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THE CARCINOGENIC ACTION OF TANNIC ACID EFFECT OF CASEIN ON THE DEVELOPMENT OF LIVER TUMOURS

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It was recently published (*Korpássy* and *Kovács* 1949) the successful production of liver cirrhosis in white rats by prolonged subcutaneous administration of tannic acid solutions. Already then these investigations had indicated that the progressive process produced in the liver of rats by regular tannic acid administration does not come to an end with the characteristical distortion of the liver-architecture. In the liver of some of the rats namely, which survived the 100th day of treatment, there appeared small hepatomas and cholangiomas. These investigations suggest a blastomogenic action of the tannic acid.

Experimental

The experiments here to be reviewed can be divided into two parts : examination of local and remote effect of the tannic acid.

1. *Parenteral administration.* Tannic acid solution was subcutaneously injected in 58 young (two month-old) white rats. At the beginning 150 mg., later 200 mg. of tannic acid per kg. body weight, in 1,5—2 per cent aqueous solution was subcutaneously administered, usually every 5th day, on the back of all the animals. The tannic acid used was obtained through the Pharmacy of this University (Acid. tannic U. S. P. Johnson, Hendon, London).

Rats thus treated were divided into two groups. In group A the rats received a mixed diet of waste food from the hospitals. This food seemed sufficient as a great number of untreated white rats from the same strain, being fed in the same way grew and reproduced normally.

In group B the food consisted of 50 per cent milk-bread, or rice boiled in milk and 50 per cent lean fresh cheese, the contents of the latter in *Sós*'s opinion is protein 18,2 per cent, out of which casein 16,5 per cent, fat 2,2 per cent, carbohydrate 3,9 per cent. Rats of both groups were further fed on oats forming 20 per cent of all the foods.

Beginning the experiment the average weight was in group A 68 gm., in group B 50 gm. Weight of the rats was systematically controlled and in case of considerable loss of weight in order to avoid early death the treatment was discontinued for some days. The weight-curve showed a steady rise until the 150th day of the experiment when the animals of both groups reached an average weight of 175 gm. Then the body-weight in group A ranged from 150 to 180 gm. In group B the average body-weight, apart from slight fluctuation, continued to grow until the 230th day, when it reached 210 gm., later, however, it had hardly changed.

The rats, as it happens, were of the same own bred strain as those used by *Korpássy* and *Kovács*. In both groups were in identical number males and females.

In the first third of the treatment from group A 5. from group B 10 rats died. 100 day treatment was survived by 23 from group A and 19 from group B. 200 days by 12 from group A and 7 from group B. 300 days only by 5 rats from group A and by 1 rat from group B.

Treatment was discontinued on the 290th day, rats surviving this period received altogether 49 tannic acid solution injections. From group A one rat was killed on the 358th, one on the 363rd and an other on the 388th day: from group B only one rat was killed on the 388th day. The other animals perished spontaneously before the 388th day. From group A one and from group B six were devoured by their mates and so they were not autopsied. These 7 rats, however, died before the 100th day of treatment.

2. *Skin painting*. In order to study the local effect of the tannic acid on the one hand and the generale remote effect of the skin ulcers on the other hand, the skin on the back of 39 white rats was burned with a glowing spatula in the size of a 2 shilling piece and when the scab peeled off, it was tried to inhibit the healing of the ulcers. The ulcers of 20 rats were daily painted with 5 per cent fresh aqueous tannic acid solution while the ulcer of 19 rats, also daily, with 5 per cent hydrochloric acid. If healing of the ulcers in spite of this treatment advanced, the skin has been burned again or treated with concentrated hydrochloric acid. This treatment had to be repeated at intervals of six weeks. From the group treated with tannic acid 14 rats survived 300 days, 11 rats 400 days: from the group treated with hydrochloric acid 10 rats survived 300 days and 9 animals 400 days. Two animals of each group had been killed on the 505th day. The perished animals died in intercurrent diseases (otitis media, pulmonary abscess, enterocolitis).

In this experiment adult (6 months old) white rats were used, their average weight at the beginning was: tannic acid group 161 gm., hydrochloric acid group 171 gm. The strain and diet of the animals were the same as in experiment 1.

All the rats were dissected the soonest possible. For histological examination the tissues were fixed in 4 per cent formaldehyde, embedded in paraffin and stained with haematoxylin-eosin, van Gieson stain, and Gömöri's stain for reticulum. The livers were examined in all cases, other organs only in the few instances in which a macroscopic lesion was present.

Changes produced

Local effect. Having repeatedly employed subcutaneous administration of 1,5—2 per cent aqueous tannic acid solution skin necrosis were observed on the injected place when, however, the necrotic parts came off there arose ulcers (1. experiment). Although in animals treated for longer period fairly large skin-ulcers were formed changing, however, the place of treatment or having occasionally discontinued it, the ulcers in general rapidly and unexceptionably healed and did not seem to influence the state of health of the animals. In no case arose a tumour either from the margin of the ulcers or from the healed scars.

The local treatment with tannic acid and with hydrochloric acid of the skin-ulcers induced by burning (2. experiment) yielded no result at all. The skin i. e. skin-ulcer of 11 rats was daily painted with 5 per cent tannic acid solution over a period of 400 days (2 animals were killed on the 505th day of

the treatment) but no change of any kind appeared on the place of the administration in any of the cases.

Remote effect. The liver of several of the 41 rats surviving the 100th day of subcutaneous tannic acid treatment (1. experiment) showed the signs of an early or advanced diffuse nodular cirrhosis and associated with such changes or without them there arose hepatic tumours variable in size and structure.

On the other hand in the liver of the animals with skin-ulcers painted with tannic acid or hydrochloric acid solution for a longer while no change,

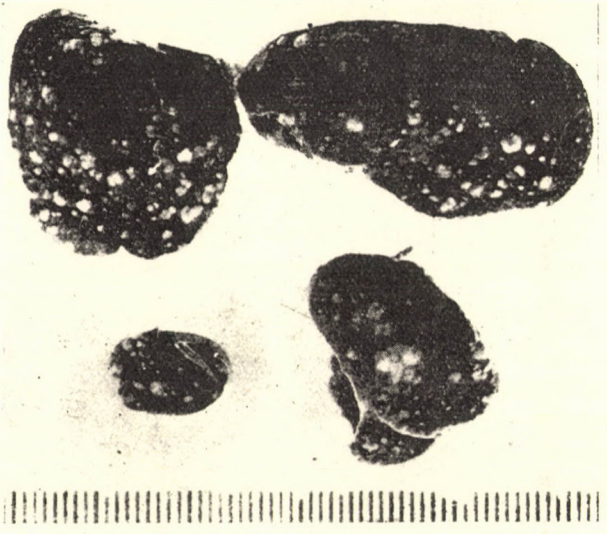

Fig. 1.
Rat o/13. Treated with 4250 mg. per kg. body weight tannic acid, administered in 22 doses.
Died on 122nd day.

which could be connected with the treatment, was observed. Cirrhotic or praecirrhotic changes were observed in none of the animals, nor was possible to state an increase of the reticulum fibers in the liver. Similarly in no case occured a tumour in the liver of these rats, though they were 4 months older at the beginning of the experiment and the survival time of the greater part was 100—200 days longer than that of those treated with subcutaneous tannic acid solutions.

Changes in the liver. Findings referred to below are solely concerned with animals having had *subcutaneous tannic acid solution treatment* (1. experiment). The first definite naked-eye changes were seen in a rat died on the 109th day, in whose liver there appeared a few minute nodules, of greyish-white colour.

Fig. 2.
Rat o/21. 48 injections, total 9700 mg. tannic acid per kg. body weight. Died on 278th day.

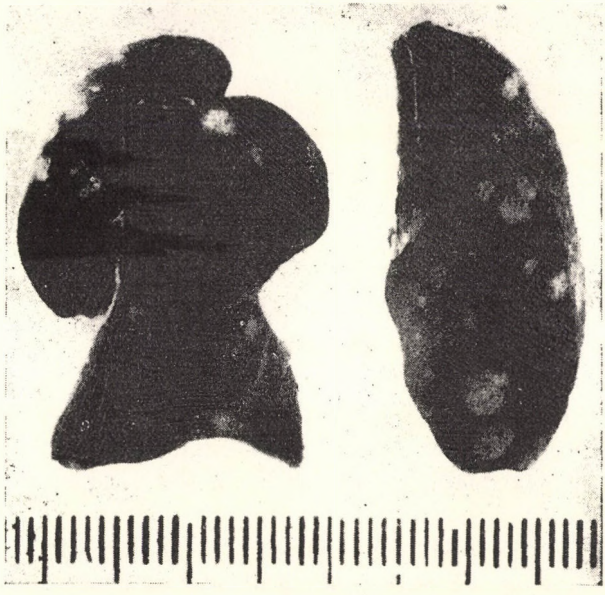

Fig. 3.
Rat M/19. 29 injections, total 5700 mg. tannic acid per kg. body weight. Died on 165th day.

On the 121st and 122nd day 6 rats were lost from group A and in the liver of one there were well marked and advanced changes : the surface was rendered granular by a large number of nodules in the size of a millet or pease, of greyish white colour. Beside such nodules in one of the lobules there arose a fairly solid tumour of $5 \times 6 \times 4$ mm., sharp limited and of greyish-white colour too (Fig. 1). This picture completely differs from the diffuse nodular cirrhosis produced by the tannic acid (see fig. 2). The liver of the rat died on the 287th day (from group A) showed the most marked diffuse nodular cirrhosis (Fig. 2).

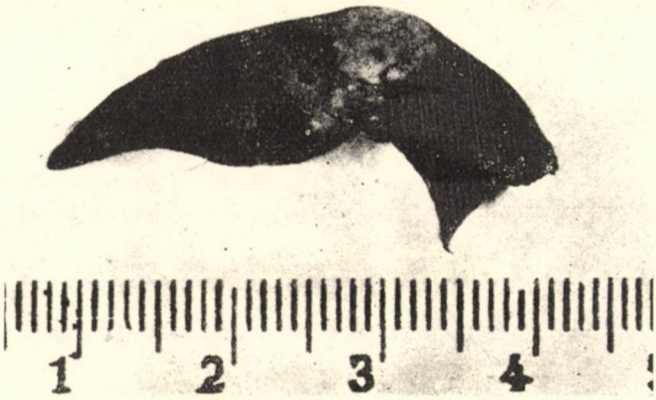

Fig. 4.
Rat M/28. 46 injections, total 9000 mg. tannic acid per kg. body weight. Died on 274th day.

In the liver of a rat belonging to group B and died on the 165th day, several greyish-white nodules of 2—5 mm diameter size were developed (Fig. 3). In the liver of another rat also of group B, and died on the 274th day of treatment one solide nodule in the size of $8 \times 6 \times 6$ mm of palc-greyish colour was developed with several smaller ones (Fig. 4).

After 290 days when the administration of tannic acid solution was discontinued, the macroscopical appearances of the livers — apart from the tumours — gradually reverted to normal. After this time 9 animals died or were killed and in 6 of these the livers showed solid tumours. They were not larger than those described above.

In some of the animals, especially in which the cirrhosis seemed to be the most advanced, there appeared ascites but varices of the portal-systematic venous anastomoses were never detected. No metastases were seen.

Histological examination. In the liver of the rats died during the first two months of the parenteral tannic acid treatment was extensive necrosis involving the central one-third to one-half of the lobules. There were numerous mitotic figures in the viable periphereal portion of the lobules and regeneration was evident through the greater part of tannic acid administration. The proliferation of fusiform cells seemingly arising from the sinusoids was rather obvious already in the first third of the treatment. They may be considered immature reticuloendothelial cells (*Korpássy* and *Kovács* 1949). Bile duct proliferation was well marked mostly about the end of the second month.

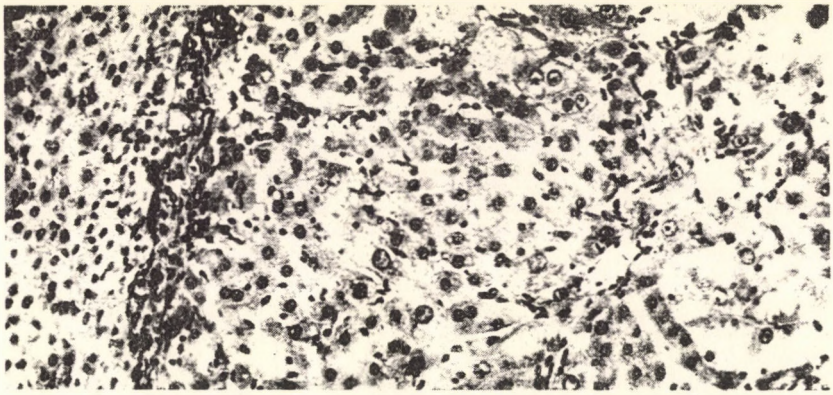

Fig. 5.
Rat o/27. 49 injections. total 9950 mg. tannic acid per kg. body weight. Killed on 358th day. Hepatoma and adjacent hepatic tissue. Haematoxylin and eosin. × 200.

In many animals surviving the 100th day of tannic acid administration the architecture of the liver was greatly disturbed. There was an appreciable increase in reticulum and sometimes in connective tissue, distributed mainly in the vicinity of the portal areas but also extending away from these zones in an irregular fashion setting off the parenchyma into "lobules" of irregular size and shape. These irregularly scattered hyperplastic nodules rarely showed necrosis.

Hepatic tumours. The induced tumours of the liver although showing a variety of structures could be divided histologically into two main groups, namely hepatoma and cholangioma.

Of the *hepatomas* two types can be distinguished, a well-differentiated and a less differentiated one, the former as a rule appeared in smaller nodules. The well-differentiated type was sometimes encapsulated, compressing the adjacent hepatic tissue which often showed a local increase in reticulum (Fig. 5).

The characteristic feature of the hepatomas was that the epithelial cells were arranged in cords which were separated from endothelial-lined sinuses by delicate slips of reticulum. In the well-differentiated form the tumour cells

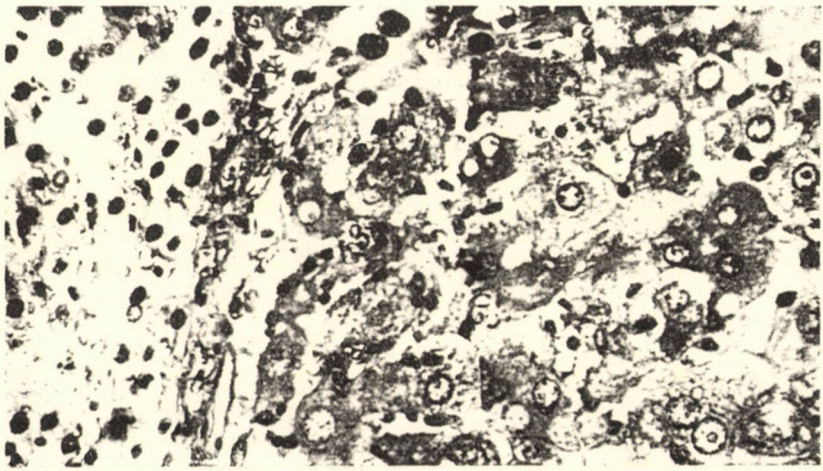

Fig. 6.
Rat o/27 (see fig. 5). Hepatoma and adjacent hepatic tissue. Liverlike cells with prominent nucleoli. Haematoxylin and eosin. × 350.

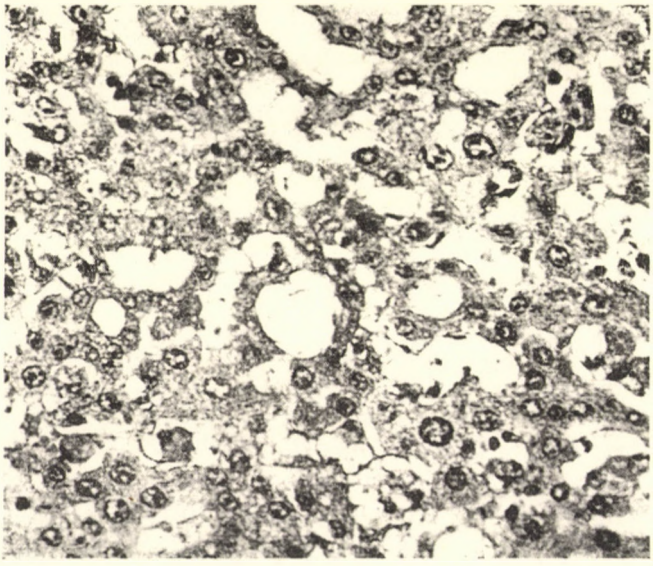

Fig. 7.
Rat o/13. (see fig. 1). Acinar formation in hepatoma. Haem. and eo. × 300.

had prominent cell margins, granular, occasionally vacuolated acidophilic cytoplasm and often irregularly enlarged vesicular nuclei with one to three prominent often acidophil nucleoli (Fig. 6).

In the larger hepatomas the cordlike arrangement of the tumour cells has been disorganised or took shape of cords of variable thickness. Acinar structures were not infrequently encountered that were formed by cells closely resembling hepatic parenchymal cells (Fig. 7). The cell-margins became dim, the cytoplasm stained faintly and was usually basophilic. The hepatoma cells

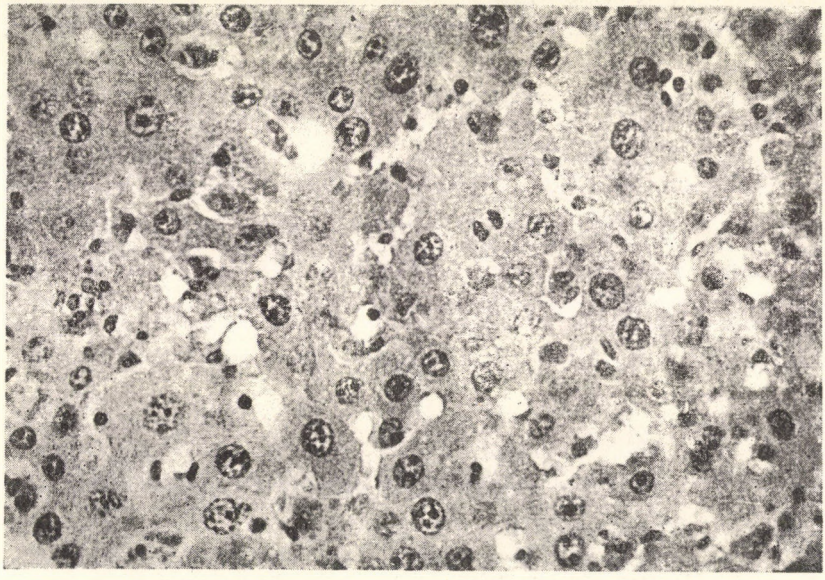

Fig. 8.
Rat o/27 (see fig. 5). Mitosis in hepatoma. Haem. and eo. × 350.

showed considerable variety in size; mitotic figures varied considerably in number and some were atypical (Fig. 8). A centrally placed vessel or bile ducts were not found. Fatty degeneration was slight and focal necrosis was seen in some larger hepatomas. The connective tissue of the tumours was usually scanty. In some instances there was an invasion of hepatic blood vessels at the periphery of the tumours (Fig. 9, 10).

The proliferation of the bile-ducts can be ascertained for the most part already before the 100th day. In rats surviving the 100th day of treatment not infrequently small areas of bile-duct proliferation were formed, showing an abundance of dense reticulum separating the ducts. *Opie* (1944) described this change in the liver of rats treated with p-dimethylaminoazobenzene and called it cholangiofibrosis.

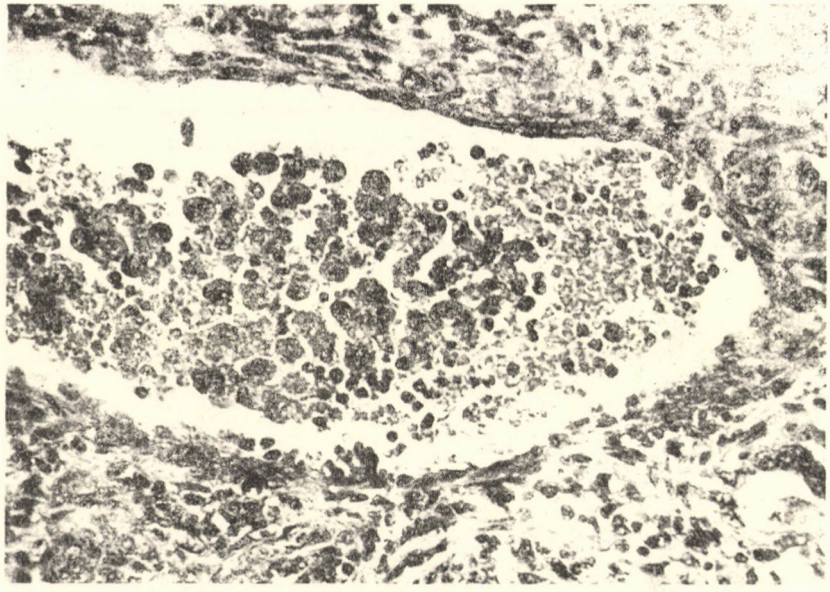

Fig. 9.
Rat o/13 (see fig. 1). Invasion of a large blood vessel.
Haematoxylin and eosin. × 300.

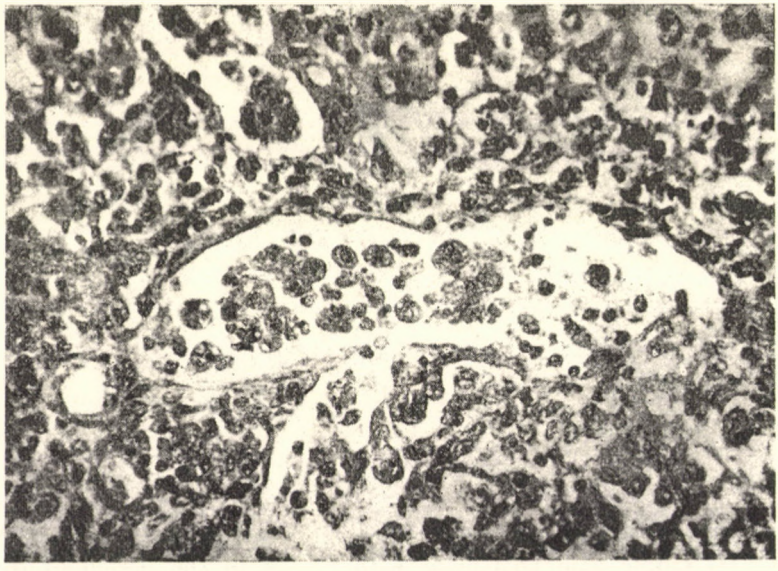

Fig. 10.
Rao/23. 49 injections, total 9950 mg. tannic acid per kg. body tweight. Died on 294th day. Invasion of a blood vessel.
Haematoxylin and eosin. × 350.

Beside these changes, also a more extensive and markedly irregular bile-duct proliferation could be observed in quite a number of cases (Fig. 11, 12). The shape and width of the tubules may be very varied and they are surrounded by but very few connective tissue. Still more marked irregularities may be shown by the epithelial lining : the cuboid-, or columnal epithelial cells are of markedly polymorph in shape and size, their nuclei sometimes vesicular, sometimes quite extended and hyperchromatic and, layering of the cells sometimes occurred (Fig. 13). Moreover, increase in the nuclear-cytoplasmic ration, the presence

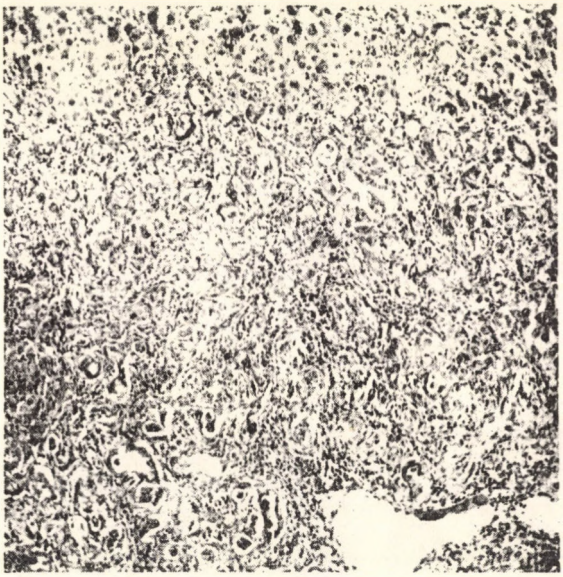

Fig. 11.
Rat T 18. 28 injections, total 750 mg. tannic acid. Killed on 141st day. Large area of proliferated bile ducts. Haematoxylin and eosin. × 40.

of solid acini, numerous and atypical mitoses are the features which distinguish from the non-neoplastic bile-duct proliferation. Finally, the fact that not infrequently smaller liver-cell islets can be found incorporated in the mass of the proliferating bile-ducts, proves the infiltrative growth of such tumours (Fig. 14).

Effect of casein on the incidence of induced liver tumours and cirrhosis. The frequency of hepatomas and cholangiomas in groups A and B produced by parenteral tannic acid treatment with regard to the period of treatment is represented in Table I. When taking in consideration only those animals which survived the 100th day of the treatment then in group A out of 23 in 13 (i. e. round 56 per cent), in group B out of 18 in only 3 (i. e. round 16 per cent)

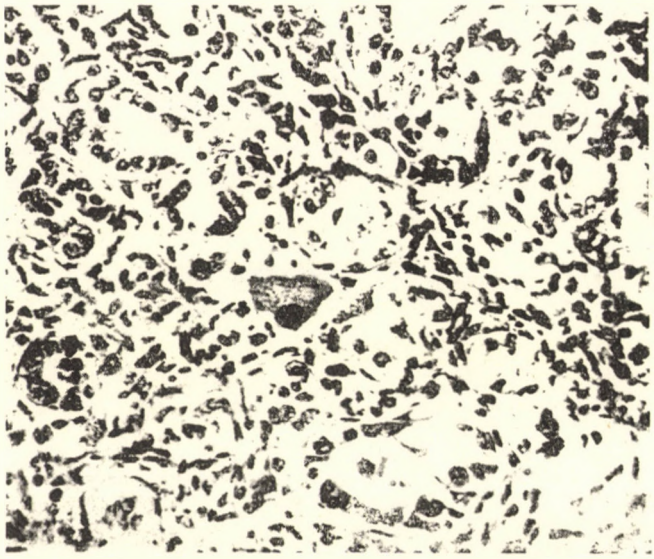

Fig 12.
Rat o/16. 28 injections. total 5500 mg. tannic acid per kg. body weight. Died on 166th day. Cholangioma. × 150.

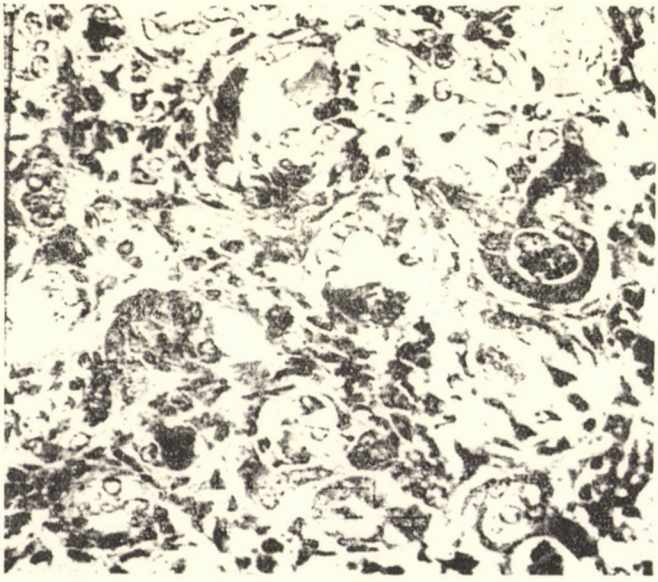

Fig. 13.
Rat T/18 (see fig. 11). Low grade adenocarcinoma. Haematoxylin and eosin. × 350.

were hepatic tumours induced. These results suggest that high casein diet (group B) protected rats to a considerable extent from the development of liver tumours. We would, however, mention that only macroscopic nodules whose neoplastic feature was of course histologically established, have been taken in consideration. There are authors who went further e. g. *Crabtree* (1949) investigating the carcinogenic effect of aminoazotoluenes regarded both the

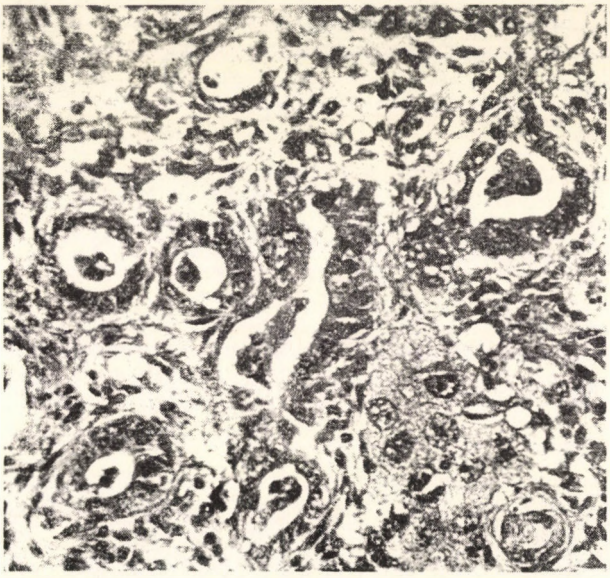

Fig. 14.
Rat o/13 (see fig. 1). Cholangioma with incorporated hepatic cells. Haematoxylin and eosin. × 350.

microscopic nodules of hepatoma and beginning of cholangioma as a tumour produced by his treatment.

Table II. shows the relation between cirrhosis and hepatic tumours. In group A out of 23 rats, surviving the 100th day of treatment in the liver of

Table I.

Days on experiment	Number of rats examined		Average survival time, days		Number of rats with liver tumours						Total	
					Hepatoma		Cholangioma		Hep.+.Chol.			
	A	B	A	B	A	B	A	B	A	B		
5— 87	5	5	36	38	—	—	—	—	—	—	—	—
103—195	11	11	128	146	1	1	3	—	1	—	5	1
201—295	7	6	265	230	2	1	2	—	—	—	4	1
320—388	5	1	353	388	2	1	1	—	1	—	4	1
Total	28	23	—	—	5	3	6	—	2	—	13	3

15 animals, there appeared a cirrhosis of different grade. In 9 animals, contemporaneously with cirrhosis liver tumours were formed, while in 4 animals with liver tumours no cirrhosis could be determined. In group B (high casein diet) out of 18 rats surviving the 100th day in only 5 developed an early cirrhosis. The sex of the animals seemed not to influence the incidence of induced liver tumours and cirrhosis.

Table II.

Days on experiment	Number of rats examined		Number of rats with liver cirrhosis		Cirrhosis in tumour rats	
	A	B	A	B	A	B
5— 87	5	5	—	—	—	—
103—195	11	11	8	4	4	1
201—295	7	6	5	1	4	—
320—388	5	1	2	—	1	—
Total	28	23	15	5	9	1

Incidence of spontaneous tumours. The albino rat strain which has been used for years in all our experiments, our own bred, is scarcely susceptible of spontaneous tumours. Although quite a large number of untreated old rats was autopsied, spontaneous liver tumour could be noted only in a single case. In experiment 2. of this paper in the liver of one rat, died on the 425th day of hydrochloric acid skin painting, a characteristic cysticercus sarcoma appeared with extensive omentum and pulmonary metastases. Spontaneous hepatoma, cholangioma, benign- or malign tumours arising from any other organ, however, were in no case observed.

Transplantation. Subcutaneous and intrahepatic transplants of tumour tissues were made by the trocar technique into rats of homologous strain. From a bean-sized liver tumour of one animal subcutaneous transplantation has been made into 5 white rats. No tumour is palpable even after 5 months. From a hazelnut-sized liver tumour of an other animal, transplantation into the liver of 5 white rats and in other 5 subcutaneously, has been made. Transplantation, even after several months, seems to fail.

Discussion

The best known of the hepatic tumours producing chemical substances are the azo-dyes. *Sasaki* and *Yoshida* (1935) were the first to succeed in producing liver cancer in rats fed o-aminoazotoluene. The most effective of the carcinogenic azo-dyes is p-dimethylaminoazobenzene (butter-yellow). The carcinogenic action of this dye on the liver was discovered by *Kinoshita* (1937). *Cruz* (1948) observed four types of lesions in the liver of rats fed p-dimethylaminoazobenzene more or less in accordance with the time of survival of the rats : 1. acute serous hepatitis, 2. adenomatosis, or bile ducts adenoma, 3. fibrosis, or annular cirrhosis, 4. stage of carcinoma or hepatoma.

Eltsina (1944) painted the skin of mice with 1 per cent o-aminoazotoluene and after 9 month of treatment in the liver there appeared small hepatomas and cholangiomas. Investigations of *Shabad* and *Buvailo* dealt with the azo compounds of 9, 10-dimethyl —1,2 benzanthracene, synthetized by *Mikhailov* and *Blokhina* which also proved to be blastomogenic. *Morozenskája* (1946) succeeded in transplanting the hepatoma of a mouse fed o-aminoazotoluene. It was transplanted and grew under the skin of white mice for 37 generations. Recently *Spitz*, *Maguigan* and *Dobriner* (1950) studied the carcinogenic action of benzidine. The structure of thehepatic tumours following the administrations of benzidine was in most respect quite like those produced by butter-yellow.

Carbontetrachloride administered perorally to mice resembles much in its effect to the azo-dyes (*Edwards* 1941). According to investigations made by *Cameron* and *Karunaratne* (1936) and others the carbontetrachloride is a substance producing cirrhosis.

There is an other carcinogenic agent, the effect of which is exclusively remote, that is 2-acetyl-aminofluorene. The carcinogenic properties of this agent were discovered by *Wilson*, *DeEds* and *Cox* (1941). Most tissues that gave rise to tumours were also the sites of nodular epithelial hyperplasia and no sharp distinction could be made between these nodules and the tumours formed by similar tissues (*Cox*, *Wilson* and *DeEds* 1947).

According to our own investigations the effect of tannic acid on liver resembles very much to the substances here specified. Administered parenterally it produces serous hepatitis and acinocentral necrosis (*Korpássy* 1949). It proved hepatotoxic when administered perorally in due dosage (*Korpássy*, *Koltay* and *Horvai* 1950). By parenteral administration to rats for a longer period it produces liver cirrhosis (*Korpássy* and *Kovács* 1949).

As it is shown by the investigations published here, tannic acid has no local effect producing tumour. As to the morphology of liver tumours produced by parenteral tannic acid administration — employed for the first time by the authors — parallels that described by *Orr* (1940), *Opie* (1944) and others in rats fed butter-yellow, or by *Wilson*, *DeEds* and *Cox* (1947), *Harris* (1947) and others in rats fed acetyl-aminofluorene ; or by *Edwards* (1942), *Eschenbrenner* and *Miller* (1946) in mice fed carbontetrachloride. It is true we did not succeed as yet in producing metastases involving liver cancer with parenteral tannic acid treatment, yet invasion of hepatic blood vessels observed in two animals too, indicates that some of the tumours thus produced cannot be regarded benign any more. *Willis* (1948) states that the invasion of hepatic veins is the prelude in human carcinoma of liver to metastasis to the lungs. One of our animals in whose liver at least early hepatic carcinoma can be ascertained, died on the 122nd day. Should this animal have lived a few weeks longer, it seems probable that metastases would have formed.

As tannic acid produces necrosis at the site of the injections, the question may arise whether some product of this necrosis could be responsible for the tumours. Our experiments with burn ulcers repeatedly treated locally with tannic acid or hydrochloric acid definitely are against this suggestion. Namely the greater part of these rats with healing inhibited skin ulcers survived the 400th day of the treatment, although during this time much product of necrosis could be absorbed, but no hepatomas or cholangiomas arose in any of the animals.

One of the outstanding features of the hepatic tumours induced by azo dyes, that their development can undoubtedly be influenced by diet. The addition of casein to the diet exerts some protective action against liver cancer caused by dimethylaminoazobenzene (*Kensler*, *Sugiura*, *Young*, *Halter* and *Rhoads* 1941). The addition of fresh milk daily to a rice-carrot diet containing p-dimethylaminoazobenzene protected rats to a considerable extent (*Hoch-Ligeti* 1946). *Griffin*, *Clayton* and *Baumann* (1949) stated that both casein and methionine were effective in improving the hepatic retention of riboflavin by rats fed the azo-dyes. According to our investigations may be ascertained, that high casein diet has a certain inhibitory effect on the cirrhogenic and carcinogenic action of tannic acid.

The widely disputed question of the relation between cirrhosis and formation of hepatic tumours can not be left out of consideration. According to *Sugiura* and *Rhoads* (1942) first develops liver cirrhosis, then follows the appearance of tumours, due to the effect of p-dimethylaminoazobenzene. When, however, using o-amino-azotoluene, as a rule, no cirrhosis occurs. On the other hand *Maruya* (1940), *Miller*, *Miner*, *Rusch* and *Baumann* (1941) further *Opie* (1944) too are of the opinion that cirrhosis is not necessary for experimental liver tumour formation. *Harris*, *Krahl* and *Clowe*'s (1947) data also show that tumours develop in the liver readily in the absence of cirrhosis. *Kline* (1943) observed that addition of p-aminobenzoic-acid to the diet containing butter-yellow caused great reduction of cirrhosis without changing the incidence of liver cancer. *Eschenbrenner* and *Miller* (1946) by quantitative histologic studies stated that repeated liver necrosis and its associated chronic regenerative state are probably not necessary for the induction of tumours with carbon-tetrachloride.

Fitzhugh and *Nelson* (1948) stated that thiourea administered for longer period to white rats produces hepatic tumours without cirrhosis. On the other hand thioacetamid, a powerful nodular cirrhosis producing substance, does not at all or only exceptionally produce hepatic tumours.

Anyhow on the basis of our investigations made till now we do not wish to take a definite attitude as to the relationship between cirrhosis and formation of liver tumours. No doubt, to establish a sharp distinction between regenerative hyperplasia and neoplasia is not easy. Nodules which were not unquestionably

neoplastic were classed as non neoplastic, though numerous areas were suggestive of early neoplasm. It is, however, certain that no cirrhosis could be determined in a number of our animals with liver tumours. Wether tannic acid is the active agent in inducing hepatic tumours, or whether these tumours are merely the result of hepatic damage caused by tannic acid, awaits further study.

It is worthwhile to mention that *Morozenskaja* (1946) proved that dimethylaminoazobenzene, apart from causing growths in the liver, can also induce growths in other sites and produce characteristic changes of the mesenchyme of the liver resembling leukosis. *Vylegnsanin* (1945) observed analogous changes from the action of orthoaminoazotoluene in mice. Recently *Hoch-Ligeti* (1949) gave the account that in rats which did not develop liver tumours after receiving a diet containing butter-yellow for 17 months, three pancreatic tumours were found. Our observations, however, suggest that the tannic acid effect may be paralleled in this respect too with that of butter-yellow. We could namely state that in the lung of some of the rats a bronchial adenoma developed.

These investigations give rise to several problems. The question of the effective agent in the tannic acid and its causal role in the etiology of human liver tumours may be of the highest importance. Naturally, further investigations are needed to solve these and related problems.

Summary

1. 58 young white rats have been treated with tannic acid solution administered subcutaneously. In group A the rats received a mixed (»normal«) food, in group B a high casein diet.

2. Ulcer was produced by burning the skin of other 39 young white rats. Ulcer in 20 animals has been daily painted with 5 per cent tannic acid solution for a longer period, while the ulcer of the remaining 19 animals also daily painted, however, with 5 per cent hydroahloric acid.

3. Cirrhosis and hepatic tumours appeared merely in animals with parenteral tannic acid treatment.

4. Out of 23 rats of group A (»normal« diet) surviving the 100th day of the parenteral treatment, died or killed between days 109 and 388 of the experiment, in 13, i. e. 56 per cent were induced hepatic tumours.

5. Out of 18 rats of group B (high casein diet) which survived the 100th day of treatment and were killed or died between days 103 and 388 of the experiment, only in 3, i. e. 16 per cent developed hepatic tumours.

6. These results suggests that high casein diet inhibits to a considerable extent the blastomogenic action of tannic acid.

7. The induced tumours were always multiple, histologically hepatomas and cholangiomas, and in general benign, although the invasion into the liver veins, observed in 2 cases, likewise on the basis of atypical pattern in some cases, low grade carcinoma could be considered.

8. The local tumour producing effect of the tannic acid could not be stated and no great importance in tumour induction could be attached to skin nekrosis products.

Карциногенное действие таннина и действие казеина на развитие печеночных опухолей

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Выводы

58 молодых белых крыс получили в среднем через каждые 5 суток, вначале 1.5% -ный, а затем 2Ю-ный раствор таннина подкожно. Всем крысам давали сначала 150, а затем 22 мг таннина на кг веса. Леченных таким образом животных с точки зрения диэты разделили на 2 группы: группа А получила »нормальную« диэту, то-есть остатки госпитальной еды, а группу Б держали на равном количестве молочного хлеба и творога.

Во втором опыте выжигали на спине 39-ти молодых белых крыс кожную язву, величиной 2-форинтной монеты. Обжигаемые таким образом, животные также разделялись на 2 группы: язву 20-и животных ежедневно смазывали 5%-ным раствором таннина, а язву остальных 19-и животных ежедневно смазывали 53-ной соляной кислотой. Животные, подвергаемые этим опытам, держали на »нормальной« диэте.

В печени крыс, переживающих 100-ый день лечения, возникли изменения разной степени: ранняя или более поздняя стадия цирроза, а также опухоли. Вызываемые этим способом, опухоли имели размер от 2–8 мм белосероватыми буграми и вырисовались среди окружающей печеночной ткани. Гистологическое строение этих опухолей соответствовало картине гепатомы и холангиомы.

Опухоли были множественные и, в общем, доброкачественные. В двух случаях, однако, мы наблюдали вторжение опухоли в более крупные печеночные вены, а, вместе с тем, выраженный полиморфизм ядер. В этих случаях мы с полным правом говорили о раннем раке печени. Два раза мы попытались пересадить гепатому, вызванную таннином (под кожу и в печень), но до сих пор безрезультатно.

Из группы А 23 крысы переживали 100-ый день лечения, а среди них у 13-и крыс (56%) удалось вызвать печеночную опухоль. В то-же время среди 18 крыс, бывших на диэте, богатой казеином (группа Б) и переживающие 100-ый день лечения, только у трех (16%) появилась опухоль в печени. Таким образом кажется, что питание, богатое казеином известной степени, предохраняет животных от опухолей.

На коже животных, смазываемых таннином или соляной кислотой (второй опыт), опухоли совсем не возникли. Но ни в печени, ни в других внутренних органах опухоли не встречались. Это опровергает предположение, что при подкожном лечении таннином рассасывание некротических тканевых продуктов играет роль при возникновении цирроза или опухолей печени.

Стихийное возникновение опухоли в печени у этой, воспитанной нами-же линии крыс совсем не наблюдалось, хотя мы исследовали многочисленных крыс более старого возраста.

Циррогенное и карциногенное действие таннина, описанное авторами в первые, ставится ими в один ряд с бластомогенным действием масляного желтого, о-амино-азотолуола, четырех-хлористого углерода и ацетил-амино-флуорена.

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