



读 书 报 告

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Growth performance, intestinal histomorphology and growth-related gene expression in response to dietary *Ziziphus mauritiana* in Nile tilapia (*Oreochromis niloticus*)

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ABSTRACT

The present study was conducted to investigate the effects of *Ziziphus mauritiana* leaf powder (ZLP) on growth performance, intestinal histomorphology and growth-related gene expression of Nile tilapia (*Oreochromis niloticus*). ZLP was incorporated into the basal diet at 5, 10 and 20 g per kg diets (w/w) and fed to fish for 12 weeks. The obtained results revealed significantly ($P < .05$) improved average body weight, weight gain percentage, feed intake, feed efficiency ratio, protein efficiency ratio and apparent protein utilization in fish fed ZLP as compared to fish fed ZLP free diet with the highest being in fish fed 10 g per kg diet. The highest carcass total protein and the lowest moisture and lipid contents were observed in fish fed ZLP at 20 g/kg as compared to the control, while the lowest ash content was observed in fish fed ZLP at 10 g/kg. Serum ion Na^+ and Ca^{++} significantly increased ($P < 0.05$) in all groups fed with ZLP compared to the control group, while an increase in the serum K^+ ion concentration was recorded at 5 and 10 g ZLP fed groups. Significant upregulations in insulin-like

目录



01

研究背景

02

材料与amp;方法

03

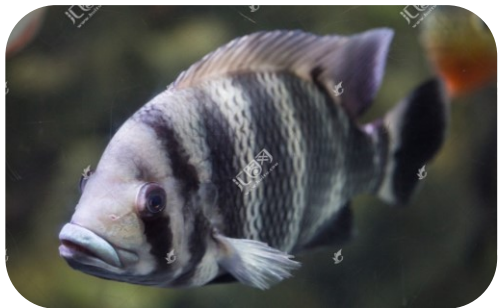
结果分析

04

结论



研究背景



罗非鱼，是全球重要经济养殖鱼类之一。

在集约化养殖系统中，它易受水质影响而被微生物感染。





抗生素通常被用做控制鱼疾病的药物，然而，它的滥用导致了细菌的耐药性，药物残留和水体污染。

开发一种抗生素替代物，是控制疾病和提高集约化养殖产量的一种有效策略。



酸枣是一种具有药用价值的植物。它的不同组织部位可治疗各种疾病，如哮喘、过敏、溃疡和炎症等。

酸枣叶片中碳水化合物和粗蛋白含量较高，易被动物消化吸收。据报道，酸枣枣叶可提高鲤鱼的免疫功能。但它作为一种饲料添加剂，在罗非鱼中还未有报道。





提出问题

酸枣叶作为一种饲料添加剂，是否能提高罗非鱼肠道功能？它对罗非鱼的生长又会产生怎样的影响？

目录

01

研究背景



02

材料与amp;方法

03

结果分析

04

结论

罗非鱼($9.63 \pm 0.05\text{g}$)

↓
酸枣叶粉

0

5g/kg

10g/kg

20g/kg

↓
饲养12周

取全鱼, 抽取血液, 取胃、肝脏、肠, 肌肉。

体成分

血液指标

生长指标

摄食和生长相关基因

形态学分析

↓
阐述酸枣叶粉对罗非鱼肠道形态和生长的影响

目录

01

研究背景

02

材料与amp;方法



03

结果分析

04

结论



饲料配方及引物

Table 1

Ingredients and chemical analysis of the experimental diets (on dry matter basis) containing different concentrations of powdered leaves of *Z. mauritiana* (ZLP).

Ingredients	ZLP (g/kg diets)			
	0	5	10	20
Fish meal (72%)	104.0	104.0	104.0	104.0
Soybean meal (44%)	429.8	429.8	429.8	429.8
Ground corn	203.2	203.2	203.2	203.2
Wheat bran	154.9	154.9	154.9	154.9
Cod fish oil	23.1	23.1	23.1	23.1
Corn oil	15.0	15.0	15.0	15.0
Vitamins premix ^a	10.0	10.0	10.0	10.0
Minerals Premix ^b	20.0	20.0	20.0	20.0
Starch	40.0	35.0	30.0	20.0
<i>Z. mauritiana</i> dry leaves powder (ZLP)	0.0	5.0	10.0	20.0
Total	1000	1000	1000	1000
Chemical analysis (g/kg)				
Dry matter	910.1	912.9	915.9	917.2
Crude protein	301.4	302.1	303.5	302.8
Ether extract	85.5	84.9	84.8	83.9
Total ash	86.5	88.6	86.6	87.6
Crude fiber	51.0	50.0	50.0	49.0
Nitrogen free extract ^c	475.6	474.4	475.1	476.7
Gross energy (Kcal/100 g) ^d	4441.6	4454.9	4464.8	4458.9

Table 2

Sequence of primers used for quantitative real-time PCR of ghrelin and IGF-1 genes.

Gene	Primer	Sequence (5'-3')	Annealing Temp.
β-actin	Forward	TGGCATCACACCTTCTATAACGA	60 °C
	Reverse	TGGCAGGAGTGTGAAAGGTCT	
Ghrelin	Forward	GTGGTGCAAGTCAACCAGTG	59 °C
	Reverse	CATGGCTTGGCGACCAATTC	
IGF-1	Forward	GTTTGTCTGTGGAGAGCGAGG	60 °C
	Reverse	GAAGCAGCACTCGTCCACG	



生长性能和饲料利用指标

Table 3
Growth performances and feed utilization of Nile tilapia fingerlings fed different *Z. mauritiana* (ZLP) levels for 12 weeks.

Items	ZLP (g/kg diets)			
	0	5	10	20
Initial weight (g)	9.63 ± 0.04	9.67 ± 0.01	9.68 ± 0.04	9.64 ± 0.02
Final weight (g)	28.79 ± 0.25 ^d	33.89 ± 0.20 ^c	38.30 ± 0.49 ^a	36.19 ± 0.25 ^b
Weight gain (g)	19.16 ± 0.25 ^d	24.21 ± 0.20 ^c	28.62 ± 0.45 ^a	26.55 ± 0.26 ^b
SGR (% g/day) ¹	1.30 ± 0.01 ^d	1.49 ± 0.01 ^c	1.63 ± 0.01 ^a	1.57 ± 0.01 ^b
Feed intake (g feed /fish)	34.85 ± 0.07 ^d	39.85 ± 0.16 ^c	42.42 ± 0.31 ^a	40.87 ± 0.26 ^b
FER ² 饲料系数	54.97 ± 0.65 ^d	60.77 ± 0.63 ^c	67.45 ± 0.75 ^a	64.97 ± 1.04 ^b
PER ³ 蛋白质效率比	2.00 ± 0.02 ^c	2.20 ± 0.04 ^b	2.42 ± 0.02 ^a	2.34 ± 0.05 ^a
APU (%) ⁴ 表观蛋白质利用	9.10 ± 0.50 ^c	10.82 ± 0.58 ^b	12.28 ± 0.25 ^a	11.71 ± 1.88 ^{ab}
EU (%) ⁵ 能量利用率	5.59 ± 0.44 ^b	6.29 ± 0.45 ^a	6.09 ± 0.86 ^a	6.25 ± 1.19 ^a

¹ Specific growth rate (SGR) = 100 (Ln W2 - Ln W1) / T.

² Feed efficiency ratio (FER) = 100 (weight gain / feed intake).

³ protein efficiency ratio (PER) = weight gain / protein intake.

⁴ apparent protein utilization (APU) = 100 [protein gain in fish(g) / protein intake in diet (g)].

⁵ energy utilization (EU) = 100 [Energy gain in fish (g) / energy intake in diet (g)]. Where W1 and W2 are the initial and final weights, respectively, and T is the experimental period (days). Data are presented as mean ± SEM. Means with the same letters in the same row are not significantly different at $P < .05$, using Tukey post hoc test.



体成分分析

Table 4
Whole body composition (g/kg, on fresh weight basis) of Nile tilapia fingerlings fed different *Z. mauritiana* (ZLP) levels for 12 weeks.

Items	ZLP (g/kg diets)			
	0	5	10	20
Moisture	741.0 ± 0.31 ^a	733.8 ± 0.22 ^{ab}	738.0 ± 0.18 ^{ab}	731.9 ± 0.21 ^c
Crude protein 粗蛋白	160.5 ± 0.21 ^b	167.6 ± 0.28 ^{ab}	167.3 ± 0.56 ^{ab}	171.0 ± 0.63 ^a
Total lipids	52.4 ± 0.26 ^a	49.8 ± 0.64 ^{ab}	48.9 ± 0.32 ^{ab}	48.2 ± 0.71 ^b
Total ash	46.1 ± 0.05 ^a	45.0 ± 0.62 ^{ab}	43.3 ± 0.06 ^b	44.9 ± 0.13 ^{ab}

酸枣枣叶可促进罗非鱼对饲料的摄入量，进而影响其体成分。



血液离子指标

Table 5

Serum ion concentrations (mEq/l) of Nile tilapia fingerlings fed different *Z. mauritiana* (ZLP) levels for 12 weeks.

Item	ZLP (g/kg diets)			
	0	5	10	20
Na ⁺	160.28 ± 0.48 ^b	164.00 ± 1.02 ^a	164.76 ± 0.68 ^a	164.86 ± 1.20 ^a
K ⁺	03.23 ± 0.14 ^b	04.27 ± 0.18 ^a	04.13 ± 0.09 ^a	03.30 ± 0.20 ^b
Ca ⁺⁺	140.52 ± 1.64 ^b	146.09 ± 0.63 ^a	146.67 ± 0.60 ^a	149.86 ± 1.81 ^a

处理组血清离子浓度显著增加，可能是植物的活性物质具有抗氧化作用，从而增加微绒毛对矿物质和营养物质的吸收。



摄食和生长相关基因表达

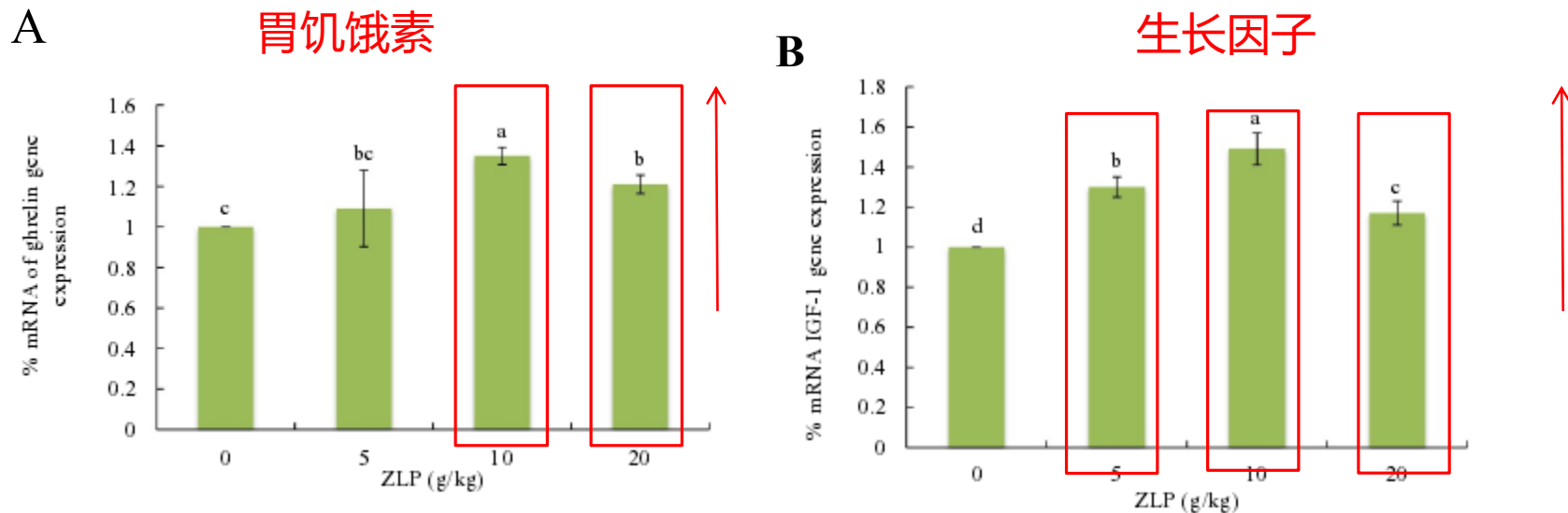
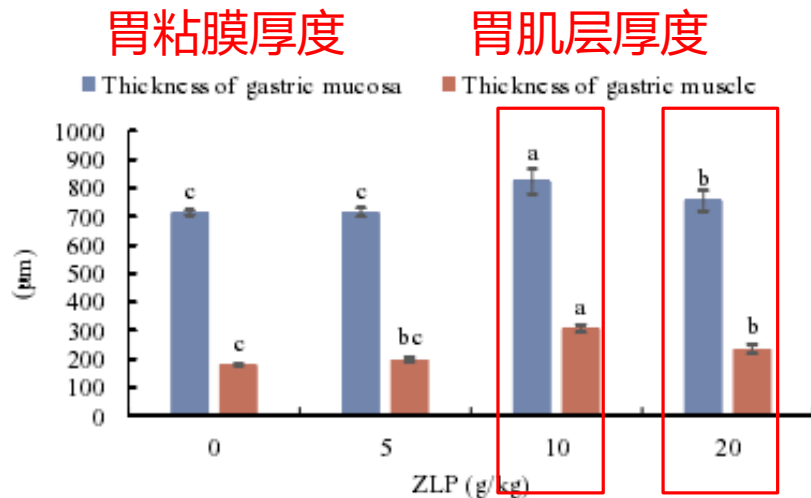


Fig. 1. The relative mRNA expression of (A) ghrelin gene in the stomach (B) insulin like growth factor (IGF-1) gene in the liver of Nile tilapia fed different concentrations of *Z. mauritiana* (ZLP) for 12 weeks. Data are presented as mean \pm SEM ($n = 4$). Means with different letters are significantly different at $P < .05$, using Tukey post hoc test.



胃形态学分析



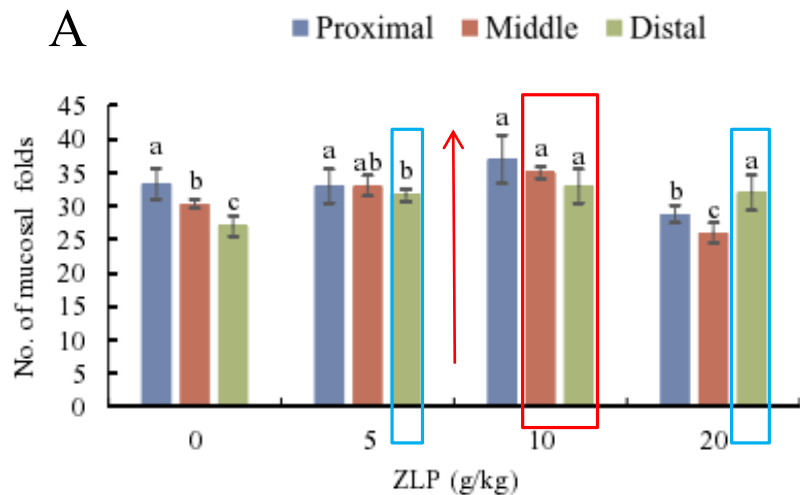
中浓度和高浓度处理组，均可显著增加胃粘膜和胃肌层厚度。

Fig. 2. Effect of different concentrations of *Z.mauritiana* (ZLP) on Nile tilapia gastric morphometric measurements; thickness of the gastric mucosa and thickness of the gastric muscle layer. Data are presented as mean \pm SEM ($n = 3$). Bars with the same colour with different letters are significantly different at $P < .05$, using Tukey post hoc test.

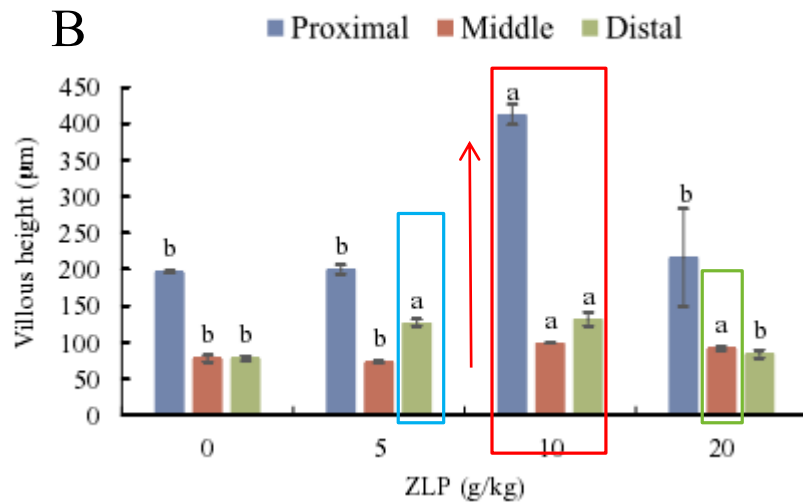


肠道形态学分析

粘膜褶皱数量

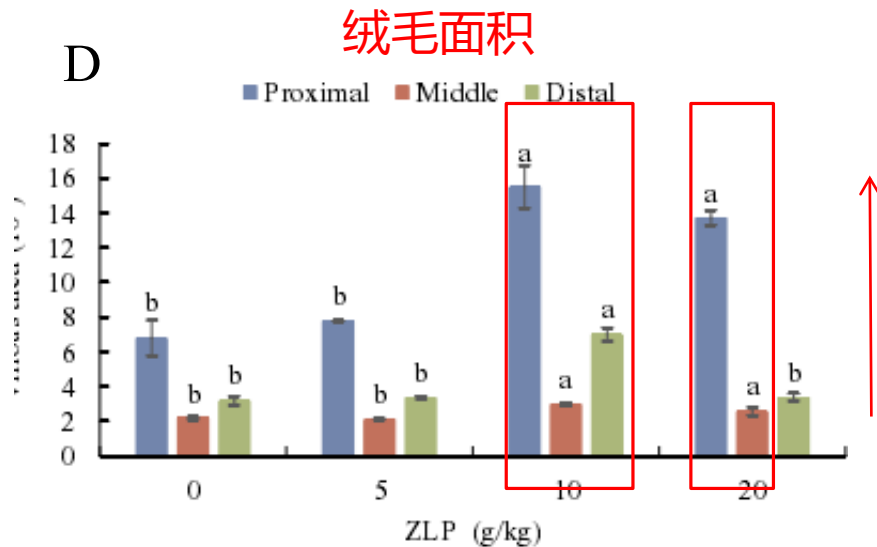
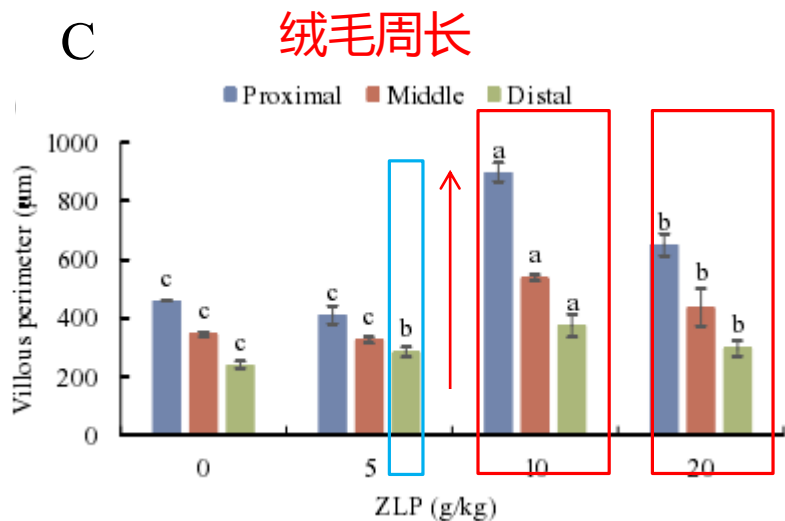


绒毛高度





肠道形态学分析

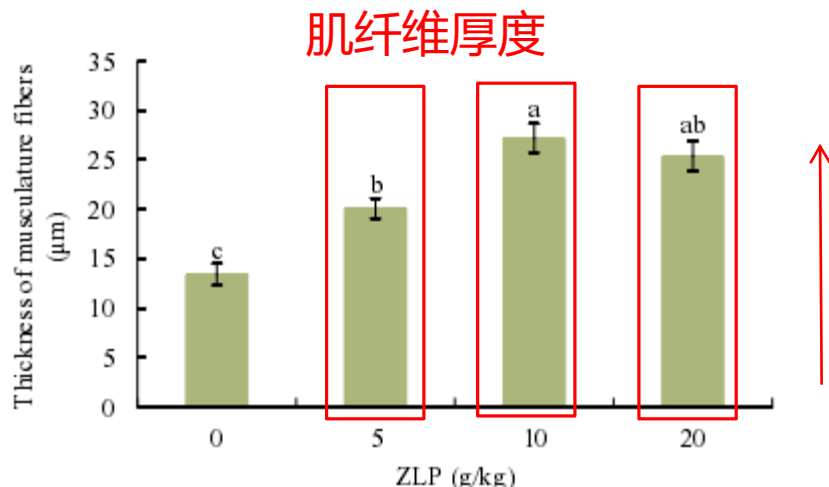


在低浓度处理组，肠道远端绒毛周长显著升高，在中、高浓度处理组的远端绒毛周长均显著升高。

在中浓度处理组，肠道绒毛面积显著升高，在高浓度处理组，近端和中端绒毛面积显著升高。



肌肉形态学分析



饲料中添加酸枣叶粉，可显著增加肌纤维厚度，这可能是酸枣叶中某些活性物质所造成的。

Fig. 4. Effect of different concentrations of *Z. mauritiana* (ZLP) on Nile tilapia musculature fiber thickness. Data are presented as mean \pm SEM ($n = 3$). Means with different letters are significantly different at $P < .05$, using Tukey post hoc test.



胃上皮细胞电镜图

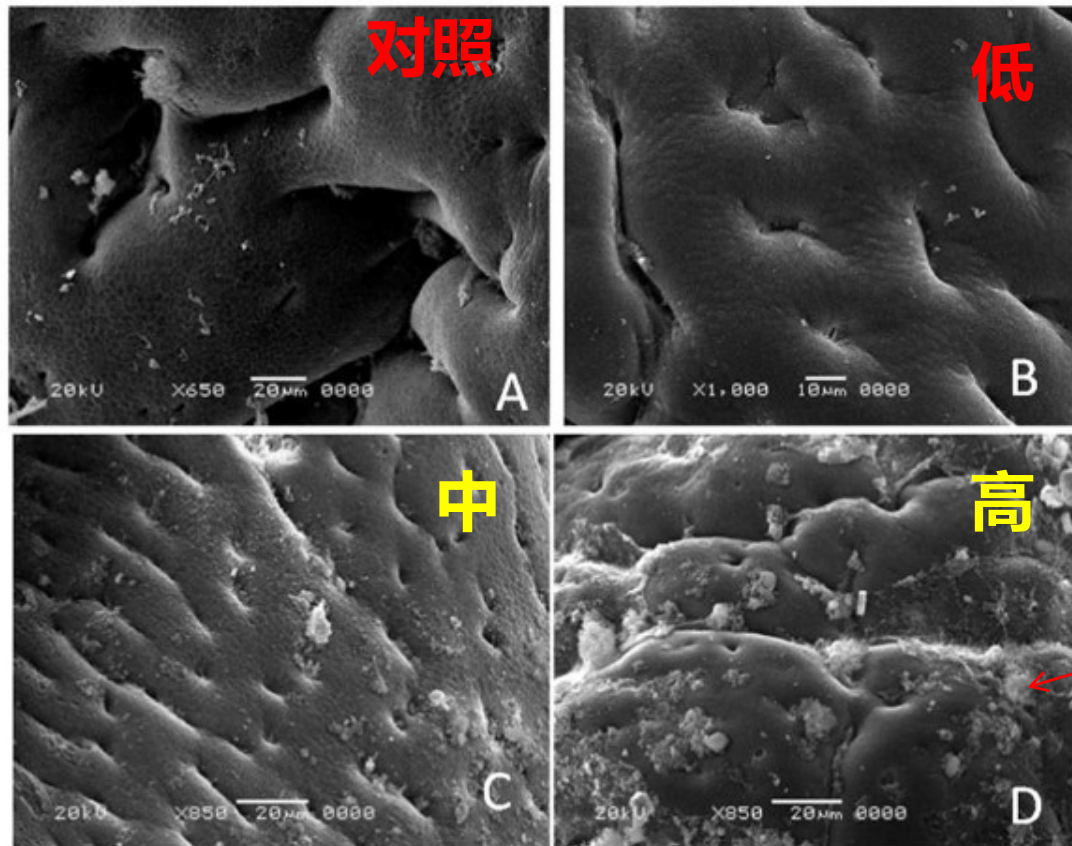


Fig. 5. Micrograph of the stomach of Nile tilapia at the control group (A), (B-D) obtained from fish received different concentrations of *Z. mauritiana* (ZLP) fed groups. (A), showing normal and regular oval or rounded columnar epithelial cells (CEC) lining mucosal folds and gastric pits ($\times 650$); (B) stomach of fish received *Z. mauritiana* at 5 g/kg diet showing normal and regular CEC with partially opened pits ($\times 1000$); (C) stomach of fish received *Z. mauritiana* at 10 g/kg diet showing normal and regular CEC with partially opened pits with accumulation of mucous ($\times 850$); (D) stomach of fish received *Z. mauritiana* at 20 g/kg diet showing normal and regular CEC with partially opened pits and severe mucus secretion ($\times 850$).

粘液



肠上皮细胞电镜图

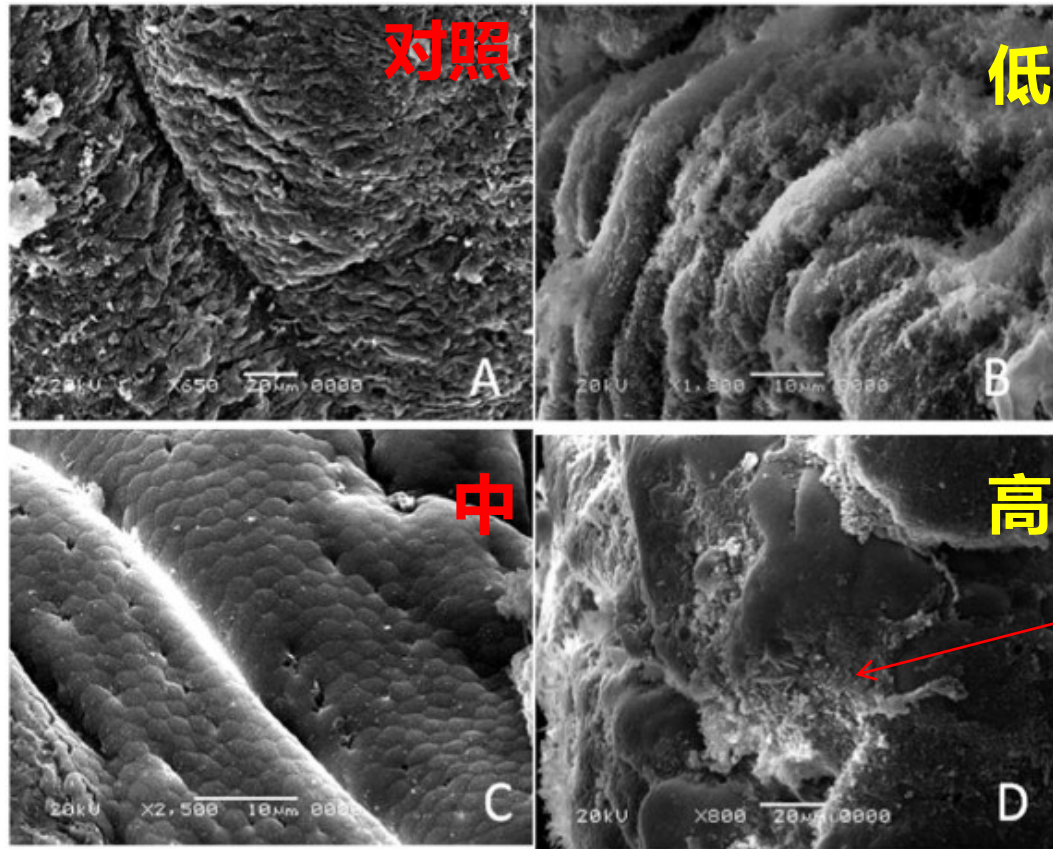


Fig. 6. Micrograph of the intestine of Nile tilapia at the control group (A), (B-D) obtained from fish received different concentrations of *Z. mauritiana* (ZLP). (A), showing normal and regular mucosal folds supported by oval or rounded CEC ($\times 650$); (B), intestine of fish received *Z. mauritiana* at 5 g/kg diet showing normal mucosal folds covered with excessive mucous ($\times 1000$); (C), intestine of fish received *Z. mauritiana* at 10 g/kg diet showing normal mucosal folds packed with oval or rounded CEC ($\times 500$); (D), intestine of fish received *Z. mauritiana* at 20 g/kg diet showing slight degeneration of CEC of mucosal folds covered with excessive mucous ($\times 800$).

酸枣叶粉可刺激
胃肠道中粘液分泌，
促进物质的消化吸收。

目录

01

研究背景

02

材料与amp;方法

03

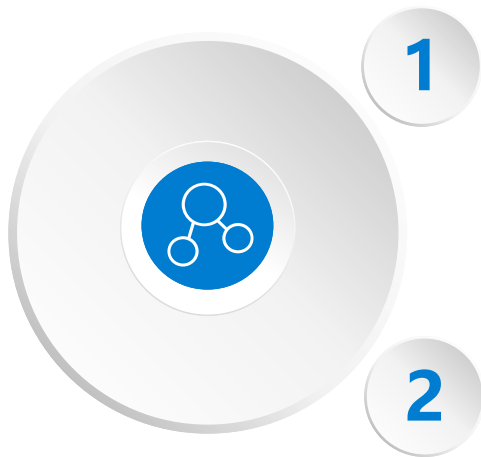
结果分析

→ 04

结论



结论



1
在饲料中添加酸枣叶粉，
可改善罗非鱼肠道健康。

2
在饲料中添加酸枣叶粉，
可促进罗非鱼生长。

谢 谢 聆 听

Thanks for your listening

