Comparative Effects of Thyroxin Analogues as Hypcholesteremic Agents

By Maurice M. Best, M.D., and Charles H. Duncan, M.D.

DRIED THYROID SUBSTANCE or the pure natural hormones, L-thyroxin and L-triiodothyronine, effect a reduction in the serum total cholesterol of euthyroid individuals when administered in sufficient amount.1-3 Their general clinical use to reduce elevated serum cholesterol levels in the hope of favorably influencing the course of atherosclerosis has been limited by two factors. When given in moderate dosage, dried thyroid substance effected a prompt reduction in serum cholesterol, but despite continuation of the hormone the reduction was not sustained.4 This “escape” is presumed to be due to suppression of thyrotropin secretion by the administered thyroid and consequent decrease in production of endogenous thyroid hormone. When the dose of administered hormone is increased to maintain a reduced serum cholesterol level, hypermetabolism manifested by an increased basal metabolic rate, tachycardia, and weight loss may occur. An ominous consequence of thyroid hormone administration to patients with coronary artery disease is the frequent increase in severity of angina pectoris.5

If some modification of the chemical structure of L-thyroxin or L-triiodothyronine resulted in an analogue that largely retained the effect of the natural hormones on cholesterol metabolism but was sufficiently less active in its other metabolic effects, a way out of this dilemma would be available.

In the rat the formic acid analogue of thyroxin, tetraiodothyroformic acid, has been shown to exert an effect on cholesterol metabolism that is disproportionate to its effect on oxygen consumption, growth, and thiourea-induced goiter.6-8 More recent studies in the rat of a series of thyroxin analogues have shown that the D-isomers of thyroxin and triiodothyronine also exert a disproportionate effect on cholesterol metabolism.9

In view of these observations in the experimental animal that replacement of the L-alanine side chain of the natural thyroid hormones with either a carboxyl group or D-alanine resulted in analogues displaying a degree of dissociation of effects, these analogues were compared with L-thyroxin as hypcholesteremic agents in the euthyroid human subject. To study further the general metabolic effects of these analogues their ability to serve as thyroid replacement therapy in human myxedema was also determined.

Material and Methods

Seventeen euthyroid patients were observed for periods up to 4 years. Ten were male and seven female; ages ranged from 28 to 77, with a mean of 56 years. Three had elevated serum cholesterol levels without evidence of cardiovascular disease (patients 1, 2, and 3). The remaining 14 patients had arteriosclerotic heart disease; 12 had myocardial infarction 3 or more months prior to study (patients 4 to 15), four had moderate to severe angina pectoris (patients 13 to 16), and three had congestive heart failure controlled by digitalis (patients 4, 5, and 17). Serum protein-bound iodine levels of the patients ranged from 4.7 to 6.9 µg. per 100 ml.

Mean serum cholesterol levels of the individual patients before treatment ranged from 244 to 394 mg. per 100 ml., the mean for the group being 286. Patients with wide fluctuations in serum cholesterol, such as occur in idiopathic hyperlipemia, and patients with overt diabetes mellitus were excluded. Major criteria for inclusion were the ability and willingness of the subjects to cooperate in the study as demonstrated during the initial control period. Six patients were hospitalized throughout the period of study.

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and consumed a fairly constant diet; the remaining patients were seen as outpatients and continued their usual diets.

All patients were seen at 2- or 3-week intervals, the visits being scheduled at approximately the same time of day. After a 20-minute rest period the pulse rate and body weight were recorded, blood was drawn, and the clinical state of the patient was evaluated. Serum total cholesterol was determined by the method of Abell et al.10

The L-thyroxin and analogues were given orally, in single or twice daily doses. The total daily dose of each hormone is indicated in figure 1. During the periods immediately preceding and following those of drug administration the patients received placebos that were identical in appearance to the active drug. Whenever a change in the appearance of the medication or the frequency of administration was necessitated by the form available, it was accomplished during the placebo period.

Thirty-four treatment periods of 6 weeks to 10 months were completed in the 17 patients, each being preceded and followed by a period of placebo administration. Five patients received only one drug, the other patients two to four.

Seven myxedematous patients were also observed for periods ranging up to 5 years. Six were female and one male; ages ranged from 37 to 74 years, with a mean of 55 years. In three the etiology of the myxedema was unknown; in the remaining four it was the result of thyroid surgery or therapeutic radioiodine. Prior to treatment basal metabolic rates ranged from minus 39 to minus 18 per cent, serum protein-bound iodine from 0.5 to 1.7 μg. per 100 ml., and serum total cholesterol from 260 to 450 mg. per 100 ml.

After the diagnosis of myxedema was established and replacement therapy was instituted with L-thyroxin or L-triiodothyronine, the patients were observed as described for the euthyroid subjects. In addition, special attention was directed to the clinical evidences of the thyroid status, and determinations of the basal metabolic rate were made at frequent intervals in the four patients in whom the results were most reproducible. Each of the analogues under study was then substituted for the natural hormone in three to seven of the patients for periods of 6 to 12 months each. The doses employed ranged from one-half to one and one-half those employed in the euthyroid subjects, and were adjusted in accordance with the individual responses of the patients.

Figure 1

Thyroid hormones and analogues studied in human subject. The goiter-inhibiting activities, with L-thyroxin assigned a value of 100, are those determined in our laboratory. The daily dose is that given the euthyroid patients.

<table>
<thead>
<tr>
<th>Relative Activity</th>
<th>Daily Dose (mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rat Goiter Assay</td>
<td></td>
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<tr>
<td>L-Thyroxin</td>
<td>L-T4 100</td>
</tr>
<tr>
<td>D-Thyroxin</td>
<td>D-T4 20</td>
</tr>
<tr>
<td>L-Triiodothyronine</td>
<td>L-T3 533</td>
</tr>
<tr>
<td>D-Triiodothyronine</td>
<td>D-T3 27 1</td>
</tr>
<tr>
<td>Tetraiodothyroformic Acid</td>
<td>TFA-4 0.09 200</td>
</tr>
<tr>
<td>Diiododimethyl-thyroformic Acid</td>
<td>TFA-2 0.23 100</td>
</tr>
</tbody>
</table>

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Results

Nine of the 17 euthyroid patients received tetraiodothyroformic acid (TFA-4) for periods of 2½ to 10 months of continuous administration (fig. 2). In all patients there was a reduction in the mean serum cholesterol during the treatment period. For the group the mean control serum cholesterol level was 287 mg. per 100 ml.; the mean treatment level was 235, a mean reduction of 52 mg. per 100 ml. or 18 per cent of the control level. When a placebo was substituted for the TFA-4, there was a prompt return to the pretreatment range of serum cholesterol.

Dimethyldiiodothyroformic acid (dimethyl-TFA-2), which in the rat was found to have a disproportionate effect on cholesterol metabolism comparable to that of TFA-4 and to possess twice the activity of the latter analogue (fig. 1), was given to six patients. Due to the limited supply the periods of administration were only 1½ to 4 months. The effect on serum cholesterol (fig. 3) was very similar to that of TFA-4; with the substitution of a placebo there was again a prompt return to the pretreatment range.

To date the studies of the effect of D-thyroxin (D-T4) have been completed in only four of the 17 patients (fig. 4). The mean control serum cholesterol level of these patients...
TPHYROXIN ANALOGUES

DIMETHYLDIIODOTHYROFORMIC ACID
(DIMETHYL TFA-2)
100 mg./day

PLACEBO

Figure 3

The mean control serum cholesterol of patients receiving dimethyl-TFA-2 was 284 mg. per 100 ml. Responses to drug and placebo administration are indicated as in figure 2.

Patients 5 to 7, 11, 13, and 16.

was 294 mg. per 100 ml., the mean level during the 4- to 8-month treatment period was 219, a reduction of 26 per cent. Again the serum cholesterol returned to the pretreatment range following the substitution of a placebo.

Eight patients received D-triiodothyronine (D-T3) for periods of 3 to 10 months (fig. 5). The administration of 0.5 mg. of D-T3 twice daily resulted in a fall in mean serum cholesterol from 294 to 225 mg. per 100 ml., the reduction in the individual patients ranging from 42 to 94 mg. per 100 ml. In the six patients in whom a placebo had been substituted for the D-T3 at the time of this report, a return of the serum cholesterol to the control range occurred within 6 weeks.

The mean control serum cholesterol of the seven patients receiving L-thyroxin (L-T4) was 312 mg. per 100 ml. (fig. 6). Although the response of the individual patients was somewhat more variable than that to the analogues, the mean reduction of 15 per cent of the control level is not appreciably different from that resulting from the formic acid analogues (fig. 7). No tendency to escape from this effect was observed during the mean treatment period of 7½ months; substitution of placebo again resulted in a prompt return to the pretreatment range.

Undesirable side effects occurred in several of the patients while they were receiving the formic acid analogues. Diarrhea and cramping abdominal pain were experienced by two
patients during the administration of TFA-4 and by the same two patients during the administration of dimethyl-TFA-2. Another patient developed an extensive acneiform eruption and a fourth patient had salivary gland swelling during TFA-4 administration, both reactions being typical of iodium. In three of these four patients the side effects were sufficiently severe to necessitate discontinuation of the medication; all subsided when a placebo was substituted. No toxic effects on liver, kidney, or bone marrow were observed with any of the analogues.

None of the hormones at the dose employed had an appreciable effect on body weight. Mean resting pulse rate was 6 per minute higher during the period of L-T4 administration than during the control period; no increase in mean resting pulse resulted from the analogues. Increase of angina pectoris occurred in two patients during the study, in both instances during L-T4 administration. In the myxedematous patients a clinically euthyroid state was maintained by each of the analogues when given in appropriate dosage. The mean daily amount of each compound required to achieve this result was as follows: L-T4, 0.3 mg.; L-T3, 0.075 mg.; D-T4, 4 mg.; D-T3, 0.75 mg.; dimethyl-TFA-2, 100 mg.; and TFA-4, 220 mg. That all the analogues studied are calorigenic when given in adequate amount is indicated by the basal metabolic response of one patient who was hospitalized throughout the 5-year period of study.
THYROXIN ANALOGUES

Serum D-TRIIODOTHYRONINE (D-T3) 1 mg./day

Placebo

The mean control serum cholesterol of patients receiving D-T3 was 294 mg. per 100 ml. Responses to drug and placebo administration are indicated as in figure 2. Patients 1, 3, 4, 7, 9 to 11, and 15.

Discussion

Each of the four analogues studied, TFA-4, dimethyl-TFA-2, D-T4, and D-T3, resulted in a mean reduction of serum total cholesterol of the euthyroid patients. Although the subjects displayed the usual variability in serum cholesterol during both control and treatment periods, the consistency with which the mean level during treatment was reduced and the return to the pretreatment range when a placebo was substituted would seem to eliminate the possibility that these changes in serum cholesterol levels were due to spontaneous fluctuation.

The limited supply of dimethyl-TFA-2 prevented a sufficiently long period of administration to determine if its hypocholesteremic effect would be sustained. With the other analogues, no tendency of serum cholesterol to return toward control levels during continued administration for 6 to 10 months was observed. The "escape" from the initial hypocholesteremic effect of dried thyroid observed by Strisower et al.4 was evident by 6 weeks and essentially complete by 24 weeks. Thus it seems likely that if escape were to occur with these analogues it would have been observed during the present study. In the case of D-T4,
Figure 6

The mean control serum cholesterol of patients receiving L-T4 was 312 mg. per 100 ml. Responses to drug and placebo administration are indicated as in figure 2. Patients 1 to 3, 5, 13, 14, and 17.

the maintenance of reduced serum cholesterol levels throughout the period of its administration to euthyroid subjects has been noted by Starr et al.\textsuperscript{11} and by Jones.\textsuperscript{12} In view of the presumed mechanism of this escape phenomenon, its failure to occur with these analogues may be due to their relatively lesser inhibition of thyrotropin secretion.\textsuperscript{9}

The analogues, unlike L-T4, did not induce or aggravate symptoms in any of the 14 patients with arteriosclerotic heart disease. None developed or experienced intensification of angina pectoris or congestive heart failure during therapy with the analogues. One patient did have a recurrent myocardial infarction during the placebo period after treatment. Boyd and Oliver\textsuperscript{18} also observed no intensification of angina with a 10-mg. daily dose of D-T4, but unlike our experience they did observe an increase in angina with D-T3 at a daily dose of 1 mg. It is noteworthy that in their study about one half of the patients who experienced intensification of angina pectoris during administration of various thyromimetic compounds displayed no concomitant increase in basal metabolic rate.

Attempts early in our study to evaluate any possible effects of the analogues on basal metabolic rate of the euthyroid patients were abandoned, the variability of repeated determinations, even in the hospitalized patients, being sufficient to obscure any moderate change. The lack of effect of the analogues on resting pulse rate and body weight suggests that they
did not cause any very pronounced hypermetabolism. It should not be inferred that any of these analogues are without calorigenic activity, since such activity was clearly demonstrated in the myxedematous patients. It would appear, however, that the dissociation of cholesterol-lowering and calorigenic effects, though by no means complete, was of such a degree as to permit a dose that will predictably lower serum cholesterol without undesirable complications in the majority of patients with arteriosclerotic heart disease.

L-thyroxin was included in the study with the expectation of observing effects essentially similar to those reported with moderate doses of dried thyroid, that is an initial hypocholesteremic effect with subsequent tendency to escape. These expectations were only partially realized; serum cholesterol did fall moderately in the early weeks but the anticipated escape did not occur, the reduced level being maintained throughout the period of hormone administration, which was 6 or more months in all but one of the patients. Confirmation of this apparent difference in the effects of dried thyroid and L-thyroxin needs further study with comparable doses of the two materials.

At the doses employed, the hypocholesteremic effect of the L-T4 was less than that of the D-T4 and D-T3. The mean increase in pulse rate suggests that the L-T4 at this dose did have some calorigenic effect, and both patients in this group who had angina pectoris experienced an increase in symptoms.

The high dosage of TFA-4 and dimethyl-
L-thyroxin and four thyroxin analogues were administered to a group of hypercholesteremic euthyroid patients, the majority with arteriosclerotic heart disease, and the effects on serum total cholesterol compared. L-thyroxin and each of the analogues studied, tetraiodothyroformic acid, dimethylidiothyroformic acid, D-thyroxin and D-triiodothyronine, were given to four to nine patients for periods up to 10 months.

At the doses employed each of the analogues resulted in a reduction in the mean level of serum cholesterol without obvious evidence of hypermetabolism or aggravation of angina pectoris or congestive heart failure. The reduced level of serum cholesterol was sustained throughout the period of hormone administration, and upon the substitution of a placebo returned to the pretreatment range.

L-thyroxin, at the dose employed, also effected a modest sustained reduction in serum cholesterol but both patients with angina included in this group experienced an increase in severity of symptoms.

Except for four patients who developed acneiform dermatitis, salivary gland swelling, or gastrointestinal symptoms during the administration of the formic acid analogues, no toxic or undesirable effects were observed from any of the analogues.

To evaluate better the general metabolic effects of the analogues each was administered to three or more of a group of seven myxedematous patients for periods of 6 to 12 months. Given in sufficient amount all were observed to increase basal metabolic rate and to maintain a clinically euthyroid state.

From these observations, it is concluded that the D-isomers of thyroxin and triiodothyronine, while not without general metabolic effects, are tolerated by the majority of euthyroid patients with coronary atherosclerosis in amounts sufficient to maintain a reduced serum cholesterol level.

Acknowledgment

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Summary

Figure 8

Response of the basal metabolic rate to L-T4 and three of the analogues in a 35-year-old patient with myxedema induced by 131I for alleviation of congestive heart failure due to mitral insufficiency. Each hormone was given for a minimum period of 6 months. Basal metabolic rates indicated are the means of determinations made at approximately monthly intervals. Congestive failure was well controlled throughout the period except during the administration of L-T4, 0.3 mg. daily. As in all the myxedematous subjects, the serum cholesterol was more responsive to the thyroid hormones than in the euthyroid subjects; initial pretreatment control was 302 mg. per 100 ml., mean levels during periods of replacement therapy ranged from 155 to 220 mg. per 100 ml.

TFA-2 necessitated by their low activity probably precludes their general use because of the hazard of iodism in the sensitive individual. This limitation does not apply, however, to D-T3 and D-T4, which are effective in daily amounts of 1 to 4 mg.

A possible explanation for the disproportionate effect of these dextro isomers of thyroxin and triiodothyronine on cholesterol metabolism is offered by the recent observation of differences in their tissue distribution as compared to the L-isomers. The D-isomers were found in a relatively higher concentration in the liver than in skeletal and cardiac muscle as compared to the natural hormones.14, 15 In view of the dominant role of the liver in the regulation of plasma cholesterol, the disproportionate effect on serum cholesterol of the D-isomers may be attributable to their relatively higher concentration in this organ.

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Laboratories for d-thyroxin, and to Smith, Kline & French for d-triiodothyronine.

References


Leopold Auenbrugger
1722—1809

We know to-day how important to medical science was Auenbrugger’s discovery. In retrospect it is easy enough to perceive this. Nevertheless, we should be unjust to reproach Auenbrugger’s contemporaries for having failed to recognize forthwith the value of percussion. Pathological anatomy was a new science, which had not yet secured general acceptance. The majority of doctors still regarded illnesses as essentially general, and thought that local conditions were subsidiary. Decades had still to elapse before anatomical outlooks secured general acceptance. As we shall see, it was left for French physicians in the beginning of the nineteenth century to realise that percussion was one of the most important methods of examination of the sick, as it has remained unto this day.—Henry E. Sigerist, M.D. The Great Doctors. New York, W. W. Norton & Co., Inc., 1933, p. 242.
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