The first 4.2 ml of the effluent was colorless. The next 1.8 ml, fraction 1, was slightly colored; the next 1.8 ml, fraction 2, was deep reddish brown; a third 1.8 ml, fraction 3, was slightly colored. No more of the effluent, up to a total volume of 22.2 ml, was colored.

The three colored fractions were submited to Karl Folkers, of Merck and Company, for analysis. He reported the following: fraction 1, 80 microbiological units per mg; fraction 2, 2000 units per mg; fraction 3, 240 units per mg.

As the color accompanies the activity, it is easy to know what portion of the effluent to collect. The method offers a way of obtaining B_{12} active material from commercially available sources, which already contain the activity in a conveniently small volume.

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Fat Absorption and Atherosclerosis[1]

G. H. Becker, Jacob Meyer, and H. Necheles

Department of Gastro-Intestinal Research, Medical Research Institute of Michael Reese Hospital, Chicago

In our work on the effect of age on fat absorption (*1*) we have observed a phenomenon which may be fundamental in the pathogenesis of atherosclerosis.

Using a modification of Frazer's chylomicron dark field technique (*1, 2*), we have studied the absorption of fat in thirty young and in thirty old subjects with average ages of 18 and 76 years respectively. The fasting subjects were given a standard fat meal of ½ g of oleomargarine/kg of body weight on 2 oz of white toast together with a cup of tea. Samples of finger blood were drawn before and after the meal at regular intervals as indicated in Fig. 1. The number of chylomicrons in the serum of each specimen was determined and chylomicrographs were constructed.

As shown in Fig. 1, the chylomicron counts of young subjects reached a peak at 2½–3 hr, and returned to fasting levels by the end of the 5th hr. The counts of the old group on the other hand did not reach their peak until 8–12 hr, and they did not return to fasting levels until 24 hr had elapsed. In addition, the total number of chylomicrons was found to be consistently and considerably higher in the old than in the young group.

Previous work by Gage and Fish (*4*), and Frazer (*2*) has established that the chylomicron curve serves as an index of postabsorptive lipemia. From our results it is obvious that a definite delay in the rate of absorption and a definite increase in the total absorption of corpuscular fat exist in aged individuals as compared with a group of young subjects.

The observations of Hueper (*6, 7*) and of Moreton (*9, 10*) seem to indicate that the alimentary hyperlipemia and its accompanying high concentrations of chylomicrons

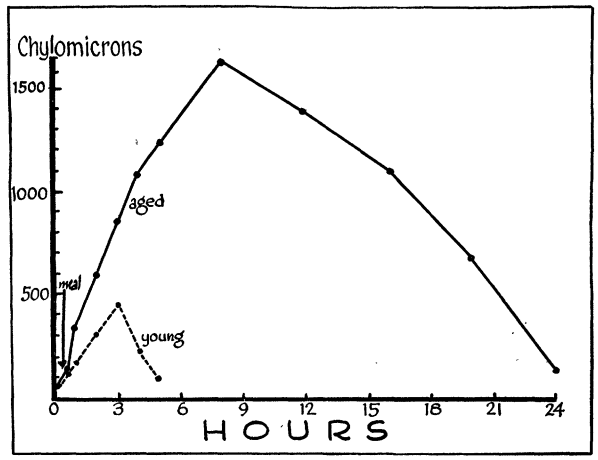

FIG. 1.

in the peripheral blood, occurring in normal individuals, are indistinguishable from the sustained hyperlipemia and hyperchylomicronemia of pathological and experimental origin which have been found to be characteristic of the known causative conditions of atherosclerosis.

Hueper (*6, 7*) and Moreton (*9, 10*) state that macromolecular substances can be deposited and can damage the internal layers of arteries. Chylomicrons are macromolecular bodies containing largely neutral fat and some cholesterol (*2, 3, 9, 10*). The neutral fat, according to Moreton (*9, 10*), disappears rapidly from the intima and subintima, while the cholesterol remains and accumulates gradually, attracting macrophages, giving rise to foam cells, and ending in atherosclerosis.

The question is still controversial, whether endogenous or alimentary cholesterol produces atherosclerosis in man. This cholesterol is in true solution and it is possible that chylomicrons, which are macromolecular aggregates, are the source of irritation and degeneration of arterial walls, rather than cholesterol, fats, or other lipids in the dissolved state (*6, 7, 9, 10*). In the chicken, endogenous cholesterol seems to play a large role in the genesis of arteriosclerosis (*5*).

Increased chylomicronemia following fat-containing food occurs at every age. However, it lasts only a relatively short time and it is only of moderate intensity in young persons. With increasing age, and particularly above 50 years of age, chylomicronemia is of greater intensity and it is practically permanent. If the chylomicrons play a role in atherosclerosis, the fundamental physiological basis of the mechanism of the disease may lie in this fact.

Another significant observation made was that oral ad-

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ministration of lipase[2] or of a detergent (Tween 80)[3] with the fat meal reduced the hyperchylomicronemia of older persons to practically the level of the younger age group. The chylomicron count in young persons following a fat meal was not influenced materially by the administration of lipase with the fat meal.

We have found earlier (*11*) that pancreatic lipase secretion and blood lipase levels are significantly lower in older persons than in young ones. The effect of lipase or of a detergent in reducing the hyperchylomicronemia in old subjects to levels of young subjects seems to support the assumption that the mechanisms of fat digestion or of fat absorption, probably both, change with aging. It is not probable that hyperchylomicronemia is due to delayed disposition of circulating fat, because intravenous injection of equal volumes of hyperlipemic plasma into young and old subjects yielded similar chylomicron curves (*1*). It is premature to speculate whether administration of lipase or of detergent to normal persons may prohibit the development of atherosclerosis, or whether the progression of the disease can be interrupted by the administration of lipase or of detergent. Animal experiments may answer this question.

In persons over 50 years of age ingestion of a small amount of oleomargarine was followed by a practically 24 hr increase in the chylomicron count in the serum. In younger persons the chylomicron curve returned to fasting levels within 5 hr. Since all people eat some fat at least once a day, increased numbers of fat particles circulate in the blood of older persons practically permamently. If it is true that particulate fat, circulating in the blood, leads to atherosclerosis, the condition leading to that degenerative disease has been found.

Administration of lipase or of detergent with the fat meal reduced the chylomicron counts and the duration of increased counts of old subjects to levels of young subjects.

Work on animals will show whether atherosclerosis can be influenced by drugs affecting digestion and absorption of fat.

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[2] Kindly supplied by Dr. D. Klein, Wilson Company, in the form of concentrated pancreatin.

[3] Kindly supplied by Dr. G. R. Hazel, Abbott Laboratories.

A Vibrating Tissue Slicer[1]

Samuel P. Bessman

Children's Hospital, Washington, D. C.

Some recent studies (*5*) of the metabolism of brain cortex slices required six or eight parallel experiments on the same brain. Neither the freehand method (*4*), nor the microtome method (*3*) yields sufficient material from brains of guinea pigs or rabbits. We have found that a vibrating blade with freehand manipulation can provide 500 mg of cortex slices from a full-grown guinea pig brain. The source of vibration is a mechanical wood carving tool (Burgess Vibro-Tool), which develops a push-pull vibration at a frequency of 120 per sec. There is only slight lateral vibration. In the chuck, we fastened a split metal adapter tightened by a wing nut, which can hold 2-in. pieces of Stadie blades or single-edge razor blades.

TABLE 1

	Hand cut		Cutter	
	Wet weight in mg	Qo_2	Wet weight in mg	Qo_2
Mouse liver	122	5.77	78	6.08
	94	6.28	82	5.58
	112	5.79	92	6.06
Average		5.95		5.92
Guinea pig brain cortex	52	9.18	57	10.20
	72	9.72	71	9.29
	53	10.10	63	9.56
Average		9.67		9.68

The platform upon which the cerebral hemisphere is placed is made by filling a 50-mm crystallizing basin with ice and water to overflowing, sliding the bottom of a larger basin over it, and inverting the basins. The ice floats to the top, causing the platform to be chilled. The cerebral hemisphere, with meninges and blood vessels removed, is placed on a small square of moistened filter paper on this platform.

The cutter, with the edge facing the operator, is grasped firmly in both hands, one arm resting on the table edge for added stability. The vibrating blade is drawn slowly through the tissue, toward the operator, with a wrist motion. The slice lies flat on the blade, and the plane of the cut can be varied for thickness of slice and for contour of the tissue without the necessity of guiding and sawing simultaneously, which is inherent in the freehand method. The slice can be picked off the blade with a fine forceps or can be removed by dipping the vibrating blade into a small beaker of chilled Ringer's solution. The blade must be moistened slightly if the

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