NATURE OF THE FATS AND ALLIED BODIES IN CHYLOUS URINE.

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It is only rarely that the opportunity of examining chylous urine, especially in this climate, is presented, and as such urine was offered me by Dr. Dearing from a patient of his in large enough amounts for a comprehensive analysis, it seemed worth while to make a careful research into the nature of the fats comprising this emulsion, as such urine really is. All the facts I could learn concerning the patient were: sex, female, and the fact that she had spent many years in a tropical climate. The urine had the usual white, milky, opaque appearance, and after three weeks standing on ice, showed no tendency to clear up by precipitation of any of its suspended constituents. The peculiarities of the urine were undoubtedly due to parasitical influences (filiaria sangunis). This urinary fat must come, according to Franz Erben,[1] from (*a*) Excessive degeneration of the epithelium of the urinary tract; (*b*) Secretion of the fat by the renal epithelium; (*c*) Direct passage of fat from blood to urine; (*d*) Direct passage of fat containing lymph into the urine. The first proposition can be excluded because the fat is only found during digestion, and after twelve hours of fasting is reduced to a minimum, and the same phenomenon is noticed when the patient is kept upon a fat-free diet. A second proof of this rests upon the fact that when the fat of the food was colored with Sudan III. the fat in the urine was also colored, while in animals where the fatty tissue is colored by feeding Sudan III. the fat produced by fatty degeneration (phosphorus feeding) is uncolored. The possibility of secretion of fat may be eliminated by the fact that the fat changes with the nature of that fed; for instance, if olive oil be given exclusively the fat from the urine at once becomes fluid at an ordinary temperature. Nor can it be the result of direct sepation from the blood, because in two cases of chyluria in which the amount of fat in the blood was determined it was found to be respectively .328 per cent. and .378 per cent., while the urine contained .687 per cent. and 1.032 per cent fat, and in my own case the urine contained in two different analyses .523 per cent. and .621 per cent., all of which amounts are vastly greater than those found in the blood, while we would expect a similar or perhaps slightly greater amount (due to the absorption of water in the straight tubules of the kidney) than that found in the blood. Hence we are limited to the chyle as the source of the fat in the urine, for it cannot come from ordinary lymph containing from 0.2 to 0.4 per cent. fat for the same reasons adduced for its not coming from blood. In regard to other efforts made in this direction, we note that Hensen[2] found that the fluid from a lymph fistula contained 2.25 per cent. fat which on a limited fat diet fell to .62 per cent. containing .018 to .102 per cent. cholesterin and some soaps. Eggel[3] isolated a fat from chylous urine which contained fatty acids, neutral fat, cholesterin, and lecithin. Brieger[4] obtained from five and one-half liters of chylous urine 8.93 grams fat, containing .189 grams cholesterin, lecithin, and 6.73 grams fatty acids, melting-point 31° C. Grimm's case[5] (filaria chyluria) had a urine containing fat which consisted of 55–67 per cent. palmitin and 33–45 per cent. olein.

To proceed to the process of isolation: 500 c.c. of urine were evaporated to dryness at a low temperature with a small amount of washed sea sand, by which a tough, leathery dark-brown substance resulted, which was removed, finely divided by scissors, and placed for a short time in the drying oven, where as much moisture was removed as could be at a temperature of 50° C. This mass was then placed in a Soxhlet apparatus and extracted three days with ether, and the residue three days with absolute alcohol; the alcohol solution filtered hot, evaporated, and the residue again extracted with ether and both ethereal extracts united. This collective ether and alcohol extract was shaken with a weak solution of sodium carbonate and again extracted with ether, which removed the neutral fats, leaving the soaps. The watery solution containing the soaps was acidified lightly with sulphuric acid, and the freed fatty acids filtered through a dry-weighed filter and washed with water until no further acid reaction was found, then dried and weighed. Afterwards they were dissolved in alcohol-ether titrated with $\frac{N}{10}$ NaOH (alcoholic solution). After removal of the fatty acids the ethereal solution, containing neutral fats, cholesterin, etc., was evaporated to dryness and saponified with alcoholic potassic hydrate solution, the alcohol driven off, redissolved in water, and shaken again with ether, the ether driven off in small portions in a weighed watch crystal which showed by microscopic examination almost pure crystals of cholesterin. This was further purified by washing with cold dilute alcohol to which a drop of hydrochloric acid had been added, dried over sulphuric acid, and weighed. The watery solution of the soaps was now acidified with sulphuric acid and warmed until the ether was driven off, then lead acetate solution added as long as a precipitate formed. Again this was shaken with ether in a separating funnel to remove the lead oleate, and the ethereal solution shaken with hydrochloric acid and water, which precipitated the lead, leaving the oleic acid in the ether. The ether was driven off and the oleic acid was converted into sodium soap by cooking with soda, and after evaporation to dryness, extracted by means of boiling alcohol from the lead carbonate, brought into watery solution, the palmitic and stearic acids freed by sulphuric acid, evaporated to dryness, again extracted with alcohol-ether and weighed. The amount obtained was so small in both instances that no attempt was made to separate the latter acids, and they were calculated together. The result of two examinations made on the urine, taken at two periods, can be expressed in a short table, as follows:

	No. 1. grms.	No. 2. grms.	
Total ether and alc. extracts	2.6173	3.126	
Fatty acids	.005	.0032	In 500 cc.
Cholesterin	.015	.0167	urine.
Oleic acid	.1764	.201	
Palmitic and stearic acids	.031	.040	

From this table it can be readily seen that the free fatty acids are present only in small traces, the most of the fat existing in the form of the glycerides, or neutral fats. This is to be expected in accordance with the theory of origin, and corresponds very closely with the findings of Munk and Rosenstein, who had the opportunity of examining the fluid discharged from a lymph fistula. It is not impossible that a portion of these fatty acids may have been normal but in much smaller amounts than found here. S. Hibbenette[6] found in each ten liters of normal urine from .0165 to .025 grams of fatty acids which consisted chiefly of oleic acid, but also contained palmitic and stearic acids. Salkowski mentions carbohydrates as the only food substance to which there is a limit of assimilation, but refers to Brieger's demonstration that some individuals have a limited assimilative power for egg albumen, the excess appearing in the urine; but we may have also a limited assimilative power for fats, as evidenced in this instance, by the appearance of non-volatile fatty acids in the normal urine.

Unfortunately no account was taken of the soaps which must have been present in large quantities, as by means of these the neutral fats are kept in emulsion.

The amount of cholesterin shows how abundant this substance is in the digested fats; what its function is, what part it plays in the economy of the body is not known, but we find it in large quantities in the brain and nerves, in fact in all structures in which cells predominate; as a very insoluble constituent of bile we may recognize it in the formation of gall stones and the coprosterin, — a hydrated cholesterin, formed, according to the latest investigations, by bacterial action in the intestines. On account of its great insolubility except in cholic acid, its natural solvent in the bile, we may safely take it for granted that this is not soluble in the urine, and that the renal cells have the power of removing this substance by a selective vital process and not by the usually accepted action of osmosis.

Of the total fatty acids found, 85.05 per cent. in one case and 83.4 per cent. in the other is oleic acid, while the remainder is made up of stearic and palmitic acids, which were formerly known as margaric acid. These results correspond fairly closely with the quantities which L. Langer[7] found in the fluid fat of the adult human adipose tissue, 89.8 per cent. oleic acid and the remainder palmitic and stearic acids. A peculiarity of the oleic acid thus extracted and isolated from the urine is, that it remains solid at a temperature of 29° C., while it should normally be liquid at any temperature above 14° C. It is a well-known fact that the addition of a fat having a low melting-point to one or two having a higher will reduce the melting-point of the others, but there seems to be no reason why oleic acid from its association with its companions of a higher melting-point in this urine should, when isolated, have its own raised. They differ, however, very much from Erben's results, who found that the oleic acid comprised 58.40 per cent. of the total fatty acids, while the palmitic and stearic comprised the rest. This may very well happen, however, since the chyle is of no fixed composition, the latter depending on the nature of the fat taken as food. There was found here .0052 per cent. and .0057 per cent., respectively (of cholesterin), while Erben found 1.715 per cent. of the same substance, as compared with the total fats extracted. In addition to the fat, sugar and protein bodies are sometimes found in these urines. In this case no proteins were found, nor could sugar be detected in the original urine, but from the alcoholic extract of the residue obtained by evaporation of the urines, when converted to a watery solution, a body, reducing both Fehling and Nylander's solution, was found having a levogyrous power which, when calculated as dextrose, would give .080 grams in 500 cc. of urine. This body was not fermentable, nor was its rotatory power increased by splitting with acids, nor would it give an osazon with phenylhydrazin. It would seem to correspond to a sugar isolated by Leo.[8] Brieger[9] (quoted in Naunyn) also mentions the finding of sugar in chylous urine, but does not specify the nature of the sugar fully.

With all our knowledge of the nature and origin of the fat, we are still at a loss as to the manner in which the filaria act upon the chyle; whether by mechanically plugging the lymph vessels, or by establishing a limit to the assimilation of fat, remains to be determined. The latter supposition has some basis in the analogous limited assimilation of sugar in malaria, though the latter condition is explained by the fact that the malarial poison destroys the glycogen-forming function of the liver cells. The elimination of sugar by the kidneys only during the period of digestion and absorption in hepatic glycosuria is also similar to the elimination of fat during the same period. The mechanical obstruction theory is also opposed by the limited elimination of sugar and albumin, while the fat is extracted in such large quantities. If the cause were obstruction only, there is no reason why all bodies should not be retarded equally and appear in equal quantities in the urine; in fact one would regard fat as being the least easily eliminated by the kidney and hence appear in least amount. Furthermore when true obstruction does appear, as in regurgitant heart disease, emphysema, cirrhosis of the liver, or pregnancy, the appearance of albumin in the urine is not limited to the period of digestion and absorption as in the case of the fat.

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