Designer Eggs: Nutritional and Functional Significance

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Abstract The rapid decline in per capita consumption of eggs over the past 50 years is one of the most challenging problems the industry is facing today. The negative perception related to the high cholesterol content in the egg is undoubtedly one of the major contributing factors. The long stigmatized fears on salmonella problems attached to egg products have played a significant role. In order to improve the consumer's perception, Dr. Sim has been interested in designing eggs free of these negative factors by manipulating chicken diets and/or the immune system of laying chickens. Dr. Sim's research has successfully introduced the Designer Eggs to combat the cholesterol problems to the world food markets. The Immune-Powered Eggs against Salmonella problems is just about to commercially be introduced in the food market. Now, the declining trend of egg consumption pattern has been reversed in North America, and has begun to rise since 1996. This change was attributed to new market surge of specialty eggs, like Designer Eggs. Dr. Sim's Designer Egg concept has served as a catalyst to both the scientific and egg sector industry community. The Canadian Designer Eggs concept has contributed to recapture consumers who have given up eating eggs because of cholesterol-phobia, to increase the overall per capita table egg consumption, to nurture the Canadian egg industry competitive in the world food market, to diversify egg industry by developing a variety of new products (functional foods) and, to stimulate the sagging egg industry, with spin off returns to the animal feed industry and even to the oilseed growers (flaxseed) of Canada.

Inroduction

In the industrialized world, animal products contribute more than 60% of total lipids, 70% saturated fats and 100% cholesterol of the diet. Consumer preference for animal products will likely continue. Thus it would be of national strategic importance in the fight against heart disease to design/modify animal products in such a way that dietary risks are minimized. Both epidemiological and clinical intervention studies have demonstrated a decrease of coronary heart disease mortality in people consuming relatively small amounts of omega-3 fatty acids (0.5g/day) over a long period of time. One large Designer Egg supplies more than 600 mg of most needed omega-3 PUFA and 6 mg tocopherols; it would still have additional beneficial effects for egg consumers due to its balanced ratios of PUFA/SAFA (1:1) and omega-6/omega-3 PUFA (1:1). Therefore, designer egg may offer an alternative choice of food product to today's nutrition-health conscious egg consumers around the world.

Similar efforts should be made to design other animal products to achieve the ultimate goal of minimizing the dietary risk of coronary heart disease. Consumers have begun to take control of their own health. They are driving the market for a new category of foods with the potential for health promotion well beyond the traditionally recognized benefits. It is clear that this rapidly emerging area of designer egg production has enormous

market potential. An increasingly competitive world market environment requires that industry concentrates on producing what the market needs rather than simply supplying what we produce. The egg industry has been very responsive in seeking new technology to improve the consumers negative perception on the egg associated with cholesterol, salmonella, and even allergy problems attached to egg products. In this paper I would like to introduce the Designer Egg Concept and its nutritional and health implications, and to present data pertaining to egg lipids and cholesterol stability in omega-3 PUFA enriched egg yolk and the ways of preventing from autooxidation.

Food Lipids and Egg Industry

The rapid decline in per capita consumption of eggs over the past 30 years (Figure 1) is a most challenging problem facing the egg industry in many parts of the world. The negative perception related to the high cholesterol content, 195 - 250 mg/egg (1) is undoubtedly one of the major contributing factors. Consumer's attitudes towards lipid in general have changed their attitude towards egg consumption because of fear that egg cholesterol will raise their blood cholesterol levels. Eggs therefore have been singled out by diet-heart advocates as a food to be avoided (2) even though the egg contains the best and least expensive high quality protein and balanced distribution of minerals and vitamins, except vitamin C (3).

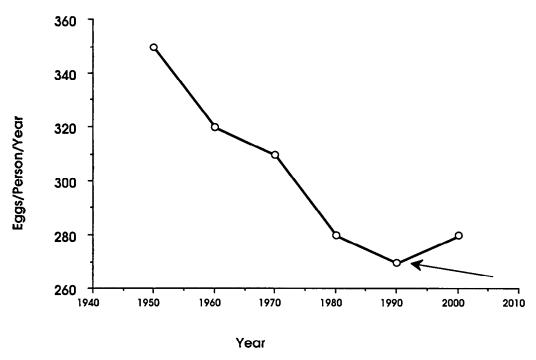


Figure 1. Egg Consumption Pattern in US during the last 50 Years. Now, the declining trend of egg consumption has been reversed in North America, and has begun to rise since 1996 (arrow). This change was attributed to new market surge of specialty eggs, like Designer Eggs

The first response to "cholesterol phobia" was the extensive investigation into factors, genetic, dietary and pharmacological in nature that would reduce cholesterol content of eggs. However, various attempts to reduce the cholesterol content (5) or produce cholesterol-free products have met with no success. Due to the lack of success in attempts to significantly reduce cholesterol levels in the egg, researchers began to investigate alternative manipulation to improve the nutritional quality of the egg and re-establish its position as a healthy and safe food item.

According to the diet-heart hypothesis, the amount and type of dietary lipids influence plasma and lipoprotein lipid levels, which in turn, increase the risk of coronary heart disease (6). The principal nutrient-related health problems in North America arise from the over consumption of lipid, mainly of animal origin (57-75 %). The National Institute of Health (7) and Health Canada (8) have adopted recommendations and target dietary guideline to limit lipid intake and modify the type of lipid consumption (Figure 2). Thus, animal agriculture must respond to the perceived needs of consumers by producing foods that are more healthful and reflecting the national nutritional guidelines. The egg industry in particular has been greatly encouraged to intensify efforts in developing and marketing products that would facilitate adherence to the dietary guidelines or national recommended target levels for fat and cholesterol.

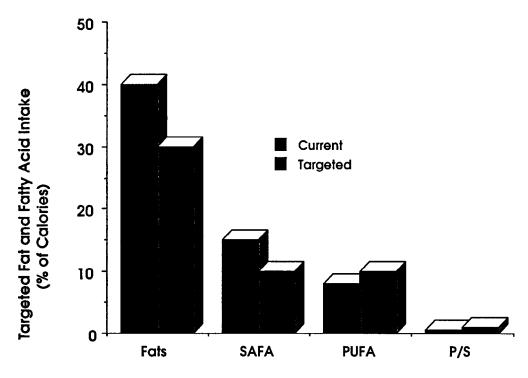


Figure 2. National Health Organizations Recommended Dietary Guidlines or Target Levels of Dietary Fats by Reducing Total Fat Intake to less than 30, SAFA intake to less than 10 and PUFA intake to 10 % of total caloric intake.

Omega-3 Fatty Acids

The pioneering discovery that omega-3 fatty acids protect against coronary heart disease in Greenland Eskimos consuming fish (9) has generated much research over the past decades on the various health benefits of dietary omega-3 fatty acid from fish oils. Researchers around the world have focused on the health effects of the dietary supply of omega-3 fatty acids, partly because those fatty acids have been reported to protect against cardiovascular and inflammatory diseases, as well as certain types of cancer (10, 11) and partly because it has been shown that omega-3 fatty acids are essential nutients for adults and children (12, 13). The benefits of dietary omega-3 PUFA include, among others, reduction in plasma triglycerides, blood pressure, platelet aggregation, throbosis and atherosclerosis particularly in diabetics, tumor growth, skin disease and enhanced immunity. The Canadian Government (8) adopted a recommendation that omega-3 polyunsaturated fatty acids are essential nutrients, thus recommending that the dietary supply should be at least 0.5% of the energy intake as linolenic acid. When the diet of infants contains no EPA and DHA, then LNA should be supplied as 1% of energy intake. This recommendation was made on the basis of the fact that North American diets are depleted of the omega-3 fatty acids.

The egg industry has stepped in to fill this gap and has begun to return the omega-3 fatty acids into the food supply. The ratio of omega-6 to omega-3 fatty acids is important, and the current high ratio should be reduced to less than 4 to 1 (14). To date the main supply of omega-3 fatty acids in human diet has been fish and fish oil. The major omega-6 and omega-3 PUFA are summarized in the following Table.

Table 1. Major omega-6 and omega-3 fatty acids

Linoleic acid (LA)	C18:2w6
Arachidonic acid (AA)	C20:4w6
Linolenic acid (LNA)	C18:3w3
Eicosapentaenoic acid (EPA)	C20:5w3
Docospentaenoic acid (DPA)	C22:5w3
Docosahexaenoic acid (DHA)	C22:6w3

Designer EggsTM

The fatty acid composition of yolk fat can readily be modified by diet (15, 16). In recent years several researchers have investigated the ability of the hen to enrich the egg with omega-3 fatty acids. The incorporation of omega-3 PUFA into egg yolk fat was easily accomplished by feeding laying hens diets containing flaxseed (17, 18, 19, 20) and fish oils (21, 22). The egg lipid composition is the result of a combination of de novo lipogensis and incorporation of lipid components from the diet. Another factor regulating the quantity and

the type of fatty acid deposition is the feed back inhibition of dietary long chain PUFAs (23). Therefore, it is feasible to alter the fatty acid composition of poultry products through dietary manipulation of long chain PUFAs and to design food products reflecting the nutritional guidelines, supplying omega-3 fatty acids while having optimal PUFA:SAFA and omega-6:omega-3 fatty acid ratios.

Sim et al (24, 25, 26) have carried out a series of studies to enhance the value of chicken eggs enriched in omega-3 fatty acids (500-600 mg/egg) with a significantly elevated PUFA:SAFA ratio (from 0.6 to 1.02), and lowering the omega-6:omega-3 fatty acid ratio (from 10:1 to 1:1). One large egg can supply about 600 mg of total omega-3 fatty acids (balanced with DHA, DPA and EPA) equivalent to approximately 100 g serving of fish (Figure 3). A consumer survey indicated the public's interest in omega-3 fatty acid-enriched eggs as a dietary alternative to fish (27).

Although LNA was the major omega-3 fatty acids deposited in the egg yolk, a considerable amount of longer chain omega-3 fatty acids were also incorporated into the phospholipid fractions of the yolk lipids. The hens fed flaxseed produced eggs enriched with omega-3 fatty acids (7-12% of yolk lipids) in the following order: LNA > DHA > DPA >EPA (28). This indicates that laying hens can convert dietary LNA to EPA, DPA and DHA via the desaturase and elongase enzyme systems (29). AA, the metabolite of linoleic acid (omega-6 fatty acid), was significantly reduced. Consequently, the ratio of omega-6 to omega-3 fatty acid was significantly decreased in the omega-3 fatty acid enriched eggs (from 10:1 to 1:1).

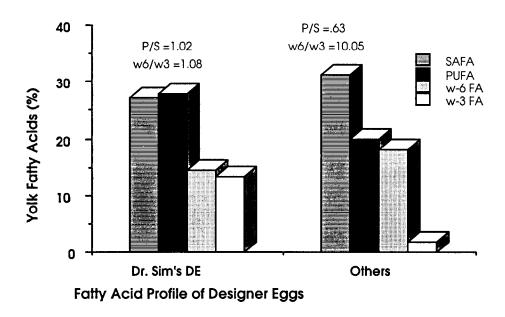


Figure 3. Fatty acid profile comparison of omega-3 fatty acid-enriched egg (DE) and regular egg (others). P/S = Polyunsaturated/Saturated, SAFA=Saturated fatty acids, PUFA = Polyunsaturated fatty acids, w-6 FA = omega-6 fatty acid, w-3 FA=omega-3 fatty acid.

Nutritional Significances

A series of studies were carried out to examine the influence, if any, omega-3 PUFA enriched eggs have on plasma cholesterol and tissue fatty acid modification in animals and human. Plasma and liver tissue cholesterol levels and fatty acid composition were analyzed after feeding omega-3 fatty acid enriched eggs to rats (30), as an animal model, and humans (31), as egg consumers.

Plasma Lipids

The eggs were hard boiled, yolks were removed, pulverized and dried. Dry yolk powders were incorporated into a semisynthetic diet at a 15% level and fed to weaning female Sprague-Dawley rats for 4 weeks. The blood and liver cholesterol levels and fatty acid composition were determined at the end of the feeding period. Feeding omega-3 PUFA enriched eggs reduced both plasma and liver total cholesterol contents by 20 and 38%, respectively (Figure 4).

Twenty-four healthy male student aged 18 - 32 were recruited and randomly divided into two groups of 12 each. Each subject had two eggs at breakfast. Before the start and at the termination of the 18-day study, subjects fasted for more than 12 hours to take blood samples at the University Health Clinic. There are clear patterns to state that consumption of Designer Eggs does not provoke plasma cholesterol despite their high inherent cholesterol content, increases HDL-cholesterol, suppresses LDL-cholesterol, and produces a marked reduction of plasma triglyceride levels (Figure 5). Consuming Designer Eggs also enriches body tissue lipids with omega-3 fatty acids, in particular phospholipids. Similar results were reported by other researchers. Ferrier et al. (32) found that human consumption of LNA enriched eggs decreased serum triglycerides and increased omega-3 fatty acids, particularly DHA, which accummulated in platelet phospholipids.

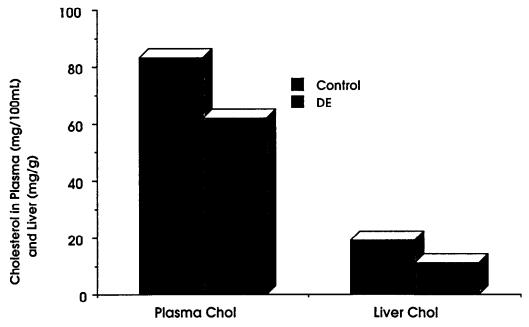


Figure 4. Plasma and liver cholesterol levels of rats at the end of 28-days of being fed yolk powder with and without omega-3 PUFA enrichment. Source: Jiang and Sim (30)

The results demonstrate that the cholesterolemic and lipidemic properties of chicken eggs can be modified by designing the fatty acid composition of egg yolk lipids through chicken diets. The Designer Egg concept offers an alternative choice of food production to today's nutrition-health conscious consumers around the world. Eggs, which have been considered an atherogenic food, are now safely consumed if enriched with omega-3 fatty acids.

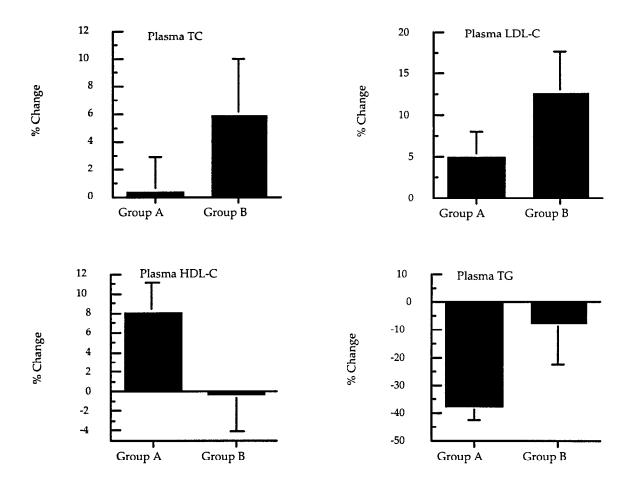


Figure 5. Percentage change in plasma total cholesterol (TC), low density lipoprotein cholesterol (LDL-C), high density lipoprotein cholesterol (HDL-C), and plasma triglycerides (TG) levels in human subjects after consuming 2 omega-3 PUFA enriched DE (A) or RE (B) with their habitual diets for a period of 3 weeks. Source: Jiang and Sim (31).

Mother's Milk

Aware of the importance of omega-3 PUFA to infants (37), the magnitude of changes in the fatty acid composition of breast milk and plasma fatty acids of eight nursing women were studied upon consumption of two Designer Eggs for a period of six weeks. Consuming two eggs as a part of their normal daily meal for 6-wk resulted in a significant deposition of total omega-3 fatty acids at 3.6 % compared with 1.9 % for the pretest milk and a reduction in omega-6:omega-3 fatty acid ratio from 6.7 to 3.0. The longer chain EPA and DHA comprised 1.2 % compared with 0.4% in the pretest milk. Consuming omega-3 enriched eggs did not alter the AA content in the milk. This phenomenon made the milk fat more favourable by increasing the DHA to AA ratio from 0.75 to 1.2 (Figure 6). Total

palsma cholesterol and triglyceride levels were not affected (38). The omega-3 PUFA-enriched eggs (DE) contained about 690 mg of total omega-3 fatty acid with 165 mg of long chain n-3 fatty acids (EPA, DPA and DHA). Assuming the intake of a 1-month old infant to be 794 mL, infants nursed from women consuming Designer Eggs could have over 300 mg of long chain omega-3 fatty acid such as EPA, DPA and DHA. Thus, a diet supplemented with Designer Eggs or their egg oils should provide an alternative way of supplying omega-3 fatty acids for breast fed infants. These differences in the omega-3 fatty acid content of breast milk could have implications for the development of suckling infants.

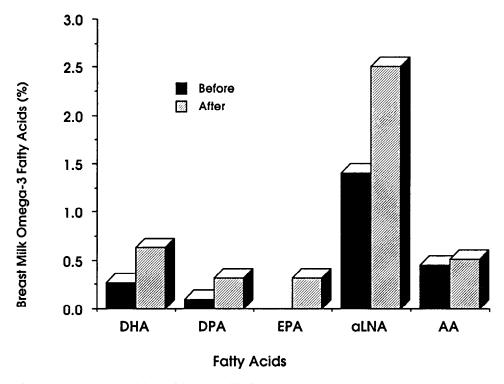


Figure 6. Fatty acid composition of breast milk from nursing mothers before (Basal) and after (6th Wk) consuming 2 Designer Eggs daily for a period of 6-weeks.

Infant Food Oils

The egg yolk consists of lipids and protein. More than 66 % of the total dry yolk mass are fats. An average egg provides about 6 g of lipids which are contained exclusively in the yolk. One Designer Egg can supply about 600 mg of total omega-3 PUFA to the human diet with elevated levels of DHA and AA in the egg. Both the DHA and AA are essential for proper brain development of infants (40). The designer egg is a potential vehicle to provide the much needed DHA and AA, which more closely mimics the fatty acid composition of human breast milk (Figure 7). In contrast to human milk, most commercial infant formulas are based on soybean oil and contain about 53% LA and 7% LNA. Most infant formulas contain LA and LNA in amounts comparable to those in human milk, but they lack long chain omega-3 PUFA contents, DHA and AA in particular (39). It is crucially important to supply the preformed essential long chain PUFA both omega-3 DHA and omega-6 AA in an adequate balance (37). Currently available infant formulas do

not contain fatty acids above 18 carbons (Table 2). The primary goal of infant formulas is to mimic the growth and development of the breast-fed infant (39).

Table 2. Essential fatty acid profile of commercial infant formulas (Survey conducted)

<u>%</u>	Commercial. Infant Formulas					<u>D E Oils</u>	<u>HBN</u>
LA	13.9	22.0	23.6	31.2	31.1	13.0	9.9
LNA	1.9	3.1	3.5	4.4	4.1	9.3	2.4
AA						1.6	0.4
DHA						2.2	0.3

LA (18:2w6), LNA (18:3w3), AA (20:4w6), DHA (22:6w3), DE (designer egg), HBM (human breast milk)

One approach to improve a milk formula is to match the composition of human breast milk, which contains both DHA and AA (Figure 7). Designed egg yolk oils provide an adequate amount of omega-3 and omega-6 precursors and LC PUFAs, including DHA, while still sustaining a significant level of AA in the egg with various ratios of omega-6/omega-3 ranging from 20 to 1. Industry and scientific community alike are searching for a new oil source supplying both long chain omega-6 and omega-3 PUFA fatty acids. Since the fatty acid make-up of designer eggs resembles that of human milk fat, yolk oil may be regarded as essential oil base for infant food industry.

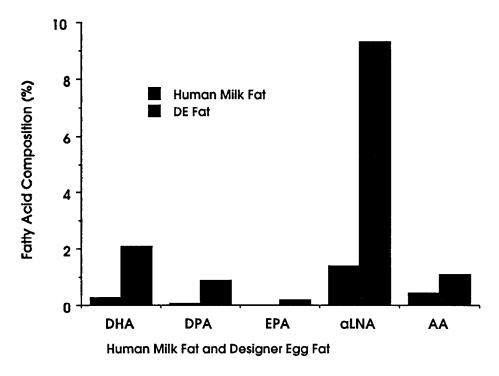


Figure 7. Long chain (LC) n-6 and n-3 fatty acid concentrations in human milk and Designer Egg Oil.

Designer Egg Oil

An average egg also provides about 6 g of lipids which are contained exclusively in the yolk. More than 66 % of the total yolk mass are fats, thus the yolk from these n-3 PUFA enriched eggs can be regarded as a potential oil crop rich in long chain polyunsaturated fatty acids, the both DHA and AA essential for infants. Designer Egg yolk oil may be regarded as an essential oil base for infant formula because it resembles the fatty acid composition of human milk. Designed egg yolk oils provide an adequate amount of n-3 and n-6 precursors and LC PUFAs including DHA still sustaining a significant level of AA in the egg with various ratios of n-6/n-3 in a range of 22.1 to 1.4.

There is an excellent possibility and a unique opportunity to diversify egg uses. Egg oil extraction-purification technologies developed at the University of Alberta is presently available to be exploited for an industrial application. A bench top model of egg oil extracting technology directly from fresh egg yolk using an aqueous solvent system and, subsequently partitioning into the neutral oil and lecithin fractions by cold precipitation technique was devised and patented as a potential technology to be exploited (Figure 8). Subsequently, a scaled-up process has been jointly undertaken in collaboration with food industry partners (Technology of egg oil extraction and Fractionation of lecithin) from fresh egg yolk (Canadian Patent Application No. 612,411, September 21, 1989, European Patents, Japan, South Korea, Finland under patent cooperation treaty, PCT No. 8150, July 10, 1990).

Oxidative Stability

There are potential risks associated with high-level of long chain PUFAs in foods. Autoxidation of long chain polyunsaturated fatty acids occurs in feeds and egg products. Cautions are warranted in scaling up the production of omega-3 PUFA enriched eggs, egg oils and their infant food applications, since the susceptibility of food lipids to oxidation is closely associated with the degree of unsaturation. A series of experiments to investigate the lipid stability from lipid oxidation and ways of preventing them from autooxidation has been conducted.

Chicken Feed

In our earlier study, feeds containing large amount of flaxseed has been associated with a fish flavor or lower sensory quality of egg products during storage. This was attributed to a combination of lipid rancidity in the feed and lipid peroxidation in the chicken tissues and eggs (33). Therefore, we conducted an experiment to monitor the chemical changes in the flaxseed, the very source of dietary omega-3 fatty acid in the poultry feed under various storage conditions. The changes in the content and stability of alpha-linolenic acid found negligible in the early stage of storage but oxidation potential markedly increased after the period of 60 days. Results indicated that the intact flaxseed (whole) is well protected from lipid oxidation by the presence of its intrinsic tocopherol content, and further supplementation of tocopherol significantly extends its stability even when flaxseeds are physically broken open for feed compounding and storage. This study confirm that flaxseed contains a sufficient amount of natural antioxidants as tocopherols, and protect dietary omega-3 fatty acids in the chicken feed from autooxidation within 60 days (41).

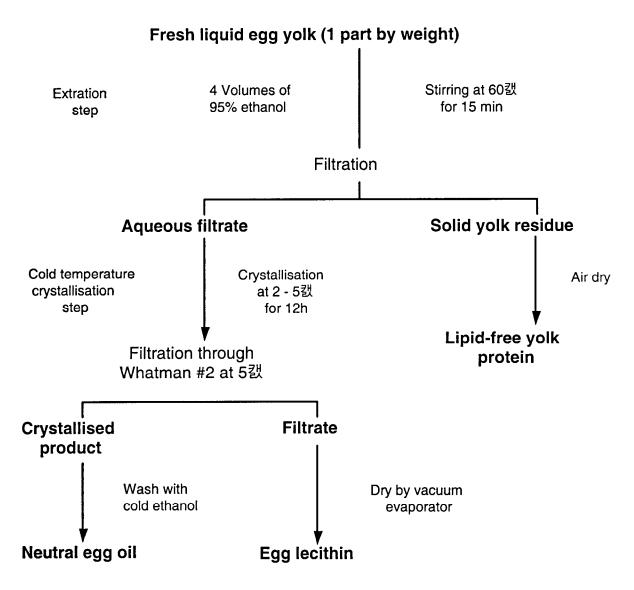


Figure 8. Schematic flow chart of sequential extraction, fractionation, and purification procedure for egg oil and lecithin from fresh yolk.

Antioxidants Tocopherols

In the early developing stage of Designer Egg, an off-flavor (fish-taint) from eggs enriched with omega-3 PUFAs was experienced. We suspect that the fish flavor generation could be the result of rancidity of omega-3 fatty acids either in feeds and/or animal products. This problem was dealt by stabilizing the dietary source of omega-3 PUFA with a natural form of tocopherols before incorporating into chicken feeds (42). A significant reduction in off-flavor, volatile compounds, cholesterol oxidation products and TBA-reacting substances was achieved in both feed and egg products by stabilizing the dietary fats before incorporating into chicken feed with natural form of tocopherols as antioxidants (34, 35, 36). Supplementing antioxidants into chicken feed not only effectively eliminates the off-flavor problem, but also greatly improves stability of egg products. The tocopherol

concentrations of egg yolk increased linearly with increasing levels of dietary tocopherol levels (Figure 9).

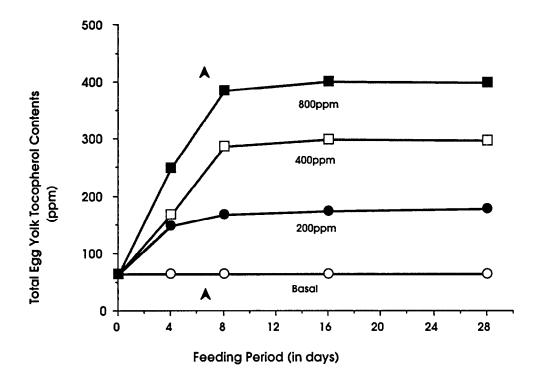


Figure 9. Supplementation of natural tocopherols at various levels into chicken feed reaches a plateau at 8th day of feeding. Tocopherol deposition into egg yolk is proportional to the dietary levels.

With increasing levels of dietary tocopherol supplementation in laying hen diets, malondialdehde contents in the egg yolk decreased from 41 to 18 nmol / g of egg yolk (Figure 10). This proves that lipid stability could be improved by increasing tocopherol contents of eggs. Reduction of the oxidation products and increasing the tocopherol concentration (antioxidants) in the yolk were major technological breakthroughs eliminating the preceding sensory and off-flavor problems. Thus, Designer Eggs not only supply stable form of essential omega-3 PUFAs, but natural form of tocopherols including Vitamin E to today's most health conscious consumers.

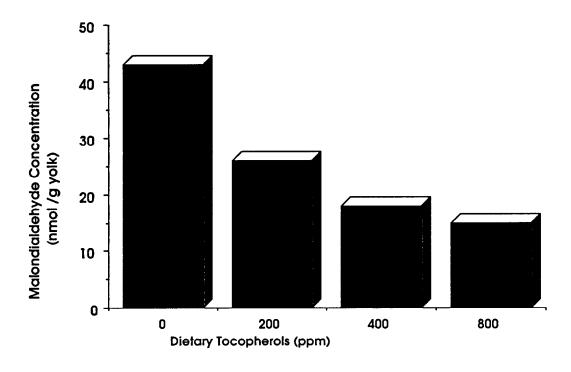
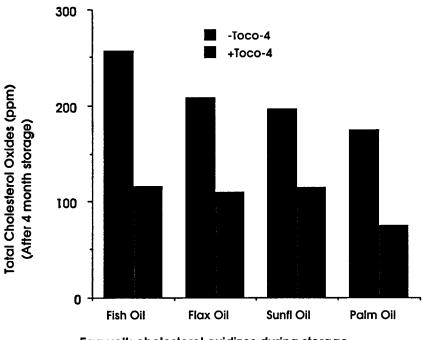


Figure 10. Effect of tocopherol supplementation on lipoperoxide level in the egg yolk. Egg yolk lipid stability was greatly improved by increasing tocopherol contents of eggs.

Cholesterol Oxidation

Cholesterol undergoes autooxidation in the presence of light and molecular oxygen, through a free-radical reaction, and forms cholesterol oxide products which are considered potent atherogens and carcinogens to human (43, 44). The presence of several cholesterol oxide products in commercial egg products have been reported, but little information about cholesterol stability in omega-3 fatty acid-enriched eggs and ways of prevention is available. Considering the importance of PUFAs and tocopherols as antioxidants in lipid oxidation, the effect of feeding flax, sunflower, palm and fish oils, with and without tocopherols, to laying hens was investigated on oxidative stability of cholesterol in the egg volk. Results show that cholesterol oxidation is accelerated by the presence of long chain polyunsaturated fatty acids in the order of fish oil > flaxseed oil > sunflower seed oil > palm oil. The initial levels of cholesterol oxides were 7 - 10 ppm and reached over 200 ppm within 4-month storage period. Cholesterol oxide formation was further accelerated by heat at 110°C for 22 hours. Feeding tocopherol supplements to laying hens increased intrinsic tocopherol content in eggs (Figure 9), and the presence of increased tocopherols significantly reduced the formation of cholesterol oxides in the egg yolk regardless of its fatty acid profiles (Fig 11). Feeding laying hens with tocopherol supplemented diets can delay or prevent the cholesterol oxidation potentials of omega-3 PUFA-enriched eggs during storage and processing. This study suggests that cholesterol oxide formation in omega-3 PUFA-enriched eggs can be prevented by adding Vitamin E or natural form of tocopherols, which would benefit the food industry and human health.



Egg yolk cholesterol oxidizes during storage.

Tocopherol protects cholesterol from oxidation.

Figure 11. Effects of dietary fatty acids and tocopherol supplementation on cholesterol oxidation in egg yolk. The cholesterol oxidation was significantly reduced in the presence of tocopherol.

Salmonella Problem

It has been recognized that chickens are common hosts to Salmonella as a food borne bacterium. If ingested in large numbers, humans can be infected, resulting in nausea, vomiting, abdominal cramps, diarrhea, fever, and headache. Fear of Salmonella poisoning is widespread in the public's perception associated with poultry meat and eggs, and efforts have been made by the regulatory agencies and poultry related food industries to control and prevent further outbreaks (45).

Twelve 40 wk-old Single Comb White Leghorn (SCWL) hens were immunized with lipopolysaccharide (LPS) antigens from Salmonella typhimurium. The immune response of laying hens resulted in the production of antibodies specific to LPS, a major constituent of the outer membrane of Gram-negative bacteria. Measuring antibody activities in egg yolk by ELISA monitored the immune response of laying hens to LPS antigens from Salmonella. The level of anti-LPS IgY activity increased after a booster injection (Fig 3). It was shown that immunization of chickens with LPS antigens led to a strong immunogenic reaction, producing a large amount of LPS-specific IgY. It was also found that the activity of antibodies against a LPS fraction containing lipid-A was higher than that of those against a LPS fraction lacking lipid-A. The results indicated that IgY specific for the LPS fraction might be useful in the prevention of Salmonella adhesion and diseases (45).

SCWL chickens also showed relatively strong immune response against both Salmonella enteritidis and S. typhimurium whole cells. The change of activity of IgY in egg yolk water soluble fraction (WSF) was monitored by ELISA using bacterial whole cells as an antigen during the immunization period (up to 9 wks), when the booster injections were given at wk 2 and 4 after the first injection. The activity of IgY against S. enteritidis increased continuously 1 wk after the initial immunization and reached a peak (OD at 405nm of samples diluted 1:1,000 was 1.02) at wk 8. The average (\pm standard deviation) concentrations of protein and total IgY in egg yolk WSF were 32.87 ± 1.25 and 7.08 ± 1.24 mg/ml, respectively, which indicated 21.5% of purity of IgY in WSF. Those of egg yolk WSF containing IgY against S. typhimurium were 33.08 ± 1.75 and 7.09 ± 1.40 mg/ml, respectively. The purity of IgY (21.4%) was almost the same as that of IgY against S. enteritidis. S. typhimurium-specific IgY also showed increasing activity 1 wk after the initial injection. The level of activity fell slightly between wk 4 and 5 and then rose again to the maximum of approximately 0.74 at wk 7. The maximum value of S. enteritidis-specific IgY activity was 1.38 times higher than that of S. typhimurium-specific IgY.

Immune-Powered Designer Eggs

S. enteritidis was incubated with IgY in a liquid medium to observe the growth inhibitory effect of IgY. After 6h incubation, there was the difference of 1 log CFU/ml in the colony counts of bacteria with specific IgY compared to those of bacteria with control IgY. Specific IgY powder with growth inhibitory effect contained 129 mg/g of total IgY (21.9% of purity). The concentration of specific IgY, which was effective in the growth inhibition, was 1.8 mg/ml of media. The growth inhibition by specific IgY at the concentration of 0.9 mg/ml of media was also observed resulting in the difference of the colony counts (0.9 log CFU/ml) between specific and control group after 6h incubation.

The growth of *S. typhimurium* was inhibited by *S. typhimurium*-specific IgY as well. The growth of bacteria with specific IgY was not observed after 4h of incubation time while that of bacteria with control IgY increased by 1.6 log CFU/ml. The purity of IgY powder was 23.1% and the concentration of specific IgY was 20 mg/g of powder. Specific IgY had an inhibitory effect on the bacterial growth at the concentration of 0.9 mg/ml of media.

Anti- S. enteritidis IgY showed high cross-reactivity with S. typhimurium by 50% of activity when reacting with S. enteritidis by 100% of activity (measured by ELISA). Anti-S. typhimurium IgY also cross-reacted with S. enteritidis by 44% of activity. The results indicated that IgY specific for S. enteritidis or S. typhimurium could inhibit the growth of both bacteria although the bacteria interacted with IgY differed from the antigen used to produce IgY.

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