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Effect of some natural by-products on fish performance, health, and physiological parameters

Nasreen M. Abdulrahman1*, Omed I. Abid2, Nasrin H. Aziz3, Tabin T. Awrhaman4, Przha A. Omar4
[1]College of Veterinary Medicine/ University of Sulaimani/ Sulaimaniya/ Iraq,
[2]Sulaimani veterinary directorate, Sulaimani/Iraq
[3]College of Agricultural sciences/ University of Sulaimani/ Sulaimaniya/ Iraq
[4]College of Education/ University of Sulaimani/ Sulaimaniya/ Iraq
Corresponding Author* : nasreen.abdulrahman@univsul.edu.iq

Abstract:
This experiment was conducted at the zoology research Laboratory, Biology department, College of Education/ University of Sulaimani/ Sulaimaniya/ Iraq. 150 common carp individuals used with an average body weight between 52.55 g were used. The duration of the study was eight weeks. A totally randomized design was organized into seven groups and two replicates were organized. Each group, containing seven individuals, receives different treatments, as follows: T1 No supplements as control, T2 with 5 g/kg carrot juice by-products powder, T3 with 10 g/kg carrot juice by-products powder, T4 with 5 g/kg cucumber by- products powder, T5 with 10 g/kg cucumber by- products powder, T6 with 5 g/kg orange juice by- products powder, and T7 with 10 g/kg orange juice by- products powder. Each tank was stocked with seven individuals assigned randomly, for reducing disparities between treatments. During the first week, fish from each tank fed in quantities corresponding to 3% of their body weight, twice a day, and they weighed twice a week. The feeding levels were then recalculated using the new weights. The feeding experiment lasted eight weeks. The protein ratio was 28% and the fat was 2.98. The carp growth parameters, hepatosomatic, Spleenosomatic, Kidneysomatic indices, MCV, MCH, MCHC, PLT, and MPV were higher significantly in G7 with 10 gm orange juice by-product, Carp in G7 with 10 gm orange juice by-product ate little than other groups in a mean of feed conversion ratio (FCR) and fat efficiency ratio, the feed efficiency ratio was higher in group 2 and 3 in adding the Carrot juice. The addition of Carrot juice by-product in both levels increased the Gillsomatic Index. Cucumber peel in both levels increased the Intestine Length, Intestine Length/ Length, Intestine Weight Indices, and Condition Factor. Orange juice by-product increased the Fish meat indices in weight without Head and without Head & Viscera Indices in both levels, WBC counts, Lymphocytes, and Monocytes percent. The Carrot juice by-product increases the Granulocytes percent in both levels. The control group was higher significantly in each of HGB, RBC, and HCT.

Keywords: Carrot juice, Orange juice, Cucumber peel, common carp, health, performance, physiological parameters

Introduction:
Fruit and plant fibers, which are by-products of the fruit juice industry, have gained popularity because they can be valued as food ingredients. Bioactive compounds have been studied in this regard, but little attention has been paid to their remaining volatile components. Today, the concept of nutrition has shifted from foods that provide energy to our bodies to diets that provide physiological benefits in treating and preventing disease. Ensuring well-being for the consumer requires a balanced dietary intake for the

metabolism of the human body, to prevent deficiency or excess of certain components, and the idea of functional food preceded the conception mentioned above (Rezzadoriet al., 2012, Rafiq et al., 2016).

Peels make up between 50-65% of the total weight of the fruit and are left as the primary by-product, if not further processed they get wet, create odor, soil pollution and shelter for insects and can cause serious environmental pollution (Wannissornaet al., 2005 and Mandalariet al., 2006).

Sweet orange (Citrus sinensis) is one of the most important and oldest horticultural products in many tropical and subtropical regions (Pourhossein et al., 2015). Orange is a citrus fruit that is consumed in large quantities around the world in peeled form and as juice. During the production of orange juice, large amounts of residues (peel, pulp, seeds, orange leaves and whole orange fruits) that do not meet the quality requirements are generated as waste (Abdel-Moneimet al., 2014). Orange peel is a primary by-product of the fruit processing industry. Agricultural waste is the most abundant and renewable material produced on earth. Agricultural and industrial wastes are among the causes of environmental pollution. Converting them into useful products can alleviate the problems they cause (Olaniyi, 2014).

The amount of world orange production is about 69 million tons; that is 57% of the total citrus production. About 31% of the world's orange production is processed (FAO, 2016). Orange creates 55-60% peel waste in the processing phase. This means that around 10-15 million tons of waste are generated. Orange peel essential oil has many advantages for animal feed use, due to high D-limonene content (84-96%), sustainable feed, inexpensive raw material, recycling of waste products and direct integration into animal feed production (Gültepe2018).

Several researchers reported that OEO has antimicrobial and antifungal effects (Dambolena et al., 2008; Chee et al., 2009). In addition, the positive effects of essential oils extracted from orange peels on growth and immune system for tilapia were determined (Acar et al., 2015).

According to results of Gültepe(2018) WG, SGR and PER values of 1‰ OEO fed group were significantly increase than the all groups ($P<0.05$). Effect of feeding with OEO added was started from the second week onwards on the WG. FCR value of 1‰ OEO fed group was significantly decrease than other groups ($P<0.05$) FCR values of Control, 0.05‰, 1‰ and 3‰ OEO groups were found that 1.46±0.06, 1.16±0.03, 1.06±0.02 and 1.25±0.03, respectively. Similarly, Acar et al. (2015) found that 1 OEO addition of fed tilapia significantly affected WG, FCR, and SGR scores of tilapias.

There are insufficient studies on the effects of OEO on animal growth parameters. Carrot (Daucus carota L.), one of the most common vegetables in human nutrition, was chosen as the vehicle because it is rich in beta-carotene, ascorbic acid and tocopherol and is classified as a vitaminized food. Carrots are a good source of carbohydrates, calcium, phosphorus, iron, potassium, magnesium, copper, manganese, and sulfur, but are lacking in protein and fat.An increased intake of carrots can favor the massive synthesis of vitamin A, since it has been reported that 100 g of carrots contain 6-15 mg of carotenoids, mainly -carotene (2-10 mg). The presence of these carotenoids and other antioxidants may protect people against certain types of cancer and cardiovascular disease, and boost the immune system, as well as protect against stroke, high blood pressure, osteoporosis, cataracts, arthritis, heart disease, bronchial asthma, and urinary tract infections(Aly et al., 2004).

In addition, the allergenic effect of carrot is very low or absent and fermentation makes it more suitable by removing any anti-nutritional factors that may be present. Therefore, carrot can be consumed by people who are intolerant to dairy products (Szilárd et al., 2008, Aliyev et al., 2021).

The volatile fraction of carrot fiber contained terpenoids (35.3%) as the main compound group. Other studies have reported that these compounds make up 97% of the total volatile fraction of fresh carrot samples; The lower percentage found in the sample analyzed could be explained by the loss of volatiles during the washing and drying treatment applied during industrial fiber processing. The most abundant constituents of carrot fiber were - and -ionone at 8.1 and 9.8%, respectively. The link between processing-induced carotenoid degradation and the production of degrading terpenes such as ionones has been described (Marsol-Vall, et al., 2021).

The result of Rafiq et al., (2016) makes carrot juice a promising vehicle for probiotic bacteria and can serve as an alternative for people who cannot consume probiotic dairy products due to allergic reactions and lactose intolerance. Further research is needed to improve the taste, storage stability and packaging of such probiotic juices.

Feed costs are the most important part of aquaculture production, accounting for 40-60% of operating costs. The decline in natural fish stocks and the consequent increase in the cost of fishmeal has prompted feed manufacturers and researchers to look for new and/or alternative raw materials for fish feed production, so the aim of this study is to evaluate the respective waste of oranges, cucumbers and carrots in aquaculture. The second objective is to assess the impact of these bowls on the performance and health aspects of carp.

Materials and Methods

This experiment was conducted at the Zoological Research Laboratory, Department of Biology, College of Education/University of Sulaimani. 150 common carp individuals with an average body weight between 52.55 g were used. They were fed commercial pellets and acclimated for about 27 days prior to conducting the feeding experiment. The duration of the study was eight weeks.A fully randomized design organized into seven groups and two replicates. Each group, consisting of seven people, receives different treatments, as follows: T1 No supplements as control, T2 with 5 g/kg carrot juice by- products powder,T3 with 10 g/kg carrot juice by- products powder,T4 with 5 g/kg cucumber by-products powder,T5 with 10 g/kg cucumber by- products powder,T6 with 5 g/kg orange juice by-products powder,T7 with 10 g/kg orange juice by- products powder.

Fourteen plastic tanks (70L) were used for the development of the study, which involves testing seven treatments performed in two replicates. A Hailea ACO-318 air compressor (power: 45 watts, air flow: 75 l/min) provided adequate continuous ventilation to each tank. Each tank is staffed with seven people who are randomly assigned to reduce differences between treatments. Daily cleaning with the pumping method performed to remove the waste from the system.

The experimental ratio included common feeds available in Sulaymaniyah municipal markets, as well as carrot, orange juice and cucumber by-products. The pellets were treated and dried at room temperature for four days before being ground into tiny particles using Kenwood multiprocessors. During the first week, fish from each tank were fed twice daily at 9:00 am and 2:00 pm at an amount equal to 3% of their body weight and weighed twice weekly. The feeding amounts were then recalculated using the new weights. The feeding trial lasted eight weeks. Protein ration 28% and fat 2.98.

Growth and feed utilization parameters.

Weight gain and daily weight gain calculated using the following equations:

1. Weight gain (g /fish) = W2–W1

   Where W_1: Fish weight (g) at the beginning of the experimental period and W_2: Fish weight (g) at the end of the experimental period.

2. Daily weight gain (DWG) (g/day) = Weight gain/ Experimental period, = W2–W_1/ T

   Where T: time between W2 and W1 (70 days).

Relative growth rate calculated according to the followings:

Relative growth rate (RGR %) = Weight gain/Initial weight x 100 = W2–W_1/ W_1 x 100

- Specific growth rate calculated according to the method described by Abdulrahman et al., (2018) as follows:

  Specific growth rate (SGR) % = (Ln W2–Ln W1)/ T x 100

Where:Ln W2 - final body weight; Ln W1initial body weight); T - experimental period

- Feed conversion ratio calculated as follows:

  Feed conversion ratio (FCR) = Total feed fed (gm.)/ Total wet weight gain (g).

- Feed efficiency ratio calculated as previously described by Uten (1978) as follows:

  Feed efficiency ratio (FER) = Total weight gain (g)/Total feed administered (g)

- Protein efficiency ratio calculated as follows:
  Protein efficiency ratio (PER) = Total wet weight gain (g/fish)/amount of protein administered (g/fish).

Biological indices: At the end of the experiment, five fish from each tank are randomly selected and anesthetized with clove powder (Hassan et al., 2016). After determining the weight and length of each fish, the fish were dissected and the liver, spleen, gills, viscera, kidney and intestine were weighed.The following organ-somatic indices were calculated

- Fulton condition factor (K) = 100 (fish weight, g)/ (fish length, cm)^3 (Lagler, 1956)
- Hepatic somatic index (HSI, %) = 100 (liver weight (g) / fish weight (g). (Lagler, 1956)
- Gills somatic index (GSI, %) = 100 (gills weight (g) / fish weight (g) (Abdulrahman et al., 2018)
- Kidney somatic index (KSI, %) = 100 (kidney weight (g) / fish weight (g) (Abdulrahman et al., 2018)
- Spleen somatic index (SSI, %) = 100 (spleen weight (g) / fish weight (g) (Abdulrahman et al., 2018)
- Intestine Length index (ISI, %) = 100 (Intestine length (cm.) / fish weight (g) (Abdulrahman et al., 2018)
- Intestine Length index (ISI, %) = 100 (Intestine length (cm.) / fish length (cm.)(Abdulrahman et al., 2018)
- Intestine weight index (IWI, %) = 100 (Intestine weight (g) / fish weight (g) (Abdulrahman et al., 2018)

Blood examination: Three fish from each experimental group that were withdrawn at the end of the trail. Aspiration of the caudal peduncle to obtain blood samples. The BC-2800 Complete Blood Count (CBC) Hematology Analyzer. It is a small, fully automated hematology analyzer made in the USA. Whole blood samples in heparinized vials for assessment of various blood indices.

Complete Blood Count: Erythrocyte (red blood cells recommended) count (RBC x 10^{12} cells/L), mean corpuscular hemoglobin (MCH, pg), mean corpuscular hemoglobin concentration (MCHC, g/dL), mean corpuscular volume (MCV, fL), hemoglobin (Hb, g/L) and platelets (PLT x 10^9 cells/ L. Leukocyte White Blood Cells (WBC x 10^9 cells/L), granulocytes (%), lymphocytes (%) and monocytes (%).

Biochemical Parameters: Alanine aminotransferase activity (ALT), aspartate aminotransferase activity (AST, units/L), Cytokines (CKI, units/mg), total proteins (g/dL), globulin (g/dL), albumin (g/dL) and albumin/ globulin (A/ G) ratio.

Data analysis and interpretation: Data subjected to analysis of variance (one-way ANOVA), using the General Linear Model procedure of XLSTAT 2016 Version 02.28451. Differences between means compared by Duncan's multiple range test and at $P<0.05$.

Results and Discussion

Table (1) shows the effect of different concentrations of some by-products on carp performance. Carp growth parameters were significantly higher in G7 with 10g of orange juice as a by-product. Carp in G7 with 10 g orange juice as a by-product ate less than other groups on average feed conversion rate (FCR) and fat efficiency rate, the feed efficiency rate was higher in groups 2 and 3 with the addition of carrot juice

The addition of 10 g orange juice as a product in G7 improves the hepatosomatic, spleenosomatic and renal somatic indices. The addition of carrot juice as a product at both levels increased the Gillsomatic Index as shown in Table (2). Product orange juice increased fish meat indices in weight without head and without head & viscera indices at both levels.

Table (3) show that Cucumber peel in both levels increased the Intestine Length, Intestine Length/ Length, Intestine Weight Indices and Condition Factor.

Table (4) shows that the addition of orange juice as a product increased WBC counts as well as lymphocyte and monocyte percentages. The by-product carrot juice increases the percentage of granulocytes in both levels.

The control group was higher significantly in each of HGB, RBC and HCT. The Group 7 with 10 gm orange juice by product increased each of MCV, MCH, MCHC,PLT and MPV as seen in table (5).

Lymphocytes and neutrophils are commonly used in studies evaluating the overall immune response due to ease of sample collection and cost-effectiveness. Lymphocytes are important white blood cells involved in the coordination of specific and non-specific immune components, and their abundance in the blood is usually a long-term (chronic) response, and this could be the reason for the improvement of some bold parameters in the recent study, as in Table (6 and 7).

The result of Nader and Abdulrahman (2017) showed that the red blood cell (RBC) data of the control group were significantly higher than other treatments. The hemoglobin of the fish in the 2.5 and 10 g/kg diet groups differed significantly from other treatments. Mean corpuscular hemoglobin (MCH) values were significantly different, resulting in mean corpuscular volume (MCV) values that showed significant differences, and these contradicted the results of a recent study comparing all by-product additives increased to the control group. White blood cell (WBC) counts were significantly different. The granulocyte percentages obtained in all treatments differed significantly from the control. Lymphocyte percentages were significantly different in all treatments compared to the 10 g/kg diet group. Monocytes were significantly different in the 2.5, 7.5 and 10 g/kg groups and these are consistent with recent results.

Abdulrahman et al., (2013) showed that the inclusion of grape seed in fish feed at 8 g/kg significantly affects the RBC ratio compared to other treatments, and this contradicts the recent study results, while the platelet ratio of the control was significantly higher than in inclusion treatments. WBC, HGB and HCT were unaffected by the treatments and are inconsistent with recent results.

The statistical analysis by Rafiq et al., (2016) showed that the optimal pH for the production of probiotic carrot juice was 6 and the optimal fermentation temperature was 30 °C. During the study of growth kinetics, a gradual change in pH, acidity and sugar concentration was observed, indicating the growth of probiotics and the production of lactic acid by them. The results suggest that fermented carrot juice may serve as a suitable medium for the growth of probiotics and this may be the result of the effect of carrot juice by-products on some immunity parameters such as leukocyte count and some differential counts.As a mixed culture of Lactobacillus acidophillus, Lactobacillus Plantarium, Lactobacillus casei and Bifidumlongum, it ferments well and survives in carrot juice under optimal conditions. The effect of probiotic strains on the immediate composition of fresh carrot juice was not found to be significant and the strains were able to be viable in carrot juice without any nutrient supplementation. This finding makes carrot juice a promising vehicle for probiotic bacteria and may serve as an alternative for individuals who cannot consume probiotic dairy products due to allergic reactions and lactose intolerance (Duda-Chodak et al., 2015; Rafiq et al., 2017).

Gültepe(2018) studied the effect of orange peel and found that the feed conversion ratio is significantly reduced, the relative growth rate, specific growth rate and protein efficiency ratio are significantly increased in the 1-rate group of orange peel essential oil supplements ($p<0, 05$). and this is consistent with the results of the most recent study. Several researchers reported that orange peel oils have antimicrobial and antifungal effects (Sharma and Tripati, 2006; Chee et al., 2009). Furthermore, the beneficial effects of orange peel essential oils on growth and immune system in tilapia (Acar et al., 2015) and rainbow trout (Gültepe 2018) were determined.

Al-Ashaab et al., (2017) examine different levels of natural aniseed, cinnamon and found that the aniseed and cinnamon levels of T3 (1% cinnamon) and T8 (2.5% aniseed + 1% cinnamon) were the best Results provided growth performance, so they advise using the above lineage in the diet of carp.

According to Marsol-Vall et al., (2017), orange and tangerine flesh samples had almost the same volatile composition and again showed a profile clearly dominated by terpenes (99.4 and 98.9% of total volatiles with a high Predominance of limonene 92.3 and 94.0%, respectively). The same behavior was observed with orange and tangerine peel fibers, which had the same individual volatile components and similar percentages of the same, and this could be the reason for the higher appetite of fish during the recent study, which gave a higher result compared to other by-products needed. And based on the total area, the residual volatile content in orange peel was higher than in the corresponding pulp, the latter being significantly higher than in lemon. a greater content of volatile compounds, especially limonene, in orange peel compared to orange flesh (Ferreira, 2009).

HS-SPME-GC-MS used to study the volatile composition of orange peel, and carrot, derived from the juice industry. Although they undergo processing that includes washing and drying, among others, and shows that the volatiles remaining in the fibers cannot be neglected. In this regard, citrus fiber contained a large amount of volatile compounds, mainly monoterpenoids (limonene). Processing to obtain fruit fiber has been shown to produce fiber with low volatile content for non-citrus products. Otherwise, the analyzed citrus fibers still showed a highly volatile composition compared to non-citrus fibers. In addition, Es-GC analyzes of the chiral volatiles present in the samples indicated that the processing involved monoterpene alcohols (terpinen-4-ol and α-terpineol) tend to show a variation in their ER,probably because of the heat applied during drying(Marsol-Vall et al., 2017) Hence, it cannot be neglected, according to the recent study on the by-products of some fruits found commercially in Sulaimaniyah markets.

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Table 1: Effect of different levels of Carrot juice by products and Cucumber peels on growth performance of common carp *Cyprinus carpio* during 45 days of rearing

Groups	Wt gain	Daily GR	Specific GR	Relative GR	FCR	FER	PER	Fat ER
Control (Group 1)	9.57 ±0.02	0.798 ±0.04	175.72 ±0.02	19.015 ±0.02	3.72 ±0.02	0.27 ±0.01	0.34 ±0.01	2.92 ±0.01
Group 2 5gm Carrot juice by product	11.38 ±0.02	0.948 ±0.03	176.54 ±0.02	22.93 ±0.01	3.27 ±0.04	0.31 ±0.03	0.40 ±0.01	3.46 ±0.03
Group 3 10gm Carrot juice by product	14.83 ±0.04	1.236 ±0.02	181.88 ±0.05	27.34 ±0.01	2.71 ±0.01	0.37 ±0.02	0.53 ±0.01	4.51 ±0.02
Group 4 5gm Cucumber peel	10.59 ±0.02	0.883 ±0.05	177.15 ±0.02	20.64 ±0.04	3.96 ±0.03	0.26 ±0.02	0.32 ±0.06	3.21 ±0.04
Group 5 10gm Cucumber peel	14.85 ±0.01	1.238 ±0.03	183.65 ±0.01	25.98 ±0.01	2.98 ±0.04	0.24 ±0.01	0.31 ±0.01	4.32 ±0.02
Group 6 5 gm orange juice by product	17.98 ±0.03	1.87 ±0.01	188.98 ±0.04	29.65 ±0.03	3.82 ±0.03	0.17 ±0.01	0.43 ±0.01	5.32 ±0.02

Group 7 10 gm orange juice by product	20.76 ±0.03	2.06 ±0.03	196.54 ±0.02	35.87 ±0.02	2.12 ±0.01	0.21 ±0.03	0.49 ±0.03	

Table 2: Effect of different levels of Carrot juice by products and Cucumber peels on some physio-biological parameters of common carp Cyprinus carpio during 45 days of rearing

Groups	hepatosomatic index	Spleenosomatic index	Kidneysomatic index	Gillsomatic Index	Fish weight without Head Index	Fish weight without Head & Viscera Index
Control (Group 1)	2.09±0.01	0.28±0.01	1.21±0.01	5.46±0.02	70.98±0.06	43.67±0.04
Group 2 5gm Carrot juice by product	2.68±0.03	0.30±0.02	1.39±0.03	6.52±0.03	75.87±0.04	45.32±0.03
Group 3 10gm Carrot juice by product	2.73±0.01	0.33±0.01	1.34±0.02	6.48±0.01	77.87±0.08	47.63±0.07
Group 4 5gm Cucumber peel	2.79±0.05	0.60±0.01	1.34±0.02	4.39±0.03	80.34±0.04	50.89±0.04
Group 5 10gm Cucumber peel	2.86±0.02	0.71±0.02	1.36±0.03	4.6±0.02	83.13±0.07	57.21±0.03
Group 6 5 gm orange juice by product	2.98 ±0.02	0.81 ±0.02	1.37 ±0.01	5.32 ±0.02	89.84 ±0.04	59.73 ±0.04
Group 7 10 gm orange juice by product	3.01 ±0.02	0.88 ±0.02	1.41 ±0.01	6.09 ±0.05	88.64 ±0.04	65.52 ±0.04

Table 3: Effect of different levels of Carrot juice by products and Cucumber peels on some biological parameters of common carp *Cyprinus carpio* during 45 days of rearing

Groups	Intestine Length Index	Intestine Length/ Length Index	Intestine Weight Index	Condition Factor
Control (Group 1)	30.90±0.07	90.43±0.07	1.76± 0.03	1.23±0.03
Group 2 5gm Carrot juice by product	32.23±0.05	92.45±0.05	1.99 ±0.02	1.26±0.02
Group 3 10gm Carrot juice by product	33.41±0.04	96.74±0.04	2.09±0.07	0.95±0.04
Group 4 5gm Cucumber peel	37.03±0.06	109.23±0.08	3.33±0.06	0.94±0.05
Group 5 10gm Cucumber peel	39.06±0.02	118.14±0.06	3.21±0.04	0.99±0.04

Group 6 5 gm orange juice by product	31.87 ±0.06	94.87 ±0.05	3.03 ±0.03	1.32 ±0.05
Group 7 10 gm orange juice by product	33.90 ±0.06	98.72 ±0.03	3.05 ±0.05	1.95 ±0.04

Table 4: Effect of different levels of Carrot juice by products and Cucumber peels on differentia white blood cells of common carp *Cyprinus carpio* during 45 days of rearing

Groups	WBC	Lymphocytes	Monocytes	Granulocytes
Control (Group 1)	135.34±0.08	45.32±0.04	29.25±0.02	23.39±0.06
Group 2 5gm Carrot juice by product	140.49±0.07	41.48±0.09	32.09±0.05	40.28±0.08
Group 3 10gm Carrot juice by product	148.15±0.05	38±0.03	34.81±0.04	44.75±0.03
Group 4 5gm Cucumber peel	116.05±0.04	34.4±0.02	35.65±0.03	25.04±0.05
Group 5 10gm Cucumber peel	112.4±0.03	32.5±0.07	37.3±0.07	27.2±0.03
Group 6 5 gm orange juice by product	115.95 ±0.08	30.94 ±0.07	39.05 ±0.06	32.95 ±0.03
Group 7 10 gm orange juice by product	100.63 ±0.07	28.94 ±0.03	41.05 ±0.04	31.59 ±0.04

Table 5: Effect of different levels of Carrot juice by products and Cucumber peels on complete blood counts of common carp *Cyprinus carpio* during 45 days of rearing

Groups	HGB	RBC	HCT	MCV	MCH	MCHC	PLT	MPV
Control (Group 1)	77.39 ±0.04	0.61 ± 0.04	18.04 ±0.06	198.39 ±0.03	121.92 ±0.01	339.03 ±0.02	26.59 ±0.03	11.38 ±0.01
Group 2 5gm Carrot juice by product	74.89 ±0.02	0.69 ±0.03	14.35 ±0.04	200.23 ±0.03	120.39 ±0.04	419.9 ±0.03	30.94 ±0.01	10.92 ±0.07
Group 3 10gm Carrot juice by product	70.5 ±0.04	0.58 ±0.02	12.95 ±0.02	228 ±0.03	122.85 ±0.02	431± 0.02	32.5 ±0.03	10.35 ±0.01

Group 4 5gm Cucumber peel	68.98 ±0.03	0.39 ±0.07	10.12 ±0.06	228.98 ±0.02	154.93 ±0.03	600.34 ±0.01	32.05 ±0.01	10.20 ±0.05
Group 5 10gm Cucumber peel	62.5 ±0.01	0.25 ±0.01	5.75 ±0.03	230.45 ±0.02	249.45 ±0.02	1088 ±0.08	33.5 ± 0.03	10.1 ±0.03
Group 6 5 gm orange juice by product	61.94 ±0.03	0.34 ±0.03	6.87 ±0.04	246.72 ±0.03	253.09 ±0.03	734.91 ±0.08	38.91 ±0.02	11.02 ±0.03
Group 7 10 gm orange juice by product	57.62 ±0.05	0.28 ±0.04	6.82 ±0.02	279.61 ±0.02	285.72 ±0.02	812.01 ±0.01	41.03 ±0.04	12.04 ±0.05