucose concentration	EPS (µg ml^{-1})
0	157.5 ± 0.04
1	194.4 ± 0.18
3	307.7 ± 0.01
5	315.3 ± 0.01
7	345.3 ± 0.19

Table S3 Fatty acid composition of the four thraustochytrid isolates (mean of triplicate analysis, SD⩽10%)

Isolate no.	Fatty acids (% TFA)									
	14:0*	**15:0**	**16:1**	**16:0**	**17:0**	**18:1**	**18:0**	**20:4**	**20:5**	**22:6**
PKU#Mn4 (*Shizochytrium* sp.)	2.5	0.5	0.2	51	0.2	0.2	0.8	---	0.3	44.3
PKU#Sed1 (*Aurantiochytrium* sp.)	4.3	0.7	0.3	51.9	---	---	0.9	---	0.7	41.2
PKU#SW1 (*Thraustochytrium* sp.)	14.7	10.7	2.1	36.4	1.6	3.1	0.5	0.1	0.3	30.3
PKU#SW2 (*Thraustochytrium* sp.)	21.8	9.7	2.5	35.2	0.8	1.9	0.3	0.03	0.28	27.3

***14:0- myristic acid; 15:0- pentadecyclic acid; 16:1- palmitoleic acid; 16:0- palmitic acid; 17:0- margaric acid; 18:1: oleic acid; 18:0: stearic acid; 20:4(n-6): arachidonic acid; 20:5(n-3): eicosapentaenoic acid (EPA); 22:6(n-3): docosahexaenoic acid (DHA).**