coupling effect on citrate oxidation. This would also account for the absence of any effect on succinate oxidation. Furthermore, this effect suggested that the inhibition of citrate and isocitrate oxidation by PTE did not result from altered mitochondrial permeability of these substrates.

Since the precise mechanism of isocitrate oxidation in mitochondria is still unsettled, the relationship of DPN and TPN effects on PTE inhibition is difficult to describe. The results certainly indicated the absence of available endogenous co-factor for isocitrate oxidation resulting from PTE injection. Several possibilities could be offered to account for this effect. Possibly PTE inhibited synthesis or stimulated breakdown of co-factor and thereby depleted the endogenous level in mitochondria. Another consideration would involve an inhibitory effect of PTE on the oxidation or reduction of endogenous co-factor. An effect on the permeability or "leaking" of these co-factors from mitochondria is still another possibility. Obviously the mechanism of the PTE effect on endogenous co-factor remains to be established.

Although these studies demonstrate a profound effect of PTE injection at a biochemical level, its relation to PTH must be established. With an *in vivo* system as described we cannot at present ascribe with certainty the nature of the inhibition as due to a direct effect of PTE or a secondary effect such as ionic changes. The experimental animals in all cases were hypercalcemic at the time of sacrifice. However, as described earlier (6), the washed mitochondria did not appear to differ significantly in the calcium concentration of control and experimental preparations suggesting the absence of a direct effect of elevated calcium. In addition, the extract effect conceivably could be the result of constituents other than PTH although the use of the controls would eliminate any effects of the vehicle. However, this inhibitory effect of PTE is certainly consistent in many respects with known effects of PTH and experimental resultst of others related to citrate levels (e.g., 10–12). Particularly, this inhibition could account for reported increased citrate production in urine (13) and in kidney (14).

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Effect of Dietary Carbohydrate on Serum Cholesterol Levels[1,2]

Quantitative nutritional studies at the National Institutes of Health have demonstrated that completely chemical diets, composed of 18 essential and nonessential L-amino acids, the required vitamins, the pertinent salts, glucose, and ethyl linoleate as the source of essential fat, will support normal growth, life-span, and reproduction in experimental rats (1–7), and positive nitrogen

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balance in man (8). Since chemical diets possess high nutritive efficacy in ultra compact form, an NASA-funded clinical study was initiated with the ultimate purpose of evaluating their long-term adquacy as a source of nutriment for man in space. Such study was conducted at the California Medical Facility, Vacaville, California, during the period of September, 1963, through February, 1964; healthy adult males were used as the experimental subjects. The clinical findings revealed that suitably formulated chemical diets will effectively sustain man, in the absence of physiological or psychological complications, over a period of several months (9).

The basic composition of the diet employed in the above study was: *amino acids:* L-lysine·HCl, 3.6 gm; L-histidine·HCl·H_2O, 1.6 gm; L-arginine·HCl, 2.6 gm; L-tryptophan, 0.7 gm; L-phenylalanine, 1.8 gm; L-leucine, 3.8 gm; L-isoleucine, 2.4 gm; L-threonine, 2.4 gm; L-methionine, 1.8 gm; L-valine, 2.7 gm; L-alanine, 2.6 gm; glycine, 1.7 gm; sodium L-asparate, 6.4 gm; L-proline, 10.3 gm; L-glutamine, 9.1 gm; ethyl L-tryosinate·HCl, 6.8 gm; L-serine, 5.3 gm; ethyl L-cysteinate·HCl, 0.9 gm. *Minerals:* ammonium molybdate·$4H_2O$, 0.42 mg; cobaltous acetate·$4H_2O$, 1.67 mg; cupric acetate·H_2O, 2.50 mg; ferrous gluconate, 0.83 mg; sodium glycerophosphate·$5\frac{1}{2}$ H_2O, 5.23 gm; calcium chloride·$2H_2O$, 2.44 gm; magnesium oxide, 0.38 gm; manganous acetate·$4H_2O$, 18.30 mg; potassium hydroxide, 3.97 gm; potassium iodide, 0.25 mg; sodium chloride, 4.77 gm; sodium benzoate, 1.0 gm; zinc benzoate, 2.82 mg. *Water soluble vitamins: p*-aminobenzoic acid, 416.0 mg; ascorbic acid, 62.5 mg; *d*-biotin, 0.83 mg; calcium pantothenate, 8.33 mg; choline bitartrate, 231.0 mg; folic acid, 1.67 mg; inositol, 0.83 mg; niacinamide, 10.0 mg; pyridoxine·HCl, 1.67 mg; riboflavin, 1.5 mg; thiamine·HCl, 1.0 mg; cyanocobalamin, 1.67 μg. *Carbohydrates:* glucono-δ-lactone, 17.2 gm; glucose, 555.0 gm (or glucose, 416.0 gm plus sucrose, 139.0 gm). The quantities denoted are the amount of each component dissolved in one liter of aqueous dietary solution. These solutions were provided at four intervals daily to each experimental subject, who ingested a total of 2100–3700

TABLE I

SERUM CHOLESTEROL VALUES OF SUBJECTS ON CHEMICAL DIETS (MG%)[a]

Subject code No.	Weeks on chemical diets								
	Phase I[b]				Phase II[b]			Phase III[b]	
	0	1½	2½	4	5	6	7	8	19
A-1	176	—	184	144	168	190	186	155	145
A-3	178	160	146	120	178	190	213	155	—[c]
B-1	152	134	130	120	140	162	157	122	—[c]
B-2	210	148	138	134	150	176	160	131	117
C-1	220	180	172	170	172	206	224	184	151
C-2	232	160	162	155	168	167	180	138	133
C-3	189	178	174	170	220	200	247	173	160
D-2	189	123	146	142	154	160	164	131	143
D-3	258	228	228	202	250	267	308	254	201
E-2	240	148	176	146	158	188	180	168	151
E-3	262	195	170	136	166	196	214	189	143
F-2	354	269	226	260	260	294	318	290	214
F-3	310	269	252	242	244	263	285	262	211
G-1	223	136	160	114	130	144	158	148	120
G-2	257	173	180	184	198	212	226	200	—[c]
G-3	155	120	120	124	128	128	140	108	105
H-1	220	125	130	134	140	151	160	136	128
H-3	262	225	170	180	184	209	226	200	140
Mean	227	175	170	160	178	195	208	175	151

[a] The normal range of serum cholesterol levels, according to the procedure employed, is 150–260 mg per cent.

[b] Carbohydrate composition of diets: Phase I, 100 weight per cent glucose; Phase II, 75 weight per cent glucose, 25 weight per cent sucrose; Phase III, 100 weight per cent glucose.

[c] Subject terminated participation prior to sample withdrawal.

calories, based on his subjective need; each subject ingested an amount of diet daily that was sufficiently constant to eliminate caloric intake as a variable. In addition, 2 gm of pure ethyl linoleate (which incorporated the fat-soluble vitamins: vitamin A acetate, 3.64 mg; calciferol, 0.057 mg; α-tocopherol acetate, 57.29 mg; and menadione, 4.58 mg) was given to each subject as a separate daily supplement. Ingestion of anything else, with the exception of distilled water, was prohibited. Each subject followed a daily activity schedule that included regulated, moderate physical exercise.

During the course of the clinical study, it was observed that chemical diets may exert pronounced effects on serum cholesterol levels, which effects are governed by the nature of the carbohydrate present in the diet. Fasting blood samples, drawn once weekly, revealed that during the first 4 weeks of the experiment, when 100 weight per cent of the sugar in the diet was glucose, the serum cholesterol levels of each of the 18 subjects fed this diet exhibited a progressive and dramatic drop (Table I), with the mean average for the group as a whole showing a decrease from the pre-experimental level of 227 mg per cent to 160 mg per cent after the fourth week (Fig. 1). Substitution of 25 weight per cent of the glucose in the otherwise identical diet with an equal weight of sucrose, at the end of the fourth week, was accompanied by a progressive increase in the total serum cholesterol levels to a mean average of 208 mg per cent at the end of the 3 weeks that this particular diet was provided. Finally, replacement with an equal weight of glucose of the sucrose component, in this latter diet, was followed by a precipitous drop in the total serum cholesterol levels to a mean average of 175 mg per cent at the end of 1 week and 151 mg per cent at the end of the 19th week of the experiment. Upon return of the subjects to natural foodstuffs, a sharp rise in the serum cholesterol of each occurred without exception, with an average mean level of 233 mg per cent shown by the group as a whole after 4 weeks. No correlation was observed between weight change and serum cholesterol levels over the 19-week period.

On the basis of a statistical analysis of the mean values of the serum cholesterol levels at the end of the 4th, 7th, 8th, and 19th weeks (see Table I), the following conclusions were drawn (95% confidence level): (a) each of the two progressive *decreases* in serum cholesterol level with the diet containing glucose as the sole sugar is statistically significant, and (b) the progressive *increase* in serum cholesterol level upon partial substitution of the glucose with sucrose is also statistically significant.

Since the chemical diet employed in the aforementioned clinical study contained less than 0.5% ethyl linoleate as the sole source of dietary fat, experimental data are now available which unequivocally demonstrate an important relationship between the nature of the dietary carbohydrate and serum cholesterol levels. These data are of special interest in view of (a) the postulated

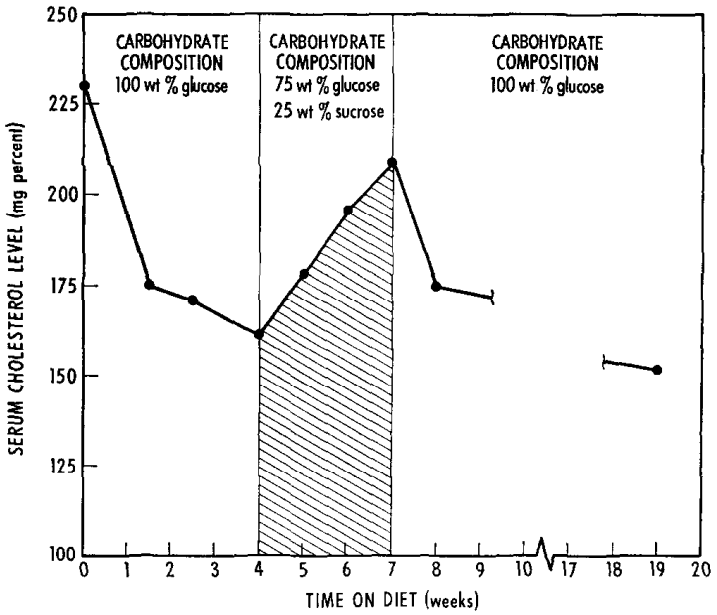

FIG. 1. Variation of total serum cholesterol levels with carbohydrate composition of complete chemical diets. Values given are mean values of 18 subjects.

relationship between blood cholesterol levels and coronary disease, (b) the recent report (10) that ". . . a high consumption of sugar is an important factor in the causation of myocardial infarction and peripheral arterial disease," and (c) the present intensive search for relationships between specific dietary components and the incidence of cardiovascular disease.

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