# Influence of Conjugated Linoleic Acid (CLA) on Establishment and Progression of Atherosclerosis in Rabbits

**David Kritchevsky, PhD, Shirley A. Tepper, Scott Wright, Patrick Tso, PhD, and Susanne K. Czarnecki, PhD**

*The Wistar Institute, Philadelphia, Pennsylvania (D.K., S.A.T., S.W.), Department of Pathology, University of Cincinnati, Cincinnati, Ohio (P.T.), Department of Chemistry, Chestnut Hill College, Philadelphia, Pennsylvania (S.K.C.)*

**Key words: atherogenesis, conjugated linoleic acid, regression of atherosclerosis, experimental artherosclerosis, rabbits**

**Objective:** To determine effects of conjugated linoleic acid (CLA) on establishment and progression of experimentally-induced atherosclerosis in rabbits.

**Methods:** For establishment of atherosclerosis, New Zealand White rabbits were fed a semipurified diet containing 0.1% to 0.2% cholesterol for 90 days. Some groups were fed diet and CLA. For effects on progression of atherosclerosis, rabbits with established atherosclerosis were fed a semipurified diet±CLA for 90 days.

**Results:** At dietary levels as low as 0.1%, CLA inhibited atherogenesis. At dietary levels of 1%, CLA caused substantial (30%) regression of established atherosclerosis. This is the first example of substantial regression of atherosclerosis being caused by diet alone.

**Conclusion:** Dietary CLA is an effective inhibitor of atherogenesis and also causes regression of established atherosclerosis.

## INTRODUCTION

Conjugated linoleic acid (CLA) has been reported to exhibit antioxidant properties *in vivo* [1] and *in vitro* [2]. It is known that oxidized derivatives of cholesterol cause atherosclerosis and/or arterial injury in rabbits [3,4] and that oxidized low density lipoprotein (LDL) has atherogenic properties [5]. The possibility then arose that the antioxidant properties of CLA might influence the course of experimental atherosclerosis. A preliminary study showed that CLA (0.5% of the diet) could inhibit cholesterol-induced atherosclerosis in rabbits [6]. Nicolosi *et al.* [7] have reported that CLA inhibits development of aortic sudanophilia in cholesterol-fed hamsters. To confirm our earlier finding we carried out further studies of the influence of CLA on atherogenesis in rabbits. We also looked into the effects of CLA on pre-established atherosclerosis to see if it could prevent continued development of arterial lesions or even cause reversal of atherosclerosis. Our findings form the basis of this report.

## MATERIALS AND METHODS

Male rabbits of the New Zealand White strain were used throughout. For establishment of lesions the animals were fed a semipurified diet containing cholesterol and including various levels of CLA (Table 1). In studies of regression of lesions a large group of rabbits was fed the atherogenic diet. After 90 days the rabbits were bled and divided into three groups of equal average serum cholesterol level. One group was necropsied to provide a baseline value for atherosclerotic involvement. The assumption was made that groups with similar average cholesterol levels would exhibit equal average atherosclerosis. The two surviving groups were placed on a cholesterol-free diet or the same diet containing CLA. These groups were necropsied 90 days later. At necropsy, plasma total and HDL cholesterol and triglycerides were determined using appropriate kits (Sigma, St. Louis, MO). Livers were removed and weighed and aliquots analyzed for free and total cholesterol [8] and triglycerides [9]. Aortas were removed, cleaned and severity of lesions graded visually using a 0–4 scale [10]. Animals were maintained in individual stainless-steel cages in an air-conditioned, humidified room maintained on a 12-hour light/dark cycle. Food and water were provided *ad libitum*. The diets were prepared to our specifications and pelleted by Dyets, Inc. (Bethlehem, PA). Experimental procedures were approved by the Wistar Institutional Animal Care and Use Committee (IACUC).

Abbreviations: CLA=conjugated linoleic acid, LDL=low density lipoprotein.

Address reprint requests to: David Kritchevsky, PhD, The Wistar Institute, 3601 Spruce Street, Philadelphia, PA 19104. kritchevsky@wistar.upenn.edu

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**Table 1.** Composition of Conjugated Linoleic Acid (CLA) Isomer Mixture Used in this Study*

| Fatty Acid | % |
|---|---|
| Palmitic (16:0) | 0.65 |
| Oleic (18:1 *c*9) | 5.80 |
| Linoleic (18:2 *c*9, *c*12) | 0.32 |
| CLA (18:2 *c*9, *t*11) | 43.29 |
| CLA (18:2 *t*10, *c*12) | 44.07 |
| CLA (18:2 *c*9, *c*12) | 1.46 |
| CLA (18:2 *c*10, *c*12) | 0.96 |
| CLA (18:2 *t*, *t*9, 11 and 10, 12) | 1.48 |
| Other | 1.97 |

* The CLA used was a gift of Natural Lipids Ltd. AS, Hovdebydga, Norway. Composition determined by gas-liquid chromatography. The *c*9, *t*11, and *t*10, *c*12 isomers represent 47.03% and 47.88%, respectively, of total CLA isomers present.

**Table 2.** Atherogenic Diet (I)

| Ingredient[1] | % | % Calories |
|---|---|---|
| Casein | 25.0 | 26.2 |
| L-arginine HCl | 0.6 | |
| DL-methionine | 0.2 | |
| Sucrose | 19.46 | 20.4 |
| Corn starch | 19.3 | 20.3 |
| Coconut oil | 12.0 | 28.3 |
| Corn oil | 2.0 | 4.7 |
| Cellulose | 15.0 | |
| Vitamin mix | 1.0 | |
| Mineral mix | 5.34 | |
| Cholesterol | 0.1 | |

[1] CLA added at expense of sucrose in test diet.

## RESULTS

In our earlier first study groups of six rabbits (three male, three female), each were fed a semipurified diet containing 0.1% cholesterol for 22 weeks (Table 2). The diet of the test group was augmented with 0.5% CLA. Total plasma cholesterol and triglycerides were 1175 mg/dL and 165 mg/dL in the controls compared with 1000 mg/dL and 140 mg/dL in the test group. The ratio of plasma LDL/HDL cholesterol was 10.9 in

**Table 3.** Atherogenic Diet (II)

| Ingredient[1] | % | % Calories |
|---|---|---|
| Casein | 25.0 | 26.0 |
| DL-methionine | 0.2 | |
| Sucrose | 19.48 | 20.3 |
| Starch | 20.0 | 21.0 |
| Coconut oil | 12.0 | 28.0 |
| Corn oil | 2.0 | 4.7 |
| Cellulose | 15.0 | |
| Mineral mix | 5.0 | |
| Vitamin mix | 1.0 | |
| Choline bitartrate | 0.12 | |
| Cholesterol | 0.2 | |

[1] CLA added at expense of starch in test diet.

**Table 4.** Regression Diet

| Ingredient[1] | % | % Calories |
|---|---|---|
| Casein | 24.0 | 27.3 |
| DL-methionine | 0.3 | |
| Sucrose | 30.58 | 34.8 |
| Starch | 20.0 | 22.7 |
| Corn oil | 6.0 | 15.3 |
| Cellulose | 14.0 | |
| Mineral mix | 4.0 | |
| Vitamin mix | 1.0 | |
| Choline bitartrate | 0.12 | |

[1] CLA added at expense of corn oil in test diet.

**Table 5.** Necropsy Data-Rabbits Fed 0.2% Cholesterol±1% CLA for 90 Days (10/gp)

| | Group | |
|---|---|---|
| | Control | CLA |
| Weight gain (g) | 822±1.22 | 742±87 |
| Liver wt. (g) | 109±5 | 111±7 |
| Liver (% body wt.) | 3.13±0.14 | 3.24±0.18 |
| Serum (mg/dL) | | |
| Cholesterol (C) | 430±40 | 559±53[a] |
| % HDL-C | 6.1±0.44 | 3.6±0.51[a] |
| Triglyceride | 77±6 | 135±36[a] |
| Liver (g/100 g) | | |
| Cholesterol, total | 2.50±0.32 | 1.55±0.14[a] |
| % Ester | 86.0±1.50 | 84.0±1.73 |
| Triglycerides | 0.84±0.06 | 0.83±0.06 |
| Aorta (0–4 scale) | | |
| Arch | 2.39±0.47 | 1.65±0.37 |
| Thoracic | 2.35±0.36 | 1.40±0.40 |
| Area (%) | 49±10 | 30±10 |

[a] $p<0.05$.

**Table 6.** Necropsy Data-Rabbits with Established Atherosclerosis Fed Corn Oil±1% CLA for 90 Days (10/gp)

| | Group | |
|---|---|---|
| | Control | CLA |
| Weight gain (g) | 461±65 | 246±37[a] |
| Liver wt. (g) | 99±9 | 83±6 |
| Liver (% body wt.) | 2.33±0.20 | 2.31±0.13 |
| Serum (mg/dL) | | |
| Cholesterol (C) | 73±10 | 140±24[a] |
| % HDL-C | 13.5±1.02 | 10.6±0.81[a] |
| Triglyceride | 77±13 | 57±5 |
| Liver (g/100 g) | | |
| Cholesterol, total | 0.80±0.12 | 0.78±0.17 |
| % Ester | 82.0±2.66 | 77.7±2.17 |
| Triglyceride | 0.77±0.05 | 0.76±0.06 |
| Aorta (0–4 scale) | | |
| Arch | 2.35±0.35 | 1.65±0.26 |
| Thoracic | 2.30±0.40 | 1.65±0.22 |
| Area (%) | 51±11 | 34±6 |

[a] $p<0.05$.

**Table 7.** Necropsy Data—Rabbits Fed 0.2% Cholesterol±0.1%, 0.5% or 1.0% CLA for 90 Days (8/gp)

| | Group | | | |
|---|---|---|---|---|
| | Control | 0.1% CLA | 0.5% CLA | 1.0% CLA |
| Weight gain (g) | 104±46 | 3±108 | 67±43 | 50±79 |
| Liver wt. (g) | 68±5 | 77±6 | 66±4 | 78±6 |
| Liver (% body wt.) | 2.73±0.16 | 3.22±0.35 | 2.63±0.12 | 3.35±0.32 |
| Serum (mg/dL) | | | | |
| Cholesterol (%) | 983±118 | 1281±116 | 1263±104 | 1103±134 |
| % HDL-C | 5.0±0.9 | 3.3±0.54 | 3.3±0.58 | 5.0±1.14 |
| Triglyceride | 190±32 | 246±47 | 205±48 | 216±38 |
| Liver (g/100 g) | | | | |
| Cholesterol, total | 1.30±0.97$^{ab}$ | 1.02±0.07$^{a}$ | 0.99±0.06$^{b}$ | 1.06±0.08 |
| % Ester | 53.9±1.1$^{abc}$ | 73.7±3.4$^{a}$ | 72.5±3.0$^{b}$ | 72.6±3.3$^{c}$ |
| Triglyceride | 1.37±0.21 | 1.19±0.18 | 1.28±0.25 | 1.23±0.23 |
| Aorta (0–4 scale) | | | | |
| Arch | 2.36±0.39$^{ab}$ | 1.69±0.23$^{c}$ | 0.88±0.20$^{ac}$ | 1.00±0.28$^{b}$ |
| Thoracic | 2.21±0.42$^{de}$ | 1.31±0.28 | 0.75±0.21$^{d}$ | 0.94±0.27$^{e}$ |
| Area (%) | 44±12$^{f}$ | 32±7$^{g}$ | 11±4$^{fg}$ | 18±6 |

Values in horizontal rows bearing same letter are significantly different.

the test group and 16.5 in the controls. Histological examination of the aortas showed maximal plaque thickness (mm) in the abdominal aorta of the CLA-fed rabbits to be 27% smaller than in the controls. Maximum plaque thickness in the thoracic aorta was the same in the two groups. Lipid deposition and connective tissue development were less severe in the aortas of the CLA-fed rabbits [6]. Using the TBARS (thiobarbituric acid reactive substances) assay, which is a test for products of oxidation, plasma peroxides were found to be similar in the two groups. A second experiment was designed to compare the apparent antiatherogenicity of CLA and also to examine its effects on progression or regression of established atherosclerotic lesions. Thirty rabbits were fed a semipurified atherogenic diet (control) containing 0.2% cholesterol (Table 3), and ten rabbits were fed the same diet augmented with 1% CLA. After 90 days the rabbits on the control diet were bled and randomized into three groups of ten rabbits each of equal average serum cholesterol levels. The CLA-fed rabbits and one of the control subgroups were necropsied. The two remaining groups were placed on a cholesterol-free semipurified diet (Table 4) with or without added CLA (1%). These groups were necropsied 90 days later.

Serum lipid levels were higher in the CLA-fed groups, but liver cholesterol levels were lower in that group ($p<0.05$ by $t$ test). Baseline cholesterol levels in New Zealand White rabbits are in the range of 50 to 70 mg/dL. Liver triglycerides were similar in the two groups. Rabbits fed 1% CLA had 31% less severe atherosclerosis in the arch and 40% less severe atherosclerosis in the thoracic aorta (Table 5). In the regression phase of this experiment (Table 6), serum cholesterol levels had fallen by 83% and 67%, respectively, on the control and CLA diets,

**Table 8.** Necropsy Data—Rabbits with Established Atherosclerosis Fed Corn Oil±0.1, 0.5 or 1% CLA for 90 Days

| | Group | | | |
|---|---|---|---|---|
| | Control | 0.1% CLA | 0.5% CLA | 1.0% CLA |
| No. | 7 | 6 | 7 | 6 |
| Weight gain (g) | 312±99 | 265±104 | 298±84 | 242±73 |
| Liver wt. (g) | 52±3 | 58±6 | 64±5$^{a}$ | 47±3$^{a}$ |
| Liver (% body wt.) | 1.82±0.07$^{b}$ | 2.05±0.16 | 2.28±0.20$^{bc}$ | 1.75±0.13$^{a}$ |
| Serum (mg/dL) | | | | |
| Cholesterol | 128±38$^{ab}$ | 140±19$^{cd}$ | 295±48$^{ac}$ | 309±46$^{bd}$ |
| Triglyceride | 47±7$^{ef}$ | 61±8 | 124±28$^{e}$ | 105±20$^{f}$ |
| Liver (g/100 g) | | | | |
| Cholesterol, total | 1.01±0.10 | 1.24±0.14 | 1.17±0.10 | 1.05±0.07 |
| % Ester | 67.8±2.7 | 72.4±4.8 | 73.3±3.1 | 71.2±2.9 |
| Triglyceride | 0.63±0.11 | 0.72±0.13 | 0.66±0.11 | 0.65±0.11 |
| Aorta (0–4 scale) | | | | |
| Arch | 2.64±0.28 | 2.25±0.28 | 2.50±0.29 | 1.95±0.40 |
| Thoracic | 2.29±0.36$^{d}$ | 2.33±0.44 | 2.00±0.15$^{e}$ | 1.25±0.17$^{de}$ |
| Area (%) | 53±7 | 53±10 | 49±5 | 30±10 |

Values in horizontal rows bearing same letter are significantly different.

**Table 9.** Regression of Atherosclerosis in Rabbits (Effect of Diet)

| Regimen[1] | No. | Duration | Grade[2] | Ref. |
|---|---|---|---|---|
| A) 6g C/wk | 39 | 90 d | 2.46±0.22 | 11 |
| R) Chow | 17 | 22–26 wk | 2.41±0.36 | |
| A) 2% C/6% CO | 49 | 8 wk | 1.75±0.11[a] | 12 |
| R) Chow | 41 | 8 wk | 2.80±0.15 | |
| 5% sat'd fat | 39 | 8 wk | 2.45±0.19 | |
| 5% unsat. fat | 44 | 8 wk | 2.35±0.18 | |
| A) 2% C/6% CO | 37 | 8 wk | 1.33±0.13[a] | 13 |
| R) Chow | 30 | 8 wk | 2.04±0.19 | |
| 6% CO | 35 | 8 wk | 1.65±0.12 | |
| Peanut oil | 32 | 8 wk | 2.39±0.20 | |

[1] A=atherogenic regimen, R=regression regimen, C=cholesterol, CO=corn oil.
[2] Graded on 0–4 scale.
[a] Average grade (aortic arch+thoracic aorta)/2.

and HDL cholesterol had risen by 121% and 74% in the control and CLA groups. Triglyceride levels were unchanged in the control group, but had fallen by 53% in the rabbits fed CLA. In the regression phase, the effects of CLA on severity of atherosclerosis were striking: whereas, severity of lesions in the control group was virtually unchanged from that seen at the cessation of cholesterol feeding, severity of atherosclerosis in the arch and thoracic aorta of the CLA-fed rabbits had been reduced by 31% and 30%, respectively.

The next study was designed to confirm previous findings and to investigate effects of different levels of dietary CLA for their effects on establishment and progression of atherosclerosis. Accordingly, 40 rabbits were fed the atherogenic diet described in Table 3. Other groups of eight rabbits each were fed the same diet augmented with 0.1%, 0.5% or 1.0% CLA. The findings are summarized in Table 6. Serum cholesterol and triglyceride levels were higher in all CLA fed groups, and

**Table 10.** Regression of Atherosclerosis in Rabbits (Drug Effects)

| Regimen[1] | No. | Duration | Grade | Ref. |
|---|---|---|---|---|
| A) 1% C/3% CO | 9 | 3 mo | 2.26±1.31[a] | 14 |
| R) 3% CO | 8 | 4 mo | 2.65±1.17 | |
| 2.5% S/3% CO | 7 | 4 mo | 3.01±0.91 | |
| A) 1% C/3% CO | 9 | 3 mo | 54±22[b] | 15 |
| R) 3% CO | 5 | 4 mo | 83±16 | |
| 2% DHC/3% CO | 5 | 4 mo | 82±22 | |
| A) 2% C/6% CO | 34 | 8 wk | 1.70±0.15[a] | 16 |
| R) Chow | 24 | 8 wk | 2.50±0.19 | |
| 0.3% CPIB/Chow | 29 | 8 wk | 2.28±0.16 | |
| A) 1% C/5% CSO | 18 | 9 wk | 17.1±3.1[b] | 17 |
| R) 5% CSO | 13 | 9 wk | 35.2±4.8 | |
| DES/5% CSO | 13 | 9 wk | 17.8±3.7 | |
| 56% CR | 13 | 9 wk | 41.6±6.4 | |

[1] A=atherogenic regimen, R=regression regimen, C=cholesterol, CO=corn oil, DHC=dihydrocholesterol, S=sitosterol, CPIB=ethyl p-chlorophenoxyisobutyrate, DES=diethyl stilbesterol, 2 mg/kg subq., 3×/wk, CR=caloric restriction.
[a] Graded 0–4 scale.
[b] Sudanophilic area.

**Table 11.** Regression of Atherosclerosis in Rabbits[3] (Effect of Thyroid Drugs)

| Regimen[1] | No. | Duration | Grade[2] |
|---|---|---|---|
| A) 2% C/6% | | | |
| CO | 14 | 6 wk | 2.00 |
| R) Chow | 11 | 6 wk | 2.80 |
| D-thyroxine | 13 | 6 wk | 2.95 |
| L-thyroxine | 13 | 6 wk | 2.55 |
| DT3 | 12 | 6 wk | 3.45 |
| LT3 | 8 | 6 wk | 3.25 |
| A) 2% C/6% | | | |
| CO | 11 | 8 wk | 1.80 |
| R) Chow | 10 | 8 wk | 2.90 |
| D-thyroxine | 10 | 8 wk | 2.65 |
| L-thyroxine | 8 | 8 wk | 2.50 |
| DT3 | 10 | 8 wk | 2.60 |
| LT3 | 9 | 8 wk | 3.05 |

[1] A=atherogenic regimen, R=regression regimen, C=cholesterol, CO=corn oil, T3=triiodothyronine.
[2] Graded 0–4 scale.
[3] After reference Kritchevsky *et al.* [18].

percentage of HDL cholesterol was lower in all but the group fed 1% CLA. Liver cholesterol levels were highest in the control group, as were triglyceride levels. The amount of esterified liver cholesterol was lowest in the control group. The severity of atherosclerosis in the aortic arch was reduced by 28%, 63% and 58% in rabbits fed 0.1%, 0.5% or 1.0% CLA, respectively, and that in the thoracic aorta was reduced by 41%, 66% and 57% in the same groups. Analysis of variance (ANOVA) showed the differences in the aortic arch, thoracic aorta and involved area to be significant at $p<0.003$, $p<0.011$, and $p<0.03$, respectively. The rabbits with established atherosclerosis were fed the regression diet or the same diet augmented with 0.1%, 0.5% and 1.0% CLA for another 90 days. The findings are presented in Table 7. Serum lipids were higher than control levels in the sera of the rabbits fed CLA, but liver lipids were the same. The aorta data show that there was no regression of atherosclerosis in the rabbits fed 0.1% or 0.5% CLA. However, compared to the initial control (Table 8), severity of lesions in the aortic arch and thoracic aorta of

**Table 12.** Regression of Atherosclerosis in Rabbits[3] (Thyroid Drugs—Mode of Administration)

| Regimen[1] | No. | Duration | Grade[2] |
|---|---|---|---|
| A) 2% C/6% | | | |
| CO | 20 | 8 wk | 1.25 |
| R) Chow | 18 | 8 wk | 2.75 |
| D-thyroxine | 17 | 8 wk | 2.05 |
| L-thyroxine | 17 | 8 wk | 1.85 |
| 5% CO | 19 | 8 wk | 2.00 |
| D-thyroxine | 13 | 8 wk | 2.05 |
| L-thyroxine | 18 | 8 wk | 2.65 |

[1] A=atherogenic regimen, R=regression regimen, C=cholesterol, CO=corn oil.
[2] Graded 0–4 scale.
[3] After reference Kritchevsky and Tepper [19].

**Table 13.** Regression of Atherosclerosis in Rabbits[4] (Drugs and Oxygen)

| Regimen[1] | No. | Duration | Grade[2] |
|---|---|---|---|
| A) 0.4% C/10% SFO | 12 | 14 wk | 65±8.7 |
| R) Chow | 12 | 10 wk | 66±7.5 |
| a) 100% $O_2$ (2 h/d; 5 d/wk) | 7 | 10 wk | 50±11.0 |
| b) 1% cholestyramine | 7 | 10 wk | 37±13.0 |
| c) a+b | 7 | 10 wk | 29±10.0 |
| d) Estradiol[3] | 7 | 10 wk | 37±17.0 |
| e) a+d | 7 | 10 wk | 31±11.4 |

[1] A=atherogenic regimen, R=regression regimen, C=cholesterol, SFO=safflower oil.
[2] Graded 0–4 scale.
[3] 1,332 mg estradiol benzoate, subq. in 0.4 ml sesame oil; 5 d/wk.
[4] After reference Vesselinovitch *et al.* [20].

rabbits fed the CLA-free diet had risen by 11% and 4%, respectively. In contrast, and consistent with the previous experiment, severity of atherosclerosis in the aortic arch and thoracic aorta of rabbits fed 1% CLA had fallen by 17% and 43% respectively. Thus, levels of CLA as low as 0.1% of the diet could inhibit atherogenesis, but only at 1% of the diet did CLA reduce progression of established lesions.

## DISCUSSION

The experiments we have described confirm earlier observations that CLA can inhibit atherogenesis in rabbits. The finding that even at a level of 0.1% CLA inhibits atherosclerosis by 34% is important since it brings the effective level down to the amount of CLA one might obtain by diet alone.

The most striking finding is the influence of CLA on progression of established atherosclerosis. This area of effort has a long history with little evidence of success. Once established, aortic lesions in rabbits will regress only under unusual circumstances. Usually, lesions become more severe after cessation of cholesterol feeding, and most treatments may inhibit only slightly the exacerbation of lesions. Studies of diet or drug effects in experimental atherosclerosis are carried out for one of two reasons: to see if the treatment can reduce severity of the lesions or to investigate the possibility that the treatment will cause diminution of severity of pre-established lesions. The former aim has been achieved by many substances, but attempts to induce regression have rarely been successful. The ensuing discussion will describe the generally unsuccessful efforts to cause regression of atherosclerotic lesions in rabbits using diets or drugs which have been shown to inhibit atherogenesis.

**Table 14.** Regression of Atherosclerosis in Rabbits

| Regimen[1] | No. | Duration | Grade[2] | Ref. |
|---|---|---|---|---|
| A) 0.5% C | 10 | 60 d | 34.4±4 | 21 |
| R) Chow | 7 | 90 d | 38.8±5 | |
| Chow+protein[a] | 7 | 90 d | 17.8±4 | |
| A) 0.5% C | 12 | 12 wk | 32.3±5.1 | 22 |
| R) Chow | 12 | 12 wk | 40.8±5.5 | |
| Fluvastatin[b] | 12 | 12 wk | 33.6±6.1 | |

[1] A=atherogenic regimen, R=regression regimen, C=cholesterol.
[2] Lesion area.
[a] 50 mg HDL-VHDL protein/wk (from normolipemic rabbit).
[b] 2 mg/kg/d.

In 1955 McMillan *et al.* [11] examined the effects of returning rabbits to a commercial ration for 26 weeks after establishment of lesions. The extent of regression was 2% (Table 9). Adding fat to the diet during the regression phase had a slight effect in one case [12], but peanut oil was found to increase atherogenic involvement by 45% compared to corn oil [13] (Table 9). Beta sitosterol [14], dihydrocholesterol [15] and Clofibrate [16] had virtually no effect when added to the diet, but diethyl stilbesterol, given subcutaneously, inhibited exacerbation of lesions, but did not cause regression [17] (Table 10). Neither D- or L-thyroxine or D- or L-triiodothyronine have significant effects regardless of fat in the diet or mode of administration [18,19] (Tables 11, 12). Estradiol benzoate and cholestyramine give significant reductions in sudanophilia when combined with maintaining the rabbits in 100% oxygen for two hours daily [20] (Table 13). Intravenous injection of HDL protein will reduce severity of lesions by 48% [21], but dietary administration of fluvastatin has virtually no effect [22] (Table 14).

We have demonstrated that CLA, even at levels as low as 0.1% of the diet, will inhibit atherogenesis. At 1% of the diet, CLA will cause significant regression of atheromata. This is a unique and important effect. The mechanism by which CLA exerts its effects on establishment and regression of atherosclerosis is unclear.

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