A Survey on Pigment Content in Commercial Formula Feeds for Poultry in Japan

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It is well known that dietary xanthophyll pigment content has a direct influence on yolk colour of eggs. Some of the Japanese consumers has been complaining that most of yolks of commercial table eggs sold in the present Japanese market are too light in colour. However, little information is available indicating how dark the colour of the yolks is in commercial table eggs, or showing what is the colour of the yolks which satisfy the consumers complaining about it.

Results of a survey in 1968 indicated that the colour of the yolks in eggs sold in retail stores in Gifu area in Japan was, on average, 8.99 by Roche Yolk Colour Fan (RCF)¹⁾. In winter, the colour was somewhat darker, being 9.94, than that in summer, being 8.54. Another survey in 1980 of one of the authors²⁾ showed that members of Japanese Society of Food and Nutrition in Tokyo area was satisfied with yolk colour of 9.3 by RCF. The eggs tested in the latter survey were laid by hens fed a formula feed containing 18.0 ppm of total xanthophyll in National Institute of Animal Industry.

In this paper, results from a survey of the pigment content in commercial formula feed for poultry in Japan are presented.

Materials and Methods

One sample of chick starter feeds, 3 samples of broiler finisher feeds, and 6 samples of feeds for laying hens were taken in each of 4 seasons randomly from feed manufacturing factories in Tokyo, Osaka, and Kyushu areas in Japan. The samples were packed in coloured polyethylene bag under nitrogen and sent to Japan Food Research Laboratories, where the contents of total xanthophyll as described by KOHLER *et al.*³⁾ and of non-epoxide pigmenting xanthophyll as given by LIVINGSTON *et al.*⁴⁾ were analyzed. The sampling was started on October, 1981, and ended on August, 1982.

The data were firstly analyzed statistically as randomized block design to compare contents of total xanthophyll and non-epoxide pigmenting xanthophyll in 120 samples as blocks. Then, the data were analyzed as three factorial design with uneven but proportional replications⁵⁾. Types of feed, seasons and areas of sampling were the 3 factors. In this factorial design, values of total xanthophyll and non-epoxide pigmenting xanthophyll were analyzed individually, then they were treated as duplicated values of the pigment. An electronic computer ACOS 800II of the Computing Center for Research in Agriculture, Forestry and Fishery was used in the analysis of variance with a program written by one of the authors.

Results and Discussion

Xanthophyll results are given in Table 1. On average there was no significant difference between the total (12.1 mg/kg) and the non-epoxide (12.2 mg/kg) xanthophyll content of feeds; the correlation between the two sets of results was 0.975. The two sets of results were treated as duplicates in the factorial analysis.

In Table 2, analyses of variance of the three-factor factorial design are presented; the error variance, e_2 , was from between duplicate values. In this design, factor A, *i. e.*, the effect among types of feed, was treated as a fixed effect, while factors B and C, *i. e.*, effects among seasons and among areas of sampling, were treated as random-effects. The main effect A was F-tested as MS_1/MS_2 ; the main effects B and C were F-tested against interactions, AxB and AxC, respectively. The main effects A and B were significant, while the

Table 1. Content of pigment (mg/kg) in samples

Area	Type of feed		Total xanthophyll				Non-epoxide xanthophyll			
			Autumn	Winter	Spring	Summer	Autumn	Winter	Spring	Summer
Tokyo	Chick	1	11.0	9.1	11.0	7.6	12.0	8.6	11.3	8.0
		2	7.7	12.6	9.5	8.1	8.1	11.7	10.1	8.8
	Broiler	1	8.5	10.6	14.0	7.6	8.7	10.3	14.2	7.7
		2	9.4	7.7	12.2	9.7	9.0	7.1	12.3	10.4
	Layer	1	13.7	10.3	18.7	7.9	13.8	10.0	19.6	8.3
		2	12.3	14.8	14.6	9.6	12.7	14.5	14.6	11.0
		3	12.2	16.0	14.3	12.4	11.0	13.8	14.2	13.8
		4	11.1	16.3	17.5	14.4	11.2	15.5	17.6	15.2
		5	11.0	16.2	18.8	11.7	10.9	18.5	19.2	12.0
		6	10.1	12.2	10.7	8.7	11.7	11.9	10.3	8.9
Osaka	Chick	1	11.4	15.9	13.1	7.4	11.9	15.3	13.1	8.0
		2	11.1	15.9	12.4	10.2	12.6	16.2	12.3	10.2
	Broiler	1	10.1	10.9	12.6	7.6	10.2	9.9	13.1	7.6
		2	9.8	11.0	12.2	7.4	11.4	10.9	12.6	7.1
	Layer	1	20.2	11.9	15.5	7.4	20.3	12.5	15.9	8.0
		2	12.5	11.1	16.6	10.7	12.4	11.4	16.9	11.3
		3	15.1	9.0	14.3	12.0	14.9	9.5	14.2	9.9
		4	11.7	9.6	9.9	11.9	12.1	9.9	9.8	12.5
		5	16.1	8.8	12.9	12.8	16.4	9.9	13.1	14.1
		6	12.6	10.3	13.8	14.6	12.5	11.0	14.5	13.5
Kyushu	Chick	1	12.6	7.2	12.2	9.5	12.5	7.9	12.4	10.4
		2	10.6	9.5	13.7	9.0	10.4	9.4	14.5	8.9
	Broiler	1	8.6	11.8	13.3	7.9	9.0	12.9	13.8	6.2
		2	10.2	9.8	12.3	8.2	10.1	8.7	12.2	8.1
	Layer	1	8.8	9.3	16.5	7.7	8.5	8.6	17.1	9.0
		2	12.7	10.0	15.2	13.9	12.4	10.3	15.9	13.3
		3	14.6	10.4	15.2	13.2	13.9	11.3	15.7	13.0
		4	15.8	14.7	13.4	13.4	15.1	15.3	13.9	13.3
		5	12.3	21.2	17.8	14.6	12.1	22.1	18.2	14.4
		6	16.3	11.9	20.4	13.4	15.7	12.9	20.8	11.7

Table 2. Analysis of variance

Source of variation	Degrees of freedom	Mean square		
		Total xanthophyll	Non-epoxide xanthophyll	Mean of xanthophyll
Type of feed A	2	111.42** ¹⁾	111.12**	111.22**
Season B	3	73.04**	80.56**	76.66**
Area C	2	4.05	3.43	3.68
A × B	6	3.92	2.92	3.27
A × C	4	7.76	7.23	7.46
B × C	6	9.35	7.97	8.12
A × B × C	12	8.03	6.28	7.07
R (ABC)	84	6.05	6.36	6.10**
e ₂	120			0.249
MS ₁ ¹⁾	2	119.46**	117.40**	118.30**
MS ₂	7	11.69	10.15	10.73

1) Significant at 1% level.

2) $MS_1 = MS_A + MS_{A \times B \times C}$, $MS_2 = MS_{A \times B} + MS_{A \times C}$.

Table 3. Pigment content (mg/kg) of feeds by season and ration

Pigment Type of feed	Season				Mean ¹⁾
	Autumn	Winter	Spring	Summer	
Total xanthophyll:					
Chick	11.7	10.7	12.1	8.2	10.7 ^b
Broiler	9.6	11.1	12.5	8.4	10.4 ^b
Layer	13.3	12.6	15.3	11.7	13.2 ^a
Non-epoxide xanthophyll:					
Chick	12.1	10.6	12.3	8.8	11.0 ^b
Broiler	9.9	10.8	12.8	8.3	10.5 ^b
Layer	13.2	12.7	15.6	11.8	13.4 ^a
Mean of xanthophyll:					
Chick	11.9	10.7	12.2	8.5	10.8 ^b
Broiler	9.8	10.9	12.6	8.4	10.4 ^b
Layer	13.2	12.7	15.5	11.8	13.3 ^a
Mean ¹⁾	12.1 ^b	12.0 ^b	14.3 ^a	10.4 ^c	

1) Values having different superscript were different at 1% level.

main effect C and all of the interactions were not significant. The variance of replication, R(ABC), indicated the variation among samples of the same type of feed from different feed manufacturing factories at the same season in the same area. The variance was F-tested against chemical error, e₂, and was highly significant.

In Table 3, average contents of total xanthophyll and non-epoxide xanthophyll in each type of feed for each season are presented. On average, formula feeds for laying hens contained 13.3 mg/kg of xanthophyll; this was significantly higher than the levels in feeds for starting chicks and for broilers. Ration xanthophyll levels of 13 mg/kg would result in the production of eggs with yolk colours of about 7-8 by RCF^{6,7)}. Levels as high as 18 mg/kg would be required to achieve a satisfactory yolk color of 9.

Additional pigment can be supplied either by replacing cereals having no pigment such as milo and rice with yellow corn, or by supplementing with pigmenting agents. The two major supplementary sources of pigment in Japan are alfalfa (lucerne) meal (range in xanthophyll content 150 to 300 mg/kg for 17% crude protein product) and corn gluten meal (180 to 400 mg/kg xanthophyll⁶⁾). Leaf meal from China has also been used in rations but this product is no longer available. A further potential pigment concentrate for feed formulation in Japan is leaf protein concentrate prepared from green herbage. This product has a high xanthophyll content (range 950 to 1560 mg/kg⁷⁻⁹⁾). Studies have shown the efficiency of utilization by poultry of these xanthophylls to be similar to^{7,8)} or greater than⁹⁾, that for alfalfa meals. From the very high relative xanthophyll potency of 1092 mg/kg⁸⁾, it is calculated that only 1.6% of leaf protein concentrate would be required to provide a total ration level of 18 mg/kg. Recent developments in the production of leaf protein concentrates in France, the USA and New Zealand suggest this product may become a substitute for dwindling supplies of leaf meals in Japan.

The seasonal variation of pigment content in poultry feeds was unexpected, and the reasons for this are not clear. Similar trends were observed in all types of feeds and in all the areas sampled, the variation was not due to a change in formulation in a certain feed manufacturing factory in a certain area. Yellow corn imported from the United States, from which most yellow corn for feed use in Japan has been imported, was judged to be much lighter in colour than usual, and was suspected to be one of the reasons for the drop in xanthophyll content in the summer season. Another factor may be release of rice stocked by the Government after last May. In contrast, however, about 200,000 tons of yellow corn was imported from Thailand and South Africa, where dark-coloured yellow corn is produced. Most of the corn was included in feeds produced in the spring season, and this may be one of the reasons for the rise in xanthophyll in this season. The proportion of yellow corn and alfalfa meal, two major natural pigment sources, included in poultry feeds remained virtually unchanged in the last year. This seasonal variation in xanthophyll content indicates the necessity of more careful use of pigmenting agents in formula feeds, particularly for laying hens.

Finally, it is noted that results for xanthophyll and non-epoxide xanthophyll were, on average, indistinguishable. This may have been for two reasons; firstly the low levels of pigment in the materials being analysed and inherent variation in the procedures, and secondly because of a high proportion of corn and corn based products in the rations which have a high proportion of total xanthophylls comprised of lutein and zeaxanthin⁶⁾. Regardless, both procedures seemed to be effective for determining xanthophyll content of formulated poultry feeds.

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Reference

- 1) BRANT, A. W., S. SENDA, and T. TAKAMURA (1966) Egg quality in Gifu city, Japan : Poul. Sci.,

- 48 : 1968—1976.
- 2) YOSHIDA, M. (1981) Questionnaire on peelability of shell, taste of egg yolk and albumen, and yolk colour of hard-cooked eggs (in Japanese) : Japan. Poult. Sci., **18** : 360—365.
 - 3) KOHLER, G. O., KNOWLES, R. E., and LIVINGSTON, A. L. (1967) An improved analytical procedure for the determination of xanthophyll : J. A. O. A. C., **50** : 707—711.
 - 4) LIVINGSTON, A. L., R. E. KNOWLES and G. O. KOHLER (1973) Comparison of two methods for the analysis of pigmenting xanthophylls in dried plant materials : J. A. O. A. C., **56** : 1378—1381.
 - 5) YOSHIDA, M. (1975) Design of experiments for animal husbandry (in Japanese) : Yokendo, Tokyo, Japan.
 - 6) MARUSICH, W. L. and BAUERNFEIND, J. C. (1981) Oxycarotenoids in poultry feeds. In oxycarotenoids as colourants and vitamin A precursors (Technological and nutritional applications). Ed. by J. C. Bauernfeind. Academic Press. p. 319.
 - 7) BLUM, J. C. and GUILLAUMIN, S. (1979) Effectiveness of xanthophylls from different sources in egg and chicken colouring. Matieres Premieres et alimentation Des Volailles. October **19**. p. 7—21 (Translated from French).
 - 8) YOSHIDA, M. and H. HOSHII (1982) Nutritive and economic evaluation of leaf protein concentrates produced in New Zealand (in Japanese) : Japan. Poult. Sci., **19** : 45—52.
 - 9) KUMICKY, D. D., LIVINGSTON, A. L., KNOWLES, R. E., KOHLER, G. O., GUENTHER, E., OLSON, O. E., and CARLSON, C. W. (1977). Xanthophyll availability of alfalfa leaf protein concentrate for broilers and laying hens. Poult. Sci., **56** : 1504—1509.

日本で市販されている養鶏用飼料中の色素含量に関する調査

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昭和56年秋, 冬および昭和57年春, 夏の4季にわたって, 東京, 大阪および九州の3地域で生産されて市販された養鶏用飼料の色素含量を調査した。

サンプルは, 各地域ごとに, 4季それぞれについて, 育すう用飼料1点, ブロイラー仕上用飼料3点, 産卵鶏用飼料6点, 計10点ずつ, 飼料工場から無作為に採集した。総点数は120点となる。サンプルは, 日本食品分析センターにおいて, 総キサントフィルと非エポキサイド・キサントフィル含量を測定した。3元配置法で反復数が等しくないが比例している場合として分散分析した。

同一飼料の総キサントフィルと非エポキサイド・キサントフィルの含量には, 統計的に有意な差は認められなかった。どちらの場合でも, 飼料間差と, 季節間差が有意であったが, 地域間差と, すべての交互作用は有意ではなかった。

そこで, 飼料ごと, 季節ごとの平均値を求めた。育す

う用, ブロイラー用, 産卵鶏用の各飼料の色素含量の1年間平均は, それぞれ10.8, 10.4および13.3 ppmで, 産卵鶏用飼料で高かった ($P < 0.01$)。一方, 秋, 冬, 春, 夏の季節ごとの平均は, それぞれ12.1, 12.0, 14.3および10.4 ppmであって, 秋と冬の含量に比し, 春では高く, 夏に低くなっており, この差は有意であった ($P < 0.01$)。

消費者に好まれる卵黄の着色度を調査した結果によれば, ロツシュ・ヨーク・カラー・ファンの評点で9であり, この着色度の鶏卵を生産するためには, 飼料中の総キサントフィル含量は18 ppmが必要とされている。これと比較すると市販されている養鶏用飼料の色素含量は十分とはいえない。季節間の色素含量の変化の原因について考察した。

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