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The Preservation of Chicken Meat. II. Bacteriological Changes

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THIS WORK was carried out in the Faculty of Agriculture, Cair- University, to investigate the effect of antibiotic (10 ppm chlo mpheni col) and packaning in polyethylene bags on the bacteriological changes of chicken meat at chilling and freezing temperatures The following results were obtained:

Under refrigeration conditions, the total, psychrophilic and proteolytic bacterial counts in the breast and leg meat decreased during the first half of the storage period in both treated and untreated carcasses due to the effect of chilling. However, during, the second half of the storage period, the bacterial counts increased gradually in both treated and untreated carcasses as the storage time progressed. However, the untreated carcasses had more total, psychrophilic and proteolytic bacterial counts in their tissues than the treated ones at any period of storage. Therefore, it can be concluded that, dipping the carcasses in antibiotics increase the shelf - life of these carcasses.

Under frozen conditions, the total, psychrophilic and proteolytic bacterial counts , were higher in the breast and leg meat of the un-, packaged carcasses, whether treated or not, until 90 days of storage, than the packaged ones. Afterwards, higher numbers of bacteria. were observed in the packaged carcasses than the unpackaged ones This could be due to that wrapping the carcasses in polyethylene bags reduced dehydration and this increased bacterial multiplication, while the unpackaged ones lost lot of humidity and this reduced bacterial growth. Dipping the carcasses in antibiotics also gave lower, number of bacteria than the untreated ones at any period of freezing whether the carcasses were packaged or not.

Using 10 ppm aureomycine (chlortetracycline, CTC) in a dip solution resulted in the up take of appreciable amounts of antibiotic which exerted a bacteristatic effect on the microorganisms present on carcasses tissue. Those carcasses had a slightly lower bacterial count and remained fresh for a greater period of time than controls(Anderson *et al.*,1958).On the other hand,Vaughn *et al.* (1957), stated that, fryers dipped in 10 ppm of CTC did not exhibit a longer shelf-life than control fryers, when both groups were stored at O°.

It appears that unless the initial concentration of bacteria is relatively low, the use of CTC would be ineffective in controlling bacterial growth. Holleck *et al.* (1958) found that, the bacterial growth seemed to be specific for the packaging material. The average bacterial counts were significantly higher in meat packaged in polyethylene than those packaged with enameled cans and cellophanepliofilm laminate. While, there was no significant difference in microbial counts as influenced by packaging material, but also bacterial counts in polyethylene were generally higher, when carcasses were packaged in either high (polyethylene) or low (vinylidene) gas permeability film and stored at 0-.5° or 6° (Thomson, 1970). Freezing brought about a marked production in viable bacterial populations and a progressive reduction in bacterial counts after storage at-23.3° up to 14 months (Conner *et at.*, 1953).

Almost 50% of the additional storage life of the antibiotics treated carcasses was due to the antibiotic of the psychrophilic bacteria on the carcasses during chilling and freezing. According to these results, the numbers of psychrophiles on the untreated carcasses increased ten fold during storage, while there was no increase on the CTC treated carcasses (Barnes and Shrimpton, 1958). Proteolytic and lipolytic changes in chickens stored at 5° was directly related to the availability of oxygen provided by the packaging procedure. Bacterial numbers paralleled the increase in biochemical indices of deterioration (Rey and Kraft, 1971).

Material and Methods

This work was carried out at the Poultry Experimental Centre, Animal Prodution Department, and Food Science Department, Faculty of Agriculture, Cairo University. A total of 140 Fayoumi chickens were used in this experiment. The chickens were used to study the effect of chloramphenicol and packaging on the shelf-life of chicken meat at chilling and freezing temperatures. The chicks were raised in the floor brooder house from hatch up to eight weeks of age, then removed to the broiler house until sixteen week old. The slaughter was done at 16 weeks, that averaged O. 90 to O. 95 kg in live weight. The chickens were dressed under conditions that would be found in most dressed plants. The eviscerated birds (average weight 0.6 to 0.7 kg) were divided into two groups.

Chilling storage

Forty carcasses were taken randomly and kept under chilling conditions. The carcasses were divided into two sub-groups.

Sub-group 1, 20 carcasses were put in iced water at 5° for 20 min. In sub-groups 2, 10 ppm of chloromyctein (Chloramphenicol) were added to the iced water, where 20 carcasses were put for 20 min. Each carcass in the two sub-groups was banded and weighted to the nearest gramme (Original, weight) and packaged individually in polethylene bags and tied firmly.Pack-

aged carcasses in the two sub-groups were stored in home type refrigerator at about 5± 1°. For the duration of the testing period (15 days). Four carcasses from each sub-group were tested microbiologically at 3, 6, 9, 12 and 15 days of storage.

Freezing storage

Ninety-six caracasses were taken and kept under freezing conditions. These carcasses were divided into four sub-groups.

Sub-group 1, consisted of 24 carcasses, which were put in iced water at 5° for 20 min (untreated-unpackaged). Sub-group 2, after being chilled as in sub-group 1, the carcasses were packaged individually in polyethylene bags and tied tightly (untreated-packaged). In subgroup 3, 10 ppm of chloramphenicol were added to the iced water, where 24 carcasses were put for 20 min. (Treat-, ed-unpackaged). The 24 carcasses of subgroup 4, after being chilled as in sub-group 3, were packaged individually in polyethylene bags and tied tightly (Treated-packaged). All subgroups were stored in a freezing room at about — 10 ± 1°. for 180 days. four carcasses from each sub-group were sampl-ed every 30 days from freezing storage. Each carcass was banded and weighed individually to the nearest g (Original weight).

All carcasses in the two groups were packaged in polyethylene bags either at chilling or freezing temperatures. These bags were 40 cm long and 22 cm wide. When the caracasses were packaged, excess air was removed by hand pressing on bags, no mechanical evacuation was used.

Control samples of fresh carcasses (4 carcasses) were taken for bacteriolo-gical tests. These were considered to be zero time (at slaughter).

Bacteriological tests

Four carcasses were removed from each storage condition (Time of sampl-ing) and were immediately prepared for bacterial examination. From each carcass a separate 10g sample of breast and leg meat, was taken respectively. The samples were aseptically removed from carcasses and were grinded for 5 min with 90 ml of sterile water in a Waring Blender to give 1/10 dilution, and further dilutions were prepared as needed (Margolf *et al.* 1956). These dilutions were plated out to determine the following :

1. *Total plate count*

Nutrient ager (Difco) was used as a medium for the plate counts. Colon-ies were counted after the plates had been incubated at 37° for 48 hr (Kotula and Kinner, 1967).

2. *Psychrophilic bacterial count*

Also, nutrient agar (Difco) was used as a medium for psychrophilic bacteria counts. Plates were incubated at 1° for 14 days (Barnes and Impey, 1968), then colonies which appeared were counted.

3. *Proteolytic bacterial count*

The proteolytic bacteria were tested for liquifying gelatin. Nutrient gelatin was used as a medium to determinate the proteolytic activity of bacteria. The plates were incubated at 20° for 48 hr (Kazanas, 1968). Colonies which appeared with a clear hollow around it were considered to be proteolytic and those colonies were counted.

Statistical analysis

Statistical analysis were carried out according to Steel and Torrie (1960).

Results and Discussion

1. *Total bacteria counts in the breast and leg meat*

Under the chilling conditions, total bacteria counts in the breast and leg meat during the first half of the storage period (Table 1). However, during the second half of the storage period, the bacterial counts increased in both treated and untreated carcasses as the storage time progressed, but the untreated carcasses had more total bacterial counts in thier tissues than the treated ones at any period of storage. These results indicate that, dipping the carcasses in antibiotic solutions gave a slight increase in the shelf-life of these carcasses. These results are in agreement with Anderson *et al.* (1958). Analysis of variance showed that there were highly significant effect of treated and storage periods on total bacterial counts in the breast and leg meat (Table 3).

Under the frozen conditions, mean bacterial counts were higher in the breast and leg meat of the unpackaged carcasses either treated or not until 90 days of storage than the packaged one (Table 2). Afterwards, total counts were higher in the packaged carcasses than the unpackaged ones. Packing reduced dehydration and this caused increased bacterial multiplication. These results are in agreement with (Ingram and Shewan, 1960). On the other hand, the treated carcasses had lower numbers of bacteria than the untreated ones at any period of freezing whether the carcasses were packaged or not.

Analysis of variance showed that there were highly significant effect of treated, packaging and storage periods on total bacterial counts during frozen storage (Table 4).

2. *Psychrophilic bacterial counts in the breast and leg meat*

Psychrophilic bacterial counts in the breast meat decreased slowly at the first 6 days in both treated and untreated carcasses due to the sudden chilling.

This was following by a continuous increase from 9 days to the end of the experiment. However, psychrophilic bacterial counts in the leg meat increased slightly after storage for 9 days. Afterwards, the increase in psychrophiliic bacteriawas highly observed (Table 1). According to Barnes (1960), the spoila ge of poultry meat stored under chilling conditions is caused mainly by the growth of few groups of psychrophilic bacteria such as pigmented and non-pigment strains of pseudomonas. The difference in the psychrophilic bacterial counts in the breast and leg meat due to treatment, storage periods and the interaction between them were highly significant (Table 3).

During frozen storage, the psychrophilic bacterial counts in the breast and leg meat showed similar trends like that observed in the total bacterial countsin the breast and leg meat in the packaged and unpackaged groups whether treated or not (Table 2). These results are in agreement with Barnes and Shrimpton, (1958). They found that, almost 50% of the additional storage life of the antibiotics treated carcasses was probably due to the prevention by the antibiotics of multiplication of the psychrophilic bacteria on the carcasses during chilling and freezing. Analysis of variance showed that there were highly significant effect of treated, packaging and storage periods on psychrophilic bacterial counts during frozen storage (Table 4).

3. *Proteolytic bacterial counst in the breast and leg meat*

The effect of antibiotic on this type of bacteria and its proteolytic role is shown in Table 1., from which it can be seen that, in the breast and leg meat-the untreated samples had more proteolytic bacterial counts than the treated ones at any period of storage. In the breast meat either treated or not and the untreated leg meat, there was a slight decrease in proteolytic bacteria at 6 days, followed by a clear increase until the end of the experiment at 15 days of storage. The decrease in proteolytic bacteria during the first days of storage could be explained on the basis that the antibiotic used was effective in inhibiting the microbial growth. Subsequently, the increase in proteolytic bacteria numbers during prolonged storage could be explained by the subsequent growth of the surviving microorganisms. Analysis of variance showed that there were highly significant effect of treated and storage periods on proteolytic bacterial counts in the breast and leg meat during chilling conditions (Table 3).

The proteolytic bacteria numbers in the breast and leg meat of the frozen carcasses decreased gradually by the increase of freezing time until 90 days of storage, followed by a clear increase until the end of the experiment at 180 days of storage (Table 2). The treated carcasses had lower numbers of proteolytic bacteria than the untreated ones at any period of freezing weather the carcasses were packaged or not. The unpackaged carcasses being of lower humidity had lower proteolytic bacteria counts than the packaged ones. Packing preserve humidity which in turn encourage bacterial growth and the premeability of polyethylene bags gave relatively suitable conditions for bacterial growth since some of proteolytic bacteria are aerobic, and also the permeability permitted growth at optimal rates until dehydration or mold growth interfered. This result was also suggested by Holleck *et al.* (1958).

The difference in the proteolytic bacteria in the breast and leg meat due to packaging, treatments, storage periods and the interactions between them were highly significant, except in the leg meat. The interaction between packaging and treated was not significant (Table 4).

TABLE 1. Effect of chilling on total bacterial counts, psychrophilic bacterial counts and proteolytic bacterial counts in breast and leg meat.

Items	Storage periods (Days)					
	At slaughter	3	6	9	12	15
T.B.C. (1) (X 10^3) in breast meat Tr.)	29.50	11.50	9.50	16.00	505.00	8800.00
„ „ (Untr.)	29.50	23.00	19.00	305.00	8800.00	110800.00
T.B.C. (X 10^3) in leg meat (Tr.)	22.00	7.50	9.50	13.50	345.00	7422.50
„ „ (Untr.)	22.00	12.50	17.00	16.00	427.50	106000.00
Psych. B.C. (2) (X 10^3) in breast meat (Tr.)	17.00	9.00	8.75	9.50	335.00	8000.00
„ „ „ (Untr.) . .	17.00	15.50	15.00	225.00	7050.00	90000.00
Psych.B.C. (X 10^3) in leg meat (Tr.)	3.50	8.00	8.75	10.00	327.50	5375.00
„ „ (Untr.)	3.50	5.00	14.00	15.00	337.50	88700.00
Prote.B.C.(3) (X 10^3) in breast meat (Tr.)	0.725	0.150	0.130	0.170	0.320	6.100
„ „ (Untr.)	0.725	0.500	0.150	0.250	2.100	8.500
Prote.B.C. (X 10^3) in leg meat (Tr.)	0.350	0.100	0.450	0.140	0.290	5.500
„ „ (Untr.)	0.350	0.400	0.135	0.220	1.500	7.000

1. Total bacteria counts in one g of meat.
2. Psychrophilic bacteria counts in one g of meat.
3. Proteolytic bacteria counts in one g of meat.

TABLE 2. Effect of freezing on total bacterial counts, psychrophilic bacterial counts and proteolytic bacterial counts in breast and leg meat.

Items	Storage periods (Days)						
	At slaughter	30	60	90	120	150	180
T.B.C.(1) ($X10^3$) in breast meat (Pack., Tr.)	29.50	4.50	3.20	1.15	3.90	9.30	18.30
T.B.C. ($X\ 10^3$) in breast meat (Pack., Untr.)	29.50	7.10	6.00	5.20	15.10	131.00	564.80
T.B.C. ($X\ 10^3$) in breast meat (Unpack., Tr.)	29.50	7.60	6.00	4.80	2.86	2.95	1.88
T.B.C. ($X\ 10^3$) in breast meat (Unpack., Untr.)	29.50	7.90	6.00	5.50	4–13	4.30	5.68
T.B.C. ($X\ 10^3$) in leg meat (Pack., Tr.)	22.00	3.00	2.40	1.>	2.67	8.00	15.00
T.B.C. (X 10) in leg meat (Pack., Untr.)	22.00	3.70	3.56	2.70	14.90	102.00	203.30
T.B.C. ($X\ 10^3$) in leg meat (Unpack., Tr.)	22.00	7.60	5.00	4.00	2.39	2.29	1.42
T.B.C. ($X\ 10^3$) in leg meat (Unpack., Untr.)	22.00	7.75	6.50	5.00	3.78	3.96	4.30
Psych. B.C. (2) ($X\ 10^3$) in breast meat (Pack., Tr.)	1.70	2.50	3.30	1.05	4.70	9.50	19.00
Psych.B.C. ($X\ 10^3$) in breast meat (Pack., Untr).	1.70	4.50	4.20	4.25	16.20	115.00	512.00
Psych.B.C. ($X\ 10^3$) in breast meat (Unpack., Tr.)	1.70	5.30	5.00	5.50	2.11	2.45	1.64
Psych.B.C. ($X\ 10^3$) in breast meat (Unpack., Untr.)	1.70	6.00	4.50	4.10	3.90	3.50	4.25
Psych. B.C. ($X\ 10^3$) in leg meat (Pack., Tr.)	3.50	2.00	2.30	1.40	2.45	6.50	14.00
Psych. B. ($X.\ 10^3$) in leg meat (Pack., Untr.)	3.50	2.80	2.50	1.20	13.75	125.00	200.00
Psych. B.C. ($X\ 10^3$) in leg meat (Unpack., Tr.)	3.50	5.00	5.00	4.50	2.28	2.23	1.39
Psych. B.C. ($X\ 10^3$) in leg meat (Unpack., Untr.)	3.50	5.00	5.50	5.50	3.60	3.20	4.00

TABLE 2 (*Cont.*)

	Storage periode (Days)						
	At slaughter	30	60	90	120	150	180
Prote.B.C (3) (X 10^3) in breast meat (Pack., Tr.)	0.725	0.250	0.175	0.100	0.150	0.300	0.500
Prote.B.C(3) (X 10^3) in breast meat (Pack., Untr.)	0.725	0.250	0.200	0.150	0.175	0.400	0.600
Prote.B.C.(3) (X 10^3) in breast meat (Unpack., Tr.)	0.725	0.225	0.080	0.075	0.100	0.150	0.220
Prote.B.C(3) (X 10^3) in breast meat (Unpack., Untr.)	0.725	0.200	0.125	0.110	0.150	0.250	0.350
Prote.B.C(X10^3) in leg meat (Pack.,Tr.)	0.350	0.200	0.125	0.100	0.120	0.200	0.420
Prote.B.C(X 10_6) in leg meat (Pack ,- Untr.)	0.350	0.250	0.150	0.100	0.120	0.250	0.520
Prote.B.C(X 10^3) in leg meat (Unpack., Tr.)	0.350	0.210	0.070	0.060	0.095	0.120	0.200
Prote.B.C(X 10^3) in leg meat (Unpack., Untr.)	0.350	0.130	0.120	0.110	0.130	0.190	0.240

(1) Total bacteria counts in one g of meat.

(2) Psychrophilic becteria counts in one g of meat.

(3) Proteolytic bacteria counts in one g of meat.

TABLE 3. ANOVA for total bacterial counts, psychrophilic bacterial counts and proteolytic bacteral counts in breast and leg meat at chilling temperature.

Segments	S.V	d.f	M.S.	F
T.B.C. in breast meat	Between treatments	1	7.14	3570.00**
	Between periods	4	17.14	8570.00**
	Interaction	4	0.51	255.00**
T.B.C. in leg meat	Between treatments	1	0.25	125**
	Between periods	4	16.20	8100**
	Interaction	4	0.002	460**
Psych. B.C. in breast meat	Between treatments	1	4.16	1386.67**
	Between periods	4	15.43	5143.33**
	Interac tion	4	1.67	557.50**
Psych. B.C. in leg meat	Between treatments	1	0.07	23.33**
	Between periods	4	16 4	5480.00**
	Intera tion	4	1.09	363.33**
Prote.B.C in breast meat	Between treatments	1	1.46	48.67**
	Between periods	4	4.13	137.50**
	Interaction	4	0.16	5.17**
Prote.B.C. in leg meat	Between treatments	1	1.09	181.67**
	Between periods	4	3.82	636.67**
	Interaction	4	0.18	30.42**

** Highly significant ($P<0.01$)

TABLE 4. ANOVA for total bacterial counts, psychrophilic bacterial counts and proteolytic bacterial counts in breast and leg meat at freezing temperature.

Segments	S.V.	d.f.	M.S.	F
T.B.C.(1) in breast meat	Between packagings (P)	1	3.31	33100**
	Between treatments (T)	1	4.63	46300**
	Between periods (Pe)	5	1.05	10500**
	Interaction P x T	1	2.09	20900**
	P x Pe	5	2.09	20900**
	T x Pe	5	0.47	4700**
	P x T x pe	5	0.24	2400**
T.B.C. in leg meat	Between packagings (P)	1	2.00	153.85**
	Between treatments (T)	1	3.00	276.92**
	Between periods (pe)	5	0.77	59.23**
	Interaction P x T	1	0.89	68.46**
	P x Pe	5	1.90	146.15**
	T x Pe	5	0.12	9.23**
	P x T x Pe	5	0.38	29.23**
Psych. B.C.(2) in breast meat	Between packagings (P)	1	3.98	199.00**
	Between treatments (T)	1	3.78	186.50**
	Between periods (Pe)	5	1.15	57.80**
	Interaction P x T	1	1.86	93.00**
	P x Pe	5	2.41	120.50**
	T x Pe	5	0.44	21.80**
	P x T x Pe	5	0.16	7.90**

TABLE 4. (*Cont.*).

Segments	S.V.	d.f.	M.S.	F
Psych B.C. in leg meat	Between packagings (P)	1	1.92	64.00**
	Between treatments (T)	1	3.77	125.67**
	Between periods (Pe)	5	0.98	32.60**
	Interaction P x T	1	0.58	19.33**
	P x Pe	5	0.75	91.67**
	T x Pe	5	0.70	23.33**
	P x T x Pe	5	0.10	3.33**
Prote. B. C.(3) in breast meat	Between packagings (P)	1	0.83	138.33**
	Between treatments (T)	1	0.33	55.00**
	Between periods (Pe)	5	0.70	116.67**
	Interaction P x T	1	0.05	8.33**
	P x Pe	5	0.04	6.67**
	T x Pe	5	0.02	4.00**
	P x T x Pe	5	0.00	0.00
Prote. B. C. in leg meat	Between packagings (P)	1	0.63	157.50**
	Between treatments (T)	1	0.23	57.50 *
	Between periods (Pe)	5	0.62	156.00**
	Interaction P x T	1	0.00	0.00
	P x Pe	5	0.04	10.00**
	T x Pe	5	0.03	7.50**
	p x T x pe	5	0.03	7.50**

(1) Total bacterial counts.
(2) Psychrophilic bacterial counts.
(3) Proteolytic bacterial counts.
** Highly significant ($p < 0.01$).

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دراسات على تخزين ذبائح الدجاج الفيومى
ثانيا ـ التغيرات البكتريولوجية

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أجرى هذا البحث بكلية الزراعة ، جامعة القاهرة على ١٤٠ من دجاج الفيومى وذلك لدراسة تأثير غمر الدجاج فى محلول ١٠ جزء فى المليون من المضاد الحيوى كلورامفينكول (كلورومايستين) والتعبئة فى أكياس البولى ايثلين على أطالة مدة حفظ اللحوم للدجاج عند درجات حرارة التبريد (+٥م°) لمدة ١٥ يوم والتجميد (−١٠م°) لمدة ١٨٠ يوم ـ ذبح الدجاج عند عمر ١٦ أسبوع أخذت العينات للتقديرات البكتيريولوجية كل ٣ ، ٣٠ يوم فى التبريد والتجمد على التوالى وقد قدرت أعداد البكتيريا الكلية والمحبة للبرودة للبروتين فى لحم صدر وفخذ الذبائح .

أعداد البكتيريا الكلية والبكتيريا المحبة للبرودة والمحللة للبروتين فى لحم صدر وفخذ الذبائح :ـ

كان للمعاملة بالمضاد الحيوى تأثير كبير على خفض المحتوى البكتيرى فى صدر وفخذ الذبائح أثناء فترتى التبريد والتجميد ، لوحظ أثناء فترة التجميد أن المحتوى البكتيرى كان أقل فى صدر وفخذ الذبائح المغلفة سواء كانت معاملة أو غير معاملة عن الذبائح الغير مغلفة وذلك حتى ٩٠ يوم من فترة الحفظ وبعد ذلك لوحظ أن الذبائح المغلفة أصبحت تحتوى على أعداد أكبر من البكتيريا عن الذبائح الغير مغلفة حتى نهاية فترة الحفظ وهذا يرجع الى أن التغليف يحافظ على رطوبة صدر وفخذ الذبائح مما يزيد من تكاثر ونمو البكتيريا بينما الذبائح الغير مغلفة فقدت كمية كبيرة من رطوبتها مما جعل البيئة غير ملائمة لتكاثر البكتيريا خاصة بعد ٩٠ يوم من فترة التجميد .

لحم الصدر دائما يحتوى على أعداد أكبر من البكتيريا عن لحم الفخذ فى أى فترة من فترات الحفظ وتحت أى معاملة وهذا يرجع الى ارتفاع نسبة الرطوبة فى الصدر والتى تجعل بيئة لحم الصدر أكثر ملائمة لتكاثر البكتيريا عن لحم الفخذ .