Egyptian Journal of Rabbit Science, 18 (1) 67 – 82(2008)

EVALUATION OF SOME NATURAL ADDITIVES AS GROWTH ENHANCERS IN RABBITS DIETS

M. N. Ali; A. A. Mahrous and Fatma G. Ahmed
Animal Production Research Institute, ARC, Ministry of Agriculture, Dokki, Giza, Egypt.

Seventy two weanling New Zealand White (NZW) rabbits averaging 753.7 g body weight age were randomly distributed to six groups (12 each). First group was fed the basal diet (control). The other groups were fed the control diet supplemented with either 0.2% Curcuma longa, 0.2% lemon extract, 0.2% citric acid, 0.2% Curcuma longa + 0.2% lemon extract or 0.2% Curcuma longa + 0.2% citric acid. The experimental period extended for 8 weeks. The results showed that dietary supplementation with 0.2% Curcuma longa + 0.2% citric acid showed the highest live body weight and dressing percentage. The differences in digestibility coefficients between the experimental groups were not significant. The experimental additives significantly (P<0.05) decreased total count of harmful microorganisms (Escherchia coli, Salmonella and Colstredium spp) in caecum. The blood constituents differed significantly (P<0.05) between groups and the total cholesterol decreased with dietary supplementation with (Curcuma longa, leamon extract and citric acid) alone or with combination enhances growth on rabbits.

Key words: Digestibility, feed additives, performance, rabbits.

Growing opposition to the prophylactic use of antibiotics as additives in livestocks feed has prompted the search for effective alternatives, such as probiotics, prebiotics, dietary acids and plant extracts (Maertens *et al*. 2004). Citric acid, like many other organic acids, elicits antimicrobial activity by various mechanisms (Sharma, 2000). El-Kerdawy (1996) reported that supplementation of citric acids to rabbit diets improved their nutritive value. Also, Abdel-Azeem *et al*. (2000) found that addition of citric acid to the diet containing high level of starch prevented the lethal effects of this diet. Since citric acid can act as a sequestering agent of divalent ions such as Ca2+ and Mg2+ and have a disrupting effect on the organic matter of gram-negative bacteria (Ocana-Morgner and Dankert 2001). Mendez-Albores *et al*. (2005) showed that the efficacy and safety of acidification with citric reduced aflatoxin levels in maize. On the other hand, Barnhart *et al*. (1999) showed that an apparent synergistic

combination of d-limonene and citric acid was observed when evaluating the potential to eliminate salmonella in presence of organic material. Plant extracts and oils have been used for food preservation, pharmaceuticals, alternative medicine and natural therapies for many years (Jones 1996, Lis-Balchin and Deans, 1997). Herbs have been found to have anti-microbial activity and anti-viral properties (Hammer *et al.*, 1999 and Smith-Palmer *et al*., 1998). Also, plant extracts were found to have immunomodulatory (Antony *et al.*, 1999) and antimutagenic activities (Soni *et al.*, 1997). Sohn *et al.*(2000) proposed that the growth-promoting effects of polysaccharides were partly due to their immunostimulating effects, thus reducing bacterial and viral infection. *Curcuma Longa* a perennial herb which is known as turmeric, is a member of zingibeaceae. *Curcuma Longa* or turmeric is a medicinal plant widely used and cultivated in tropical regions. *Curcuma Longa* has antioxidant properties (Osawa *et al*., 1995), inhibit spore count and aflatoxin synthesis (Gowda *et al*., 2004). Curcuma spp. contain turmeric (a water-soluble peptide), essential oils (such as zingiberene) and curcminoids including curcumin (Sharma *et al.*, 2005). Curcumin have many biological properties including antioxidant (Toda *et al.*, 1985), anti-inflammatory (Satoskar *et al*., 1986) properties. Also, Sung-Eun *et al*. (2001) showed that curcumin are potent inhibitors of aflatoxicol formation by chicken liver reductases.

The objective of this study was to evaluate the effects of feeding *Curcuma longa*, lemon extract, citric acid, and their combination on nutrients digestibility, nitrogen utilization, some plasma parameters, microbial count in caecum and rabbits performance.

MATERIALS AND METHODS

The present study was carried out at Gezeeret El-Sheir Station, El-Kanater El-Khyria, Kalyobia Governorate, Egypt.

A total of seventy two of New Zealand White (NZW) rabbits weighing in average about 753.7g were randomly distributed into six comparable groups, (12 rabbits in each). Animals housed in individual cages (25 x 40 x 40 cm) were provided with diets and water *ad lib*. The experiment of duration was 8 weeks rabbits. A commercial balanced diet shown in Table 1 was used as the basal diet (The feed additives added the basal diets before pelleted. The animals were kept under the same managerial, hygienic and environmental conditions and were fed on the following six diets:

- Group 1: The basal diet (Control).
- Group 2: The basal diet + 0.2% *Curcuma longa.*
- Group 3: The basal diet + 0.2% lemon extract.

Table 1: Ingredients and chemical composition of the basal diet.

Ingredients	%
Berseem hay	30.0
Yellow corn	23.7
Wheat bran	27.0
Soybean meal	12.5
Vennas	5.0
Salt	0.5
Calcium carbonate	1.0
*Mineral & Vitamins	0.3
Total	**100**
Chemical Composition:	
Crude protein (%)	16.38
Crude fiber (%)	14.06
Ash (%)	5.13
**DE (kcal/kg)	2747

* **Each kilogram contains**: Vit. A 2000000 IU, vit. D3 150000 IU, vit. E 8.33g, Vit. K 0.33g, Vit. B1 0.33g, Vit. B2 1.0g, Vit. B6 0.33g, Vit. B12 1.7mg, Vit. B5 8.33g, Pantothenic acid 3.33g, Zn 11.79, Fe 12.5, Cu 0.5g, Co 1.33 mg, Se 16.6 mg, Mg 66.79 mg, Niacin 8.33 mg, Biotin 33 mg, Folic acid 0.83g, Choline chloride 200 g, and Mn 5g.

- Group 4: The basal diet + 0.2% citric acid.
- Group 5: The basal diet + (0.2% *Curcuma longa* + 0.2% lemon extract).
- Group 6: The basal diet + (0.2% *Curcuma longa* + 0.2% citric acid).

The citric acid was obtained from Egyptian Company for Laboratory Services, Cairo, Egypt. *Curcuma longa* and lemon fruits were purchased from local market, Cairo. The blendered lemon was prepared by cutting the lemon fruits into chips and blended for 2 minuets. At the end of the experimental period, three rabbits of each treatment were housed individually cages for ten days (five days as a preliminary period and 5 days as collection period) to determine the digestion coefficients and nutritive values of the experimental diets. Both feed and feces were chemically analyzed according to A.O.A.C (1990). The microbial content was studied in their selective media, as described by (Difco, 1989) for total bacterial counts, *E. coli* and *Clostridium* Spp. Salmonella were estimated according to the method described by A.O.A.C (1990). Technique of colony forming unit (CFU) was adopted and incubation took place at 30º C for 2-7 days. Blood samples were collected at slaughtering time into dry

RESULTS AND DISCUSSION

Digestibility coefficients:

Data in Table 2 showed that there were insignificant differences between treatments in digestion coefficients and nutritive values except nitrogen retention ($P<0.05$). These results disagree with those obtained by El-Kerdawy (1996) who found that citric acid increased the digestibility coefficients of CP, CF and improved nutritive values of the tested diet. The rabbits fed 0.2% *Curcuma longa* + 0.2% lemon extract or those fed 0.2% *Curcuma longa* + 0.2% citric acid recorded lower CF digestibility coefficient than those fed the control diet. These results can be explained as a result of reduction of total count bacteria in the caecum and consequently the digestion of fiber and protein by microflora decreased. In this respect, Ivanova *et al.* (1999) found that virginiamycin significantly decreased digestibility of fiber and increased digestibility of fat. The effect of essential oil on digestibility of nutrients found to be different between various animals. For example, with broiler chickens, Jang *et al.* (2007) found that total and specific activates of pancreatic trypsin significantly increased in birds fed essential oil. In contrast another study showed that increasing amount of dietary lactic acid and fumaric acid did not offer protection from caecal salmonella colonization or carcass contamination following oral challenge with salmonella typhimurium (Waldrop *et al.*, 1995). On the other hand, Castillejos *et al.* (2006) found that limonene at 50 and 500 mg/L reduced total VFA concentration *in vitro* (-4.5 and -5.6%, respectively), and suggested that these doses were toxic to rumen bacteria and they found that thymol at 500 mg/L reduced the digestion coefficient of DM, OM, NDF and ADF *in vitro.* These results show clearly that natural feed additives like *Curcuma* longa or lemon extract can alter the total account of bacteria and consequently alter the digestion coefficients of some nutrients with or without negative effect of performance. For example the rabbits fed 0.2% *Curcuma longa* + 0.2%

Table 2: Effect of dietary supplementation of different natural substances on digestion coefficients and nutritive value of growing rabbits.

Items	Control	*C. longa*	Lemon extract	Citric acid	*C. longa* + Lemon extract	*C. longa*+ Citric acid
Digestibility Coefficients%:						
DM	82.68	82.89	77.45	82.66	79.02	75.83
	±2.63	±1.62	±4.13	±1.81	±1.47	±2.32
OM	83.08	83.66	78.60	83.31	78.15	76.58
	±2.66	±1.55	±5.78	±1.86	±1.38	±2.34
CP	84.24	85.48	79.74	84.40	79.09	76.82
	±2.11	±1.96	±5.12	±1.67	±1.37	±2.22
CF	46.62	44.70	47.21	44.73	46.87	45.82
	±4.51	±3.58	±5.44	±3.36	±2.16	±4.21
EE	96.53	96.78	93.85	95.56	96.74	93.17
	±1.04	±0.90	±1.91	±0.39	±0.18	±2.80
NFE	85.33	86.15	81.86	86.76	81.66	81.19
	±2.58	±1.12	±5.07	±0.72	±1.29	±2.05
Nutritive value %:						
TDN	76.91	77.73	73.52	80.35	73.89	71.90
	±2.43	±1.46	±5.23	±2.37	±1.26	±2.16
DCP	14.53	14.84	13.92	14.75	13.70	13.04
	±0.36	±0.34	±0.89	+0.29	+0.23	±0.37

[a-b] Means in the same row with different letters, differ significantly ($P< 0.05$).

citric acid recorded the highest live body weight, while recorded lower total counts bacteria and lower CP, CF digestion coefficients and nitrogen retention.

Growth performance:

The average of initial and final body weight, body weight gain, feed intake and the calculated feed conversion ratio are presented in Table (3). The results indicated that there were significant differences ($P<0.01$) of live body weight between treatments at 70 days of the experimental period, the rabbits fed 0.2% C. *longa* + 0.2% citric acid recorded the best live body weight by 15.02% compared with control diet, while the group which fed 0.2% lemon extract recorded the lowest live body weight. Where, the addition of lemon extract + *Curcuma longa* did not alter the body weight and these may be due to the lemon extract contain limonene (Turner *et al*., 1999). Lemon extract alone was not suitable additive for rabbit and this may be due to the effect of limonene on the microflora of the caecum. However, Oh *et al.* (1967 and 1968) observed that high doses of different plant essential oils decreased the production of gas and total VFA in *in vitro* fermentations of mixed rumen microorganisms, suggesting that high doses resulted in a general inhibition of rumen microbial activity. In this respect, (Castillejos *et al.*, 2006) concluded that there was no benefit to

Table 3: Growth performance as affected by dietary supplementation of natural feed additives to growing rabbits.

Items	Control	*C. longa*	Lemon extract	Citric acid	*C. longa* + Lemon extract	*C. longa* + Citric acid
Initial weight	750.60 ±9.59	746.16 ±19.18	740.00 ±27.33	752.85 ±26.69	765.0 ±34.27	767.8 ±20.83
42 days	1351^{b} ±60.35	1373^{b} ±34.89	1266^{b} ±60.56	1337^{b} ±62.55	1346^{b} ±30.27	1554^{a} ±28.68
70 days	1973^{ab} ±18.8	1872^{b} ±36.26	1865^{b} ±48.82	1925^{b} ±64.50	1840^{b} ±75.80	2066^{a} ±19.39
Body weight gain						
0 – 42	601.2^{b} ±30.0	627.1^{b} ±30.91	526.75^{b} ±65.21	585.00^{b} ±46.22	581.75^{b} ±36.87	786.00^{a} ±25.34
42 – 70	622.0 ±32.0	499.0 ±27.71	599.0 ±60.74	534.6 ±36.34	474.6 ±25.50	512.8 ±33.66
0 - 70	1223^{ab} ±22.35	1126^{bc} ±27.35	1125^{bc} ±27.84	1144^{bc} ±60.18	1026^{c} ±56.63	1299^{a} ±36.10
Feed intake						
0-42	2827^{c} ±116.0	3083^{bc} ±51.6	3136^{b} ±83.69	2917^{bc} ±95.4	3122^{b} ±160.0	3454^{a} ±47.9
42-70	3107^{cd} ±43.62	3143^{bcd} ±76.97	3638^{a} ±29.12	3218b^{c} ±50.8	3013^{d} ±60.0	3311^{b} ±54.1
0 – 70	5934^{b} ±128.9	6226^{b} ±108.4	6775^{a} ±71.75	6244^{b} ±118.79	6135^{b} ±209.3	6766^{a} ±69.00
Feed conversion						
0-42	5.05 ±0.45	5.07 +0.29	6.41 ±0.53	7.49 ±0.66	5.57 ±0.54	4.43 ±0.10
42-70	5.24 ±0.38	6.51 ±0.37	6.65 ±0.82	6.32 ±0.41	6.78 ±0.49	6.78 ±0.57
0 - 70	4.86 ±0.11	5.58 +0.21	6.03 ±0.12	5.59 +0.24	6.40 ±0.40	5.25 ±0.14

[a-c] Means in the same raw with different letters, differ significantly (P<0.05).

using limonene as an additive to modify rumen microbial fermentation. The addition of either *Curcuma longa* or citric alone did not alter live body weight. In this respect, Sharma (2000) showed that citric acid like many other organic acids, elicits antimicrobial activity by various mechanisms. Martin *et al*. (2002) suggested that dietary organic acid can influence the intestinal environment to less appropriate conditions for pathogenic bacteria growth or killing some pathogens. The data showed clearly that the addition of 0.2% *Curcuma longa* + 0.2% citric acid is the best additive under the condition of this study. Many researcher mentioned that Gram positive bacteria are more sensitive to essential oil compounds (Busquet *et al*., 2005 and Smith-Palmer *et al*., 1998) while citric acid can act as a sequestering agent of divalent ions such as Ca2+ and Mg2+ and have a disrupting effect on the organic matter of Gram-negative bacteria (Ocana- Morgner and Dankert, 2001), then the addition of 0.2% *Curcuma longa* + 0.2% citric

acid may have a synergetic effect on either gram-positive or negative bacteria. On the other hand, *Curcuma longa* known to be a good antioxidant and citric acid may create a synergistic relationship with *Curcuma longa* by chelating metal ions. In this respect, Sharma (2000) suggested that citric acid may create a synergistic relationship with antioxidants by chelating metal ions. Finally, Lippens *et al*. (2005) showed that organic acids and plant extracts work differently so, it might be postulated that the combination of both groups of products could give an additive effect. We hypothesis that a successful feed additive for rabbits that decreases total count of bacteria in digestive tract without negative effect on performance.

It is clear that all suggested additives improved the weight gain especially with citric acid as reported by El-Kerdawy (1996) who found that fumeric, citric and malic acids at rate of 0.5% increased weight gain and return of rabbits. While, El-Allawy (2001) found that citric acid (0.5 g / kg diet) had no effect on rabbit weight gain but decreased feed intake. Moreover, feed conversion ratio. The improvement in growth rate with organic acids may be due to decrease pH value of the cecum and ileum to level not favorable for pathogens also may be that it severs as substrates in the intermediary metabolites which increase gastric proteolysis (Kirchgessner and Roth, 1988).

There were significant ($P<0.01$) differences between treatments in feed intake (Table 3) in first period (0-42 days). The animals fed *Curcuma longa* + citric acid recorded the highest value while the rabbits fed 0.2% citric recorded the lowest value, while in the second experimental period (42-70 days), showed significant ($P<0.01$) differences between experimental groups where animals fed 0.2% lemon extract recorded the highest value while the animals fed 0.2% lemon extract + 0.2% *Curcuma longa* recorded the lowest feed intake. Finally, the data showed significant differences ($P<0.01$) between treatments in feed intake all over the tested period (0-70 days). The rabbits fed the control diet recorded the lowest value, while the rabbits fed *Curcuma longa* + citric recorded the highest value of feed intake.

There were insignificant differences between experimental groups in feed conversion (FC) ratio. The animal fed the control diet recorded the best value, while feed additives used in this study did not improve the FC. The same results reported by (Ivanova *et al.*, 1999).

Carcass characteristics:

Results in Table 4 indicated that the values of carcass, liver, heart, kidney, lung, stomach, giblets and total edible parts relative to the live body weight were not significantly affected by different supplementations.

Table 4: The effects of dietary supplementation of natural feed additives on carcass characteristic to growing rabbits.

Items	Control	*C. longa*	Lemon extract	Citric acid	*C. longa* + Lemon extract	*C. longa*+ Citric acid
Carcass (%)	52.45	53.09	51.33	54.59	55.22	56.42
	±0.49	±1.58	±1.33	±1.14	±2.14	±0.82
Liver (%)	2.93	2.63	2.52	2.56	2.30	2.34
	±0.08	±0.22	±0.16	±0.19	±0.06	±0.12
Heart (%)	0.28	0.29	0.25	0.31	0.36	0.32
	±0.05	±0.02	±0.02	±0.02	±0.02	±0.06
Head (%)	6.58	6.46	6.40	6.26	6.28	6.76
	±0.61	±0.19	±0.16	±0.31	±0.19	+0.27
Kidney (%)	0.71	0.72	0.75	0.59	0.62	0.67
	±0.04	±0.04	±0.07	±0.02	±0.01	±0.04
Stomach (%)	4.60	5.08	3.79	4.27	4.35	3.63
	±0.14	±0.66	±0.60	±0.18	±0.55	±0.37
Caecum	29.66	29.33	31.00	32.33	27.66	29.00
length (cm)	±0.33	±1.73	±1.00	±1.76	±0.33	±1.15
Giblets (%)	3.93	3.65	3.53	3.48	3.30	3.35
	±0.01	+0.24	+0.20	+0.20	±0.09	±0.05
Dressing (%)	56.38	56.74	54.87	58.07	58.52	59.78
	±0.50	±1.62	±1.13	±1.23	±2.05	±0.80

The same trend was also found with the values of caecum length (cm). Also, El-Kerdawy *et al.* (1996) found that carcass traits including giblets, carcass and dressing percentages were insignificantly affected by dietary supplementation with 0.5% fumaric, malic, citric or a mixture of these acids. However, Ibrahim (2000) found a significant increase in total giblets weight in groups received 0.25 and 0.50% as growth promoters. El-Allawy (2001) found that citric acid (0.5 g/kg diet) had no effect on rabbit dress.

Caecum microflora:

The data in Table 5 showed that all experimental diets significantly ($P<0.01$) decreased the counts of total bacterial, *Escherchia coli,* Salmonella and *Colsterdium* sp. compared to control diet. The rabbits fed 0.2% C. *longa* + 0.2% lemon extract and 0.2% *Curcuma longa* + 0.2% citric recorded the lowest values of bacteria numbers which indicate a synergetic effect between *Curcuma longa* with either Lemon extract or citric acid. In this respect, Martin *et al*. (2002) reported that dietary organic acid can influence the intestinal microbiota by changing the intestinal environment to less appropriate conditions for pathogenic bacteria. Hammer *et al*. (1999) reported that herb extracts such as lemon grass, oregano, hay and thyme have antimicrobial activity. Also, Namkung *et al*. (2004) found a reduction in fecal coliform bacteria counts of pigs with antibiotics, herb and acid treatments. However, Mehall *et al.* (2002) concluded that acidification of formula with citric acid is equally protective

Table 5: Caecum microbial counts (log^{-1}cfu/ml) as affected by dietary supplementation of natural feed additives to growing rabbits.

Items	Control	C. *longa*	Lemon extract	Citric acid	C. longa + Lemon extract	C. longa+ Citric acid
Total bacterial counts	6.38^{a} ±0.09	4.91^{c} ±0.05	5.16^{b} ±0.03	5.19^{b} ±0.01	4.25^{e} ±0.02	4.59^{d} ±0.02
Eschrechia coli	7.19^{a} ±0.02	5.81^{d} ±0.02	6.15^{c} ±0.02	6.37^{b} ±0.03	3.56^{f} ±0.03	5.14^{e} ±0.03
Salmonella	5.33^{a} ±0.03	4.66^{d} ±0.02	5.07^{c} ±0.01	5.19^{b} ±0.01	3.85^{f} ±0.03	4.27^{e} ±0.03
Colstredium Sp.	2.61^{a} ±0.04	2.24^{c} ±0.02	2.33^{bc} ±0.02	2.37^{b} ±0.04	1.65^{e} ±0.02	1.79^{d} ±0.05

$^{a-f}$ Means in the same row with different letters, differ significantly ($P \leq 0.05$).

against bacteria but better tolerated than acidification with hydrolic acid. With broiler Jang *et al.* (2007) found that essential oil decreased *E. coli* in ileo-cecal digesta. The synergistic mechanism between *Curcuma longa* with either lemon extract or citric acid can be explained by our hypothesis that essential oil affect gram positive bacteria, while citric or limonene (in the lemon extract) affect gram-negative bacteria and the combination between them may reduce the numbers of either gram-negative and positive bacteria. On the other hand, Langhout (2000) suggested that a combination of organic acids and essential oils would be beneficial as the organic acids appear to be particularly active in the feed, crop, and gizzard, whereas essential oils appear to work more in the later segments of intestinal tract.

Also, Cross *et al.* (2007) found that the caecal coliform populations in the birds fed some of herbal had higher count than those fed the control diet and indicated that these dietary herbs may select more against gram-positive rather than gram-negative bacteria and may be the conditions become suitable for the presence of *Campylobacter* sp. or *Salmonela* sp.

Dorman and Deans (2000) demonstrated the antimicrobial activity of limonene, mainly against gram-negative bacteria. On the other hand, gram positive bacteria are more sensitive to essential oil compounds (Busquet *et al*., 2005 and Smith-Palmer *et a*l., 1998). The addition of both lemon extract + C. *longa* may have a synergetic effect on either gram-positive or negative bacteria and/or *Curcuma longa* decrease the bad effect of lemon extract by unknown mechanism.

Blood constituents:

Data in Table 6 indicated that there were significant ($P<0.01$) differences between treatments in plasma total protein, the rabbits fed 0.2%

Table 6: Some blood constituents as affected by dietary supplementation of natural feed additives to growing rabbits.

Items	Control	C. *longa*	Lemon extract	Citric acid	*C. longa* + Lemon extract	*C. longa* + Citric acid
Total protein (g/dl)	6.70^{cd} ±0.70	7.76^{c} ±0.91	8.32^{bc} ±0.59	10.05^{ab} ±0.51	10.69^{a} ±0.67	5.46^{d} ±0.47
Albumin (g/dl)	3.45^{d} +0.26	4.34^{b} +0.29	3.83^{c} +0.26	3.86^{c} ±0.16	4.67^{a} ±0.37	3.68^{d} ±0.43
Globulin (g/dl)	3.24^{c} ±0.55	3.57^{bc} ±0.54	4.48^{abc} ±0.52	6.18^{ab} ±0.36	6.62^{a} ±0.91	1.77^{c} ±0.10
Total lipids (g/L)	3.10^{a} ±0.37	2.17^{c} ±0.13	2.51^{b} ±0.33	2.41^{b} ±0.10	2.49^{b} ±0.07	2.15^{c} ±0.18
Total cholesterol (mg/dl)	107.37^{a} ±10.90	86.20^{c} ±3.40	90.55^{b} ±10.20	100.20^{a} ±9.00	95.80^{ab} ±0.94	60.99^{d} ±3.50

$^{a-d}$ Means in the same row with different letters, differ significantly ($P \leq 0.05$).

Curcuma longa + 0.2% lemon extract recorded the highest value, while the rabbits fed 0.2% *Curcuma longa* + 0.2% citric recorded the lowest values. There were significant ($P<0.05$) differences between treatments in plasma globulin with the same trend of plasma total protein. As we mentioned before, many investigators reported that gram positive bacteria are more sensitive to essential oil compounds (Busquet *et al*. 2005 and Smith-Palmer *et al*., 1998) while citric acid can act as a sequestering agent of divalent ions such as Ca2+ and Mg2+ and have a disrupting effect on the organic matter of gram-negative bacteria (Ocana-Morgner and Dankert, 2001). The addition of 0.2% *C. longa* + 0.2% citric may have a synergetic effect on either gram-positive or negative bacteria and consequently the immune cost will be decreased .It has been reported that the immunoglobulin A secreted across the intestinal mucosa accounts for >70% of total antibody production of an animal (Macpherson and Uhr, 2004). For example the adult human secretes more than 5 g of immunoglobulin. A each day, most of which binds to the intestinal microbiota and to dietary antigens (Macpherson *et al.*, 2000 and Kraehenbuhi and Corbett, 2004). Richards *et al.* (2005) showed that micrflora-specific immunoglobulin A and immunoglobulin G secretion alone can cost the animal several hundred grams of protein over a lifetime that is not directed towards growth. Macpherson *et al*. (2000) reported that the back-ground of immunoglobulin-A against bacterial antigens is directed toward the individual's established gut flora with a further or renewed response to antigen change within that established gut flora. Richards *et al.* (2005) showed that reducing changes in the gut microflora by controlling species that either from the food or the environment should

reduce the additional response of the gut immune system according to changes in microflora antigenic stimulation. Sandberg *et al*. (2007) showed that the more virulent pathogen may have stimulated a greater immune response, rather than causing more damage to the host and through a larger resource requirement of the greater immune responses had greater reductions in growth. From previous discussion we can indicate that the addition 0.2% *Curcuma longa* + 0.2% citric acid decrease globulin synthesis and consequently saved protein which is directed towards growth. The data in Table (6) showed that there were significant differences ($P<0.05$) between treatments in plasma total lipids and total cholesterol, the rabbits fed 0.2% C. *longa* + 0.2% citric acid recorded the lowest value, while those fed 0.2% citric recorded the highest value. There was insignificant differences between rabbits fed control diet and rabbits fed 0.2% citric and these results agree with those obtained by El-Kerdawy (1996) who found that addition of 0.5% citric acid significantly decreased the levels of total lipids and total cholesterol .The addition of 0.2% C. *longa* alone or with citric acid numerically decreased the level of plasma total cholesterol compared with those fed control diet. Letje Wientasih *et al*. (2002) found that dietary curcuma fed to rabbits decreased cholesterol and triglyceride concentration in blood. However, it is known that most components of essential oils inhibit hepatic 3-hydroxu-3-methylglutaryl ceonzyme A (HMG-CoA) reductase activity (Crowell, 1999) which is a key regulatory enzyme in cholesterol synthesis.

Conclusively, the results obtained in this study showed that addition of 0.2% C. *longa* + 0.2% citric acid improved the rabbits performance and that the experimental additives which used in this study significantly decreased total count of caecum bacteria, *Escherchia coli*, *Salmonella* and *Colstredium sp*. All the feed additives except citric acid tended to decrease the digestibility of nutrients. The addition of *Curcuma longa* + citric acid numerically decreased plasma total protein, while *Curcuma longa* decreased cholesterol level compared to control diet. Finally, the combination of *Curcuma longa* and citric acid is the most successful feed additives for under this study.

REFERENCES

Abdel-Azeem, F., El-Hommosany, Y.M. and Ali, G. M. (2000). Effect of citric acid in diets with different starch and fiber levels on productive performance and some physiological traits of growing rabbits. *Egyptian Journal of Rabbit Science*, **10**:121-145

Antony, S. Kuttan, R. and Kuttan, G. (1999). Immunomodulatory activity of curcumin. *Immunological. Investigation*., **28**:291-303.

A.O.A.C. (1990). Association of Official Analytical Chemists. *Official Methods of Analysis*. 15th ED., Published by the A.O.A.C. *Washington, D.C.*

Barnhart, E. T., Sarlin, L.L., Caldwell, D. J., Byrd, J. A., Corrier, D. E. and Hargis, B. M. (1999). Evaluation of potential disinfectants for preslaughter broiler crop decontamination. *Poultry Science*. **78:**32-37.

Busquet, M., Calsamiglia, S., Ferret, A. and Kamel, C. (2005). Screening for effects of plant extracts and active compounds of plants on dairy cattle rumen microbial fermentation in a continuous culture system. *Animal Feed Science and Technology*, **123-124:**597-613

Castillejos, L. Calsamiglia, S. and Ferret, A. (2006). Effect of essential oil active compounds on rumen microbial fermentation and nutrient flow in *in vitro* systems. *Journal of Dairy Science Association,* **89**:2649-2658.

Cross, D. E.., Mcdevitt, R. M., Hillman, K. and Acamovic, T. (2007). The effect of herbs and their associated essential oils on performance, dietary digestibility and gut microflora in chickens from 7 to 28 days of age. *British Poultry Science*, **48:**496-506

Crowell, P. L. (1999). Prevention and therapy of cancer by dietary monoterpenes. *Journal of Nutrition,* **129**:775S-778S.

Difco, M. (1989). *Dehydrated Culture Media Regents for Microbiology.* 10th edition, pp 689-691, Difco Laboratories incorporated Detroit *Michigan, USA.*

Dorman, H. J. D. and Deans, S. G. (2000). Antimicrobial agents from plants: antibacterial activity of plant volatile oils. *Journal of Applied Microbiology*, **88:**308-316.

Duncan, C. B. (1955). Multiple ranges and multiple F test. *Biometrics*, **11:**1-42

El-Allawy, H. M. (2001). The use of bio-feed additives to improve rabbit performance. Ph. D. Thesis, Cairo University, Egypt.

El-Kerdawy, D. M. A. (1996). Acidified feeds for growing rabbits. *Egyptian Journal of Rabbit Science*, **6:** 143-156.

Gowda, N. K. S, Malathi, V. and Suganthi, R. U. (2004). Effect of some chemical and herbal compounds on growth of *Aspergillus parasiticus* and aflatoxin production. *Animal Feed Science and Technology*, **116**:281-291.

Hammer, K.A., Carson, C.F. and Riley, T.V. (1999). Antimicrobial activity of essential oils and other plant extracts. *Journal of Applied Microbiology*, **86**: 985-990.

Ibrahim, Sh. A. M. (2000). Clove and cinnamon as growth promoters and detoxificants against cadmium chloride in growing rabbits. *Egyptian Journal of Rabbit Science*, **10:**89-104.

Ivanova, V., Marounek, M. and Klein, P. (1999). Effects of virginiaymycin and salinomycin on performance, digestibility of nutrients and mortality of rabbits. *Animal Feed Science and Technology*, **77**:139-147

Jang. I. S., Ko, Y. H., Kang, S.Y. and Lee, C. Y. (2007). Effect of a commercial essential oil on growth performance, digestive enzyme activity and intestinal microflora population in broiler chickens. *Animal Feed Science and Technology*, **134:** 304-315.

Jones, F. A. (1996). Herb-useful plants. Their role in history and today. *European. Journal of Gastroenterol. Hepatol***., 8**: 1227-1231.

Kirchgessner, M. and Roth, F. (1988). Ergotrope effekte durch organisch sauren in der ferkelaufzucht und schweinemast. *Ubersichten zur Tierenahrug*, **16** (93): 1-8.

Kraehenbuhi, J. P. and Corbett, M. (2004). Keeping the gut microflora. *Science***, 303**:1625.

Langhout, P. (2000). New additives for broiler chickens. *World Poultry*, **16** (3): 22-27.

Letje-Wientarsih, Sebastian Chakeredza and Udo ter Meulen (2002). Influence of curcuma (Curcuma Xanthorrhiza Roxb) on lipid metabolism in rabbits. *Journal of the Science of Food and Agriculture***, 82**:1875-1880.

Lippens, M., Huyghebaert G. and E. Cerchiari (2005). Effect of the use of coated plant extracts and organic acids as alternatives for antimicrobial growth promoters on the performance of broiler chickens. *Arch.Geflugelk*, **69:**261-266.

Lis-Balchin, M. and Deans, S. G. (1997). Bioactivity of selected plant essential oils against *Listeriamonocytogenes. Journal of Applied Bacteriology*, 125-136.

Macpherson, A. J. and Uhr, T. (2004). Induction of protective Ig A by intestinal dendrite cells carrying commensal bacteria. *Science*, **303**: 1662-1665.

Macpherson, A. J., Gatto, D., Sainsbury, E., Harriman, G., Hengartner, H. and Zinkernagel, R. (2000). A primitive T cell-independent mechanism of intestinal mucosal Ig A responses to commensal bacteria. *Science***, 288**:2222-2226.

Maertens, L., Aerts J. M. and De Boever, J. (2004). Degradation of dietary oligofructose and inuline in the gastro-intestinal tract of the rabbit and the effects on caecal pH and volatile fatty acids. *World Rabbit Science*, **12**:235-246.

Martin, W., Verstegen, A. and Williams, B. A. (2002). Alternatives to the use of antibiotics as growth promoters for monogastric animals. *Animal Biotechnology*, **13**:113-127.

Mehall, J. R., Saltzman, D. A. , Jackson, R. J., and Smith, S. D. (2002). Acidification of formula with citric acid is equally effective and better tolerated than acidification with hydrochloric acid. *Critical Car. Medicine*, **30:**1701-1704

Mendez-Albores, A., Arambula-Villa, G., Loarca-Pina, M. G. F., Castano-Tostado, E. and Moreno-Martinez, E. (2005). Safety and efficacy evaluation of aqueous citric acid to degrade B-aflatoxins in maize. *Food and Chemical Toxicology*, **43**: 233-238.

Namkung, H., Li, M., Gong, J., Yu, H., Cottrill, M. and De Lange, C. F. M. (2004). Impact of feeding blends of organic acids and herbal extracts on growth performance, gut microbiota and digestive function in newly weaned pigs. *Canadian Journal of Animal Sciences*, 84:697-704.

Ocana-Morgner, C. and Dankert, J. R. (2001). Induction of complement sensitivity in *Escherichia coli* by citric acid and low pH. *Journal of Applied Microbiology*, **90**: 771-778.

Oh, H.K., Jones, M. B., Longhurst, W. M. (1968). Comparison of rumen microbial inhibition resulting from various essential oils isolated from relatively unpalatable plant species. *Applied Microbiology*, **16**:39-44.

Oh, H. K., Sakai, T., Jones, M. B., Longhurst, W. M. (1967). Effect of various essential oils isolated from Douglas Fir Needles upon sheep and deer rumen microbial activity. *Appl. Microbiol.*,**15**: 777-784.

Osawa, T., Sugiyama, Y., Inayoshi, M. and Kawakishi, S. (1995). Antioxidative activity of tetrahydrocurcuminoids. *Bioscience Biotechnology. Biochemistry*, **59**:1609-1612.

Richards, J. D., J. Gong and De Lange C. F. M. (2005). The gastrointestinal microbiota and its role in monogastric nutrition and health with an emphasis on pigs: Current understanding, possible modulations, and new technologies for ecological studies. *Canadian Journal of Animal Sciences*, **85**:421-435.

Sandberg, F. B., Emmans, G. C. and I. Kyriazakis (2007). The effect of pathogen challenges on the performance of naïve and immune animals: the problem of prediction. *Animal*, **1**:67-86.

SAS. (1990). SAS Users Guide, *Statistics*. SAS Institute, Inc, *Cary, NC*.

Satoskar, R. R., Shah, S. J. and Shenoy, S. G. (1986). Evaluation of anti-inflammatory property of curcumin (diferuloylmethane) in patients with post operative inflammation. *International Journal of Clinical. Pharmacology Toxicology*, **24** :651-654.

Sharma, R. A., Gescher, A. J. and Steward, W. P. (2005). *Curcumin: The Story so far European Journal of Cancer*, **41**:1955-1968.

Sharma, R.K. (2000). Citric acid in Natural food antimicrobial systems. Pages 689-702, A.S. Naidu, ed CRC press LCC, United States of America.

Smith-Palmer, A., Stewart, J. and Fyfe, L. (1998). Antimicrobial properties of plant essential oils and essences against five important food-borne pathogens. *Letters. Applied Microbiology*, **26**:118-122.

Sohn, K. S., Kim, M. K., Kim, J. D. and Han, I. K. (2000). The role of immunostimulants in monogastric animal and fish. *Asian-Australian. Journal of Animal Science*, **13:**1178-1187.

Soni,K.B., Lahiri, M., Chackradeo, P., Bhide, S. V. and Kuttan, R. (1997). Protective effect of food additives on aflatoxin-induced mutagenicity and hepatocarcinogenicity. *Cancer Letters*, **115**: 129-133.

Sung-Eun, L., Bruce, C. C., Russell, J. M., Shin, H. and Hoi-Seon, L. (2001). Inhibitory effects of naturally occurring compounds on aflatoxin B_1 biotransformation. *Journal of Agricultural Food Chemistry*, **49**:5171-5177.

Toda, S., Miyae, T., Arichi, H., Tanizawa, H., Takino, Y. (1985). Natural antioxidants III. Antioxidative components isolated from rhizome of *curcuma longa* L. *Chem. Pharm. Bull.*, **33**: 1725-1728.

Turner, G., Gershenzon, J., Nielson, E. E., Froehlich, J. E. and Croteau, R. (1999). Limonene syntheses, the enzyme responsible for monoterpene biosynthesis in peppermint, is localized to leucoplasts of oil gland secretor cells. *Plant Physiology*, **120**:879-886.

Waldrop, A., Kaniawato, S. and Mauromoustakos, A. (1995). Performance characterеristics and microbiological aspects of broiler fed diets supplemented with organic acids. *Journal of Food Protection.*, **58**: 482-489.

تقييم بعض الاضافات الطبيعية كمحسنات نمو فى علائق الارانب

محمد نبيل على - احمد عبد الرحمن محروس - فاطمة جلال احمد
معهد بحوث الإنتاج الحيوانى - مركز البحوث الزراعية – وزارة الزراعة – الدقى – جيزه - مصر.

تم توزيع عدد 72 ارنب نيوزيلندى عمر 6 اسابيع توزيعا عشوائيا وقسمت الى 6 مجموعات (12 أرنب لكل مجموعة) غذيت المجموعة الاولى على عليقة المقارنة المتزنة الاحتياجات الغذائية وغذيت المجاميع الخمسة الاخرى على نفس عليقة المقارنة مضافا اليها الاضافات الطبيعية كما يلى:
العليقة الثانية: عليقة الكنترول + 0.2% كركم.
العليقة الثالثة: عليقة الكنترول + 0.2% مخلفات الليمون بالعصير.
العليقة الرابعة: عليقة الكنترول +0.2% حمض ستريك.
العليقة الخامسة: عليقة الكنترول + 0.2كركم +0.2% مخلفات الليمون بالعصير.
العليقة السادسة: عليقة الكنترول + 0.2كركم +0.2% حمض ستريك.
تمت التغذية لمدة 8 اسابيع لدراسة تاثير هذه الاضافات على معاملات الهضم و النمو ومواصفات الذبيحة والتغير فى العشائر الميكروبية داخل الاعور ومكونات الدم.
اظهرت النتائج ان المجموعة التى غذيت على 0.2% كركم + 0.2% حمض ستريك قد اعطت افضل قياسات للنمو وكانت الفروق بين المجاميع المختلفة معنوية ، ولم تختلف معاملات الهضم بين المجاميع معنويا. وقد اعطت المجموعة السادسة اعلى نسب تصافى وانخفض العدد الكلى الميكروبى فى كل المجاميع المحتوية على الاضافات المختبره ، ولوحظ نقص معنوى فى اعداد *E. coli* ، *Salmonella* وكذلك *Colstredium Sp* فى ميكروفلورا الاعور. وتغيرت مقاييس الدم بتغير الاضافات المختلفة ، وقد انخفضت نسبة الكوليسترول معنويا فى المجموعة السادسة.
مما سبق يتضح ان اضافة الكركم + حامض الستريك حسن من نمو الارانب وزاد من نسبة التصافى للذبيحة على عكس مستخلص مخلفات الليمون وعصيره ، و قللت كل الاضافات معنويا من اعداد الميكروبات الضارة فى الاعور ويمكن استخدام هذه الاضافات فى مزارع الارانب.