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Effect of feeding CLA on plasma and granules fatty acid composition of eggs and prepared mayonnaise quality

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ABSTRACT

Eggs rich in *trans*, *trans* conjugated linoleic acid (CLA) are significantly more viscous, have more phospholipids containing linoleic acid (LA), and more saturated triacylglycerol species than control eggs. However, the fatty acid (FA) composition of yolk plasma and granule fractions are unreported. Furthermore, there are no reports of mayonnaise rheological properties or emulsion stability by using CLA-rich eggs. Therefore, the objectives were (1) compare the FA composition of CLA-rich yolk granules and plasma, relative to standard control and LA-rich control yolks, (2) compare the rheological properties of mayonnaise prepared with CLA-rich eggs to control eggs and (3) compare the emulsion stability of CLA-yolk mayonnaise. CLA-rich eggs and soy control eggs were produced by adding 10% CLA-rich soy oil or 10% of control unmodified soy oil to the hen's diet. The eggs were used in subsequent mayonnaise preparation. CLA-yolk mayonnaise was more viscous, had greater storage modulus, resisted thinning, and was a more stable emulsion, relative to mayonnaise prepared with control yolks or soy control yolks.

1. Introduction

Conjugated linoleic acid (CLA) is an 18-carbon fatty acid, with various positional and geometric isomers, and is found mostly in dairy and beef products (Whigham, Cook, & Atkinson, 2000). CLA has been shown to possess many positive human health effects, including anti-carcinogenic properties (Cesano, Visonneau, Scimeca, Kritchevsky, & Santoli, 1998; Kim et al., 2002), the capacity to combat obesity and atherosclerosis (Feitoza, Pereira, Ferreira da Costa, & Ribeiro, 2009; Nicolosi, Rogers, Kritchevsky, Scimeca, & Huth 1997), decrease the risk of diabetes (McGuire & McGuire, 1999), and improve immune function (Bassaganya-Riera et al., 2012).

Approximately 3.2 g of CLA need to be consumed daily to realize the health benefits (Berven et al., 2000; Mougios et al., 2001). However, consumption of ruminant CLA sources are limited because they are also high in saturated fat and cholesterol (McGuire & McGuire, 1999; Mougios et al., 2001). Jain, Proctor, and Lall (2008) developed a process to produce a 20% CLA-rich soy oil by photoisomerization of soybean oil linoleic acid in the presence of an iodine catalyst. *trans*, *trans* (*t*,*t*) CLA was the predominant CLA geometrical isomer. Gilbert, Gadang, Proctor, Jain, and Devareddy (2011) demonstrated that when obese Zucker rats consumed this CLA-rich soy oil, their total serum cholesterol and LDL cholesterol were reduced by 41% and 50%, relative to rats fed a soy oil control diet. Also their liver lipid contents and weights were reduced by 35% after 30 days. Recently, *t*,*t* CLA has shown greater anti-carcinogenic and anti-atherogenic effects in mouse and rat models, in comparison to *cis*, *trans* isomers (Islam et al., 2008; Shah, Baum, & Proctor, 2014).

The CLA-rich soybean oil was subsequently fed to chickens as 1.5% of their diet to produce CLA-rich egg yolks (Shinn, Gilley, Proctor, & Anthony, 2015a). Maximum CLA yolk accumulation occurred after 12 days of feeding, with *t*,*t* CLA isomers being the most abundant, followed by *trans*-10, *cis*-12 CLA.

CLA-rich and control yolks were also produced to determine the fat content and intact triacylglycerol and phospholipid species present (Shinn et al., 2014). Extractions resulted in a total lipid content of 34.7 ± 0.05% per volume of yolk with no significant difference in egg type.

A later study on the effects of CLA-rich soy oil on hen egg quality (Shinn, Gilley, Proctor, & Anthony 2015b) showed that eggs obtained with the CLA-rich soy oil diet had similar yolk size to

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the non-CLA controls and saturated fat was increased by only 28%. Furthermore, fresh CLA-yolks were significantly more viscous than fresh control yolks, and CLA-rich yolks maintained viscosity for 20 d, while control yolk viscosity significantly decreased (Shinn et al., 2015b). CLA-rich yolk viscosity had not been previously reported. These findings were in contrast to previous CLA egg enrichment studies with *cis*, *trans* CLA mixtures which produced smaller yolks and up to a 34% increase in yolk saturated fatty acids (Aydin, Pariza, & Cook, 2001).

The yolk comprises of granules, consisting of 10% lipids, suspended in a liquid plasma, containing 90% lipids (Belitz & Grosch, 1986). Low-density lipoprotein (LDL) is present in both the granule and plasma yolk fractions and is regarded as responsible for yolk's emulsifying properties (Mine & Bergougnoux, 1999). The plasma contains predominantly triacylglycerols, while the granules contain the majority of phospholipid species (Anton & Gandemer, 1997). Mayonnaise is an oil-in-water emulsion containing at least 65% oil and egg yolk (U.S. FDA) and the yolk provides the emulsifiers to stabilize the emulsion. The "film" around the emulsified oil droplets in mayonnaise is composed of yolk plasma-LDL (Chang, Powrie, & Fennema, 1972; Ford, Borwankar, Pechak, & Schwimmer, 2004). The plasma film and yolk granule particles serve as "bridges" between the oil droplets in an emulsion, and this granule-oil droplet network holds the formula in a gelatinized structure, without lipid droplets coalescing (Chang et al., 1972; Langton, Åström, & Hermansson, 1999). Both the plasma and granule fractions play an integral role in mayonnaise emulsion stability and undoubtedly influence its rheological properties (Ford et al., 2004). However, the fatty acid compositions of these two yolk fractions is unreported. Therefore, a comparative study of the fatty acid composition of CLA-rich yolk plasma and granule fractions with those of control yolk fractions may provide further insight on how yolk fatty acid profile affects mayonnaise quality.

Furthermore, since CLA in yolks increases yolk viscosity (Shinn et al., 2015b), modifying the egg yolk fatty acid profile with CLA in yolks may also affect the rheological properties mayonnaise and emulsion stability. Mayonnaise emulsion viscosity is an important quality attribute (Weenen, Van Gemert, Van Doorn, Dijksterhuis, & De Wijk, 2003), and it is thought that the continuous phase viscosity controls mayonnaise stability and texture (Langton et al., 1999). Modifying the egg yolk lipid profile in the continuous aqueous phase of the emulsion may have significant effects on mayonnaise quality. However, there are no rheological studies reporting the effect of CLA-rich yolks on mayonnaise quality. Mayonnaise quality includes mayonnaise gelatinization, flow, and spread-ability, and these properties can be determined instrumentally by assessing oscillatory stress and storage modulus (G'), creep and recovery curves, and viscosity, respectively (Whittingstall). Mayonnaise oscillatory stress and resulting elasticity measurements (G') indicate the degree of "gel" structure and the thickness that will be encountered when the product is in a container (Whittingstall). Mayonnaise creep and recovery behavior describes how the mayonnaise responds to low steady stresses, like gravity after being spooned from a vessel (Whittingstall, 2014). Relating viscosity to shear rate results in simple flow curves that indicate mayonnaise spreadability (Whittingstall).

Additionally, emulsion stability is also an important quality attribute that indicates mayonnaise shelf-life before possible phase separation. By considering how CLA incorporation affects fatty acid composition and subsequent mayonnaise rheology and stability we may gain a better understanding of egg yolk emulsion properties and capabilities, and how these are modified by yolk lipid alterations.

Therefore, the objectives of this study are (1) compare the fatty acid composition of CLA-rich yolk granules and plasma with that of yolk granules and plasma of less viscous control yolks, (2) compare the rheological properties of mayonnaise prepared with CLA-rich eggs, relative to controls and (3) compare the emulsion stability of CLA-egg mayonnaise, relative to controls.

2. Experimental section

2.1. Materials

The refined, bleached, deodorized soybean oil used for CLA-rich soybean oil production, feed preparation, and mayonnaise formulation was provided by Riceland Foods (Stuttgart, AR). Other mayonnaise ingredients were purchased from a local supermarket. All lipid extraction solvents were analytical grade. The fatty acid methyl ester (FAME) internal standard used were purchased from Supelco (Bellefonte, PA).

2.2. Methods

2.2.1. Feed preparation and egg collection

2.2.1.1. CLA-rich soy oil production. A 15% CLA-rich soybean oil was produced by linoleic acid photoisomerization using a pilot scale unit and method of Jain et al. (2008). CLA-rich soy oil was analyzed for fatty acid composition and contained: 15% CLA, 12.2% C16:0, 0.09% C16:1(n−7), 4.2% C18:0, 25.9% C18:1(n−9), 38.5% C18:2 (n−6), and 3.6% C18:3(n−3).

2.2.1.2. Feed preparation. The 15% CLA soybean oil was combined with pelleted commercial corn and soybean meal-based finisher diet (Cobb-Vantress, Siloam Springs, AR). The commercial pelleted diet contained 2761 kcal/kg metabolizable energy and 15.43% crude protein. Feed was formulated without meat or animal by-products to meet or exceed minimum National Research Council standards (1994) for all ingredients. The CLA diet was produced by adding 10% (wt.) CLA-rich soy oil to the standard commercial feed and combined using a Hobart stand mixer. A soy oil control diet was similarly prepared, and the unmodified feed was the standard control diet.

2.2.1.3. Diet administration. One hundred single-comb white leghorn chicks were reared under standard commercial conditions. At 35 weeks, thirty-six hens were randomly chosen and assigned to single bird cages in blocks of 6 birds each, separated by 2 empty cages. All hens received the same standard commercial finisher diet until the feeding trial began at 36 weeks. Each soy oil diet (15% CLA soy oil and conventional soy oil) was randomly assigned to 2 blocks of 6 birds each, resulting in 12 birds receiving each treatment diet. The remaining twelve hens continued to receive standard commercial feed without additional soy oil and served as a standard control group. Animal procedures were approved by the University of Arkansas Institutional Animal Care and Use Committee (Protocol # 13033).

2.2.1.4. Egg collection. After 12 days of treatment feed administration, eggs were collected daily for 10 days, counted, and labeled with cage number and date. Eggs were stored in a walk in cooler at 4 °C for 24 h prior to subsequent analysis.

2.2.2. Lipid extraction and fatty acid analysis of yolk plasma and granules

2.2.2.1. Plasma and granule fractionation. Yolks were separated from albumen and rolled on a *Kimwipe* to remove any residual adhering albumen. Yolks from the duplicate treatment blocks were pooled (6 eggs = 1 sample replicate). Duplicate 20 g yolk samples from all three egg types were combined with 1 mL of 1% NaCl in deionized water and centrifuged at 10,000 rpm for 1 h (Belitz and