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Some Recent Views on the Structure and Function of the Kidney

By Gösta Glimstedt

The introduction of the punction technique of *Richards* (1939) was a turning-point in the history of renal research. By means of observations made with this method, it has been definitely established that the primary urine is formed through an ultrafiltration in the Malpighian corpuscles. Further, certain indications have been obtained regarding the function of single parts of the nephron. The *Richards* experiments were originally carried out on lower animals, but the method has since been applied to mammals by the group of investigators *Walker, Bott, Oliver* and *MacDowell* (1941). This constitutes a basic prerequisite for the general applicability of the results. However, in spite of the new-won investigation results, the usefulness of the punction technique seems to be to a certain degree restricted. So far only the superficial nephron parts of the kidney have been examined with this technique. Thus, only the urine occurring in the Malpighian corpuscle, in certain portions of the proximal and distal tubules and, perhaps, in the initial collecting ducts can be subjected to direct examination, whereas other parts of the nephron and the collecting tubules situated deep in the kidney elude direct investigation.

It has been ascertained by morphological examinations that the structure in separate parts of the nephron is highly differentiated and variable. This suggests a likewise varying function. However, a mere glance through the renal literature will show that our present knowledge of the partial functions of the nephron and the collecting tubules is extremely limited. As a rule, the demonstration of a transformation of primary urine to secondary urine by processes in the tubules has had to suffice, and the old point of dispute whether secretory or resorptive processes are active in this connection comes to the fore. Still, it is evident that the whole problem must be dealt with on a broader base so as to involve

an elucidation of the functions of the *different* parts of the nephron and the collecting tubules. Not until definite knowledge of this has been acquired, can a detailed analysis of the patho-physiology of the kidney be attempted. The *Richards* puncture technique offers certain facilities for level-diagnostics in the nephron but is, as already mentioned, limited to a certain extent. It has, therefore, seemed necessary to try to develop a new method capable of supplementing and increasing present means of investigation and permitting level-diagnosis also of the parts of the nephron and collecting tubules situated deep in the kidney. The purpose of this lecture is to draw attention to such a method and to some of our investigations performed regarding the histo-topochemistry of the kidneys during the past years. I shall here only deal with the principles of the method and give a slight orientation of the results. For further details, reference may be had to works by *Glimstedt* (1942, 1943) and *Ljungberg* (1947).

Linderström-Lang and his coworkers (1934) elaborated a method rendering possible direct comparison between cellular construction and the content of various chemical substances. The principle of *Linderström-Lang's* method has been adopted in the renal investigations concerned here. The method has been carried out as follows. Rabbits were used as experimental animals. They were killed by hanging and the kidneys carefully prepared without being exposed to pressure. One of the kidneys was immediately placed on the object slide of a freezing microtome and frozen in toto with carbon dioxide. After this, two renal cylinders were stamped out, by means of a stamping instrument with a diameter of 3 mm, close to each other from the middle point of the convex part of the kidney and perpendicularly down to the hilus. One of these cylinders was used for quantitative histological analysis, the other for micro-chemical determinations. The former was fixed in Carnoy's fluid, embedded in paraffin, microtomed in series from the surface to the renal hilus and the sections were stained in iron alum hematoxylin acc. Heidenhain and erythrosin. The latter was cut in the freezing microtome in series, the obtained sections being transferred to analysis tubes for quantitative, micro-chemical determinations. By comparing the quantitative histological structure in the sections at different distances from the renal surface at one cylinder with the amount of determined chemical substance in the corresponding sections in the other, it is possible to find the correlation between the structure and the content of various chemical substances at the different renal levels. Histo-topochemical analyses of this type permit level-diagnosis in the nephron and in the collecting tubules. Lately, the comparative histological and micro-chemical analysis has been per-

formed in one and the same cylinder, this having been fixed in isopentane dipped in liquid nitrogen and then vacuum-dried at -40°C . After embedding in paraffin, the cylinder was subjected to serial microtomy and the sections were alternately placed under quantitative histological and micro-chemical analysis. In this way, the sources of error are avoided which the variation in the quantitative histo-architectonic of the kidney in the horizontal plane may involve. This latest variation in the technique has been adopted in after-examinations of the earlier tests, with conforming results, which will be published later.

At the quantitative histo-architectonic determinations, paper reconstructions of the renal sections have been made, the various partial components in the reconstructions having been cut out and weighed. By means of this procedure, the percentage amount of histological components contained in the kidney has been determinable. Since a quantitative histological analysis takes a great deal of time, a photometric apparatus has in recent years been constructed rendering analysis of the histological material much quicker than previously. Fig. 1 shows which components have been determined and the percentage distribution of them in a typical sample.

Fig. 1. The quantitative architectonic at different levels of the right kidney in rabbit No. 22. The figures give the percentage distribution of the individual parts of the kidney examined (after *Glimstedt*, 1942).

Distance from the surface in mm	Malpighian corpuscles	Large vessels and vascular islands	Interstitial tissue	Proximal convoluted tubule		Distale convoluted tubule		Collecting tubule		The broad limb of Henle's tubule		The narrow tubule of Henle
				Epi-thelium	Lumen	Epi-thelium	Lumen	Epi-thelium	Lumen	Epi-thelium	Lumen	
0,3	3,0	1,0	6,2	77,1	4,0	8,2	0,4	—	—	—	—	—
0,6	4,0	1,1	6,2	78,5	3,8	3,7	0,2	2,4	0,2	—	—	—
1,0	3,9	1,3	5,4	78,1	4,7	3,4	0,2	2,6	0,2	—	—	—
1,4	3,3	2,0	4,5	76,9	3,6	5,5	0,3	3,5	0,3	—	—	—
1,7	3,2	2,5	5,6	76,3	3,9	5,2	0,5	2,7	0,2	—	—	—
2,1	3,3	3,0	3,3	76,6	3,7	6,7	0,4	2,8	0,3	—	—	—
2,5	2,3	6,8	6,4	66,6	3,6	9,5	0,6	3,6	0,5	—	—	—
2,9	0,7	5,9	13,6	59,1	3,1	12,3	0,8	3,7	0,8	—	—	—
3,3	—	15,4	18,9	44,0	1,7	—	—	3,6	0,7	15,1	0,8	—
3,7	—	18,7	25,7	27,1	1,1	—	—	3,6	0,7	21,9	1,1	—
4,1	—	11,2	29,7	17,3	1,0	—	—	6,2	1,3	31,8	1,5	—
4,4	—	6,3	47,5	—	—	—	—	5,6	1,2	32,1	1,7	5,6
4,8	—	5,1	47,4	—	—	—	—	7,6	2,4	30,2	1,3	5,9
5,2	—	4,3	42,6	—	—	—	—	8,7	3,1	31,4	1,7	7,9
5,6	—	6,0	43,0	—	—	—	—	10,1	3,1	28,0	1,5	8,1
6,0	—	5,6	53,4	—	—	—	—	10,6	4,3	14,1	1,2	10,7
6,4	—	6,0	58,0	—	—	—	—	11,9	4,9	3,9	0,2	15,1
6,8	—	5,6	58,0	—	—	—	—	10,8	4,4	—	—	21,0
7,3	—	4,3	55,2	—	—	—	—	12,9	5,8	—	—	21,8

Micro-chemical analyses have hitherto been performed on chlorides and total alkali. On account of the fact that the preliminary results have been submitted to detailed examination in a extensive material only as regards the chlorides, the usefulness of the method will be illustrated merely by an orientating survey of these analyses. The micro-chemical chloride determinations were performed according to a modification of *Westfall, Findley and Richards* (1934) technique (see *Glimstedt* [1942]). The determination method gives values that are a little too high. The accuracy as measured by the standard deviation was 10,38 p. c. (*Glimstedt* [1942]), and 8.9 p. c. (*Ljungberg* [1947]).

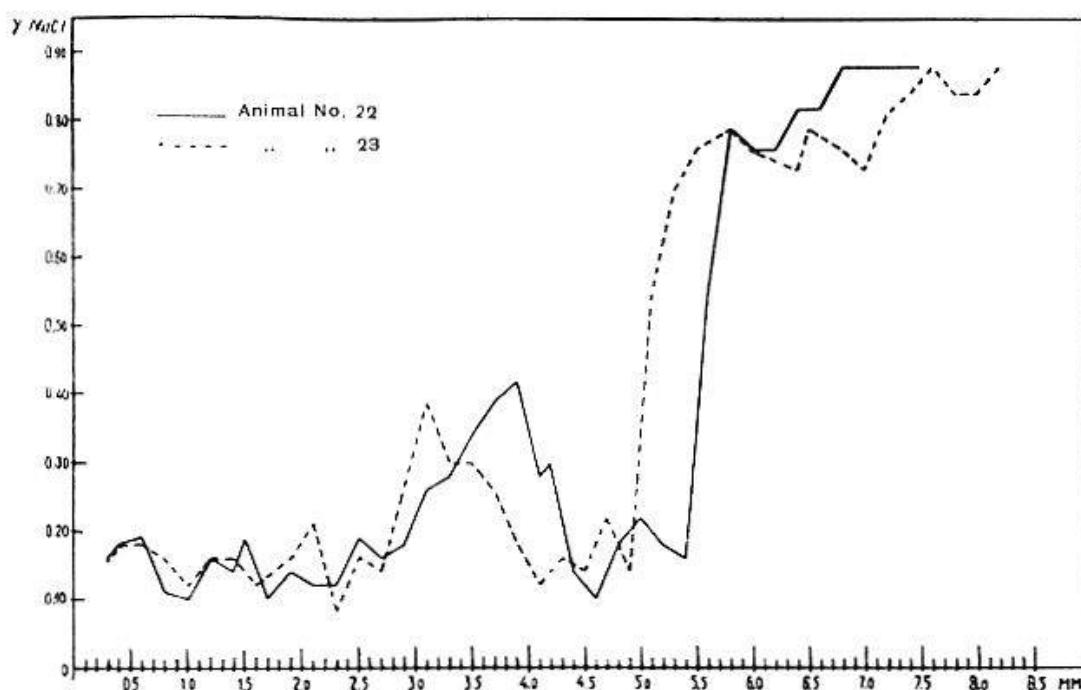


Fig. 2. The quantity of chloride at different levels of the kidneys in rabbits Nos. 22 and 23. The chloride quantities are given along the ordinate, expressed in γ NaCl in 25μ thick sections. The abscissa gives the distance from the surface of the kidney in mm (after *Glimstedt*, 1942).

These chloride curves have, in principle, the same appearance in all the examined kidneys. I shall therefore only show a picture of two typical curves of this kind (fig. 2). In the first zone nearest to the renal surface, the chloride content is low. At different levels in this zone the curve describes but slight variations which, broadly speaking, keep within the limits of error inherent in the micro-chemical determination method. Accordingly, in this region the curve mostly resembles a plateau. The chloride amount in the samples mentioned here equalled on an average 0.15 and 0.16 γ respectively, in sections 25μ thick. The chloride content then rapidly rises to values above the limits of error of the method. Within a limited region, the chloride content may then keep at a

higher level, either resembling a plateau or a peak, as shown in the plotted curves. After this rise, the chloride amount again falls to the depth of the kidney. This gives rise to a new plateau or ravine with, on an average, 0.16γ NaCl in sections 25μ thick. After this third renal zone, the chloride amount again increases. The increase is so rapid that the curve describes an exceedingly steep ascent. In the curves in Fig. 2, a value of 0.88γ NaCl in sections 25μ thick is obtained. After this, the chloride amount is subjected to only slight increases further into the depths of the kidney.

In the case of substances occurring to such an extent as the chlorides throughout the cells and tissues, it is not of course to be expected that these substances will be bound to a certain part of the kidneys. Thus, it seems obvious that the close conformity between cellular structure and enzyme distribution ascertained in earlier histo-topochemical works with regard to other organs cannot come in question here. The only factor which may throw light on the correlation between structure and function is the determination of a larger accumulation of chlorides in certain nephron or collecting tubule parts than in others. The comparative analysis between histo-architectonic and chloride content has been performed individually in the various animals. Analogous results were obtained in all. In order to give an illustration of the manner in which the analysis has been carried out, the results of the examination of animal No. 22 will be reported, which is the animal whose quantitative histo-architectonic and curve of chloride distribution has been shown in Figs. 1 and 2. In this animal, an increase in the amount of chloride will be seen on levels 3.1–4.2 mm from the renal surface. This coincides with the added number of greater vessels. At the same time, Henle's broad loop appears. On levels 3.7–4.1 mm, this nephron part greatly increases while the chloride content has here exceeded its maximum and is now decreasing. Simultaneously, the amount of larger vessels is reduced. Henle's broad loop cannot, consequently, be the cause of the added amount of chloride in the zone 3.1–4.2 mm. This is, moreover, accentuated by the fact that Henle's broad loop in the nearest deeper renal zone still occurs to a large extent whereas the chloride amount is diminishing. The collecting tubules are seen in the region 3.3–3.7 mm in the same quantities as in the renal zone situated above them. Nor are these tubules responsible for the increase of the chloride quantity. The amount of proximal convoluted tubules is strongly decreasing. Thus, it is evident that the increase of the chloride amount in the zone 3.1–4.2 mm from the renal surface is due to the occurrence there of the greater vessels. In this connection, it should be observed that, according to examinations by

Amberson, Nash, Mulder and Binns (1938), the chloride concentration in the plasma is twice as high as in the renal tissue and this irrespective of the height of the chloride level in the plasma.

Counting from level 5.2 mm to level 6.0 mm from the renal surface, the chloride amount has increased by 322 per cent. In the same region, the vascular islands, the interstitial tissue, the collecting tubules and the narrow limb of Henle's loop have also increased with the following values: 30, 25, 21 (epithelium), 39 (lumen), 26 (epithelium and lumen) and 35 per cent, respectively. The sudden marked increase of the chloride amount, accordingly, cannot in some way be correlated with the increase in the separate tissue parts. It appears obvious that the added chloride amount in this region is not due simply to the fact that the tissue or lumina situated closer to the renal surface contain chlorides in the same concentrations as here. Thus, the conclusion may be drawn that an *active* increase in the chloride quantity must have taken place. This cannot have been caused by the components occurring in this renal region, viz, the vascular islands, the interstitial tissue and Henle's broad loop. As regards this latter nephron part, this is evident from the marked decrease of Henle's broad loop between the levels concerned and, further, from the fact that when this part has entirely disappeared the chloride amount does not fall but, on the contrary, rises. Thus, the collecting tubules and the narrow limb of Henle's loop remain as the only parts which may have produced the increase in the amount of chloride. In addition, it appears that the rapid increase of the chloride content cannot have been caused by the collecting tubules or by the narrow limb of Henle's loop or, possibly, by both in their whole length, but that this capacity exists only in parts of them, viz, the parts occurring in the zone in question. Since Henle's broad loop is still in this region, the narrow limb of Henle's loop present there must, at any rate to a predominating extent, belong to the descending part. In the zone 6.0–7.3 mm, the chloride amount increases by 16 per cent. However, the narrow limb of Henle's loop increases simultaneously by 104 per cent, the epithelium of the collecting tubules by 22 per cent, their lumina by 35 per cent, and their epithelium and lumina by 26 per cent. Now, if the majority of the chlorides between the levels 5.2–6.0 mm should increase entirely through processes in the narrow limb of Henle's loop this would here be caused by a concentration of the urine. It has been impossible to ascertain by histo-chemical methods an accumulation of chloride in the cells of this nephron part. Consequently, the urine in the more distal parts of the narrow limb of Henle's loop cannot, at any rate, have a lower chloride concentration than in the proximal parts. The chloride content should

therefore increase in equal proportion to the narrow limbs of Henle's loop. However, as already proved, this does not happen. It is therefore hardly likely that the sudden strong increase in the chloride content between the levels 5.2–6.0 mm is due to processes in the narrow limbs of Henle's loop, at least not to any marked extent. On the other hand, the slight increase in the chloride content of the zone 6.0–7.3 mm coincides with the moderate increase of the lumina of the collecting tubules in the same zone. Thus, it is evident that the increase in the amount of chloride between the levels 5.2–6.2 mm must, principally, be due to active processes in the collecting tubules in the region concerned.

Such studies have now been performed on a fairly large number of animals and the results are on the whole in agreement. It may, therefore, be inferred that the collecting tubules carry out active processes in the mechanism of urine formation. The result is naturally surprising, considering that scientists have hitherto, almost without exception, been inclined to attribute merely an evacuating function to the collecting tubules, i.e. that of affording outlet for the urine which occurs in its final form in the nephron. However, the result appears probable from a teleological point of view. It seems to be a common principle in the organism to spare building material, i.e. not to create a structure which would only have such an elementary and subordinate function as that of accomodating on excretions. Many examples of this may be presented with regard to other organs.

This result derives support from an investigation by *Ljungberg* in 1947. He injected a series of rabbits intravenously with potassium cyanide and subjected the kidneys to histo-topochemical analysis with re-

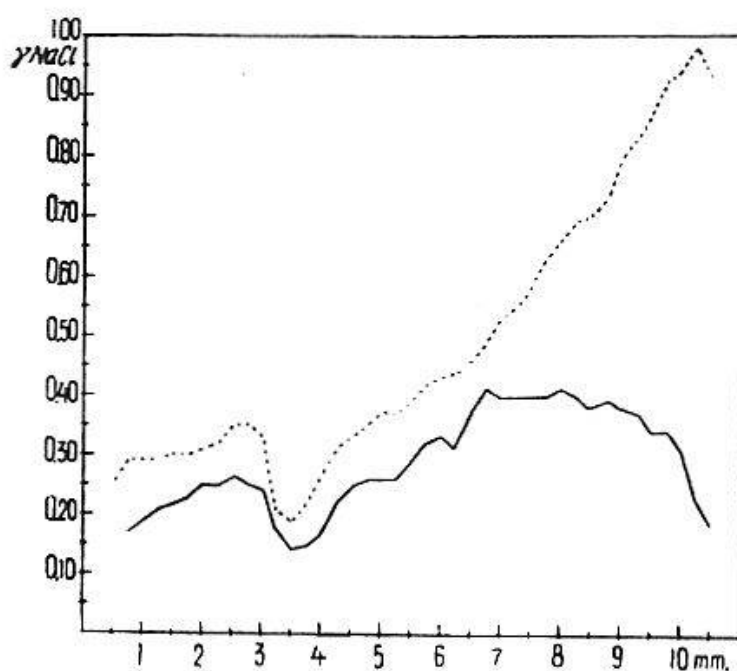


Fig. 3.
 ----- The standard normal curve.
 — The standard KCN curve made on 10 rabbits killed 1 hour after injection of KCN.
 (after *Ljungberg*, 1947).

gard to the chlorides an hour after the injection. Fig. 3 shows his standard normal curve after histo-topochemical analysis of 16 normal rabbits and his KCN curve made on 10 other rabbits. It will be seen from this diagram that the two curves run parallel up to 7 mm from the surface of the cortex, the normal rise of the chloride curve after this level failing to appear in the animals poisoned by KCN. Evidently, the cyanide has blocked the active chloride-enriching processes, as was ascertained also in the normal material.

The general conception regarding the structure of the collecting tubules, such as it has appeared in text- and handbooks as well as in special works on this subject, is that the collecting tubules consist of an undifferentiated epithelium which is lower in the more initial parts of the collecting tubules and becomes increasingly cylindrical and prismatic down towards the papillary ducts. These epithelial cells would, in the ordinary histological preparations, appear like uniform, light, clear formations without any special structure whatever. However, a perusal of the renal literature will show, in certain publications, indications of the fact that the collecting tubules have a more complex structure and should be attributed an active function in the mechanism of urine formation. Thus, *Ekberg* and *Wigert*, as early as in 1903, ascertained that in the collecting tubules of the *Rana Esculenta* a cellular specialization occurs with two cellular types, one resembling the central cells, the other the parietal cells in the stomach. Not until 1929 were questions regarding a possible cellular specialization in the collecting tubules submitted to discussion. In this year, *Okkels* discovered, inter alia, that cells occur in the collecting tubules which can be made conspicuous with Da Fano's method. This staining procedure makes specific cells appear as dark formations. Ten years later, the problem was subjected to renewed analysis by two French scientists, *Feyel* and *Vieillefosse*. They verified *Okkels*' observations and demonstrated that quantities of chlorides are histochemically ascertainable in the «cellules spéciales» of the collecting tubules. In a series of experiments on animals, they produced an elevation as well as a lowering of the bloodchloride mirror. It was found in the former group that the so-called chloride cells, as I propose to call them here, were sparse, their volume comparatively small, and that they were restricted to a fairly small part of the longitudinal extension of the collecting tubules. In the latter, i. e. in experiments with hypochloremia and the subsequent increase of the resorption of chlorides, the special cells were seen almost right down to the papillary apex and their volume was markedly increased. Thus, the cells do not seem to be functionally fixed. The ordinary epithelial cells of the collecting tubules can, evi-

dently, when so required develop into special cells. Thus, it is obvious that the epithelial cells of the collecting tubules have a functional potentiality.

Other facts exist also, indicating the active significance of the collecting tubules with regard to the formation of urine. In the case of uric acid infarcts in children and in gout nephritis, uric acid is precipitated into the collecting tubules. In some instances, also sulfonamide crystals may be precipitated. In the transfusion kidney the precipitated hematin obstructs the collecting tubules. These conditions indicate the significance of the collecting tubules with regard to water reabsorption.

Thus, it seems as though it should now be possible to obtain a more satisfactory conception of the manner in which the kidney regulates the composition of the blood as regards these chlorides. In an investigation performed by *Walker, Bott, Oliver and MacDowell* (1941) it was, inter alia demonstrated that the chlorides filtrated into the glomeruli were reabsorbed to about $\frac{2}{3}$ in the proximal convoluted tubules. They also observed that the majority of samples from the bladder urine had a lower chloride concentration than the plasma. From this, they concluded that some other part, farther down in the nephron, must reabsorb the chloride ion. Where this other nephron part is to be sought for, they were unable to state. Accordingly, it seems possible from the experiments described briefly in the present paper to point out which renal part is concerned in this respect. It must be the collecting tubules that perform this particular function. When the results of the two types of investigation dealt with here are compared, it will be noted that the main part of the chlorides is reabsorbed isosmotically in the proximal convoluted tubules. Thus, a coarse regulation takes place in these ducts. In the collecting tubules, a mechanism of fine-regulation exists the function of which is to keep the blood-chloride mirror on a normal level. Here the chlorides are enriched by so-called dry retention to a varying extent in the special cells.

The investigations briefly reported here form a first step towards a plan of research concerning the histo-topochemistry of the kidney. It is our intention further to pursue these experiments with studies of the normal distribution curves correlated to the quantitative histo-architectonic of the kidney. We shall also attempt another mode of approach where, above all, *Engströms* (1946, 1947) quantitative roentgen absorption spectrography will be a valuable supplementary aid. In due course, a teamwork with other specialists will be necessary. I particularly have in mind such problems as the influence of hormones on the specialized renal function, the attacking point of various drugs in the kidney and a

variety of similar questions. It is to be hoped that unbiased primary observations shall contribute towards a research calculated to give final results.

Summary

In spite of the great progress made, especially during the last few decennia, in the investigation of the morphology and physiology of the kidney the problem of the urine formation still conceals a number of unsolved mysteries. In the lecture an account is given of a rather new method, the histotopochemical one which seems to imply a contribution to the solution of the said problem. The principle of the method is to look for correlations between the quantitative histoarchitectonic of the kidney and the amount of different chemical substances on different levels from the surface of the kidney towards its hilum. In this way the wanted connection between structure and function is found and the method allows level-diagnostics within the nephrons and the collecting tubules. The lecture gives an account of the chloride distribution in the kidney of rabbit. The normal curve for the chloride distribution shows 4 characteristic regions, with the greatest increase in the amount of chloride in the 4th region towards the apex of the papilla of the kidney. Comparative analyses of the distribution of chloride and the histological structure, together with poisoning experiments with potassium cyanide (*Ljungberg*), show that the amount of chloride is here increased by an active process which takes place, mainly at least, in the "cellules spéciales" (*Okkels*) in the collecting ducts, where the chloride is enriched by "dry retention".

Investigations on rat and guinea-pig made by *Walker, Bott, Oliver*, and *MacDowell* showed that about $\frac{2}{3}$ of the chlorides filtrated away in the glomerules are reabsorbed in the proximal convoluted tubules. Further, this group of investigators also found that the majority of samples from bladder urine had a lower chloride concentration than blood-plasma, and inferred that some additional part of the nephron deeper down must reabsorb the chloride ion. Where this additional part of the nephron is to be found, the investigators could not say.

From our investigations related in the lecture it thus seems, as though it were now possible to point out which part of the kidney is here concerned. It seems evident that the collecting tubules must have that function. On comparison of the results of the two related types of investigations it is seen that the bulk of the chlorides is reabsorbed isosmotically in the proximal convoluted tubules. Thus, a coarse regulation takes place there. In the collecting tubules a fine-regulation mechan-

ism is situated, the function of which is to keep the blood-chloride concentration at a normal level.

Zusammenfassung

Trotz der großen Fortschritte, die während der letzten Dezennien in der Untersuchung der Morphologie und Physiologie der Nieren gemacht wurden, ist das Problem der Urinbildung immer noch nicht vollständig gelöst. Es werden Angaben über eine neue, histotopochemische Methode gemacht, welche einen Beitrag zur Lösung des Problems zu enthalten scheinen. Das Prinzip der Methode ist die Suche nach Beziehungen zwischen der quantitativen Zellarchitektur der Nieren und der Menge der verschiedenen chemischen Substanzen in den verschiedenen Schichten von der Oberfläche gegen das Innere der Nieren hin. Auf diese Weise wird die gewünschte Korrelation zwischen Struktur und Funktion gefunden, und die Methode erlaubt Schichtdiagnosen innerhalb der Nephronen und der Sammelröhren.

Die Arbeit gibt eine Übersicht über die Chloridverteilung in den Nieren des Kaninchens. Die normale Kurve der Chloridverteilung zeigt 4 charakteristische Zonen mit der größten Zunahme der Chloridmenge in der 4. Region gegen die Spitze der Nierenpapille. Vergiftungsexperimente mit Kalium-Cyanid (*Ljungberg*) zeigen zusammen mit vergleichenden Analysen der Chloridverteilung und der histologischen Struktur, daß die Chloridmenge hier infolge eines aktiven Prozesses zugenommen hat, der hauptsächlich in den «cellules spéciales» (*Okkels*) der Sammelröhren, in denen das Chlorid durch «dry retention» angereichert wird, stattfindet.

Die Untersuchungen von *Walker, Bott, Oliver* und *MacDowell* an Ratten und Meerschweinchen zeigten, daß ungefähr $\frac{2}{3}$ der Chloridmenge, die in den Glomeruli wegfiltriert wurde, im proximalen Teil der Tubuli rückresorbiert wird. Ferner fanden diese Forscher, daß die Mehrheit der Blasenurinproben eine niedrigere Chloridkonzentration aufweist als das Blutplasma und schlossen daraus, daß ein zusätzlicher, tieferliegender Teil des Nephrons die Chloridionen rückresorbieren müsse. Wo aber dieser liege, konnten die Forscher nicht sagen. Nach unseren Untersuchungen wird es möglich, den in Betracht kommenden Teil der Niere zu bestimmen. Es scheint auf der Hand zu liegen, daß die Sammelröhren diese Funktion haben müssen. Aus dem Vergleich der Resultate der beiden Untersuchungsarten geht hervor, daß die Menge des Chlorids isosmotisch im Hauptstück rückresorbiert wird. Die grobe Regulation findet somit dort statt. In den Sammelröhren hingegen ist der feinere

Regulationsmechanismus lokalisiert, dessen Funktion es ist, den Blut-chloridspiegel auf einem normalen Niveau zu halten.

Résumé

Malgré les grands progrès réalisés, spécialement ces dernières années, dans la morphologie et la physiologie rénale, le problème de la formation de l'urine comporte encore nombre de points non résolus. L'auteur décrit la méthode histo-topochimique – relativement nouvelle – qui semble fournir un appoint à la solution de ce problème. Le principe de la méthode consiste à chercher des rapports entre l'histo-architectonie quantitative du rein et la quantité de divers produits chimiques dans les différentes couches, à partir de la surface du rein jusqu'au hile. De cette manière, on peut se rendre compte des rapports existant entre la structure et la fonction, et la méthode permet le strato-diagnostic dans les néphrons et les tubes collecteurs. Le rapport fait état de la répartition des chlorures dans le rein chez le lapin. La courbe normale de la répartition des chlorures montre 4 régions caractéristiques. Le taux le plus élevé se trouve dans la 4^e couche, du côté de l'apex de la papille rénale. Les analyses comparatives de la répartition des chlorures et la structure histologique ainsi que des expériences d'intoxication par le cyanure de potassium (*Ljungberg*), montrent qu'ici la quantité de chlorures est augmentée par un processus actif ayant lieu, au moins pour la plus grande partie, dans les «cellules spéciales» (*Okkels*), dans les canaux collecteurs, où les chlorures sont augmentés par «rétention sèche».

Des recherches chez le rat et le cobaye faites par *Walker, Bott, Oliver et MacDowell* montrèrent qu'environ $\frac{2}{3}$ des chlorures qui filtrent à travers les glomérules sont réabsorbés dans les tubes contournés proximaux. En outre, ces auteurs trouvèrent aussi que la plupart des échantillons d'urine de la vessie avaient une concentration chlorée plus basse que le plasma sanguin. Ils en tirent la conclusion qu'une autre partie plus profonde du néphron doit réabsorber les ions chlore. Mais ces chercheurs ne sont pas parvenus à en préciser la localisation.

D'après nos recherches, il semble maintenant qu'il soit possible de localiser la partie du rein dont il s'agit. Ce seraient les tubes collecteurs qui rempliraient cette fonction. En comparant les résultats des deux types d'expériences ci-dessus, on voit que la plus grande partie des chlorures est réabsorbée par isosmose dans la partie proximale des tubes contournés. Il y a donc ici régulation grossière. Dans les tubes collecteurs, il existe un mécanisme de régulation fine qui a pour but de maintenir à un niveau normal la concentration chlorée du sang.

Riassunto

Malgrado tutto il progresso compiuto negli ultimi decenni nel campo delle investigazioni sulla morfologia e fisiologia renali, numerosi problemi concernenti l'escrezione dell'urina devono ancora essere risolti. Nel presente lavoro si parla di un nuovo metodo, isto-topo-chimico, che sembra poter dare un importante contributo alla risoluzione del problema. Il principio su cui si basa il metodo è lo studio delle relazioni che corrono tra la architettura istologica quantitativa del rene e la presenza delle varie sostanze chimiche disposte nei vari strati tra la corteccia e l'ilo renale. In questa maniera si mettono in luce i rapporti tra struttura e funzione e questo metodo permette una diagnosi stratigrafica rispetto ai nefroni e ai tubuli collettori. Il lavoro parla della distribuzione dei cloruri nel rene di coniglio. La curva normale di distribuzione dei cloruri dimostra 4 regioni caratteristiche, con aumento particolare della quantità dei cloruri nella quarta regione, presso l'apice della papilla renale.

Ricerche comparative sulla distribuzione dei cloruri e la struttura istologica, in correlazione con esperimenti di intossicazione con cianuro di potassio (*Ljungberg*), dimostrarono che la quantità dei cloruri è qui aumentata da un processo attivo che avviene quasi sempre nelle cellule speciali (*Okkels*) nei tubuli collettori, dove i cloruri sono aumentati a causa della «ritenzione secca». Ricerche sul ratto e sulla cavia, portate a termine da *Walker, Bott, Oliver, MacDowell*, dimostrano che $\frac{2}{3}$ dei cloruri, allontanati tramite filtrazione nei glomeruli renali, sono riassorbiti nella parte prossimale dei tubuli contorti. Ma questo gruppo di ricercatori ha anche trovato che la maggior parte dei campioni di urina estratta dalla vesica contiene una concentrazione in cloruri inferiore a quella del plasma sanguigno e che la restante parte distale del nefrone, deve riassorbire ioni cloro. Gli investigatori non erano in grado di dire in quale punto di questa parte restante del nefrone questo avvenisse. Con le nostre investigazioni, di cui parla questo lavoro, sembra quasi possibile dimostrare con sicurezza quale è la parte del rene che ha questa funzione. Sembra evidente che questa funzione sia devoluta ai tubuli collettori. Comparando i risultati di questi due tipi di ricerche che hanno dei punti in comune, risulta che la più gran parte dei cloruri è riassorbita iso-osmoticamente dalla parte prossimale del tubulo contorto. In questo punto si ha dunque una regolazione grossolana. Nei tubuli collettori invece risiede un meccanismo di regolazione delicato, che ha la funzione di mantenere costante a un livello normale la concentrazione dei cloruri del sangue.

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