

**USING OF *YUCCA SCHIDIGERA* EXTRACT (YSE ) AGAINST  
AMMONIA TOXICITY AT DIFFERENT STOKING DENSITY FOR  
IMPROVEMENT GROWTH PERFORMANCE AND  
PHYSIOLOGICAL STATUS OF NILE TILAPIA  
(*OREOCHROMIS NILOTICUS*)**

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**Abstract**

This study was based on a 2×4 factorial design with or without *Yucca schidigera extract* (YSE) and four stoking densities T1=100, T2=100, T3=150, T4=200 and T5=250 fish/m<sup>3</sup>). In this study, Nile tilapia, *Oreochromis niloticus* (L.), (2 to 3g) was distributed into the fifteen aquaria at a rate of 10, 10, 15, 20 and 25 fish/100L according to design of experiment to five groups each group three replicate (The 1st group(T1) was contain 10 fish /100 L without *Yucca* and left as control. While 2<sup>nd</sup> group , 3<sup>rd</sup> group ,4<sup>th</sup> group and 5<sup>th</sup> groups (T2, T3, T4 and T5) at density 10, 15, 20 and 25 fish /100L of aquarium and received 10mg of *Y. schidigera* extract /100L) respectively. At the end of the experimental trial, blood samples were taken to determine the different physiological variables. The growth parameters were positively affected by YSE and inversely affected by stocking density. The high growth performance (WG, SGR and RGR) of tilapia, subject at yucca with 100 and 150 fish density, respectively than the control group (10 fish density without YSE). The best feed conversion ratio was obtained with yucca plus 10 fish density .Also The highest values of protein efficiency ratio and protein productive were obtained with yucca at stock densities of 100 and 150 fish/m<sup>3</sup>.The physiological variables including erythrocyte count (RBCs), haemoglobin content (Hb), haematocrit value (Hct) and activities of aspartate aminotransferase(AST), alanine aminotransferase (ALT), total protein, albumen ,total lipids, and glucose in plasma were significantly affected by yucca extract and/or rearing density. The overall results presented here indicate that the best growth performance of Nile

tilapia was obtained when the fish reared with yucca and at a stocking density of 100 or 150 fish/m<sup>3</sup>.

**Key words:** Density of *O. niloticus*, yucca, growth, physiological parameter.

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## INTRODUCTION

Aquaculture is the most developed sector in terms of the supply of high-quality food products that meet human nutritional requirements (Ottinger *et al.*, 2016). Pervasive population growth has led to increased demand for fish as food, which has affected wild fishery stocks and altered coastal environments, causing water pollution and habitat deterioration (Yoo and Bai, 2014) The expansion in aquaculture practices accompanied by intensification of aquaculture caused pollution that is directly linked to the elevated levels of fish stressors, both environmental and physiological.

Tilapias are the world's second most important fish species for aquaculture after the carp and this is due to their high growth rates, being prolific breeders, completing their life cycle in captivity, tolerance to environmental stress and high market demand (El-Sayed, 2002.).

The effect of population density is usually seen to be either density dependent or density independent (Wiener and Hameman, 1982). They suggested that stocking density that negatively affects fish growth is density dependent. Stocking density is an important parameter in fish culture as the health, growth and survival of fish depend upon this factor [Backiel and Lecren, 1978]. Higher stocking density reduces the growth and survival rates during fish culture [Shugunan, 1997]. Increasing stocking density results in stress (Leatherland and Cho, 1985). Hengsawat *et al.* (1997) which leads to enhanced energy requirements causing reduced growth and food utilization Stocking density is one of the most important factors in aquaculture because it directly influences Growth, survival behavior, health, feeding and production of fish under farmed conditions [Rahman and Rahman , 2003)

Water pollution is one of the most common issues encountered around the world in aquaculture by nitrogenous compounds on. The major hazardous

nitrogen metabolic product in aquaculture is ammonia, constituting about 70% of nitrogenous waste excretion in fish (Benli and KBksal, 2005). Due to those conceivable impacts on fish health and survival, ammonia accumulation is of specific concern in aquaculture (Evans *et al.*, 2006). Ammonia is the end product of the protein catabolism by living organisms, Total Ammonia Nitrogen (TAN) is a total result of NH<sub>3</sub> (non-ionized) and NH<sub>4</sub><sup>+</sup> (ionized). Only the NH<sub>3</sub> is considered the TAN's most toxic form as it excreted freely through fish gills (Silva *et al.*, 2013).

Besides affecting land usability that subsequently impacting on profitability of an aquaculture venture, stocking density is believed to affect growth performance and survival of fish species stocked. (Ugando *et al.*, 2014). They showed that a negative correlation between stocking density and growth rate was recorded. Survival was lowest with high stocking densities, 87% at 4000 fry/m<sup>3</sup> and 82.9 at 5330 fry/m<sup>3</sup>. in addition to determining the economic viability of production system in intensive aquaculture (Huang and Chiu, 1997). Stocking densities of Nile tilapia fry normally range from 3000 to 4000 fry/m<sup>3</sup>. However a stocking density as high as 20,000 fry/m<sup>3</sup> is also practical given good water quality, Food and Agricultural Organization (FAO, 2014).

Aquatic animals excrete ammonia as a product of protein metabolism, the mineralization of feed waste and organic nitrogen metabolism in fishes (Avnimelech and Ritvo, 2003). The main concern regarding excessive nitrogen loads in fish ponds is the transformation of ammonia to the toxic form (Páez-Osuna, 2001) Many plant products, in addition to being used for medicinal purposes, exhibiting anti-parasitic properties in large animals and fish ((Wang *et al.*, 2005), have been reported to arouse appetite, act as immune-stimulants and increase weight gain. *Yucca schidigera* extract was found to act as an anti-protozoa substance with the ability to trap ammonia and improve the feeding value of low-quality roughage (Cheok *et al.*, 2014). Direct application of yucca extract in rearing facilities or rations (Cheeke and Otero, 2005) has been used in the livestock (Holtshausen *et al.*, 2009).

Numerous studies have been carried out on the dynamic effect of yucca extract on various aquatic species, including common carp (*Cyprinus carpio*) (Francis *et al.*, 2002), Nile tilapia fingerlings (*Oreochromis niloticus*) (Gaber, 2006). Therefore, it is importance of optimize the stocking density for the target species in aquaculture for desired level of growth and production. Knowing the best densities for a species is a critical factor for good husbandry practices and creating efficient culture system. Considering this aspect the present study was conducted to investigation of the effect of *Y. schidigera* extract on ammonia reduction in aquaculture, survival, growth indies and physiological aspects in Nile tilapia (*Oreochromis niloticus*) at different densities.

### MATERIALS AND METHODS

The experimental work of this study was carried out indoor wet Lab. in Central Laboratory for Aquaculture Research, Abbassa , Abo-Hammed, Sharkia, Egypt. Average weight of fry Nile tilapia "*O. niloticus*" of average weight 2-3 gm were acclimated in laboratory conditions for 2 weeks before the begging of the experimental work. Fish were distributed in fifteen glass aquaria of about 100-liter capacity each and stocked at a rate of 10, 10, 15, 20 and 25fish/ aquarium. The glass aquaria were supplied with dechlorinated tap water and continuous aeration was adapted by using an air pump and air stones. Average water temperature was maintained at 27+ 2 C. The aquaria were divided into 5 groups with three replicates per group. The 1st group (T1) was kept as control and contain 10 fish/ aquarium without yucca. While The fish distributed in 2<sup>nd</sup> group , 3<sup>rd</sup> group ,4<sup>th</sup> group and 5<sup>th</sup> groups (T2, T3, T4 and T5) at 10, 15, 20 and 25 fish /aquarium and received 10mg of *Y. schidigera* extract /l ) respectively. These groups are illustrated in Tables (1). Fish of the experimental groups were fed on a pelleted fish diet containing 32 % CP. (Table 2) and the diet was fed at a rate of 3 % of live body weight twice daily for 90 days. Semi-dynamic method for removal of excreta was used every day by siphoning a portion of water from the aquarium and replacing it by an equal volume of water.

**Table 1.** Design of experimental.

Treatment	Yucca extract	Stoking density
T1	-	10 fish
T2	10mg/100L	10 fish
T3	10mg/100L	15 fish
T4	10mg/100L	20 fish
T5	10mg/100L	25 fish

**Table 2.** Chemical analysis of commercial diets, used in the experiment (on dry matter basis).

Item	Precent
Dry matter (DM %)	93.12
Crud protein (CP %)	29.65
Ether extract (EE %)	6.23
Crud fiber (CF %)	6.67
Ash	12.12
NFE	45.33
GE	412.7
Kcal / 100g	
P / E ratio	71.84

**Measurements of fish growth:**

At the end of the experimental period, the following growth and feed utilization indices were calculated: weight gain (WG), specific growth rate (SGR), food conversion ratio (FCR), feed efficiency (FE) and protein efficiency ratio (PER) using the following formulae: according to Jauncey and Rose (1982).

WG = Final average weight (g) – initial average weight (g);

SGR (% d – 1) =  $100 \times (\ln W_t - \ln W_0) / t$

Where  $W_t$  and  $W_0$  represent final and initial body weights of fish, respectively, and  $t$  represents the duration of the feeding trial;

FCR = Dry weight of feed (g) / wet weight gain by fish (g); PER = weight gain by fish (g) / protein intake (g), RGR = weight gain / initial weight, FER = weight gain / feed intake \* 100 and PPV = relative protein / protein intake \* 100

Where protein intake (g) = Protein (%) in feed  $\times$  total weight (g) of diet consumed / 100

**Physiological Analyses:**

Blood samples were taken from the caudal vein of no anaesthetized fish by sterile syringe containing EDTA as an anticoagulant. Erythrocyte count according to Dacie and Lewis (1984), hemoglobin content according to Van kampen (1961) and hematocrit value according to Britton (1963) were detected.

Plasma was obtained by centrifugation of the blood at 3000 rpm for 15 min and the non haemolyzed plasma was stored in a deep freezer at -20 °C till analysis. Plasma protein content was determined by Biuret method described by Wotton (1964). Glucose concentration was measured according to Trinder (1969) using Boehring Mannheim kits. Total lipids were determined calorimetrically using kits supplied by El Nasr Pharmaceutical Chemical Co. according to Joseph *et al.* (1972). Aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were determined colorimetrically using kits supplied by Diamond Diagnostics according to Reitman and Frankel (1957).

**Statistical analysis:**

One way analysis of variance (ANOVA) was conducted to test the effect of *yucca* extracts on fry Nile tilapia during different stoking density. This analysis was done using the computer program SPSS and least Significant difference (LSD) post hoc were done to determine significant differences (Tamhane and Dunlop, 2000).

**RESULTS AND DISCUSSION**

Water pollution is one of the most common issues encountered around the world in aquaculture by nitrogenous compounds. The major hazardous nitrogen metabolic product in aquaculture is ammonia, constituting about 70% of nitrogenous waste excretion in fish (Benli and KBksal, 2005).

In recent decades, many types of research have focused on studying the evaluation of the desert plant *Y. schidigera* with promising findings, such as the

aquaculture-associated reduction in ammonia levels by yucca extracts (Yu *et al.*, 2015; Hassan *et al.*, 2017 and Fayed *et al.*, 2019)

The effect of yucca extract in water at different stocking density fish to reduction ammonia and nitrogenous compounds was recorded in (Table 3) . The ammonia (NH<sub>3</sub>) concentration was decreased significantly in all treatment groups contain YSE in different stocking density fish (100, 150, 200, 250 fish/m<sup>3</sup>) at all periods when compared with control group (100 fish/m<sup>3</sup>). it is represented as significant decrease when compared to the control. The ammonia reduction might be the result of either the binding of ammonia with some fraction of *Y. schidigera* or of the transformation of ammonia to another compound. These results in agreement with those by Headon and Dawson (1990). The concentration of nitrite nitrogen was too low to be detected in the tanks during the experimental period. Ammonia concentration in all experimental tanks was kept at the same level). Similarly, (Hassan *et al.*, 2017) concluded that ammonia reduction in water by using *Yucca schidigera extract*. Amber *et al.* (2004) concluded that the dietary YSE decrease urea and ammonia levels in the blood and caeca and thus may be beneficial for improving the health, by reducing ammonia emission. Yucca Shidigera Extract (YSE) supplementation in fish diet has been proved effective reduction of ammonia in aquaculture and livestock industry (Yu *et al.*, 2015). (Khalil *et al.*, 2015) revealed that Yucca cause significant ( $p < 0.05$ ) decrease levels of ammonia (0.25, 0.5 gm\L) and nitrite in the aquaria water and increase in nitrate in water. Fayed *et al.* (2019) shows that exposure of water containing ammonia to yucca extract significantly reduces ammonia levels over time.

**Table 3.** The ammonium concentration in the water aquaria of Nile tilapia after rearing with 10 mg of *Y. schidigera* /100L and different density at different period.

Items	(different stoking density)				
	T1 ( 10 fsh)	T2 (10 fish +Y.S)	T3 (15 fish +YS)	T4 (20 fish+Y,S)	T5 (25 fish+Y,S)
After 2 week	0.009 ± 0.0007 <sup>a</sup>	0.0025 ± 0.0006 <sup>b</sup>	0.0050 ± 0.0012 <sup>b</sup>	0.0054 ± 0.0014 <sup>b</sup>	0.0035 ± 0.0003 <sup>b</sup>
After 4 week	0.0154 ± 0.0034 <sup>a</sup>	0.0029 ± 0.0006 <sup>b</sup>	0.0054 ± 0.0005 <sup>b</sup>	0.0046 ± 0.0010 <sup>b</sup>	0.0048 ± 0.0004 <sup>b</sup>
After 6 week	0.0577 ± 0.0172 <sup>a</sup>	0.0061 ± 0.0012 <sup>b</sup>	0.0069 ± 0.001 <sup>b</sup>	0.0132 ± 0.0030 <sup>b</sup>	0.0180 ± 0.0030 <sup>b</sup>
After 8 week	0.1133 ± 0.0173 <sup>a</sup>	0.0254 ± 0.0183 <sup>b</sup>	0.0082 ± 0.0005 <sup>b</sup>	0.0095 ± 0.0019 <sup>b</sup>	0.0139± 0.0011 <sup>b</sup>
After 10 week	0.1633 ± 0.0270 <sup>a</sup>	0.0069 ± 0.0013 <sup>b</sup>	0.0166 ± 0.0027 <sup>b</sup>	0.0100 ± 0.0020 <sup>b</sup>	0.014 ± 0.0017 <sup>b</sup>
After 12 week	0.1820 ± 0.0091 <sup>a</sup>	0.0073 ± 0.0009 <sup>d</sup>	0.0163 ± 0.0044 <sup>cd</sup>	0.0317 ± 0.0034 <sup>bc</sup>	0.0423 ± 0.0077 <sup>b</sup>

The same letter in the same row is not significantly different at P<0.05.

### Growth performance and feed utilization:

Results of growth performance are summarized in Table (4) after 90 days of raised fish treatment with *Y. schidigera* and with low density had a high significantly in final weight, weight gain, relative growth rate and specific growth rate than fish at same density and different levels of density with *Y. schidigera*. There were lower significantly in all parameter of growth performance of fish exposed to *Y. schidigera* with high stoking density (20 and 25 fish /100 L). Moreover, WG, ADG and SGR improved significantly with yucca . The survival (%) was up to 100% for in treatment with *Y. schidigera* and with two level of density (10 and 15/ fish / 100 L). However, these values were decreased significant in treatment (T4 and T5) with high density plus *Y. schidigera* when compared to the control group.

Fish growth and feed utilization were significantly retarded herein with increasing the rearing density. It has been demonstrated that rearing fish at high density (T2) may reduce their growth due to factors such as social interaction



and the deterioration of water quality, which can affect the feed utilization by fish (Ellis *et al.*, 2002).

These results are in agreement with that obtained by Zannatul (2014) who indicated that stocking density had a significant effect on growth and survival rates of monosex tilapia. Fry held at the highest density exhibited lowest growth and survival rates also, Costa (2017) showed that the increase in stocking density caused a decrease in the final weight of fish, weight gain, daily weight gain, standard length and survival, as well as an increase in feed conversion. However, higher densities seem to reduce the effect on weight variation. These results can be explained by the hypothesis supported by that a high density condition does not always result in increased fish stress: territorially, fish can reduce its own competitiveness assuming less injurious threat signals when submitted to great densities in stocking, as aggressive behaviors demand high energy cost and sometimes (depending on density) are ineffective. Similarly, Mansour *et al.* (2018) and Elkhayat *et al.* (2019) The effect of *Yuucca. Schidigera* extract dietary supplementation at different level (0, 0.25, 0.5 and 1 g kg<sup>-1</sup> diet) on growth performance of European sea bass *D. labrax* were investigated and the results showed an improvement of final weight by 8.16, 26.02 and 36.98% with 0.25, 0.5 and 1 g YE kg<sup>-1</sup>, respectively compared to the control. Also, growth performance of Nile tilapia, *O. niloticus*, improved significantly with feeding Y.E (El-Saidy and Gaber, 2004). High stocking density has been reported to cause growth inhibition of Amur sturgeon (Shi *et al.*, 2006) and other fish species due to crowding stress (Montero *et al.*, 1999) or the deterioration of water quality (Ellis *et al.*, 2002) or decreased food consumption (Suresh and Lin, 1992) Reductions in growth rate and food conversion efficiency of fish reared at high stocking density are attributed to an alteration in metabolism due to physiological stress (Lupatsch *et al.*, 2010)..In many cultivated fish species, growth and feed utilization are inversely related to rearing density, and this is mainly attributed to social interactions such as competition for food and/or space that can negatively affect fish growth (Irwin *et al.*, 1999). Similar results for Nile tilapia were obtained by (Ayyat *et al.*

(2011) who found that the increase of stocking density inversely affected the growth. Ridha (2006) reported that a density of 200 fish/m<sup>3</sup> decreased the growth performance significantly of Nile tilapia compared with a density of 125 fish/m<sup>3</sup>.

**Table 4.** Growth performance of Nile tilapia (*O.niloticus*) exposed 10 mg *Y. schidigera* /100L with different density .

Items Treatment	Initial weight (g)	Final weight (g)	Weight gain (g)	RGR	SGR(%day)	Survival rate
<b>T1 (10Fish)</b>	2.31 ± 0.032 <sup>a</sup>	25.46 ± 0.179 <sup>b</sup>	23.15 ±0.19 <sup>b</sup>	10.03 ± .20 <sup>b</sup>	4.29 ±0.07 <sup>ab</sup>	100.0 ± 0.0 <sup>a</sup>
<b>T2 (10 fish+ 10mgYSE/100L)</b>	2.32 ± 0.015 <sup>a</sup>	28.72 ±0.234 <sup>a</sup>	26.40± 0.003 <sup>a</sup>	11.38 ± .18 <sup>a</sup>	4.43 ± 0.04 <sup>a</sup>	100.0 ± 0.0 <sup>a</sup>
<b>T3 (15 fish+ 10mgYSE/100L)</b>	2.28 ± 0.017 <sup>a</sup>	25.10 ± 0.104 <sup>b</sup>	22.82 ± 0.10 <sup>b</sup>	10.01 ± .05 <sup>b</sup>	4.35 ±0.02 <sup>ab</sup>	100.0 ± 0.0 <sup>a</sup>
<b>T4 (20 fish+ 10mgYSE/100L)</b>	2.28 ± 0.018 <sup>a</sup>	22.91 ±0.12 <sup>c</sup>	20.63 ± 0.10 <sup>c</sup>	9.06 ± .03 <sup>c</sup>	4.23 ± 0.03 <sup>b</sup>	91.6 ±1.67 <sup>b</sup>
<b>T5 (25 fish+ 10mgYSE/100L)</b>	2.31 ± 0.023 <sup>a</sup>	21.43 ±0.18 <sup>d</sup>	19.12 ± 0.19 <sup>d</sup>	8.28± 0.15 <sup>d</sup>	4.07 ± 0.05 <sup>c</sup>	89.33 ±1.33 <sup>b</sup>

The same letter in the same column is not significantly different at P<0.

The results in Table (5) shows that feed conversion rate (FCR) was decreased significantly to  $1.68 \pm 0.02$  and  $1.71 \pm 0.02$  in *O. niloticus* rearing in 10ml yucca with different densities (T2= 10 and T3=15), respectively when compared to the control group (D1=10 fish without yucca,  $1.86 \pm 0.04$ ) and significantly different ( $P < 0.05$ ) from other treatments. However, in same Table (5) indicated that protein efficiency ratio (PER), feed efficiency ratio (FER) and protein production value (PPV) increased in fish treated with yucca and low density (T2= 10 and T3=15) when compared to the control. FCR decreased with administrated of YSE at high density fish. FER and PER are used to assess protein utilization and turnover. These results are in agreement with that obtained by El-saidy and gaber (2004) showed that the best feed conversion ratio (FCR) was achieved with Y750 fed groups. The protein efficiency ratio (PER) and feed efficiency ratio (FER) of the Y750 and Y1000 fed groups were significantly ( $P < 0.05$ ) higher than that of the control group. Also, Capeng *et al.* (2012) declines in serum concentrations of total T3 and free TH, as well as the reduction in food consumption coupled with the increase in

FCR, caused by high stocking density may have contributed to the growth inhibition .

**Table 5.** Feed intake, feed conversion ratio (FCR) , feed efficiency ratio (FER), protein efficiency ratio and protein production value (PPV) in Nile tilapia rearing at 10 mg *Y. schidigera* /100L with different density .

Items	Treatment (different density)				
	T1 (10 fish)	T2 (10 fish+Y.S)	T3 (15 fish +YS)	T4 (20 fish+Y,S)	T5 (25 fish+Y,S)
<b>Feed Intake</b>	43.05 ± 0.81 <sup>a</sup>	44.45 ± 1.03 <sup>a</sup>	39.10 ± 0.55 <sup>b</sup>	39.75 ± 0.98 <sup>b</sup>	37.38 ± 1.54 <sup>b</sup>
<b>FC</b>	1.86 ± 0.04 <sup>ab</sup>	1.68 ± 0.02 <sup>c</sup>	1.71 ± 0.02B <sup>c</sup>	1.93 ± 0.05 <sup>a</sup>	1.96 ± 0.09 <sup>a</sup>
<b>FER</b>	53.82 ± 1.27 <sup>b</sup> <sup>c</sup>	59.43 ± 0.83 <sup>a</sup>	58.38 ± 0.69A <sup>b</sup>	51.98 ± 1.43 <sup>c</sup>	51.34 ± 2.41 <sup>c</sup>
<b>PER</b>	1.82 ± 0.04 <sup>bc</sup>	2.00 ± 0.03 <sup>a</sup>	1.97 ± 0.02A <sup>b</sup>	1.75 ± 0.05 <sup>c</sup>	1.73± 0.08 <sup>c</sup>
<b>PPV</b>	15.15 ± 1.20 <sup>b</sup>	22.47 ± 0.84 <sup>a</sup>	19.04 ± 0.61 <sup>ab</sup>	14.49 ± 1.34 <sup>b</sup>	17.65 ± 1.5 <sup>b</sup>

The same letter in the same row is not significantly different at P<0.

In the present study , the results in Table (6) showed that protein content in fish body was significantly higher in fish subject with yucca at second group (D =10 fish), while the protein content at all other group of yucca was similar control group as similar Elkhayat *et al.* (2019).

Contrarily, total lipid content in fish body was decreased significantly (20.24± 0. 6) in fish subject in yucca at second treatment , while all other group contain of YSE with different density similar to control ones. Ash content was higher significantly in fish reared yucca and all different stoking density (all treatment ) when compared to the control group (Table 6). These results are in agreement with that obtained by El-saidy and gaber (2004) indicated hat proximate composition of whole body moisture, protein, and lipid and ash contents was significantly influenced by adding Yucca with different density.

**Table 6.** Chemical analysis of the experiment fish at the end of the experimental period (on dry matter basis).

Items	Initial	Stokig density fish				
		T1 ( 10 fsh)	T2 (10 fish+Y.S)	T3 (15 fish +YS)	T4 (20 fish+Y,S)	T5 (25 fish+Y,S)
DM	21.17 ± 0.35 <sup>d</sup>	23.52 ± 0.21 <sup>bc</sup>	25.03 ± 0.21 <sup>a</sup>	24.14 ± 0.11 <sup>b</sup>	23.42 ± 0.1 <sup>6c</sup>	23.72 ± 0.37 <sup>bc</sup>
CP	55.13 ± 0.06 <sup>e</sup>	57.83 ± 0.07 <sup>b</sup>	58.46 ± 0.15 <sup>a</sup>	57.49 ± 0.14 <sup>bc</sup>	57.10 ± 0.12 <sup>d</sup>	57.38 ± 0.15 <sup>cd</sup>
EE	16.75 ± 0.05 <sup>e</sup>	21.56 ± 0.19 <sup>a</sup>	20.24 ± 0.06 <sup>d</sup>	21.36 ± 0.09 <sup>ab</sup>	21.07 ± 0.12 <sup>bc</sup>	20.83 ± 0.10 <sup>c</sup>
ASH	29.12 ± 0.04 <sup>a</sup>	20.63 ± 0.26 <sup>c</sup>	21.26 ± 0.19 <sup>bc</sup>	21.17 ± 0.23 <sup>bc</sup>	21.79 ± 0.22 <sup>b</sup>	21.83 ± 0.26 <sup>b</sup>

The same letter in the same row is not significantly different at  $P < 0$ .

### Hematological parameters:

The results of erythrocyte count and hemoglobin content, hematocrit value obtained from fish exposure to yucca with four stoking density are given in (Table 7). This Table shows that the fish exposed to yucca extract in water at 10 and 15 fish density/100L capacity caused increased significantly to  $(2.37 \pm 0.044$  and  $2.27 \pm 0.044 \times 10^6/\text{mm}^3$  erythrocyte,  $8.36 \pm 0.133$  and  $8.25 \pm 0.058$ , g/100ml in Hb and  $28.17 \pm 0.105$  and  $28.09 \pm 0.206\%$  of Hct) in all blood parameter examined respectively as compared of the control values ( $2.07 \pm 0.03$ ,  $7.43 \pm 0.248$  and  $27.44 \pm 0.31$ ). In this study, the RBC and WBC counts, which are indicators of hematopoiesis, showed that yucca extract supplementation in water greatly enhanced the blood cell count along with the Hct percentage. Similar results were observed in previous studies by Güroy *et al.* (2014) reported that the growth and hematological responses of striped catfish juveniles (*P. hypophthalmus*) were greatly enhanced, and ammonia levels were considerably decreased by high levels of yucca extract. The improved hematological and immune responses of *D. labrax* juveniles are consistent with the superior growth performance observed in the present study. Many studies have shown that addition of yucca extract to diets or water significantly increased the growth proportion of channel catfish juveniles (*Ictalurus punctatus*) (Kelly and Kohler, 2003).

While, this values in all blood parameter were decrease in fish exposed to (YES) with high level of density (200 and 250 fish/ m<sup>3</sup>) .In vertebrates including fish, blood is the most frequently examined tissue in efforts to establish their health status or physiological status. Accordingly, health status such as oxygen carrying capacity has been directly determined by reference to main hematological indices including red blood cell (RBC), hemoglobin concentration(Hb), percentage of blood volume consisting of red cells and hematocrit (Hct) (Houston, 1990).

**Table 7.** Changes in erythrocyte (count x 10<sup>6</sup>/mm<sup>3</sup>), hemoglobin content (g/100ml) and hematocrit value (%) in the blood of Nile tilapia (*O. niloticus*) reared to 10mg *Y. schidigera* /100L with different density.

Items	Treatment (different density)				
	T1 (10 fish)	T2 (10 fish +Y.S)	T3 (15 fish +Y.S)	T4 (20 fish+Y,S)	T5 (25 fish+Y,S)
<b>Erythrocyte count (RBCs)</b>	2.07 ± 0.030 <sup>b</sup>	2.37 ± 0.044 <sup>a</sup>	2.27 ± 0.044 <sup>b</sup>	1.99 ± 0.020 <sup>bc</sup>	1.88 ± 0.020 <sup>c</sup>
<b>Hemoglobin (Hb)</b>	7.43 ± 0.248 <sup>b</sup>	8.36 ± 0.133 <sup>a</sup>	8.25 ± 0.058 <sup>a</sup>	7.23 ± 0.064 <sup>bc</sup>	6.99 ± 0.081 <sup>c</sup>
<b>hematocrit value</b>	27.94 ± 0.315 <sup>b</sup>	28.17 ± 0.105 <sup>d</sup>	28.09 ± 0.206 <sup>c</sup>	27.33 ± 0.130 <sup>bc</sup>	25.51 ± 0.205 <sup>a</sup>

The same letter in the same row is not significantly different at P<0.

### Biochemical parameter:

The glucose concentration in the blood in Nile tilapia was decreased significantly to (50.39± 0.165 mg/ L) in fish reared with yuacc (2<sup>nd</sup> group) as compared to the control. While, it values were increased significantly to (54.63 ±0.197 and 55.22± 0.208 mg/ L) in fish rearing with yucca extract in water and high stoking density (4and5 groups) respectively when compared to the control group (53.08 ±0.120 mg/ L (Table 8). This result suggests that high stocking density may cause stress. The primary response against stress involves the increases in plasma catechol amines and cortisol. These hormones induce secondary stress responses, characterized by increased glucose levels, mobilizing glucose to tissues for homeostasis to cope with energy-demanding processes of restoration (Barton, 2002). Stress in fish is indicated by elevated

blood glucose, cortisol levels, and indeed, a high stocking density typically leads to increased cortisol levels in a variety of fish.

The mean values of plasma total protein and albumin in *O. niloticus* after reared at different stocking density with yucca extract in water for 90 days are shown in (Table 8). It can be observed that the total protein and albumen were significantly increased after exposed to yucca extract with low density ( T2 and T3) when compared the control group. On other hand, it values were decreased significantly to (  $6.44 \pm 0.256$  and  $5.76 \pm 0.235$ , protein and  $1.76 \pm 0.035$  and  $1.66 \pm 0.044$ , albumin in fish at high density with yucca (T4 and T5) respectively as compared to the control one ( $7.36 \pm 0.13$  and  $1.84 \pm 0.06$ ). The most portion of serum synthesizes in the liver and it can be used as an indicator of liver dysfunction. The reduction in the total protein concentration is the obvious feature of many diseases and may occur due to liver disease, the absorption reduction or the loss of protein (Bernet *et al.*, 2001). The concentration of total protein in blood serum is used as a basic index for the health status of brood fish (Rehulka, 1996). In our study, total protein of serum was significantly enhanced in all experiments compared to control ( $P < 0.05$ ) which agrees with findings of Abdel-Zaher *et al.* (2009). Nayak *et al.* (2004) reported that the increase in serum total protein indicates that fish are immunologically strong. Fayed *et al.* (2019) noticed that the total serum protein, albumin, globulin, and lysozyme levels were significantly higher ( $P < 0.05$ ) among plasma samples of seabass (*Dicentrarchus labrax*) treated with yucca extract in water. These results indicate that extracts from yucca alleviate the effects of environmental stressors on fish, triggering growth and improving physiological parameters.

Hepatic enzymes activities of plasma Nile tilapia were shown in Table (8). Plasma aspartate amino transferase (AST) and alanine aminotransferase (ALT) activities indicate the level of liver damage and were significantly affected by the experimental levels of *Y. schidigera* liquid extract (Table 8). The results showed that AST and S. ALT activities significantly decreased in the groups treated with *Y. schidigera* liquid extract in water with low fish

density (T2 and T3) compared to the standard group. In the present study, AST and ALT activities considerably decreased in groups treated with yucca extract in water compared with the control group, which could be attributed to the effectiveness of yucca extract in the improvement of liver health. These results are consistent with those of (Haridas *et al.*, 2001), who showed that the active component of yucca extract reduced the stress conditions for the fish, triggering serum enzyme activity for improvement of liver health. While these values were increased significantly in fish treated with yucca with high density (T4 and T5) as compared to the control. S. ALT and S. AST, present mainly in cardiomyocytes and hepatocytes of fish, respectively, are necessary for protein metabolism. Damaged or increased permeability of liver and myocardial cells leads to increased levels of S. AST and S. ALT, resulting in elevated blood transaminase activity. Therefore, the welfare of fish can be monitored by detecting the activities of S. AST and S. ALT (Wang *et al.*, 2005).

**Table 8.** Changes in glucose concentration, total lipid, total protein albumin content, aspartate amino transferase (AST) and alanine amino transferase activities (ALT, IU/L), in the blood of Nile tilapia (*O.niloticus*) reared to 10mg *Y. schidigera* /100L with different density.

Items	Treatment (different density)				
	T1 (10 fish)	T2 (10 fish +Y,S)	T3 (15 fish +YS)	T4 (20 fish+Y,S)	T5 (25 fish+Y,S)
<b>Glucose</b>	53.08 ±0.120 <sup>c</sup>	50.39 ±0.165 <sup>d</sup>	53.45 ±0.156 <sup>c</sup>	54.63 ±0.197 <sup>b</sup>	55.22 ±0.208 <sup>a</sup>
<b>T. Protein</b>	7.36 ±0.136 <sup>b</sup>	8.32 ±0.152 <sup>a</sup>	8.12 ±0.110 <sup>a</sup>	6.44 ±0.256 <sup>c</sup>	5.76 ±0.235 <sup>d</sup>
<b>Albumin</b>	1.84 ±0.06 <sup>b</sup>	2.27 ±0.028 <sup>a</sup>	2.06 ±0.084 <sup>a</sup>	1.76 ±0.035 <sup>bc</sup>	1.66 ±0.044 <sup>c</sup>
<b>AST</b>	52.07 ±0.429 <sup>b</sup>	48.16 ±0.106 <sup>d</sup>	50.92 ±0.385 <sup>c</sup>	52.60 ±0.179 <sup>b</sup>	54.12 ±0.364 <sup>a</sup>
<b>ALT</b>	53.72 ±0.128 <sup>c</sup>	50.47 ±0.246 <sup>d</sup>	51.23 ±0.460 <sup>d</sup>	54.27 ±0.196 <sup>b</sup>	55.60 ±0.246 <sup>a</sup>
<b>T. Lipid</b>	5.35 ±0.145 <sup>bc</sup>	5.97 ±0.081 <sup>a</sup>	5.59 ±0.232 <sup>ab</sup>	5.11 ±0.107 <sup>c</sup>	5.29 ±0.076 <sup>c</sup>

The same letter in the same row is not significantly different at  $P < 0$ .

Contrarily, in Table (8) the total lipids in plasma of Nile tilapia was decreased non significantly to  $(5.11 \pm 0.107$  and  $5.29 \pm 0.076$ mg/L in exposure

fish to yucca with high density (T4 and T5) when compared to the control group ( $5.35 \pm 0.145$ ).

The results of challenge test (Table 9) revealed that mortality rate was 6.67, 13.33, 33.33 and 40 % in fish rearing at different stoking density respectively with yucca extract. The mortality rate of control group was 55.33%. the addition of yucca extract at different density in water showed the same effect on mortality rate of *O. niloticus* challenged with *A. hydrophila*. which illustrated the yucca had antibacterial activity antagonized *A. hydrophila* in fresh water as Shalaby *et al.* (2006).

**Table 9.** Challenge test of *A. hydrophila* inject I/P and pattern of mortality among *O. niloticus*) reared to *Y. schidigera* /100L with different density.

Items	Treatment (different density)				
	T1 (10 fsh)	T2 (10 fish+Y.S)	T3 (15 fish +YS)	T4 (20 fish+Y,S)	T5 (25 fish+Y,S)
No. injection fish	15	15	15	15	15
Bacteria dose ( $5 \times 10^5$ ) cfu	0.3 ml	0.3 ml	0.3 ml	0.3 ml	0.3 ml
Mortality No.	7	1	2	5	6
Mortality rate	46.67	6.67	13.33	33.33	40
Survival rate	53.33	93.33	86.67	66.67	60

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## استخدام مستخلص نبات اليبوكا ضد سمية الأمونيا الناتجة عن كثافة الأسماك المختلفة لتحسين معدلات النمو والحالة الفسيولوجية لاسماك البلطي النيلي

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### الملخص العربي

تمت الدراسة لتأثير اضافة ١٠ ملجرام من نبات اليبوكا في مياة المحتوية على كثافات من الأسماك المختلفة لاختزال الأمونيا وتأثيرها على اداء النمو وبعض التغيرات الفسيولوجية لاصباغيات البلطي النيلي المرباة باحواض زجاجية والتي تتراوح من ٢-٣ جم/سمكة، حيث تم توزيع الأسماك على ١٥ حوض زجاجي سعة ١٠٠ لتر بكثافات مختلفة هي (١٠، ١٥، ٢٠، ٢٥) سمكة / ١٠٠ لتر وتم تقسيمها الى خمسة مجموعات كالتالي: المجموعة الأولى تحتوى بمعدل ١٠ سمكة/١٠٠ لتر وبدون اى اضافات، وتركت كمجموعة ضابطة. بينما المجموعة الثانية، الثالثة، الرابعة، الخامسة تحتوى على نسبة اسماك التالية ١٠، ١٥، ٢٠، ٢٥ سمكة / ١٠٠ لتر على التوالي كما تحتوى كل منها على ١٠ مجم من مستخلص اليبوكا. وقد غذت المجموعات المختلفة على علائق اسماك متزنة تحتوى على ٣٢ % بروتين لمدة ٩٠ يوما.

اوضحت الدراسة: تحسين معدلات النمو ومعدل التحول الغذائى وكفاءة التحويل البروتينى باسماك المجموعات التى تحتوى على اليبوكا وكثافة منخفضة ومتوسطة (١٠، ١٥ سمكة/١٠٠ لتر) بينما حدث انخفاض بالمجموعات الأخرى.

كما لوحظ اختلاف فى بعض معايير الدم بشكل ملحوظ حيث اظهرت النتائج تأثير ذا دلالة على كرات الدم الحمراء ومحتوى الهيموجلوبيني والهيماتوكريت بالمجموعات المحتوية على كثافات منخفضة ومضاف اليها اليبوكا. بينما حدث انخفاض معنوي فى جلوكوز الدم والأنزيمات الكبدية (ALT , AST) بالمجموعات سابقا الذكر وتحسين محتوى البروتين فى دم اسماك البلطي المعرضة لليوكا مع الكثافة المنخفضة. كما ان هذه الأسماك التى تم تربيتها مع مستخلص نبات اليبوكا اصبحت مقاومة للأمراض البكتيرية ( الأيرومونس هيدروفيليا).

من هذه الدراسة يمكن اضافة تركيزات اعلى من الجرعة التى تم الدراسة عليها من مستخلص نبات اليبوكا بالأستزراع السمكى المكثف لقدرتها على التخلص من الامونيا الذائبة فى المياة وتحسين معدلات النمو والحالة الفسيولوجية للاسماك.