a hæmochromogen is formed which slowly loses its iron, the end-product being also hæmatoporphyrin.

It is evident that the integrity of the hæmoglobin molecule is dependent upon the quantity of secretion that the adrenals supply to the blood, and also upon the condition of that molecule at a given time. In other words, while the adrenals may be supplying their normal proportion of secretion, the hæmoglobin molecule in the red corpuscles of venous blood—*i.e.*, blood about to return to the vena cava for a fresh supply—may be compared to that of blood during insufficiency. Even as hæmoglobin, the blood-pigment is loosely combined; when approaching the end of its systemic circle, it is still nearer the state of disintegration—according to the activity of the oxidation processes which it has subserved. Starting from the lungs as oxyhæmoglobin, it returns promptly to the heart as hæmoglobin or reduced hæmoglobin, ready to absorb at once another supply of suprarenal secretion and, once in the lungs, take up its oxygen.

Blood from the head, extremities, and other structures in which the drain upon its resources has not been excessive returns such a molecule to the heart; it is still efficient as an oxygen-carrier. *But not so with the blood from any organ directly connected with the digestive system.* As is well known, all the blood from the organs of the alimentary tract—stomach, intestine, pancreas, and spleen—is not returned to the heart before it has been submitted to whatever action the liver may have upon it; then only can it re-enter the circulation through the hepatic veins, which carry the blood to the vena cava. But not all the blood may thus be rejuvenated; some has gone beyond; it has, indeed, almost reached the state of hæmatoporphyrin, the last on the list of pigments, that which appears in the most advanced stages of suprarenal insufficiency. We have seen that hæmatoporphyrin and bilirubin are similar; and, as is well known, it is bilirubin which passes out with the bile.

A salient feature of the hæmoglobin molecule is missing here, however, namely: the iron. As stated, a reducing agent, if used in the presence of oxygen, will reduce hæmoglobin in the absence of oxygen; the primary product is a hæmochromogen which gradually parts with its iron, leaving as end-

product hæmatoporphyrin. As bilirubin and hæmatoporphyrin are fundamentally identical, the presence of the former in the bile must be the result of a similar process in the liver. That such is the case is sustained by considerable collateral evidence, first of which is the invulnerability of the hæmoglobin molecule.

Paradoxical as this statement appears, it nevertheless constitutes the key-stone of the entire edifice, since it is only when *vulnerable* that the molecule becomes the prey of disintegrating influences. I have used the words "reducing agents" several times; but the hæmoglobin molecule does not yield to even moderately-strong reagents of this nature; indeed, only a powerful agent—sulphuric acid, for instance—will dissociate it: an exemplification of the wonderful binding power which the suprarenal secretion must exert upon all its constituent parts. Still, we must not overlook the fact that the oxidizing substance in the blood-plasma is identical to this binding compound. Indeed, I have accumulated so much testimony affirming the fact that the plasma *per se* is a potent source of energy, while the red corpuscles always played so secondary a rôle in the various intrinsic functional mechanisms, especially those concerned with muscular and glandular elements, that I have been led to conclude that the red disks are, after all, but servants of the blood-plasma: pack-mules, as it were, from which it can draw, as needed, enough oxidizing substance to maintain its own functional potentiality as previously stated.

Under these circumstances we can readily see how the hæmoglobin molecule, gradually deprived of its binding oxidizing substance during the inordinate metabolism which the tissues of the digestive organs undergo during their periods of activity, should yield more readily to dissociating influences. It is evident that a molecule so weakened, thrust in blood such as that of the portal vein,—a vast sewer replete with physiological waste-products and deoxidized blood-cells, all bathing in a plasma itself despoiled of its oxidizing substance,—should soon become dissociated. Transported through gradually narrowing channels, the walls of which, like all tissues, eagerly absorb any loose oxygen that it may contain, it must inevitably undergo the transformation of hæmoglobin referred

to, *i.e.*, into the primary hæmochromogen, which soon drops its iron, leaving as end-product bilirubin.

We have here the identical process that occurs in the brain or other structures when blood-clots are disorganized into hæmatoïdin preparatory to absorption. "The bile-pigments originate from hæmoglobin," says Professor Howell; "this origin was first indicated by the fact that in old blood-clots or in extravasations there was found a crystalline product, the so-called 'hæmatoïdin,' which was undoubtedly derived from hæmoglobin, and which, upon more careful examination, was proved to be identical with bilirubin. This origin, which has since been made probable by other reactions, is now universally adopted." That the influence of the suprarenal secretion rests upon as solid a foundation is illustrated by the experiments of Boinet, who found the blood of a large number of rats from which he had removed the adrenals replete with "hæmatoïdin."

To trace the itinerary of the two products, iron and bilirubin, through the liver, naturally brings the hepatic cell within the scope of our inquiry, since we have to account for the transfer of the former to the bile and the return of the iron to the general circulation.

The functional importance of iron in the hæmoglobin molecule is generally recognized. Yet, the pigments, when separated from it, are not unable to take up oxygen. Indeed, we have ample evidence of this in the formulæ of the very products of which bilirubin is the primary compound. Thus, while bilirubin is $C_{16}H_{18}N_2O_3$, biliverdin is $C_{16}H_{18}N_2O_4$, and the latter can readily be prepared artificially from the former by oxidation. "It is supposed that, when the blood-corpuscles go to pieces in the circulation," says Howell, "the hæmoglobin is brought to the liver, and then, under the influence of the liver-cells, is converted into an iron-free compound: bilirubin or biliverdin. It is very significant to find that the iron separated by this means from the hæmoglobin is, for the most part, retained in the liver, a *small portion* only being secreted in the bile. It seems probable that the iron held back in the liver is again used in some way *to make new hæmoglobin* in the hæmatopoietic organs." We have seen that *it is not under the influence of the liver-cells, as now believed, that hæmoglobin is dissociated:* an important feature, since it removes the main element of confusion from our path. Indeed, we can now easily account for the retention of the greater part of bilirubin in the liver, since we have at our disposal all the constituents for the synthetic production *within* the portal capillaries of the hepatic lobule of new hæmoglobin, and particularly oxidizing substance, brought there by the terminals of the hepatic artery.

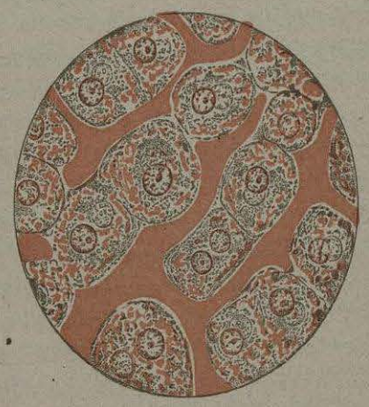
On examining, on page 329, the illustration from Piersol's work, the distribution of the hepatic artery's terminal arterioles or capillaries will be found to be unusual. Immediately above the margin of the lobule—*i.e.*, where the portal or interlobular vein breaks up into the capillary net-work of the lobule—the hepatic arteriole may be seen to open *into* the portal capillary. The inference is obvious. *The hepatic artery coming directly from the cœliac axis, brings freshly oxidized blood,—i.e., oxidizing substance,—which, mixing freely in the narrow channels of the lobule with the portal blood, at once groups bilirubin and iron, and builds up all the hæmoglobin that the constituents present (including what iron the splenic leucocytes and those from the intestinal follicles have brought) allow. What bilirubin cannot, owing to deficiency of either of the other constituents, be utilized, becomes an excretory product; and with many others it enters the hepatic* CELL *and is passed out with the bile.*

That deficiency of oxidizing substance (adrenoxidase) can increase the excretion of bilirubin has been repeatedly shown herein. I may refer, for example, to the many forms of acute poisoning and to the diseases attended with suprarenal insufficiency in which there is increased excretion, either in the urine or fæces, of hæmatoporphyrin, methæmoglobin, urobilin, stercobilin, etc.: *i.e.*, of some derivative of hæmoglobin.

I have referred to the hepatic cell as a miniature sponge. This comparison, due to Berdal, is especially warranted, since Schäfer[14] noted the existence, within this cell, of canaliculi which are in direct communication with the blood-capillaries. Having injected carmine gelatin into the portal vein, the colored substance filled this vessel and its subdivisions, besides the canaliculi, but no other structure. It may, therefore, be

[14] Schäfer: Journal of Physiology, Jan. 31, 1902.

inferred, says Professor Schäfer, "that the injection has passed directly from the blood-vessels into the liver-cells; indeed, here and there one can see what appear to be such direct communications." These can readily be seen in the annexed illustration. He refers to the conclusion reached by Browicz,[15] based on appearances, normal and pathological, "that there must exist a net-work of nutritive *canals* within the hepatic cells which are in direct communication with lobular capillaries"; this he had not as yet, however, verified by injections. Schäfer's observation probably accounts for the direct transfer of the bilirubin to the biliary capillaries, along with other



LIVER OF RABBIT INJECTED FROM THE PORTAL VEIN. THE INJECTION HAS PASSED INTO CANALICULI WITHIN THE LIVER-CELLS. (*E. A. Schäfer.*)

products of oxidation, to which we will refer later on. Indeed, J. W. and E. H. Fraser[16] are also stated to have found intracellular *passages* communicating with the blood-vessels in the hepatic cells of frogs. For the present it seems logical to conclude that one or more of the canaliculi may lead to the vacuole previously referred to as nearest the bile-capillaries, and that it is in this vacuole that bilirubin joins the bile. That even this vacuole is supplied with a canaliculus we have already

[15] Browicz: Bulletin de l'Académie des Sciences de Cracovie, 1899.

[16] J. W. and E. H. Fraser: Journal of Anatomy and Physiology, vol. xxix, p. 240, 1895.

seen; Kupffer found it to afford a direct channel between this bile-reservoir and the bile-capillaries *per se*.

The Hepatic Tissues in their Relations to Bacteria.—A prominent feature of the work so far done is the evidence furnished that several physiological processes now ascribed to the hepatic cell in no way involve this structure, and that the portal vein itself and the *intercellular*[17] capillaries are the seat of several of these processes.

Before proceeding further, however, reference must be made to the connection between bacteria and the *normal* liver. I emphasize "normal" here, because I can thus simultaneously lay stress upon a feature which plays a predominating rôle in disease: *i.e.*, the fact that anatomically, as far as bacteria go, there is no direct normal connection between the digestive system and this organ. The liver, in fact, is essentially a physiological organ in the sense that it is mainly intended to rid the system of waste-products and to economize others that may again prove useful, by preparing them for reabsorption in the intestine.

We have seen that the venules of the villi allow iron-pigment leucocytes to enter the mesenteric veins which carry their blood to the portal. A depraved condition of all the digestive structures—such as that induced by alcoholism, for instance—can so lower the functional activity of these structures as to cause these venules to lose their normal turgescence and afford passage to bacteria, alcohol in large doses being known to impair metabolism. The intestinal venules under these circumstances, surrounded by weakened protective structures, can well give passage to Adami's cirrhosis bacillus, for instance, or any other capable of coping with what prophylactic conditions may still prevail. "The portal vein can transport to the liver morbid germs from the intestinal surface," says Labadie-Lagrave. "One of the best established pathogenic connections of this kind is the influence exerted upon the development of hepatitis by dysentery; although this relationship is not constant, all observers have noted it. Phlebitis

[17] We find it necessary to give the terminals of the portal this name in order to avoid confusion; they contain blood from both the portal and hepatic channels, and in reality form part of both as extensions.—S.

starting from an ulcerated area and directed toward an hepatic focus has also been observed. When the primary portal structures are normal, transmission of the putrid material may occur through the lymphatics. While this fact seems admissible, it has not been verified." Again, pathological conditions of the stomach, pancreas, or spleen may supply the portal vein with pathogenic elements. In the normal subject, however, *the liver-tissues per se are totally isolated anatomically from any of the structures that come into contact with exogenous bacteria,* precisely as they are in other organs: the muscles, the heart, etc. That its blood-stream affords protection from disease is undoubted, however, judging from the leucocytes that are constantly entering the organ, and the perivascular lymphatic channels. That the portal vein is also an important field for the splitting of toxalbumins and their reduction to harmless bodies we shall also see. But it seems quite clear that the liver itself is not primarily a germ-killing organ, and that its attributes are essentially chemical. This removes the hepatic cell still further from the functions now attributed to it, and suggests that the oxidizing substance in the lobular blood-vessels may be the main source of the liver's functional activity.

This brings us to the consideration of the functions in which the oxidizing substance in the blood-plasma acts as a reagent. We have already reviewed, in this connection, the synthesis of hæmoglobin; we will now take up and consider two equally important subjects: *i.e.,* the origin of urea and the conversion of sugar into glycogen.

Urea and its Formation.—We will first analyze an experiment by Schröder[18] in which the liver was taken from a freshly-killed dog and irrigated through its blood-vessels by a supply of blood taken from another animal. Howell refers to this experiment in the following words: "If the supply of blood was taken from a fasting animal, then circulating it through the isolated liver was not accompanied by any increase in the amount of urea contained in it. If, on the contrary, the blood was obtained from a well-fed dog, the amount of urea contained in it was distinctly increased by passing it through the liver, thus indicating that the blood of an animal after digestion contains something that the liver can convert to urea."

[18] Schröder: Archiv für exper. Pathol. and Pharm., Bd. xv and xix, 1882 and 1885.

Considered from my standpoint, this experiment has another meaning. During digestion, especially after copious feeding, as stated above, the entire organism is, to a certain degree, involved in the digestive process, as shown by the general sensation of heat often experienced after such a meal. As liver, intestines, pancreas, and spleen, even after gastric digestion has passed, are all operating together, the suprarenal activity is doubtless enhanced. In other words, at such times the blood contains either in its corpuscles or in its serum a more or less marked increase of oxidizing substance. Conversely, the fasting dog's blood—especially if the fasting has been prolonged—is really abnormal blood, in which the oxidizing substance is unusually low, since suprarenal activity is depressed with that of the rest of the tissues. We have also seen that, under these conditions, the tissues nevertheless continue to absorb their normal supply of oxygen, the blood being thus actively depleted while insufficiently oxygenated. It seems clear, therefore, that the blood of the well-fed dog contained more oxidizing substance than that of the fasting one.

That the injected blood taken from the well-fed animal should have been the source of the urea-forming substance is unlikely. Since the liver alone receives alimentary waste-products, it is only with blood from the portal vein that such substances could have been obtained. This is not specified. The urea-forming agent must, therefore, have been in the excised liver's portal channels, and the only available agency capable of inducing the reactions involved appears to be the oxidizing substance. Experimental evidence may be adduced to show that such is the case. I consider the blood of the hepatic artery, as previously stated, as the source of supply of the oxidizing substance, since it is directly derived from the cœliac axis. Stewart states that, "although the portal vein carries a much greater supply of blood than the hepatic artery, *ligation of the latter* causes a greater diminution in the ratio of the amount of urea to the total nitrogen in the urine than

ligation of the former. This seems to indicate that oxidation plays an important part in the formation of urea in the liver (Doyon and Dufourt)."

That the substances thus oxidized reach the liver by the portal vein needs hardly to be emphasized. But Foster says: "The introduction of even a small quantity of proteid material into the alimentary canal at once increases the urea in the urine, and in the curve of the discharge of urea in the twenty-four hours each meal is followed by a conspicuous rise. . . . We have seen reason to think that the proteids of a meal are absorbed not by the lacteals, but by the portal blood-vessels, and such bodies as leucin probably take the same course. This being so, all these bodies pass through the liver and are subjected to such influences as may be exerted by the hepatic cells."

Such bodies of leucin—one of the main products of nitrogenous dissociation—naturally follow the same course. Drechsel has suggested that all bodies of this class—*i.e.*, leucin, tyrosin, glycocoll, etc.—first undergo oxidation in the tissues, and that their ammonia and carbonic acid then combine synthetically, forming ammonium carbamate, this, in turn, being carried to the liver and there transformed into urea.

It is clear that these ammonia compounds take the course outlined by Foster: *i.e.*, the venules of the villi, the mesenteric veins, and finally the portal vein. That they undergo oxidation in the blood of these vessels, however, is not likely, for they contain probably the most watery blood of the organism, and that most depleted of its oxygen.

Quite another field of activity is afforded, however, when the hepatic lobule is reached; *here the ammonia compounds meet the oxidizing substance brought by the hepatic artery's capillaries.* Taking the ammonium compound referred to by Drechsel, for example, the series of reactions outlined by him seem to follow in normal sequence: 1. In the portal vein: hydrolytic cleavage with the formation of amido-bodies, such as leucin, tyrosin, aspartic acid, glycocoll, etc. 2. In the hepatic-lobule capillaries and their oxidizing substance: oxidation, with the formation of ammonia, carbonic acid, and water, followed by the synthetic union of ammonia and carbonic acid,

forming the carbamate of ammonium. This being dehydrated, urea is formed, as shown in the following equation:—

$$CO<\begin{matrix}NH_2\\ONH_4\end{matrix} - H_2O = CO<\begin{matrix}NH_2\\NH_2\end{matrix}$$

That the conversion of ammonia compounds into urea does occur in the liver is sustained by experimental physiology. Howell refers to the experiments of Schröder, in which this is demonstrated as follows: "As further proof of the urea-forming power of the liver, Schröder found that if ammonium carbonate was added to the blood circulating through the liver —to that from the fasting as well as from the well-nourished animal—a very decided increase in the urea always followed. It follows, from the last experiment, that the liver-cells are able to convert carbonate of ammonium into urea. The reactions may be expressed by the equation:—

$$(NH_4)_2CO_3 - 2H_2O = CON_2H_4."$$

The foregoing facts, considered collectively, indicate that the formation of urea in the liver is probably accomplished in the following manner, taking Drechsel's series of reactions as standard of the numerous ones of the same class that must occur in this organ:—

Granting that the *nitrogenous* bodies are absorbed by the venules of the intestinal villi and transmitted by the mesenteric veins to the portal vein, the ramifications of which would then carry them to the hepatic lobules, *the first reaction would occur in the prelobular portal vessels: i.e., the nitrogenous bodies would undergo hydrolysis, with the formation of amides, leucin, aspartic acid, tyrosin, etc.* The second reaction would follow as soon as these bodies reached the pericellular capillaries, owing to the presence therein of the oxidizing substance supplied by the terminal branches of the hepatic artery; in other words, *further reduction of these bodies by oxidation to ammonia, carbonic acid, and water would occur in the pericellular capillaries of the lobule.* The third reaction would seem, like the first, to require comparatively inert surroundings: *i.e.*, a fluid not charged with oxidizing substance as is the blood of the pericellular capillaries. Such a medium we have, in all likelihood,

in the afferent venous channels, since it is very improbable that any oxidizing substance, so precious in all physiological functions—as I have now shown—should be wasted in vessels ultimately ending, *via* the hepatic veins, in the inferior vena cava. Hence, whether it involved a preliminary formation of an ammonic carbamate or proceed to immediate synthesis, *it appears as if the terminal reaction ending in the formation of urea occurred in the efferent venous hepatic channels.*

The salient point of this series of reactions is the fact that, contrary to the general belief, they all *occur, not in the hepatic cells, but in the blood-stream of the lobular capillaries.* The following facts show this to be the case: It is clear that, if urea is formed in transit through the vessels of the organ, it should appear as soon as, or at least soon after, its causative agencies are introduced in the portal system. We will recall the quotation from Professor Foster's text, in which he says: "The introduction of even a small quantity of proteid material into the alimentary canal at once increases the urea in the urine, and in the curve of the discharge of urea in the twenty-four hours each meal is followed by a conspicuous rise, etc." When we consider that the entire circulatory circuit occupies but twenty-six seconds, the cause of the rapid appearance of urea—heretofore unexplained—becomes apparent.

When the rôle of the oxidizing substance in the production of uric acid from the alloxuric bases was analyzed in the third chapter, uric acid was considered as the end-product of a series of reactions in which, according to modern views, these toxic nuclein derivatives were converted into benign ones. All nitrogenous products being transferred to the portal system, it now seems clear that normally the reaction must occur in the intercellular capillaries of the hepatic lobules, and that it is when this oxidation process in the liver is inadequate that the so-called "uric-acid diathesis" symptoms occur. As uric acid leaves the organism, as does urea, by the urine, it is evident that we are again dealing with a function totally disconnected from the hepatic cell *per se.* Again, we have repeatedly seen, in the preceding chapters, that the elimination of phosphoric acid was increased by the administration of suprarenal, pituitary, and other organic extracts and by various

drugs. As we have seen, increased excretion by the kidneys due to drugs always coincides with suprarenal overactivity, hence with enhanced oxidation. It thus becomes apparent that *many constituents of the urine, normal and abnormal, the origin of which is obscure, are connected with variations in the oxidation processes in the intercellular capillaries of the liver, caused by corresponding fluctuations in the functional activity of the adrenals.*

Glycogen and its Formation.—Glycogen obviously removes our inquiry from the arteriole to the hepatic cell, since this organ is that in which it accumulates; but we must not lose sight of the important fact that two processes are involved in the analysis,—(1) the formation of the glycogen and (2) its conversion into dextrose,—and that the latter reactions must occur in the vascular channels. Again, the first process seems so bound up with the formation of the bile that it becomes necessary to consider this subject simultaneously to avoid repetition.

The sponge-like construction of the hepatic cell due to its vacuoles, the delicate canals described by J. W. and E. H. Fraser, Browicz, and Schäfer, and, finally, the bile-collecting vacuole, or vesicle, leading through its own canaliculi to the perilobular bile-capillaries, does not appear to afford much room for protoplasm capable of undergoing functional metabolism, since this would have to be embodied in the partitions separating all these cavities. Yet, were it otherwise, the nucleus—often duplicated, particularly in herbivorous animals, almost one-third in size that of the entire cell, and containing nucleoli—would represent a useless structure. It seems evident, judging by the appearance of the cell as a whole, therefore, that the nucleus, which, as we have seen, is surrounded by a thin limiting layer of protoplasm, must impart its energy to this layer. This, in turn, being the central terminal of all the partitions, which, along with the cell's own pseudocovering, are protoplasmic, the vacuoles become receptacles, as it were, of the products of their walls.

Again, when we behold the minute canals so clearly shown in Schäfer's illustration (shown on page 340) a direct communication with parts external to the cell is evident in several