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基于异源 cDNA基因芯片杂交的鳜鱼肌肉组织基因表达谱初步分析

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摘要: cDNA基因芯片技术已广泛应用于生物物种间功能基因组和表达谱学研究。然而,鱼类基因芯片开发和应用相对落后。为了筛选与肉质性状相关功能基因,本研究首次试用异源斑马鱼基因 cDNA芯片,对两种肉质性状明显差异的鳜鱼和鲢鱼肌肉组织中基因表达进行了比较分析。从两种鱼肌肉组织中提取总 RNA,经 Biotin荧光标记与拥有 15617个 cDNA片段的斑马鱼基因芯片 (Affymetrix)杂交后,检测出 375个表达基因。与鲢鱼比较,鳜鱼肌肉组织锁定的基因中有 180个上调表达基因和 195个下调表达基因。在鳜鱼肌肉组织 180个上调基因中,49个为已知功能基因,131个为未知功能基因。根据基因文库同源功能基因分析,我们将 49个已知上调基因按功能大约分为七大类,其中与肌肉结构相关基因包括肌球蛋白重链基因 (MYH)、肌纤维间连接基因和细胞骨架结构基因等。同时,我们对与肉质结构性状密切相关的功能基因进行了分析,并结合与鳜鱼优良肉质结构和功能基因表达关系进行了讨论。

关键词:鳜鱼;鲢鱼;肌肉组织;基因表达;基因芯片
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基因芯片技术是目前最快捷和有效的分子生物学方法之一,可用来从全基因组水平上研究控制某一生物个体或同一个体不同发育阶段,或不同组织的基因或基因表达水平。它的基本原理是将高达一万个以上的基因片段 (寡聚核苷酸、EST、cDNA等)吸附或原位合成在同一薄片或芯片上,然后应用核酸分子碱基互补原理,用荧光素 (如 Biotin, Cy3或 Cy5)标记的 cDNA或 cRNA与之进行分子杂交,经充分洗去非特异结合后,采用计算机扫描和数据处理,通过与对照样品分析比较,找出差异表达的基因组或转录组[1,2]。基因芯片技术在鱼类研究的应用已有不少报道,如 Ton, *et al.* 构建了含有 4512个斑马鱼基因转录组谱,即 cDNA基因芯片[3]; Linney, *et al.* 采用 cDNA芯片对斑马鱼不同体节发生阶段的基因表达图谱进行了对比研究,发现在受精后 12h有 420个上调基因和 386个下调基因,而发育到 24h,上调和下调基因分别增加到 954和 766个,同时发现一些差异性表达的肌肉分化的新基因[4]。此外, cDNA基因芯片技术还被成功地应用于鱼类免疫基因的锁定[5—7]、病原微生物基因控制[8—10]以及环境毒性因子影响的基因表达[11,12]等。

鳜鱼 (Mandarine fish)又称桂花鱼,桂鱼,属鲈形目,鳜亚科,鳜属。鳜鱼肉质细嫩,味鲜美,营养价值高,已成为广受欢迎的名贵水产品之一。在我国,鳜鱼已由过去天然水域发展到规模化的集约化人工养殖,以满足社会对高档名贵水产品日益增长的需求。根据我们近期用生物化学和蛋白质组学比较研究证实,与常规养殖品种鲢鱼肉质成分比较,鳜鱼不仅蛋白质干重含量高于鲢鱼 8%左右,而且必需氨基酸和鲜味氨基酸含量也明显高于鲢鱼[13]。为什么两种硬骨鱼类品种间存在明显肉质差异?是否鳜鱼肌肉组织存在某些控制肉质性状的差异基因,或存在控制肉质性状功能基因的表达差异?针对这些问题,本研究采用斑马鱼 cDNA基因芯片分析和检测了鳜鱼和鲢鱼肌肉组织基因表达谱,并对其进行了比较分析。研究

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结果表明,与鲢鱼相比,鳜鱼肌肉组织经与斑马鱼 cDNA基因芯片的 15617个基因杂交,共检测出 375个差异表达基因,其中包括 180个上调基因,195个下调基因。此外,我们还对鳜鱼上调表达基因进行了功能分类,并结合鳜鱼肌肉结构与其相关的功能基因关系进行了分析讨论。

1　材料与方法

1.1　鱼肌肉样品的取材和处理　从长沙市水产品市场购买健康体重 500g左右野生鳜鱼和鲢鱼各 3尾,待实验室暂养 24—42h后,取其背部白色骨骼肌肌肉 50g,经用生理盐水充分洗涤后切碎或立即进行全 RNA提取或贮存于液氮中随时备用。

1.2　总 RNA的提取　准确称取鳜鱼和鲢鱼肌肉 5g,采用 Trizol方法提取肌肉组织总 RNA,所得 RNA样品质量通过 RNA凝胶电泳检测,随即进行分析或保存在 -80℃保存备用。

1.3　基因芯片杂交,cRNA的合成、标记和芯片杂交　斑马鱼 cDNA基因芯片(Zebrafish Genome Array, Part#900487)购自美国 Affymetrix公司,其 cDNA合成、Biotin标记、芯片杂交和分析方法均按公司提供的操作规程进行。主要技术线路和方法是:鳜鱼和鲢鱼总 RNA经 Qiagen公司的 RNeasy minikit纯化后,采用 Affymetrix-one cycle cDNA synthesis Kit合成第一和第二单链 cDNA。新合成的 cDNA经纯化后,Genchip IVT Labeling Kit完成 cDNA的转录合成和 Biotin荧光标记。Biotin荧光标记的 cDNA片段经纯化后,按如下成分配成芯片杂交液(cDNA片段(0.5mg/mL)30μL,Oligo B_2 50μL,20 ×Hybridization control 15μL,鱼精 DNA(9.3mg/mL)3.3μL,乙酰化 BSA(50mg/mL)3μL,2 ×杂交缓冲液 150μL,DMSO 30μL,RNase-free water 63.7μL),杂交反应总体积为 300μL。待芯片杂交液加入到芯片中,在 45℃条件下,按 60r/min迅速摇转,杂交反应持续 16h。杂交结束后,吸出芯片中杂交液,加入洗液,充分洗涤后,进行基因芯片扫描分析。

1.4　芯片扫描和数据处理分析　经杂交洗涤后的基因芯片采用 Affymetrix 418 Array Scanner进行扫描分析,所获数据应用 Affymetrix GCOS软件进行数据处理。经杂交反应与斑马鱼基因芯片的 15617个基因杂交反应所获信号采用 R/Bioconductor软件逐个分析(Http://www.bioconductor.org);数据校正和标准值(Background correction and normalization)按 Robust multi-Array Average cRMA方法进行[14,15]。鳜鱼和鲢鱼品种间肌肉组织中基因表达按 MF to SC 2.0倍信号强度进行分析,位于标准值红线以上的基因为上调基因,红线以下的为下调基因。所分析检测出的两种鱼基因分别从基因文库中检索,并按其基因性质和功能进行分类并对在鳜鱼肌肉中表达差异明显的基因进行比较,以找出与肉质结构和功能相关的特定差异基因。

2　结　果

2.1　芯片杂交反应效果

由于鳜鱼和鲢鱼 cDNA基因芯片缺乏,我们采用了 Affymetrix公司的斑马鱼 cDNA基因芯片,该基因芯片含有斑马鱼基因组的 15617个基因 cDNA或 EST片段,如图 1所示,鳜鱼(A)和鲢鱼(B)均显示出明显的杂交反应效果,即表现出基因表达强度与反应信号呈正相关性。按照误差干预值和 IQR(Interquartile Range)为 -0.653,IQR值为 0.204(图 1C);而鲢鱼干预值和 IQR值分别为 0.221和 0.15(图 1D)。针对基因数目分布分析,所获的数据均应进行标准校正(Microarray background correction and normalization)。如图 2所示,为对两种鱼基因分布密度和信号强度的校正和标准化分析。图 2A为基因分布密度和表达强度之关系。按其基因表达(Log2 expression)大多基因在 2—4区域,少量高表

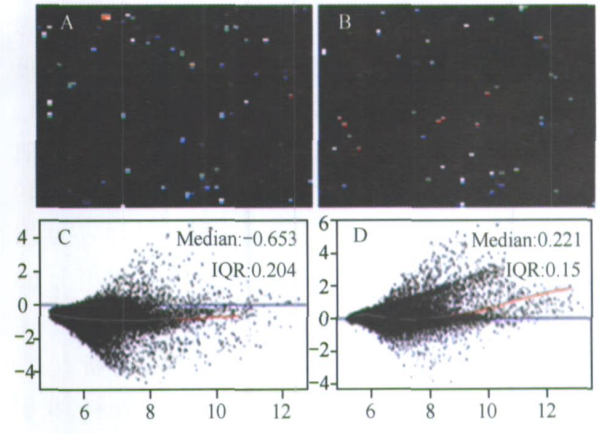

图 1　生物素标记的鳜鱼(A)和鲢鱼(B)cDNA与斑马鱼基因芯片杂交后的部分扫描图像

Fig. 1　Microarry hybridization image. Biotin labeled cDNAs from the madarin fish (A) and silver carp (B) hybridized with Zebrafish cDNA chip

白色斑点为高水平上调表达基因,红色次之,蓝色再次之,蓝黑色为不表达基因。C和 D分别为鳜鱼和鲢鱼基因表达倍数变化分布图(与误差和 IQR值比较)

Colour dots in white, red, blue and darkblue represent the hybridization intensities from the highest to lowest. C and D indicate gene distribution plots

达基因在 4以上区域。同样,采用 Boxplot分析,经校正后 50% 分布在红色框内,位于红色框上区域的为高表达基因,位于红色框下区域为低表达基因。

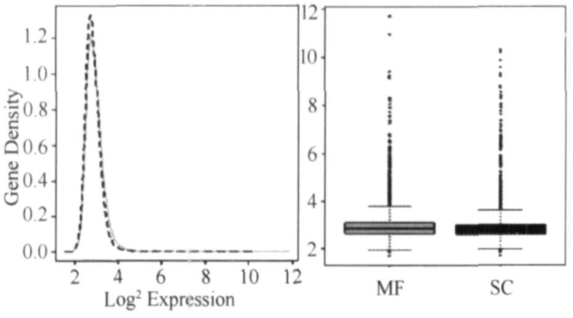

图 2 基因表达密度的方差分布与修正

Fig. 2 Expression density plot (left) and boxplot (right) after background correlation and normalization

左图为基因表达与分布密度的关系,表明大多基因分布于方差 2—4区域;右图为经 gcRMA (Robust multi-Array Average)方法校正后的基因分布区域

MF: Madarin fish; SC: Silver carp

2.2 鳜鱼基因表达谱及归类分析

经与含有 15617个 cDNA片段或 EST序列的斑马鱼基因芯片杂交分析,鳜鱼和鲢鱼肌肉组织共检测出表达基因数为 375个。以鲢鱼 2.0倍信号强度为参数,鳜鱼肌肉的 375个已检定的基因中,180个为上调表达基因,即其表达强度高于鲢鱼的基因数目,而低于鲢鱼的为 195个,即归类为下调表达基因。两种鱼所检测出的 375个基因可分为 8类 (Clusters),分别所含基因数目为 1、3、5、23、36、45、43和 219 (图 3)。与鲢鱼比较,鳜鱼肌肉上调基因为 180个,其中已知功能基因为 49个,未知功能基因为 131个;其下调的 195个基因图中,已知功能基因为 133个,未知功能基因为 62个 (表 1)。

2.3 鳜鱼上调表达基因功能分类和功能分析

根据基因文库以及相关表达的文献资料,我们选择性地对鳜鱼上调表达基因按其功能和性状分为七大类 (表 2)。与鳜鱼肌肉结构组成和发育相关的一类基因中,肌球蛋白重链基因和肌肉生长因子呈现出分别高于鲢鱼表达 32%和 15%;另一与肌肉结构相关的肌纤维连接因子 (Plectin1)亦表现出较高的表达,即高于鲢鱼 17%。组成肌肉细胞维管结构一类基因中,微管蛋白 (α-*tubulin*和 β-*tubulin*)基因的表达明显高于鲢鱼,分别为 26%和 15%;此外,构成肌肉膜组装蛋白 (BQ284848),鳜鱼高达 24%;与 DNA合成和加工的一类基因中,鳜鱼有 8个相关的高表达基因,其中值得注意的是逆转录酶基因,鳜鱼高于鲢鱼的 21%;与鳜鱼肌肉细胞发生分化相关的一类信号传导和转录因子中,多个基因表现出高表达,尤以在转录因子中,如转录因子 3 (Transcription factor 3)基因在两种鱼中均表现出高量表达 (鳜鱼和鲢鱼分别为 9.12和 8.19);在与免疫相关的一类基因中,包括有组织相溶性蛋白基因 (MHC histocompatibility)和大分子球蛋白前体基因。此外,在鳜鱼肌肉中,还检测出两个重要酶基因,即葡萄糖淀粉酶 (Glucoamylase)和组蛋白甲基化酶 (Histone demethylase)。在鳜鱼上调基因中,有 131个未知功能基因,这对鱼类功能基因的研究提供了较大的研究空间。

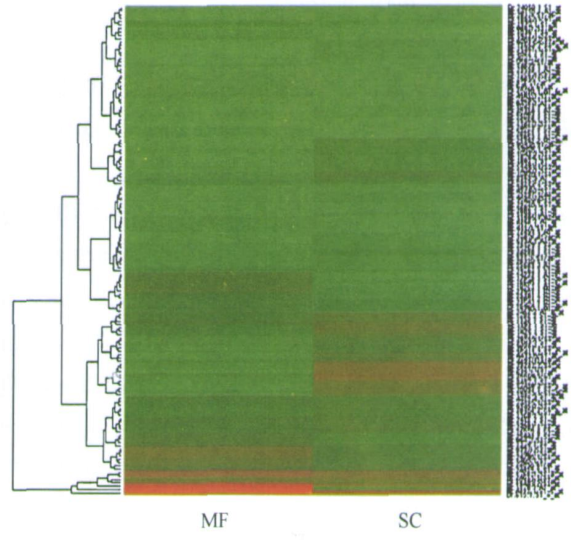

图 3 鳜鱼 (MF)和鲢鱼 (SC)肌肉中差异表达基因的分类分析

Fig. 3 Clustering of expressed genes between the two fishes

红色的为鳜鱼肌肉中高表达 (鲢鱼肌肉中相对低表达)的基因,绿色为低表达 (鲢鱼肌肉中相对高表达)的基因

Red colour: up-regulated genes; green colour: down-regulated genes

表 1 鳜鱼和鲢鱼肌肉组织基因表达数量比较

Tab. 1 Comparison of gene express between two fish species

Genes		鳜鱼 (MF)	鲢鱼 (SC)
Up-regulated	Known	49	133
	Unknown	131	62
	Total	180	195
Down-regulated	Known	133	49
	Unknown	62	131
	Total	195	180

表 2　鳜鱼上调基因的功能分类

Tab. 2　Functional classification of up-regulated genes in muscle tissues in *S. chuatsi*

ID	Genebank#	log2(MF)	log2(SC)	Median	Biological roles
Myosin structure and related genes					
Dr. 4812	NM152982	10.96	7.44	3.52	Myosin heavy polypeptide
Dr. 24997	AI831650	4.21	3.49	0.72	Cell adhesion
Dr. 9013	AY150226	3.95	3.30	0.65	Mesogenin
Dr. 17869	AI830555	3.96	3.35	0.60	Plectin 1, myofibe linker
Dr. 26173	BQ284848	4.45	3.36	1.10	Assembly of the membrane
Cytoskeleton structure and dynamics					
Dr. 11310	BG306212	5.00	3.95	1.05	Alpha-tubulin
Dr. 19061	Bg306212	4.87	4.17	0.70	Beta-tubulin
Dr. 16404	BI845781	4.79	3.69	1.10	Centrosome associated protein
Dr. 9552	AW019242	4.00	3.41	0.59	Microtubular aggregate protein
Dr. 2613	AW128192	4.07	3.48	0.59	Chromosome-associated kinesin
Dr. 14137	BC045503	4.06	3.43	0.64	Synaptonemal complex protein
DNA/RNA processing					
Dr. 12034	BI882418	5.82	5.01	0.82	Nuclear protein
Dr. 26273	AL715446	3.64	3.05	0.59	Cytidine 5-triphosphate synthase
Dr. 17164	BQ263996	3.68	3.08	0.60	GC-rich DNA-binding factor
Dr. 25530	BC046077	3.86	3.04	0.82	Nucleobindin 2
Dr. 16916	BM104181	3.83	3.11	0.71	Anti-freez protein
Dr. 12227	AA605698	4.45	3.51	0.94	Reverse transcriptase homolog
Dr. 3568	BC049408	4.39	3.55	0.83	Nei endonuclease
Dr. 6157	BM533522	4.33	3.61	0.72	Regulation of nucleobase
Signal transduction					
Dr. 5587	CB358631	4.72	3.91	0.81	SH3 domain-binding protein
Dr. 23670	AL728818	3.88	3.13	0.75	GTP-binding unclear protein
Dr. 18362	BC052113	4.21	3.53	0.68	Phospholipase A2
Dr. 13796	AI793818	4.17	3.43	0.74	Nicotinic acetylcholine receptor
Dr. 20081	BI983775	4.13	3.54	0.59	Elongation factor
Dr. 12310	BM956945	4.47	3.71	0.75	Giantin, Golgi autoantigen
Tanscription factors					
Dr. 21930	AI943061	9.12	8.19	0.93	Transcription factor 3
Dr. 2604	AW116861	4.27	3.39	0.88	Transcription complex subunit
Dr. 24623	BM026579	4.30	3.54	0.76	Retinoic acid receptor
Dr. 16462	AW018970	4.12	3.41	0.71	Peptide transport protein
Dr. 10645	BC044500	5.72	4.24	1.47	PC4 and SFRS1 related
Immune responses					
Dr. 25695	AW076914	4.63	3.86	0.78	MHC histocompatibility antigen
Dr. 25769	BG738204	3.97	3.24	0.73	MAHU macroglobulin precursor
Dr. 37	NM131835	4.18	3.52	0.65	Leukocyte derived chemotaxin
Dr. 22159	AL715280	4.71	3.82	0.89	TBP-1 interacting protein
Dr. 22711	NM131695	4.05	3.45	0.60	MHC membrane protein
Enzyme and others					
Dr. 25770	AW115530	3.68	2.99	0.69	Maltase-glucoamylase
Dr. 45	BI533287	3.49	2.81	0.69	Transmembrane protein
Dr. 4827	AL913284	4.16	3.53	0.63	Histone demethylase

3 讨 论

3.1 斑马鱼 cDNA基因芯片在鱼类基因表达分析的应用

cDNA基因芯片是用来分析研究不同生物个体基因表达,并用来从数万个基因中筛选相关目的功能基因的最快捷和有效的方法之一。至目前,硬骨鱼类基因芯片仅有斑马鱼 cDNA基因芯片 (Affymatrix)作为商业化的应用。本研究的目的是从两种肉质性状显著差异鳜鱼和鲢鱼中,比较分析研究两种鱼基因表达谱差异,力图筛选控制鳜鱼优良肉质性状相关的功能基因。由于鳜鱼和鲢鱼在系统分类地位上与斑马鱼有一定差异,这可能给杂交效果以及可能未检测出的基因数目带来限制,但是这种异源 cDNA芯片杂交和应用已在其他研究中有类似的报道,如 Rnnn, *et al*. 采用非洲丽鱼 (*Astatotilapia burtoni*)大脑 cDNA片段的基因芯片成功地用来检测 8种不同鱼类基因表达[16]。我们的研究结果证实,应用 Affymatrix斑马鱼芯片在本研究中的应用是有效和可行。我们选用两种鱼 cDNA与斑马鱼 cDNA基因芯片杂交,获得了较好的效果 (图 1A和 B)。同时,鳜鱼和鲢鱼杂交图谱 (图 1A和 B)比较,鲢鱼效果明显优于鳜鱼,这可能是鲢鱼和斑马鱼同属鲤科鱼类,亲缘关系较近所致。因此本研究采用异源斑马鱼 cDNA基因芯片用来检测鳜鱼和鲢鱼不同种间比较转录组学研究提供了可行的初探。但问题明显存在的是这类异源 cDNA基因芯片杂交对全面检测全基因表达谱带来限制,应采用其他方法进行补充研究,如 cDNA文库中的 EST序列分析,或蛋白质组学分析,尤其应结合鱼类基因库数据和生物信息学在鱼类基因开发上应用[38]。

3.2 鳜鱼肌肉结构与相关功能基因表达的关系

在锁定的鳜鱼肌肉上调表达的基因中,肌球蛋白重链基因 (Myosin heavy polypeptide)的表达量高于鲢鱼 32% (10.96/7.44)。肌球蛋白是构成肌肉的主要结构和功能的蛋白之一,它由约两条分子量为 220kD左右的重链和四条分子量为 16—20kD左右的轻链组成[17,18]。可以推断,高量表达的肌球蛋白可能与鳜鱼紧密的肉质结构和高肉含量直接相关。在硬骨鱼类,还存在一类与肌肉分化相关的转录因子[39,40]。然而本研究所检测到的转录因子仅有 mesogenin(AY150226)在两种鱼中有表达,这也可能是远源种间差异所致。在鳜鱼肌肉中锁定的另一个与肌肉结构相关的基因是 *Plactin*,它的表达量高于鲢鱼肌肉 15%。Plactin是一个分子量为 500kD左右的细胞连接蛋白 (Myofiber crosslinkers),在哺乳类肌肉细胞中首次发现[19]。该基因的主要功能是作为细胞骨架的三种纤维包括微管、微丝和中间纤维的连接蛋白,还参与细胞信号传导作用[20,21]。*Plactin*基因在鱼类肌肉细胞中表达仅见在雄性鱼中的工作[22]。该研究证实,Plactin纤维与肌纤维的紧密连接使鱼肌肉保持高度的韧性。同时,尤为重要的是 Plactin蛋白属于肌纤维间连接蛋白,它可能对紧密肉质结构有着重要的作用。因此,对于该基因的进一步分析研究有着重要的生物学意义。此外,与鳜鱼肌肉肉质紧密有关的基因包括细胞联合蛋白 (Cell adhesion)在肌肉中高度表达。本结果显示,鳜鱼肌肉细胞联合蛋白 (AB31650)比鲢鱼表达高 17%。Dumont, *et al*. 在大马哈鱼 (Trout)中研究发现,NLRR-1作为细胞联合分子在慢速肌肉细胞中表达 (Slow muscle cells)[23]。另外,在斑马鱼肌肉纤维的结构和分化中,细胞联合蛋白起着重要的作用[24,25]。综上所述,可以推断,鱼类肌肉组织的结构受多种不同基因共同作用。

3.3 细胞骨架相关基因与鳜鱼肌肉细胞结果关系分析

细胞骨架包括微管、微丝和中间纤维是构成真核生物细胞的主要结构蛋白之一,它对维持细胞结构、物质运输和细胞分裂起着重要作用[26]。相对于鲢鱼比较,构成微管的结构蛋白,如 α-*tubulin*和 β-*tubulin*在鳜鱼肌肉中高度表达,其中 α-*tubulin*表达量高于鲢鱼 21%,β-*tubulin*高于鲢鱼 17%,可以推测,微管蛋白在鳜鱼肌肉细胞内的高度表达可能与其肌肉肉质韧性直接相关。此外,还有三个与细胞分裂过程中微管聚合相关的基因,即 BB845718、AW019242和 BC045503等三个基因,它们的产物为细胞分裂过程中微管的聚合提供条件,它们与肌肉细胞分裂和分化直接相关[30—32]。Kinesin属微管运动蛋白,为微管收缩和运动提供能量[30]。在鳜鱼肌肉中高表达的 Kinesin也可能为其肌肉细胞微管的运动能量提供相关。

3.4 与鱼类肌肉细胞生理活动相关功能基因的分析

除上述肌肉细胞结构基因以外,本研究锁定的与鳜鱼肌肉细胞分化和生理活动相关的上调表达基因还包括与 DNA和 RNA代谢相关功能基因 7个,信号传导相关基因 5个,转导因子及相关基因 5个,免疫相关基因 5个以及相关酶类基因 4个 (表 2)。

值得注意的是在鳜鱼肌肉细胞中有 2个与细胞分裂和周期控制相关的基因 (BI882418)[31,32],另外,抗冻基因 (BM104148)也在鳜鱼肌肉中上调表达,这可能是鳜鱼对低温环境适应的需要[33]。在锁定的转录因子中,鳜鱼肌肉细胞转录因子 BTF_3 (CB358631)和 PC4相关基因 (BC044500)表现高表达,分别较鲢鱼高 17%和 26% (表 2)。这 2个基因直接参与 DNA 转录过程中 RNA 聚合酶的激活和启动[34,35]。这证明鳜鱼肌肉细胞仍保持活跃的基因转录表达。此外,在鳜鱼中与免疫相关基因有 AW076914、BG738204、NM131815、NM131695、AL715280[36,37]。这些基因均在脊椎动物包括鱼类中发挥作用,说明鳜鱼具有一定抗性功能。

小结:本研究首次采用异源 cDNA 基因芯片在养殖名贵鱼类研究中进行了初步尝试,从方法上基本上是可行的,但毕竟由于鳜鱼和斑马鱼亲缘关系较远的缘故,对于全面检测基因表达谱带来了限制。因此,开发研究更多鱼类基因芯片用来鱼类基因组学研究急待解决并具有重要意义。

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GENE EXPRESSION PROFILES OF THE MUSCLE TISSUES OF THE MANDARIN FISH, SINIPERCA CHUATSI L. WITH ZEBRAFISH cDNA MICROARRAY

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Abstract: The mandarin fish has been recently becoming one of the most important aquaculture fish species in China because of its good meat quality and protein composition. Improving the production of these fish is a continuing goal of the aquaculture industry, and a better understanding of the molecular control of the muscle structure and development in this commercially important species could prove beneficial for more economic rearing and quality of production. Therefore, we intended to investigate what genes control the development of large amount of high-quality white muscle in this species, which may provide foundation for the understanding of its myogensis and information to refine musculature for aquaculture.

cDNA microarray is a powerful tool for the assaying of an organism's gemone and expression profiles. However, it is relatively behind in fish because limited arrays available. In order to obtain the gene expression profile and screen differentially expressed genes in muscle tissues in the mandarin fish, we applied the Affymetrix zebrafish cDNA microarray heterologously hybridized to both the madarin fish, *Siniperca chuatsi* and the silver carp, *H. molitrix*. Total RNAs were isolated from

the muscle tissues of the two species of fish and labeled with biotin, and hybridized to Zebrafish cDNA gene chips. Hybridized microarray chips were scanned with Affymetrix 418 Array Scanner. All expression signals from the hybridization to 15617 genes in zebrafish cDNA array were analyzed by R/Bioconductor software (http://www.bioconductor.org). Background correction and normalization were done by the methods of Robust multi-Array Average known as gcRMA in R/Bioconductor. Out of 15617 genes in Zebrafish cDNA chips, total of 375 genes were identified to be significantly expressed in the muscle tissues of the two fishes. Compared to the silver carp, 180 genes are up-regulated and 195 are down-regulated in the madarine fish's muscle tissues. Among the 180 up-regulated genes, 49 genes are functionally known and 131 are unknown. The 49 known genes could be categorized into seven functional groups and several of them are considered as potential candidates related to the fish's muscle structure and development. Several genes related to the muscle structure and meat textures were identified, including myosin heavy chain gene (MYH), myofiber linker gene (plectin) and cytoskeletal structure genes (tubulin and actin). In the mandarin fish, myosin heavy chain gene is expressed extremely higher at 32% than that in the silver carp (Tab. 2) and this large amount of myosin heavy chain protein expression may reflect to the large abundance of high-quality white muscle in this fish. Plactin identified from mammalian muscle tissue is another gene directly related to muscle structure and it functions as a cytolinker to cytoskeletal proteins and signal transductor. Plactin was expressed 16.5% higher in the mandarin fish than that in silver carp ($\log_2$ ratio at 3.95/3.30). Titin gene was also highly expressed in both of the two fish. These results indicated that the muscle tissue of the mandarin fish was highly endurant and elastic. A cluster of genes functioning in cytoskeleton and dynamics including tubulin isoforms, actin and microtubule-associated protein genes were quite abundantly expressed in the muscle of the mandarin fish. This group of genes played important roles in muscle cell structure, cell division and material transportation. In comparison to the silver carp, both alpha- and beta-tubulin were expressed at 21% and 17% higher in the mandarin fish, which suggested that high amount of tubulin protein in muscle cells of the mandarin fish may be directly related to its muscle elastic feature.

This is the first study using zebrafish cDNA microarray to heterogonous hybridization of other evolutionally far from fish species, and this information can be used to help select strains of high quality of muscular species for aquaculture industry and to provide a better understanding of gene expression profiles in relation to their biological functions in muscle tissue of different fish species.

Key words: *Siniperca chuatsi*; *H. molitrix*; Muscle; Gene express; cDNA microarray