THESIS

For the Degree of M.D., Edinburgh University

ON the CHOLESTEROL CONTENT of the BLOOD in CASES of CHOLELITHIASIS

With Special Reference to the Variations it shows after Operations on the Gall Bladder and Bile Ducts.

Submitted By

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GENERAL INDEX.

PART/

PART I.

PART I.

AIMS AND SCOPE.

The variations which occur in the Cholesterol content of the blood in pathological conditions have been the subject of much investigation, and in no condition have more divergent results been obtained than in Cholelithiasis. Many workers have found a definite hypercholesterinaemia associated with the presence of gallstones, while others have been unable to confirm this.

It seemed, therefore, that it would be of great interest to estimate the blood Cholesterol in gallstone cases, both before and after operation, with the following aims:-

(i) To determine whether a hypercholesterinaemia occurs in patients who have gallstones, and if so, whether this fact is of practical value in regard to diagnosis.

(ii) To observe the effect upon the blood Cholesterol of the various operations undertaken for the relief of pathological conditions of the gall-bladder.

It was hoped that thereby some light might be thrown upon the, as yet,uncertain relationship between the gall-bladder/

gall-bladder and the metabolism of Cholesterol. The possibility that this organ may play an important part in Cholesterol metabolism has long been recognised, but although much work has been done on this subject very little that is definite has emerged. Recent work has given strong support to the belief that the gall-bladder has some more important function than that of a reservoir and pressure regulating mechanism, and if this function is concerned with the lipoid Cholesterol it is reasonable to suppose that the removal of the organ would produce consistent changes in the Cholesterol content of the blood.

IMPORTANCE.

Cholesterol occurs in all tissues and fluids of the body, and there is increasing evidence that the part which it plays is a very active one. Apart from the attempted elucidation of the problem of Cholesterol metabolism by means of pathological studies, sidelights are thrown during the course of this research upon the metabolism of other substances, especially the fats. The relationship between cholesterol/

cholesterol and the blood fats is an extremely close one, some writers believing the former to be a stage in fat metabolism. If this proves to be the case then the known facts about Cholesterol will assume even greater importance, and such knowledge as has been gained by the study of its changes in pathological conditions will aid towards the understanding of its true physiological relationship to the other lipoids and fats of the blood.

From the clinical standpoint, the estimation of the blood Cholesterol after operations upon the gall-bladder is of importance because it is known that continued bile drainage undoubtedly does deplete the body of its lipoids, and it is of great interest to correlate the findings in the blood with the general condition and symptoms of the patient, throwing light if possible upon the mechanism whereby the body adjusts itself to this loss, and learning in which cases drainage is essential, and in which it may prove unnecessary or even harmful.

TECHNIQUE/

TECHNIQUE.

METHOD.

The method used was that described by BLOOR (1), with slight modifications, and is the application of the Liebermann Burchard reaction (as developed by AUTENREITH & FUNK (2),) to an alcohol and ether extract of whole blood.

MATERIALS USED.

a) Oxalated Blood.
b) Extracting mixture - consisting of 3 parts of Anhydrous Spirit to 1 part of Pure Ether.
c) Pure Chloroform.
d) Acetic Anhydride (Analytical Reagent).
e) Pure Sulphuric Acid.
f) Standard solution consisting of 0·4 mg. of Cholesterol in 1 c.c. of Pure Chloroform.

STEPS in the ESTIMATION

1. 2 cc. of oxalated blood run slowly into 20 cc. of Extracting Mixture in a 50 cc. flask.

2. Contents of the flask brought to boiling-point.

3. Flask filled with Extracting Mixture up to 50 cc. mark, corked, contents thoroughly mixed, and left in the dark for 24 hrs.

4. 10 cc. of supernatant fluid pipetted off.

5. Evaporated almost to dryness.

6./

6. Decanted 3 times with Chloroform.

7. Chloroform added up to 5 cc.

8. 2 cc. of Acetic Anhydride and 0·1 cc. Pure Sulphuric Acid added, thoroughly mixed, and left in the dark for 30 minutes.

9. Compared with Standard in Kober Colorimeter.

DETAILED DESCRIPTION of METHOD.

All flasks, pipettes and cylinders after being cleaned were thoroughly dried before use. 2 cc. of oxalated blood was run slowly into a 50 cc. volumetric flask containing 20 cc. of the Extracting Mixture, the flask being shaken gently all the time to ensure thorough mixing of the blood with the extracting fluid. The contents of the flask were then slowly heated on top of a steam oven until boiling, then cooled, and the Extracting Mixture added up to the 50 cc. mark. The flask was then tightly stoppered with a solid rubber cork, inverted and shaken, and left in the dark for 24 hours. It was found that in this way a clear, colourless supernatant fluid was obtained, and no visible evaporation of the contents of the flask took place in that time.

10cc. of the supernatant fluid were then pipetted/

pipetted off and evaporated almost to dryness in the steam oven, a process which took about 1 hour. The residue was cooled and then 5 cc. (approx.) of Chloroform were added; this was boiled down to half its volume, and decanted into a 10 cc. graduated cylinder. Chloroform was again added up to 5 cc., and the process of boiling and decanting repeated 3 times. After decanting for the last time, the solution was made up to 5 cc. exactly with Chloroform.

1 cc. of the Standard Solution was pipetted into a 10 cc. cylinder, and made up to 5 cc. with Chloroform.

2 cc. of Acetic Anhydride were added to each cylinder, then 0·1 cc. of pure Sulphuric Acid; each cylinder was inverted to mix the contents thoroughly, taking care that none should be spilt, and the cylinders were then left in the dark for 30 mins.

The resulting green solutions were then compared in a Kober Colorimeter, the standard being set at 20.

COLORIMETRIC READINGS.

4 estimations were done together; calling them A,B,C, and D the readings were taken as follows/

follows -

4 of A, 4 of B, 4 of C, 6 of D, 4 of C, 4 of B

4 of A. The average reading for each was then calculated. It was found that the readings took from 10 - 15 minutes to be completed. During that time no apparent alteration in colour took place.

CALCULATION.

1 cc. of the Standard Solution contains 0·4 mg. of Cholesterol.

2 cc. of whole blood were used and were extracted with 50 cc. of alcohol-ether mixture.

10 cc. of the extract were taken ∴ the result represents the quantity of Cholesterol in 1/5 of 2 cc. i.e. 2/5 of 1 cc. of whole blood.

With the standard set at 20 a reading of 10 would equal $0{\cdot}4 \times \frac{5}{2} \times \frac{20}{10}$ mg. in 1 cc. of blood

i.e. 2mg. in 1 cc. of blood.

Therefore, if x is the reading on the scale of the Colorimeter the blood Cholesterol expressed in milligrammes per 100 cc. of blood is -

$$0{\cdot}4 \times \frac{5}{2} \times \frac{20}{x} \times 100$$

EXAMPLE.

Average reading was 12·26.

$$\text{Blood Cholesterol} = 0{\cdot}4 \times \frac{5 \times 20 \times 100}{2 \times 12.26} \text{ mgms. per 100 cc.}$$

$$\text{i.e. } \frac{2000}{12{\cdot}26}$$

i.e. 163·16 mgms. per 100 cc.

DIFFICULTIES/

DIFFICULTIES.

One of the chief limitations of all colorimetric methods is the impossibility of eliminating the personal factor. That individuals vary greatly in their capacity for matching shades accurately is an undoubted fact, and the extent also to which fatigue may affect this power is extremely difficult to estimate. Nevertheless it does not seem probable that these factors could materially affect the results of investigations carried out by this method if all reasonable precautions are taken to minimise their influence.

It is very important to obtain an extract free from any yellowish tinge, as otherwise it is extremely difficult to match the colours accurately.

It was found that attention to two points got rid of this difficulty:

1. The extraction was allowed to continue for 24 hours, so that the supernatant fluid was colourless;

2. Care was taken that the evaporation of the alcohol-ether extract was not carried far enough to cause charring of the organic matter present.

With due attention to these two points it was found that the tone of green produced by the reagents when/

when added to the extract was exactly similar to that produced when they were added to the standard solution of Cholesterol in Chloroform.

As regards the time taken for the colour to develop, it was frequently found that the greatest depth of colour had not appeared in 15 minutes, which is the time given by BLOOR (1) ; on the other hand it was ascertained that once the colour was fully developed, no appreciable decrease, as compared with the standard solution, had taken place up to $\frac{3}{4}$ hour after the addition of the reagents. 30 minutes was therefore taken as the time at which the colour was fully developed in all cases, and the readings were taken then.

Not more than four estimations were done simultaneously, and the readings were therefore always finished in less than $\frac{3}{4}$ hour from the addition of the reagents.

All estimations were done in duplicate, and the duplicates were always done in different sets, in order to keep a check upon the accuracy of the results.

The addition of a drop of water to the chloroform extract prevents the appearance of the characteristic/

characteristic colour change when the reagents are added to it, in the last stage of the process; so that it is of great importance to have the reagents, flasks etc. completely anhydrous.

In spite of all precautions against any moisture interfering with the reaction, it was found that occasionally no colour change took place, or only a very poor one. Therefore from every patient sufficient blood was obtained to enable the process to be repeated if a perfectly accurate result was not obtained.

It is apparent therefore, that the difficulties in connection with this method allow of its being sufficiently accurate only if very carefully checked. This makes it rather more laborious than would at first sight appear.

ACCURACY.

A. CIRCUMSTANCES under which BLOOD was obtained from PATIENTS.

Blood was obtained from all patients during the morning, and for the majority of the pre-operative estimations it was drawn off on the morning/

morning of the operation. In some cases it was not expedient to do this, therefore an experiment was carried out to ascertain whether the preparation for operation (purgative and short period of starvation) had any effect upon the Blood Cholesterol.

In 6 patients a Blood Cholesterol estimation was done the morning after admission to Hospital while still on ordinary diet; and then again on the morning of operation.

The results were as follows -

BLOOD CHOLESTEROL in MGMS. per 100 cc.

Patient	First Estimation	Second Estimation
1.	174	171
2.	187	190
3.	208	209
4.	210	202
5.	137	134
6.	163	171

It will be seen that these figures correspond very closely, so it was concluded that the preparation for operation did not cause any alteration in the Blood Cholesterol.

B./

B. ACCURACY of the ESTIMATION.

A solution was made up consisting of 20 mgms. of Cholesterol in 50 cc. of the Extracting mixture used in the estimations. Varying quantities of this solution were pipetted off into flasks and evaporated almost to dryness; the residue was then decanted 3 times with Chloroform and the rest of the estimation carried out in an exactly similar fashion to that used with the blood extract.

With the standard set at 20, a reading of 20 indicates that the solution being examined contains 0·4 mgms of Cholesterol -

$\therefore$ x is the reading for $\frac{20 \times 0{\cdot}4}{x}$ mgms.

i.e. the reading obtained divided into 8 gives the number of milligrammes of Cholesterol present in the solution being tested.

TABLE/

	AMOUNT of SOLUTION USED	CHOLESTEROL ESTIMATED (in mgms.)	CHOLESTEROL PRESENT (in mgms.)	ERROR	PERCENTAGE ERROR.
1.	1·0 cc.	0·3866	0·4000	0·0134	3·35
2.	1·1 cc.	0·4388	0·4400	0·0012	0·27
3.	1·2 cc.	0·4745	0·4800	0·0055	1·15
4.	1·3 cc.	0·4896	0·5200	0·0304	5·84
5.	1·4 cc.	0·5472	0·5600	0·0128	2·29
6.	1·5 cc.	0·5852	0·6000	0·0148	2·47
7.	1·6 cc.	0·6205	0·6400	0·0195	3·05
8.	1·7 cc.	0·6623	0·6800	0·0177	2·60
9.	1·8 cc.	0·6820	0·7200	0·0380	5·28
10.	1·9 cc.	0·7260	0·7600	0·0340	4·47
11.	2·0 cc.	0·7744	0·8000	0·0256	4·20
12.	2·1 cc.	0·8112	0·8400	0·0288	3·43

The/

The average percentage error for 12 estimations was 3·2%. As this may be either plus or minus, and some allowance must also be made for a slight experimental error occurring during the measuring out of 2 cc. of blood, it was decided to regard the experimental error as 8%.

PART II.

PART II.

THE PHYSIOLOGY OF CHOLESTEROL.

EARLY THEORIES.

Cholesterol was isolated by CONRADI in 1775 and analysed by CHEVREUL in 1815. It is an-unsaturated alcohol, closely related to the polyterpenes, and occurs in all tissues and fluids of the body, both free and in the form of esters. The proportions of these two forms varies according to the tissue or fluid in question. It is most abundant in the white matter of the brain - in which it occurs as free cholesterol - in the medullary sheaths, the lung, and the suprarenal cortex.

The comparative quantity in the different organs is the same in different species of animal. In 1862 FLINT (4) advanced the theory that the liver excreted cholesterol. He believed that the blood became laden with cholesterol after traversing the brain and the various organs which are rich in this substance, and that the liver removed the cholesterol from/

from the blood and excreted it into the bile. It has since been disproved that the blood from different parts of the body varies in its cholesterol content (19); but the majority of writers have agreed with FLINT as to the importance of the liver in cholesterol metabolism.

Some of the earliest work in connection with this lipoid was in regard to the conception that it forms an important constituent of the cell-membrane. HERMANN showed that aliphatic narcotics liberate haemoglobin from red blood corpuscles and he believed that this was due to their action on cholesterol and lecithin in the cell-membrane. OVERTON and MAYER simultaneously propounded the theory of a semi-permeable membrane of lipoid material forming the outer layer of every cell. Many difficulties in regard to this theory have been advanced from time to time, but it has formed the basis for many extremely interesting suggestions.

EXCRETION/

EXCRETION, ABSORPTION, and SYNTHESIS.

Until the beginning of this century, cholesterol was regarded as purely an excretion. The general view was that cholesterol was removed from the blood by the liver, excreted into the bile, and so into the faeces.

Little further light was thrown upon its physiology until the researches of DORÉE, ELLIS, GARDNER, and their co-workers between 1908 and 1912. To them is due the credit of establishing some fundamental principles in regard to cholesterol. They showed that in herbivora no cholesterol is normally excreted, but that if they are fed with cholesterol only about 50% appears in the faeces, the inference being that the remainder is absorbed. By means of a series of experiments of a similar nature, carried out on various species of animal, they were able to prove that cholesterol must normally be absorbed in the intestine. It is possible that some may be totally destroyed, or converted into some non-crystalline oily product, but this they consider unlikely. They formulated the following hypothesis:

"Cholesterol/

"Cholesterol is a constituent constantly present in all cells and when these are broken down in the life process, the cholesterol is not excreted as a waste product but is utilised in the formation of new cells. A function of the liver is to break down dead cells, e.g. blood corpuscles and eliminate their cholesterol in the bile. After the bile has been poured into the intestine in the process of digestion, the cholesterol is reabsorbed, probably in the form of esters, along with the bile salts, and is carried in the blood stream to the various centres and tissues for incorporation into the constitution of new cells" (5).

They confirmed PRIBRAM'S statement that cholesterol can be found in rabbits' blood after feeding them with cholesterol and they pointed out that the quantity in the blood did not depend on the amount given, but on the amount which the body could utilise (6). The mechanism of absorption will be discussed later.

ELLIS & GARDNER also showed that the sterol content of the liver was dependent on the sterol content of the food. Their work on the cholesterol/

cholesterol content of various organs during inanition is most interesting - they found a marked increase in the blood, liver, and kidney [7]. This early work foreshadows the writings of recent investigators who have attempted to trace the real connection between fat metabolism - both normal and abnormal - and cholesterol metabolism in the human body.

General opinion at this time was against the occurrence of synthesis of cholesterol. DORÉE & GARDNER [5], believed that Cholesterol is conserved and that therefore any wastage in the sweat, sebum, etc. must be made up either by absorption from the food or by synthesis. In their earlier experiments they found no evidence in favour of synthesis; but later on GARDNER & FOX [8] found that balance experiments in man showed a considerable increase of output over intake, and they therefore concluded that synthesis does take place. They were able to confirm this by experiments on normal infants [9], and numerous other workers have in recent years produced evidence that confirms their results [38]. ARTOM [10] has shown that oleic acid increases cholesterol in liver autolysis, and that cholesterol

can/

can be synthesised from oleic acid. GARDNER & FOX (11) found no evidence of either destruction or synthesis of cholesterol during autolysis of liver and spleen, but they confirmed the belief of SCHÜLTZE and of CYTRONBERG that a cholesterase exists in the corpuscles but not in the plasma.

The balance of evidence seems to be in favour of synthesis taking place, but yet we find as recently as 1922 the following statement by MCADAM, quoted by SIR BERKELEY MOYNIHAN (12) -

> "Recent work has shown conclusively that there is no synthesis of cholesterol in the body".

Some experiments by W.M. SPERRY (13) are of importance in this connection. He finds that in bile fistula dogs fed on a sterol free diet, the excretion of cholesterol is very similar to that of normal controls, showing that synthesis probably does take place. He considers that the great similarity in the proportions of fatty acids, sterols etc. excreted by normal and by bile fistula dogs makes it probable that the source is the same, thus minimising the importance of bile in cholesterol excretion.

INFLUENCE/

INFLUENCE of VARIOUS ORGANS on CHOLESTEROL METABOLISM

THE LIVER.

All the early workers believed that the liver was the organ concerned in cholesterol metabolism, but in 1892 NAUNYN (14) propounded the theory that cholesterol in the bile does not really result from general metabolism, nor is it a secretion of the liver, but is due to catarrh of the gall-bladder wall when infection is present. This theory in its entirety received little support, but gave rise to various suggestions regarding cholesterol and the gall-bladder. These will be referred to more fully later.

SCHAFER (15) suggested that lecithin and cholesterol in the bile may be associated with the destruction of red blood corpuscles which takes place in the liver; and the experiments of KOSUMOTO in connection with icterus produced by toluylene diamine poisoning, in which red blood corpuscles are destroyed and bile cholesterol is increased, are further evidence in favour of the importance of the liver in cholesterol metabolism.

Some/

Some experiments also by JANKAU[16] in NAUNYN'S laboratory showed that cholesterol is formed in the liver and not merely excreted by it.

THE SPLEEN.

ABELOUS & SOULA found a marked increase of cholesterol on autolysis of spleen, and they believed that this organ is capable of synthesing cholesterol. Recently it has been shown that splenectomy causes a rise in blood cholesterol[17], which suggests that the spleen destroys rather than forms cholesterol. Apparently the spleen removes cholesterol from the surface layer of the corpuscles, thus decreasing their resistance to haemolysis by osmosis[18].

In 1901 RANSOM showed that cholesterol could prevent haemolysis by saponin, and it is probable that the lecithin-cholesterol ratio is important in this connection. The spleen may possibly be concerned in the maintenance of this ratio.

THE SUPRARENAL GLANDS.

GRIGAUT[19] believed that synthesis of cholesterol was carried out by the suprarenals, and he and his co-workers, LA ROCHE & CHAUFFARD, consider/

consider that the corpus luteum is also concerned.

It has been suggested that increased blood cholesterol depends on increased functioning of the suprarenal cortex in a manner analogous to the increase of adrenalin in the blood which occurs in Bright's disease, and is due to increased functioning of the medulla (20). LANDAU suggests that the lipoid-containing cortex may be of importance to the Sympathetic Nervous System, as the lipoid-containing white matter is of importance to the Central Nervous System.

There is a great increase in the cholesterol in the suprarenal cortex in pregnancy (18), and also in other conditions where there is a hypercholesterinaemia. LANDAU & MCNEE (21) have shown that feeding with cholesterol increases the cholesterol content of the suprarenals, so that they more probably act as a depot than are concerned in the manufacture of cholesterol.

JOELSON & SHORR (22) found a rise of blood cholesterol after suprarenalectomy, and suggest that the suprarenals control cholesterol as insulin does the blood sugar.

ROTHSCHILD showed that in rabbits the

blood/

blood cholesterol must increase to a certain point before any increase appeared in the bile; but that after suprarenalectomy the increase of cholesterol in the bile was parallel to that in the blood (23). Removal of the thyroid is said to cause a rise of blood cholesterol, and injection of thyroid extract a fall. As adrenalin raises the basal metabolism this would correspond with the fact that removal of the suprarenals causes increased blood cholesterol (18)

Recently MAHLER (21) has suggested that the internal secretion of the pancreas has an influence upon blood Cholesterol. He found that, during ether anaesthesia, there is a rise of blood Cholesterol, roughly proportional to the rise in blood sugar. A preliminary injection of insulin prevented these changes taking place.

It is at present impossible to correlate all the facts which have emerged regarding the relationship of cholesterol to the liver, the spleen, the suprarenal glands and the pancreas.

CHOLESTEROL and FAT METABOLISM.

LEATHES (24), in comparing the fat of adipose tissue with that occurring within the organs/

organs points out that one important difference is that the latter always contains cholesterol whereas the former does not. He maintains that fats and lipoids must be considered as being just as essential to the structure of the cell as are the proteins; and his experiments and those of LANGMUIR & ADAM (25) on fatty films are most important and suggestive. Recent researches into the proportion of cholesterol, lecithin, and fatty acids in different types of cell have brought to light many interesting facts. It has been shown (3) that in each species the ratio of these substances in any organ is remarkably constant, and for all species the ratios for the different organs come out in the same order. The ratio of cholesterol to fatty acid determines the proportion of water that can be taken up by a cell if immersed, and this proportion varies directly with the cholesterol (18).

BLOOR (26) has shown that in diabetics when an alimentary lipaemia is produced cholesterol increases much more than lecithin, so that the lecithin-cholesterol ratio is decreased. It is not clear why cholesterol should increase along with lecithin, but in many reactions - tumour growth, haemolysis/

haemolysis etc. - they are antagonistic, so that it may be a protective mechanism.

Lipase is often absent from tissues concerned in fat metabolism but a lecithin-splitting ferment present, so LEATHES has suggested that lecithin is a stage in fat metabolism. BLOOR suggests that the corpuscles take fat from the plasma and convert it into lecithin. As there is known to be a cholesterase in the corpuscles it is possible that a similar process may take place with cholesterol.

As yet however, there is little real evidence in favour of the view that cholesterol is a stage in fat metabolism, although it is undoubtedly increased in the blood in many conditions in which there is increased fat metabolism, such as lipaemia and pregnancy.

THE/

THE RELATIONSHIP of BILE to CHOLESTEROL METABOLISM.

GRIGAUT (19) believed that alterations in cholesterol content of the blood were not followed by changes in the bile cholesterol, but it has since been proved that any increase of blood cholesterol is followed by an increase of bile cholesterol (27).

It has also been shown that a diet rich in cholesterol causes an increase of bile cholesterol (28). WILENSKY & ROTHSCHILD point out that in a patient with a biliary fistula, the day on which fat is added to the diet can be ascertained from the results of daily estimations of the cholesterol content of the fistula bile (29). WINDAUS suggested that the cholic acid portion of the bile salts might be derived from cholesterol and CHAUFFARD strongly supports this view (37). The general view that bile is the medium of cholesterol excretion has been questioned by SPERRY, owing to the results of his experiments on bile fistula dogs (referred to on page 21). On the other hand MUELLER (30) has shown that in the absence of bile from the intestine cholesterol absorption is diminished, and he states that/

that absence of pancreatic secretion has the same effect.

Further work on the lines indicated by SPERRY would be of value in the elucidation of this problem.

RELATIONSHIP of the GALL-BLADDER to CHOLESTEROL METABOLISM.

With the exception of NAUNYN, whose views have already been referred to, all those who have attempted to connect the gall-bladder with cholesterol metabolism have been concerned with the question of absorption. GRIGAUT (19), pointed out that the gall-bladder bile differed greatly from that in the ducts, and it has since been proved conclusively that the gall-bladder has a very great concentrating power and removes water from the bile (31).

In 1846 VIRCHOW described the filling of the cells of the gall-bladder mucosa with fat and compares the process with fat absorption in the intestine. In 1906 ASCHOFF (32), showed that the epithelium of the gall-bladder can take up neutral fat, and he states that cholesterol esters can be taken/

taken up in the same way. These results were confirmed by POLICARD (33), and more recently by BOYD (34). The latter's studies of pathological conditions of the gall-bladder and in particular of the so-called "strawberry-gallbladder" have led him to believe that the lipoid material found deposited in the wall is normally absorbed, but becomes deposited when any inflammatory condition interferes with the normal absorptive power of the gall-bladder mucosa.

It has been shown (35) that when a catheter is inserted through the ampulla of Vater and potassium sulphocyanide injected through it into the gall-bladder, this substance is absorbed and can be found in the lymphatics of the gall-bladder wall.

BOYD (34) further points out that, comparing the bile in the cystic duct with that in the hepatic duct, sodium taurocholate has become increased 16 times, sodium glycocholate 20 times, but cholesterol only 10 times, and from these facts he concludes that not only water but cholesterol has been absorbed by the gall-bladder.

In connection with the capacity of the gall-bladder for absorption, SWEET (36) has emphasised some anatomical factors. He points out that the kinking/

kinking of the cystic duct, the presence of HEISTER'S valve, and the fact that the gall-bladder is the most dependent part of the biliary system, are all evidence in favour of his belief that whatever passes into the gall-bladder does not pass out again by the cystic duct. He says - "The manner of origin "and development, complex structure, rich blood sup- "ply, and unusual lymphatic supply, all point to the "probability of an absorptive function".

BOYD (34), also, emphasises the wonderfully complex structure of the mucous membrane, and its delicate villi, which seem specially designed for absorption.

A great stumbling-block to assuming an essential function for the gall-bladder is the fact that no one has yet proved the presence of any structure which can take its place after removal.

SWEET (36) describes the sac-like appendages found in the walls of the bile ducts. These were demonstrated by BEALE in 1899, and he referred to them as "supplementary little gall-bladders appended to the ducts". These are more numerous in animals that have no gall-bladder, and SWEET found/

found that in dogs, some months after cholecystectomy, these appendages had increased greatly in size. He concludes therefore that they are capable of assuming the function of the gall-bladder after it has been removed.

His observations on the blood cholesterol of cholecystectomised dogs are of great interest and will be referred to later. As a result of these anatomical studies and post-operative experiments he believes that the function of the gall-bladder is to absorb cholesterol or some substance which can break up cholesterol esters.

CHOLESTEROL in the BLOOD.

NORMAL DISTRIBUTION AND QUANTITY.

BOUDET, in 1833, was the first investigator to demonstrate the normal presence of Cholesterol in the blood.

Cholesterol is equally distributed between the corpuscles and the plasma, but in the corpuscles it is mainly uncombined, whereas in the plasma more than half is present as esters (39). The amount present in the plasma is more variable in disease, and in most pathological conditions it is the combined cholesterol which is increased. In obstructive jaundice, however, it is the free cholesterol which is increased, suggesting that the increase is due to retention.

The amount present in the blood has been variously computed by workers using different methods. WINDAUS' Digitonin method is undoubtedly the most accurate, but it is not very convenient for clinical purposes. CAMPBELL (18) sums up the results given by various writers and concludes that 150 to 200 mgms, per 100 cc, may be taken as the normal range/

range, with 180 mgms, as the average. This corresponds closely to the values found in this investigation, although other workers using BLOOR'S method have obtained somewhat higher figures.

HYPOCHOLESTERINAEMIA.

A decrease in blood Cholesterol has been found to occur in chronic anaemia, febrile diseases (except typhoid), tuberculosis, and cachexia (20). GRIGAUT (19) has pointed out that in febrile diseases the temperature curve and the blood cholesterol curve cross one another, because the blood Cholesterol rises again as soon as defervescence occurs (40). In this connection, it has been shown that a hypercholesterinaemia accompanies the process of immunisation. McADAM & SHISKIN (41) hold that a low blood cholesterol content is of importance in genito-urinary sepsis, either as an indication of lowered resistance, or of commencing uraemia.

Although/

Although much has been written about the connection between cholesterol and immunity, nothing is as yet definitely known, and no adequate explnations are forthcoming for the low Blood Cholesterol values found in the conditions mentioned above.

PHYSIOLOGICAL HYPERCHOLESTERINAEMIA

There are 4 conditions which may be classed as physiological, in which the blood Cholesterol is increased - acute anaemia, fat ingestion, starvation, and pregnancy.

ACUTE ANAEMIA and FAT INGESTION.

BLOOR (42) showed experimentally that the changes in the blood fats and lipoids after fat ingestion and after bleeding are very similar. Probably in the former the blood is unable to deal sufficiently rapidly with the fat brought to it, whereas in the latter the decreased number of red corpuscles leads to a concentration of lipoids and their storage in the body (42). In chronic anaemia it is probable that an adaptive mechanism has been called/

called into action, and an increase in blood Cholesterol does not take place.

STARVATION.

Reference has previously been made to the increase of Cholesterol in the blood and also in various organs which takes place during starvation. Increased fat metabolism while the body is living on its own tissues is the explanation in this instance. BLOOR (quoting DADDI & TERROINE) states that in the first few days of fasting the blood Cholesterol increases and then remains constant.

PREGNANCY.

During the early months the Blood Cholesterol rise is slight, but in the later months it averages 250 mgms. per 100 cc., or even more than that. In all probability the same explanation, i.e. increased fat metabolism, applies to pregnancy as to the other conditions referred to already. It has been shown that the Blood Cholesterol of the new-born is low, and probably the milk is the chief avenue of excretion for the excess. McNEE (20) has shown that bile has an increase of Cholesterol proportionately greater than that in the blood, and this also serves as a means of excretion for the excess. The connection of these facts with the etiology of gall stones will/

will be discussed later.

PATHOLOGICAL HYPERCHOLESTERINAEMIA

There are 4 pathological conditions in which hypercholesterinaemia is found - diabetes, nephritis, arterio sclerosis, and obstructive jaundice.

1. DIABETES.

This was one of the first conditions in which a hypercholesterinaemia was observed. It is probably here also associated with the excessive fat metabolism which occurs as a sequel to defective carbohydrate metabolism, and is in some ways analogous to the hypercholesterinaemia in starvation. Very varied opinions have been expressed as to the value of cholesterol estimations in regard to prognosis, but in general it appears to be established that a high blood Cholesterol is of grave prognostic import.

2. NEPHRITIS.

Very high blood Cholesterol values are often found in chronic nephritis - both the interstitial/

interstitial and the parenchymatous varieties. BLOOR considered that there was retarded fat assimilation due to acidosis, thus bringing nephritis into line with other hypercholesterinaemic conditions; but this view has not been generally accepted, and the cause of the raised blood Cholesterol remains obscure. CHAUFFARD (19) and his fellow-workers believe that both in pregnancy and nephritis the hypercholesterinaemia is due to increased production of Cholesterol by the suprarenals.

3. ARTERIO SCLEROSIS.

Here also the cause of the raised blood Cholesterol is not known. Many interesting experiments have been carried out in the attempt to produce arterio sclerosis by feeding with Cholesterol or by injecting it into the blood-stream, but very varying and inconclusive results have been obtained. It is very improbable that Cholesterol plays any part in the etiology of arterio-sclerosis.

4. OBSTRUCTIVE JAUNDICE.

All observers are agreed that in obstructive jaundice due to gall stones the blood Cholesterol is high, but opinions differ about Cholesterol in/

in other liver diseases with jaundice. Most writers have found that in cirrhosis it is low or normal; and that in carcinoma of the liver it may be low or high, depending on whether the obstructive or destructive element preponderates.

The question as to whether the blood Cholesterol is increased in cholelithiasis per se will be discussed in the next section.

CAMPBELL (18) has pointed out that in 4 of these conditions associated with hypercholesterinaemia lipaemia is liable to occur, i.e. in acute anaemia, fat ingestion, diabetes and nephritis; and that in 4 acidosis may occur, i.e. diabetes, nephritis, pregnancy and starvation. What the connection may be between lipaemia, acidosis, and hypercholesterinaemia it is impossible to state.

PART III.

PART III.

THE ETIOLOGY of CHOLELITHIASIS.

There is a general agreement in regard to the three main etiological factors in the production of gallstones, namely infection, stasis, and hypercholesterinaemia, but very great variety of opinion as to their relative importance.

A. INFECTION.

Infection was first suggested as the cause by GALIPPE in 1866, and B.coli and B.typhosus were found in gallstones soon after this.

NAUNYN believed that the initial process was a catarrh of the gall-bladder wall, with consequent out-pouring of cholesterol-laden mucus. This excess of cholesterol became cemented together by bilirubin calcium and thus stones were formed.

It is well-known that B.typhosus can lie latent for many years in the gall-bladder, but recently the importance of this organism in gall-bladder infections has become minimised owing to the comparative rarity of typhoid fever, whereas gallstones are just as common, if not commoner, than ever/

ever.

ROSENOW has shown that certain strains of streptococcus have a special affinity for the gall-bladder, and the role that streptococci play in diseases of this organ is much greater than was at one time believed.

The role of B.coli in gall-bladder disease is a very important one. It has been pointed out (34) that in vitro B.coli causes a precipitation of Cholesterol - probably by interfering with the bile salts. It has been shown also that an increase of dissolved bile salts enables the bile to take up an enormous quantity of cholesterol, and the converse also is true. GRAHAM believes that cholecystitis always produces hepatitis, and HURST (quoted by ROLLESTON (43)) thinks that the resulting lessened production of bile acids causes Cholesterol to be precipitated in the gall-bladder. It follows from this that anything else which interfered with that particular function of the liver would tend to cause gallstones, but this seems hardly probable.

WILKIE (44) has pointed out the frequency of infection of the gall-bladder during pregnancy, owing/

owing to the tendency to constipation with consequent B.coli infection of the portal blood..

MAYO (45) states that a change in the protective colloid medium in which the bile cholesterol is suspended may cause this substance to be precipitated; such a change may be produced by excess of albumen, fats, or toxins. He believes, however, that the primary change is metabolic.

B. HYPERCHOLESTERINAEMIA.

CHAUFFARD (37) was a strong advocate of the metabolic theory, and he believed that gallstones are formed when there is an excess of cholesterol in the bile. From a different point of view ASCHOFF (46) supported the theory of aseptic cholelithiasis, basing his belief on the fact that no organisms could be cultivated from pure cholesterin stones. Many observers since, however, have come to the conclusion that an infection, originally present in the stones, may die out. WILKIE (44) considers that the necessity for infection is unproven, and this probably expresses the general belief at the present time.

MANN (47) has shown that certain hypochlorites introduced into the gall-bladder can produce aseptic/

aseptic inflammation of the mucosa and round-celled infiltration. In experiments on dogs biliary calculi have been produced in large numbers aseptically. It is natural to assume that the presence of gallstones in a sterile and perfectly healthy gallbladder would eventually cause trauma to the mucosa with a supervening infective process, so that the presence of inflammation does not preclude the possibility that the stones were there first.

CHAUFFARD (37) lays great stress upon the hypercholesterinaemia of pregnancy, and also on that occurring during convalescence from typhoid fever, and believes not only that hypercholesterinaemia is the primary etiological factor, but that it is of great value in the diagnosis of doubtful cases of cholelithiasis. He and his fellow worker GRIGAUT believe that the liver converts cholesterin into cholalic acid, and that it is a failure of this function which leads to an excess of cholesterol in the bile and consequently to aseptic cholelithiasis.

C. STASIS.

This was considered to be the main cause of gallstones, before infection and metabolic changes became the subject of so much discussion and/

and experimental work. Undoubtedly it predisposes to the deposition of stones when one or other of the two important etiological factors is at work.

The well-known association of gallstones with repeated pregnancies lends weight to the arguments in favour of both infection and hypercholesterinaemia as primary causes of gallstones. The fact that gallstones can be shown to have been produced in crops might be used in favour of the occurrence of hypercholesterinaemic crises or of bouts of infection.

The frequent occurrence of gallstones in fat women has been instanced as a fact in favour of some associated disturbance of fat metabolism. As we have already seen the connection between cholesterol and fat metabolism is a close one.

The influence of such factors as heredity, racial predisposition, and individual metabolic differences, or the diathetic factor, must all be taken into account, but space does not permit of more than the mere mention of their existence.

BLOOD CHOLESTEROL in CHOLELITHIASIS.

However great the influence of hypercholesterinaemia in the production of gallstones, it is difficult to understand why so many writers seem convinced that it ought to be still present at the time when patients seek advice for their symptoms. An enormous number of investigators have published the results of their estimations of the blood cholesterol in cases of cholelithiasis, but only those within recent years will be referred to.

McNEE (20) examined a series of 52 patients of whom 10 had gallstones - proved at operation. Not one had a high blood cholesterol. REIMAUN & MEIGON (48) examined a series of 60 cases of abdominal lesions, and found that although the figures for gallstones were somewhat above the normal, other upper abdominal lesions showed just as high figures.

SCHNABEL (49) also found inconstant values, but the average was higher than normal. MYERS (50) examined a long series of cases and decided that the blood Cholesterol was of no value in diagnosis. All these observers give a normal limit very similar to that found in this investigation.

DENIS/

DENIS [51] found the blood Cholesterol values to be normal in cases of gallstones and also in many other conditions in which other observers have found the blood Cholesterol raised, but he gives the very wide normal range of 167 to 255 mgms. per 100 cc., so that it is difficult to compare his conclusions with these of other investigators.

On the other hand FLANDIN, (quoted by CHAUFFARD [37]) found a constant hypercholesterinaemia in all cases of gallstones; SHISKIN (quoted by MOYNIHAN [52]) finds hypercholesterinaemia in 65% of gallstone cases, and also that 75% of all patients with hypercholesterinaemia have gallstones. BELL [53] & GRIFFITHS [54] both state that there is an increase. Most of these observers omit to mention whether or not their patients were jaundiced.

The most interesting work in this connection is that done by WILENSKY & ROTHSCHILD [55]. In a long series of cases they have correlated the blood Cholesterol values with the pathological findings at operation or autopsy, and they conclude that the blood Cholesterol may be normal in the following instances/

instances:-

1. In the presence of stones in a normal gall-bladder, without common duct obstruction and with or without high temperatures.

2. In the presence of a chronically inflamed gall-bladder containing stones without common duct obstruction and with or without high temperatures.

3. In the presence of any of the above with incomplete obstruction of the common bile duct.

4. In the presence of an empyema of the gall-bladder, with or without stones, with low or high temperatures, and with no obstruction in the common duct.

5. As in (4) with partial or complete obstruction of the common duct.

In addition to this the presence of other complicating factors, such as pregnancy and arteriosclerosis, must be taken into account. They came to the conclusion that the diagnostic value of the determination of the cholesterol content of the blood is a variable and dubious one.

They state that the only instance in which the blood cholesterol is of value in diagnosis is when a distinction must be made between jaundice due to obstruction, and jaundice due to cirrhosis of the liver - the cholesterol content of the blood being invariably low in the latter condition.

BLOOD CHOLESTEROL ESTIMATIONS

in a

SERIES of 115 CASES.

TABLE I.

BLOOD CHOLESTEROL

of

16 NORMAL INDIVIDUALS BETWEEN the AGES OF 20 and 30

1.	176	9.	178
2.	153	10.	202
3.	170	11.	197
4.	165	12.	157
5.	155	13.	147
6.	181	14.	155
7.	163	15.	167
8.	161	16.	151

Average is 167·2 mgms. per 100 cc of blood. It will be seen from the above table, that with the method used, the figures for the normal blood cholesterol ranged from 147 to 202 mgms. per 100 cc. It was/

was therefore decided to take from 150 to 200 mgms. per 100 cc. of blood as representing the normal limits of the cholesterol content of the blood.

TABLE/

TABLE I I.

BLOOD CHOLESTEROL

of

20 PATIENTS with VARIOUS PATHOLOGICAL CONDITIONS.

	CHOLESTEROL	AGE	DIAGNOSIS.
1.	173	58	Inguinal Hernia
2.	209	60	" "
3.	160	44	Umbilical Hernia
4.	174	54	" "
5.	153	53	Rheumatoid Arthritis
6.	136	30	Thromboangeitis Obliterans.
7.	134	45	" "
8.	154	30	Haemophilia
9.	156	45	"
10	172	28	"
11	124	19	Osteomyelitis - Chronic
12	137	45	Pyaemia Abscesses
13	202	27	Tubercular Cervical Glands.
14	210	25	Phthisis and Pyloric Ulcer.
15	150	53	Sarcoma of the Sciatic Nerve.
16	189	58	Carcinoma of the Breast.
17	165	57	Epithelioma of the Tongue.
18	163	56	" " " "
19	159	69	" " " "
20	167	52	" " " "

Average/

Average is 164·85 mgms. per 100 cc.

In the cases the rangesof blood cholesterol values is somewhat greater than in the normal group, being from 124 to 210 mgms. per 100 cc., but the average is almost identical with that of the normal cases.

TABLE/

TABLE I I I.

BLOOD CHOLESTEROL

of

21 PATIENTS with VARIOUS INTRA-ABDOMINAL CONDITIONS.

	CHOLESTEROL	DIAGNOSIS	
1.	177		
2.	170		
3.	152	Appendictis	Average is 175·5 mgms, per 100cc.
4.	221		
5.	179		
6.	154		
7.	148		
8.	156		
9.	200	Gastric or Duodenal Ulcer	Average is 181.0 mgms. per 100cc.
10	185		
11.	199		
12.	198		
13	136		
14	222		
15	139	Chronic Pancreatitis	Average is 165·75 mgms. per 100 cc.
16	166		
17	171	Carcinoma of the Stomach	
18	168	Carcinoma of the Stomach.	
19	113	Carcinoma of the Pylorus.	
20	168	Tubercular Mesenteric Glands	
21	156	Gastroptosis.	

Average/

Average is 170·38 mgms. per 100 cc.

In this group also the range is greater than normal - 113 to 222 mgms, per 100 cc. but the average is again similar.

DISCUSSION/

DISCUSSION of TABLES I, I I, and I I I.

The Blood Cholesterol content for 58 cases shows figures ranging from 124 to 222 mgms per 100cc. of blood.

The greatest variation from normal was therefore (approximately) 25 mgms. above or below the normal limit.

The cases shown in TABLE II. were taken at random from the admissions to a Surgical Ward (except the 3 haemophilics).

3 showed a Blood Cholesterol value slightly above normal, and in none of them was any known factor present to account for this. One was a case of pulmonary tuberculosis, and the Blood Cholesterol is usually low in these cases.

4 showed a low Blood Cholesterol value. In 2 of these a chronic infective condition was present; the other 2 were cases of Thrombo-angeitis Obliterans, and were both in an extremely low state of health.

Of the cases shown in TABLE III, 2 showed a high blood cholesterol; in neither case was there/

there any known reason for this.

4 showed a low blood cholesterol; in 2 of these (Chronic Pancreatitis, 139, and Carcinoma of the Pylorus, 113) the patient's general condition explains the low value found; in the other 2 there is no apparent explanation.

Thus in a series of 42 cases of various pathological conditions, the range of Blood Cholesterol values is greater than in normal individuals, but the factors governing these alterations are not clear.

TABLE/

TABLE IV.

BLOOD CHOLESTEROL

of

27 CASES of CHOLELITHIASIS without JAUNDICE

NO.	BLOOD CHOLESTEROL.	AGE	TEMP.	BLOOD PRESSURE	
31	127	38			
40	142	42	99·2°		
85	147	34			
54	154	43		105	
56	158	37	100°		
59	158	49		160	
81	162	40		110	
42	166	43	100°		
39	166	52			
75	166	62			
53	170	50		140	
48	171	46		120	
55	176	39			Average
64	178	28		120	Blood Cho-
78	184	35		120	lesterol is
60	184	50			180·56 mgms.
41	184	60			in 100 cc.
62	188	51			
34	192	43			
63	195	35			
44	199	35			
89	205	54			
61	209	57		130	
52	218	44		130	
47	222	57	98·8°	140	
69	224	41			
65	230	43		140	

DISCUSSION of TABLE IV.

TABLE IV. gives the Blood Cholesterol for 27 cases of Cholelithiasis, without jaundice, and with no abdominal complications other than Cholecystitis, which was present in all cases but one.

The figures range from 127 to 230 mgms. per 100 cc. of blood. This is greater than the normal range but very similar to that obtained for 21 Abdominal Conditions (TABLE III), i.e. 113 to 222 mgms. The average Blood Cholesterol is 180·56 mgms. per 100 cc., this being somewhat higher than the average for 21 Abdominal Cases, which was 170·38 mgms. per 100 cc., and therefore also slightly higher than the normal average, 167·2 mgms. per 100 cc.

6 cases gave values above normal. In none was there any known factor which might raise the Blood Cholesterol. The Blood-pressure was recorded in 4 of these cases, and all were normal. Case 69, with a Blood Cholesterol of 224 mgms. per 100 cc. was a healthy male, aet.41, with no signs of arterio sclerosis.

It is interesting to note that of these 6 high/

high balues, 2 were males.- Cases 69 and 47; and 1, case 52, was an unmarried woman with a healthy gall-bladder and pure cholesterin stones.

3 cases gave low values; one of these, Case 40, was acutely ill, with a perforated gall-bladder; for the other 2 there was no apparent explanation.

4 cases showed a slight rise of temperature. This would tend to reduce the Blood Cholesterol, but probably only slightly in these cases.

In all cases in this series in which the temperature is not recorded it was normal.

TABLE/

TABLE V.

BLOOD CHOLESTEROL

of

7 CASES of CHOLELITHIASIS without JAUNDICE (with ABDOMINAL COMPLICATIONS).

NO.	BLOOD CHOLESTEROL.	AGE	TEMP.	BLOOD PRESSURE	COMPLICATION
86	156	51			Liver soft and friable. p. 14.
58	172	69			Acute Pancreatitis.
77	172	55		130	Liver Enlarged.
51	180	58	100°		Liver Enlarged.
82	180	52		110	Chronic Pancreatitis.
88	208	45			Chronic Pancreatitis.
70	242	66			Chronic Appendicitis.

DISCUSSION of TABLE V.

The range is from 156 to 242 mgms per 100 cc.; and the average is 187·14 mgms per 100 cc.

2 cases gave values above the normal; in one, case 70, some arterio sclerosis was present.

None gave a low Cholesterol value.

The average here is higher than in any of the other groups so far, but this is due to the high figure in the patient with arterio-sclerosis.

TABLE/

TABLE V I.

BLOOD CHOLESTEROL

of

6 CASES of CHOLELITHIASIS with JAUNDICE.

NO.	BLOOD CHOLESTEROL	AGE	DEGREE of JAUNDICE	TEMP.	BLOOD PRESSURE	COMPLICATION
49	178	45	+++	100°		
33	187	61	+			
35	200	60	+			
43	210	28	+			
66	255	63	+++	99°		Malignant Pancreas.
72	263	47	++		120	

Average is 215·5 mgms. per 100 cc.

DISCUSSION of TABLE VI.

The range is from 178 to 263 mgms per 100cc and the average is 215·5 mgms per 100 cc. No definite correspondence between the depth of Jaundice and the hypercholesterinaemia was apparent, but the only case in which Jaundice was very intense, and the Blood Cholesterol not high was Case 49, and this patient was very acutely ill.

Average for 40 Cases of Cholelithiasis is - 186·95 mgms per100 cc

Average for 34 Cases of Cholelithiasis without Jaundice is - 181·91 mgms per 100 cc.

TABLE/

TABLE VII.

BLOOD CHOLESTEROL

of

9 CASES of CHOLECYSTITIS.

NO.	BLOOD CHOLES-TEROL	AGE	DEGREE of JAUN-DICE.	TEMP.	BLOOD PRES-SURE	COMPLICATION
68	197	57	+			
74	179	47				
38	162	48				
46	159	57	+		180	
67	148	45				
87	166	61				Chronic Pan-creatitis
83	180	52			120	Chronic Pan-creatitis.
37	175	45			150	Chronic Pan-creatitis.
84	188	33			120	Adenoma of Gall-bladder.

Average is 172·67 mgms per 100 cc

DISCUSSION of TABLE VII.

The range is from 159 to 197 mgms per 100 cc.

The Average is 172·67 mgms. per 100 cc.

Excluding 2 cases with Jaundice, the average is 171·14 mgms. per 100 cc.

All the cases show Blood Cholesterol values within normal limits.

The range is much less extensive than in any other group, but whether this is significant, or merely a coincidence, it is impossible to say.

TABLE/

TABLE VIII.

BLOOD CHOLESTEROL

of

8 CASES of OTHER CONDITIONS of the GALLBLADDER & LIVER

NO.	BLOOD CHOLES-TEROL	AGE	DEGREE of JAUN-DICE	TEMP	BLOOD PRES-SURE	DIAGNOSIS.
57	242	59			160	Cirrhosis of the Liver.
71	216	58	+++			Cirrhosis of the Liver.
36	265	43	+++	100°	120	Carcinoma of the Liver.
80	203	56	+++			Carcinoma of the Liver.
91	146	63	+			Carcinoma of the Liver.
90	147	52				Carcinoma of the Gall-bladder.
73	150	47			120	Carcinoma of the Liver.
79	170	65				Carcinoma of the Gall-bladder.
92	189	50				Liver Cyst & Ascites.

Average is 192 mgms per 100 cc

DISCUSSION/

DISCUSSION of TABLE VIII.

The range is from 146 to 265 mgms. per 100 cc.

The Average is 192 mgms. per 100 cc.

4 of these cases were Jaundiced, and excluding them the average is 179·6 mgms. per 100 cc., i.e. very similar to that obtained in cases of Cholelithiasis without Jaundice.

The range here is greater than in the other groups, which is natural considering the various diseases under consideration and their varying effect upon the patient's general condition.

6 Cases of Cholelithiasis with Jaundice averaged 215·5 mgms per 100 cc.

6 Cases of Jaundice without Cholelithiasis averaged 197·67 mgms per 100 cc.

In these 2 groups the highest figures obtained were similar (263 and 265), but the second group included 2 cases with lower values than were found in any of the first group.

No conclusion can be drawn from these facts.

TABLE/

TABLE IX.

BLOOD CHOLESTEROL in 11 CASES of CARCINOMA

	REGION INVOLVED.	BLOOD CHOLESTEROL.	
1	Breast	189	
2	Tongue	165	
3	"	163	
4	"	159	
5	"	167	
6	Stomach	171	Average is 160.2 mgms. per 100 cc.
7	"	168	
8	Pylorus	113	
9	Liver	150	
10	"	170	
11	Gall-bladder	147	

DISCUSSION of TABLE IX.

The average for 11 Cases is 160·2 mgms per 100 cc.

Very divergent opinions have been expressed concerning the blood Cholesterol values in cases of carcinoma.

Table IX. gives the values found in 11 cases; those in which any complicating factor (e.g. Jaundice) was present were omitted.

The blood cholesterol was normal except in one case, in which it was very low. The patient's general condition was poor, and might explain this low value.

It/

It has been stated that the blood cholesterol is high in cases of early malignant disease, falling later as anaemia and cachexia develop.

TABLE IX. does not bear out this statement, as some of the cases were quite early, others very advanced.

TABLE/

TABLE X.

COMPARISON

of

AVERAGE BLOOD CHOLESTEROL of DIFFERENT GROUPS.
excluding all CASES with JAUNDICE.

NO. of CASES	TYPE of CASE	RANGE of BLOOD CHOLESTEROL	AVERAGE BLOOD CHOLESTEROL in mgms. per 100 cc.
16	Normals	147 - 202	167.20
20	Various (non-abdominal) Conditions	124 - 210	164.85
21	Abdominal Cases (excluding liver and gall-bladder)	113 - 222	170.38
27	Cholelithiasis	127 - 230	180.56
7	" + other Abdominal Conditions	156 - 242	187.14
3	Cholecystitis	148 - 179	163.00
4	" + other Abdominal Conditions.	166 - 188	177.25
5	Diseases of the Liver and Gall-bladder.	147 - 242	179.60
11	Malignant Disease.	113 - 189	160.20
34	Cholelithiasis.	127 - 242	181.91
33	Other Abdominal Conditions.	113 - 242.	171.94

Cases of Cholelithiasis show an Average Blood Cholesterol value that is 8.79% above the normal average.

Cases of other Abdominal Conditions show an average Blood Cholesterol value that is 2.83% above the normal average.

OCCURRENCE. of HYPERCHOLESTERINAEMIA.

Out of 103 Cases (all patients with Jaundice being excluded) 15 or 14.56% had Hypercholesterinaemia (over 200 mgms. per 100 cc). The average rise was 17.7 mgms. per 100 cc.

Of the 15 cases of Hypercholesterinaemia -
8 or 53% were cases of Cholelithiasis.;
The Average rise was 20 mgms. per 100. cc.
7 or 47% occurred in a variety of other conditions;
The average rise was 15 mgms. per 100 cc.

Out of 34 Cases of Cholelithiasis -
8 or 23.5% had Hypercholesterinaemia;
the average rise was 20 mgms. per 100cc.

Out of 33 Cases of Other Abdominal Conditions -
3 or 9.1% had Hypercholesterinaemia;
The average rise was 28.3 mgms. per 100 cc.
A factor which has not so far been referred to is the influence of menstruation upon the blood Cholesterol. DR. SHISKIN (quoted by MOYNIHAN (52)) has shown that a considerable premenstrual rise takes place, and that during the period a gradual fall back to the normal figure occurs.

In/

In this series, the following facts are available in regard to the 15 cases of hypercholesterinaemia:

5 were males.

2 were females past the menopause.

2 blood Cholesterol estimated midway between periods.

2 blood Cholesterol estimated on first day of period.

4 Time of last menstrual period unrecorded.

It is obvious therefore that whatever influence this pre-menstrual rise might have upon the figures obtained in this series of cases would be in the direction of confirming the general conclusions and minimising the importance and frequency of hypercholesterinaemia.

DISCUSSION.

It will be seen from a consideration of the comparisons given above that Hypercholesterinaemia is of more frequent occurrence in Cholelithiasis than in other abdominal conditions without jaundice.

But when all the conditions examined are taken/

taken into consideration the occurrence of Hypercholesterinaemia is found to be almost as common in other conditions as it is in Cholelithiasis.

Further, the average degree of hypercholesterinaemia in all groups is a slight one, and a careful consideration of TABLE IX shows that although the groups consisting of cases of Cholelithiasis show a slightly higher average blood Cholesterol than other groups, the difference is not marked, and the average falls well within the normal limits.

It is noteworthy that the blood Cholesterol was high in 2 cases of cirrhosis of the liver - one without jaundice had 242 mgms. per 100 cc, and the other, with jaundice, 216 mgms per 100 cc. This is not in agreement with the results found by others (55) in this condition.

An attempt was made with this series of Cases to correlate the pre-operative Blood Cholesterol with the operative findings, but nothing definite emerged. It was decided that an account in each case of the pathological condition was unnecessary, and such operative findings as appear to have a definite bearing on the case will be discussed in connection with the post-operative Blood Cholesterol estimations.

CONCLUSIONS.

1. The BLOOD CHOLESTEROL in cases of Cholelithiasis is normal in the large majority, and is of no value in the diagnosis.

2. A slight degree of Hypercholesterinaemia occurs not infrequently in cases where no apparent reason can be found. It is evident that we have, as yet, very little knowledge of the facts governing the minor variations of Blood Cholesterol.

3. The Blood Cholesterol in cases of malignant disease shows a normal range, in the absence of jaundice.

4. The Blood Cholesterol estimations cannot be considered of value in differentiating between jaundice due to cirrhosis of the liver and that due to obstruction in the common bile duct.

PART IV.

PAGE.

EFFECT upon the BLOOD CHOLESTEROL of VARIOUS OPERATIVE PROCEDURES.

P A R T IV.

PRELIMINARY NOTE.

In the ensuing series of 57 Cases, estimations of the Blood Cholesterol were made before operation, and on the 4th, 8th and 15th days after operation. When any complicating factor, such as jaundice was present, mention of it occurs in the analysis of the table of results for the group in which the case occurred.

To each group is appended a paragraph referring to any point of importance in the operative findings or the patient's condition after operation.

Very little reference has been made in the literature to the influence upon the blood Cholesterol of anaesthetics, and in particular chloroform. MAHLER (21) states that ether causes a rise of Blood Cholesterol, but he gives no details as to how long the rise lasts. MANN (56), on the other hand, states that ether anaesthesia has no effect upon the blood Cholesterol.

Various writers point out that the Blood Cholesterol falls after any operative procedure, but do not refer to the influence of the anaesthetic.

In/

In this series of cases the majority of patients had a small quantity of Chloroform, followed by ether during the remainder of the operation.

In 3 cases scopolamine-morphine anaesthesia was used, and in 2 splanchnic anaesthesia. The behaviour of the Blood Cholesterol was not in any way different from that in the other cases, so no further reference will be made to the difference of anaesthetic.

WILENSKY (57) states that Chloroform does interfere with the power of the liver to deal with cholesterol, causing a retention of this substance in the blood, but he points out that individual reactions to this particular drug are known to vary very greatly, so that we have no means of gauging the effect of the anaesthetic in any given case.

EFFECT/

EFFECT upon the BLOOD CHOLESTEROL of VARIOUS ABDOMINAL OPERATIONS.

CASES were as follows:-

	DIAGNOSIS.	OPERATION.
1.	Chronic Appendicitis	Appendectomy.
2.	Gastric Ulcer-perforated.	Excision of Ulcer.
3.	Chronic Pancreatitis.	Laparotomy.
4.	Duodenal Ulcer.	Pyloroplasty
5.	Gastric Ulcer	Excision of Ulcer.
6.	T.B. Mesenteric Glands	Laparotomy.
7.	Chronic Appendicitis	Appendectomy.
8.	Carcinoma of the Liver.	Laparotomy.
9.	Carcinoma of the Pylorus	Pylorectomy.
10.	Carcinoma of the Liver	Laparotomy.
11.	Chronic Pancreatitis	Laparotomy.

TABLE/

TABLE XI.

BLOOD CHOLESTEROL in mgms. per 100 cc.

	BEFORE OP.	4th DAY	8th DAY	15th DAY	POST. OP. FALL.	FALL AS PER-CENTAGE of PRE-OP. FI-GURE.
1	170	181	154	115	55	32
2	156	133	150	121	35	22
3	218	210	173	189	45	21
4	199	175	-	193	24	12
5	148	136	134	171	14	9
6	156	-	146	178	10	6
7	152	163	144	171	8	5
8	170	165	167	164	6	4
9	113	123	121	123		
10	150	168	180	-		
11	166	197	213	173		

ANALYSIS/

ANALYSIS of TABLE XI.

DURING the 1st WEEK -

5, or 45.4% showed a fall of Blood Cholesterol.

3, or 27.3% " no change (experimental error 8%).

3, or 27.3% showed a rise of Blood Cholesterol.

DURING the 2nd. WEEK

Of those that had fallen -

2 had fallen still further.

2 had regained part of what was lost.

1 had risen above pre-operative figure.

Of those that had not changed,-

2 had risen.

1 had remained unchanged.

Of those that had risen -

1 had remained the same.

1 had fallen to pre-operative figure.

1 was unrecorded.

The details of those that showed a Post-Operative rise are as follows:-

Case 9	10 mgms.	9% of pre-operative figure.
" 10	30 "	20% of pre-operative figure.
" 11	47 "	28% of pre-operative figure.

Average/

Average Change in Blood Cholesterol was a fall of 10 mgms per 100 cc.

DISCUSSION.

It has been stated that the Blood Cholesterol falls after any operative procedure, but the group analysed above presents various anomalies. It is interesting to note that the greatest fall - 45 mgms - and the greatest rise -47 mgms - occurred in patients with the same condition, i.e. Chronic Pancreatitis, and the operative procedure was the same, i.e. nothing done.

Those cases in which there was a rise of blood Cholesterol will be dealt with more fully later.

The 2 cases which showed the greatest percentage fall - Cases 1 and 2 had some sepsis in the wound, and a nocturnal rise of temperature for some time. In all the others, convalescence was uninterrupted.

EFFECT/

EFFECT upon the BLOOD CHOLESTEROL. of CHOLECYSTECTOMY without DRAINAGE.

This group consists of 16 Cases, but as 4 had considerable drainage of bile from the wound they are dealt with separately. (TABLE XII A).

TABLE XII.

BLOOD CHOLESTEROL in mgms. per 100 cc.

	BE-FORE OP.	4th DAY	8th DAY	15th DAY	POST-OP. FALL	FALL as PERCENTAGE of PRE-OP. FIGURE.
57	242	216	160	200	82	34
35	200	146	150	152	54	27
47	222	213	177	201	45	20
82	180	153	148	165	32	18
54	154	148	129	131	25	16
41	184	183	161	173	23	12
75	166	158	147	-	19	11
85	147	148	132	-	15	10
84	188	169	187	189	19	10
48	171	171	162	177	9	5
37	175	169	171	174	6	4
31	127	130	125	127	2	2

Analysis/

ANALYSIS of TABLE XII.

DURING the 1st WEEK -

9, or 75%, showed a fall of Blood Cholesterol.

3, or 25% showed no change.

DURING the 2nd WEEK -

Of those that had fallen -

3 remained in statuquo.

4 had regained part of what was lost.

2 were unrecorded.

Of those that had not changed -

all remained in statuquo.

Average change in Blood Cholesterol was a fall of 27.58 mgms. per 100 cc.

OPERATIVE FINDINGS and PROGRESS NOTES.

CASE 57 developed cellulitis of the left arm at the end of the 1st week, and this septic condition may have accounted to some extent for the very marked fall in Blood Cholesterol.

CASE 75 developed a swinging temperature with frequent vomiting 10 days after operation. The Blood Cholesterol was not examined after the 8th day.

CASE 51 (TABLE XII A) had a temperature of 100° on admission and had been ill for 4 days. Temperature became normal on the 4th day and convalescence was uninterrupted, except for the free discharge of bile from the wound.

DISCUSSION/

DISCUSSION.

The results obtained in this group are more uniform than those in the previous one, and the average fall greater.

It is interesting to note that those cases with a Blood Cholesterol above 180 mgms. per 100 cc. showed a greater percentage fall than the others:

6 Cases with 180 mgms. or more showed percentage fall averaging 20.7.

6 Cases with less than 180 mgms. showed percentage fall averaging 8.

Further, the cases with higher values showed a prompter fall than the others - 5 had fallen definitely by the 4th day; whereas of the 6 lower values, 3 did not fall at all, and the other 3 not till the end of the 1st week.

Only one case (Case 35) was jaundiced, and that in a very slight degree, so that the fall of Blood Cholesterol was not due in any of these cases to relief of obstruction in the common bile duct.

TABLE XIIa.

BLOOD CHOLESTEROL in mgms. per 100 cc.

	BEFORE OP	4th DAY	8th DAY	15th DAY	POST OP FALL	FALL as PERCENTAGE of PRE-OP FIGURE.
44	199	151	124	154	75	38
51	180	155	141	171	39	22
42	166	175	135	161	31	19
68	197	171	173	-	26	13

Analysis/

ANALYSIS of TABLE XIIa.

This table shows the findings in 4 Cases of Cholecystectomy, where there was considerable drainage of bile from the wound. The results are similar to those in TABLE XII, and here again the higher Blood Cholesterol values fell more promptly and, on the average, further than the lower ones. The average change in Blood Cholesterol was a fall of 42.75 mgms per 100 cc.

EFFECT upon the BLOOD CHOLESTEROL of CHOLECYSTECTOMY WITH CYSTIC DUCT DRAINAGE.

Group III consists of 4 CASES.
\+ = moderate drainage.
++ = free drainage.
+++ = very copious drainage.

TABLE XIII.

BLOOD CHOLESTEROL in mgms per 100 cc.

	BE-FORE OP	4th DAY	8th DAY	15th DAY	22nd DAY	DRAIN-AGE	POST-OP FALL	FALL as %age of PRE-OP FIGURE.
33	187	154	108	106	135	+++	81	43
65	230	188	193	225	206	++	42	18
39	166	166	137	135	-	+	31	19
40	142	159	169	187	148	++	-	-

ANALYSIS/

ANALYSIS of TABLE XIII.

DURING the 1st week:

3 or 75% showed a fall of Blood Cholesterol

1 or 25% " " rise " " "

(rise of 45 mgms or 32%)

DURING the 2nd week;- of the 3 that fell -

2 remained in statu quo

1 rose again to normal

the one that rose - continued to rise.

DURING the 3rd week:- of the 3 that fell,

1 rose a little

1 was unrecorded

1 fell a little again.

the one that rose-fell back again.

Average Change in Blood Cholesterol was a fall of 27.5 mgms per 100 cc.

OPERATIVE FINDINGS and PROGRESS NOTES.

CASE 33.- had a slight degree of jaundice prior to operation. Temperature became normal on the 7th day. Bile drainage continued for a long time.

CASE 65.- was midway between menstrual periods, so that there was no known factor to account for the hypercholesterinaemia.

CASE/

CASE 39:- had had very frequent attacks of colic and had a chronic empyema of the Gall-bladder.

CASE 40:- had a perforated gall-bladder on admission Temperature was 99.2°. There was copious bile drainage from the wound.

DISCUSSION.

The results in Group III are similar to those in Group II. The rise of Blood Cholesterol shown by Case 40 is quite adequately accounted for by his condition on admission and the improvement subsequent to operation. The fall which occurred in this case in the 3rd week may have been due to the very persistent bile leakage which was present. The marked fall in Case 33 was to be expected, as the patient was jaundiced before the operation, and bile drainage was copious and long-continued.

EFFECT/

EFFECT upon the BLOOD CHOLESTEROL of CHOLECYSTECTOMY with COMMON BILE DUCT DRAINAGE.

Group IV. consists of 3 Cases.

TABLE XIV.

	BE-FORE OP.	4th DAY	8th DAY	15th DAY	DRAINAGE	POST. OP. FALL	FALL as %age of PRE-OP. FIGURE.
77	172	165	143	169	++	29	17
81	162	164	157	155	+	7	4
43	210	239	220	207	+++		

ANALYSIS of TABLE XIV.

DURING the 1st WEEK. -

1, or 33.3%, showed a fall of Blood Cholesterol.
1, or 33.3% " no change
1, or 33.3% " a rise of Blood Cholesterol.
(rise of 29 mgms. or 14%)

DURING/

DURING the 2nd WEEK -

1 that had fallen, rose to normal.

1 that had not changed, remained in statu quo.

1 that had risen, fell to normal

Average Change was a fall of 2.3 mgms per 100 cc.

OPERATIVE FINDINGS and PROGRESS NOTES.

CASES 77 and 81 - nothing to note.

CASE 43 - this patient was just recovering from an acute attack of gallstone colic. Temperature was normal; there was slight tinge of jaundice, which disappeared after the operation. Drainage was very copious. Monthly period started the day before operation.

DISCUSSION.

The case which showed a rise is of special interest because all the circumstances pointed to the probability of a post-operative fall - relief of jaundice, copious drainage, relationship to menstruation, and high post operative blood Cholesterol. No explanation/

explanation of this anomalous result is forthcoming.

The other cases need no particular comment, except that CASE 81 is illustrative of a point already referred to, namely that lower Blood Cholesterol values tend to change less after operation than the higher ones.

EFFECT/

EFFECT upon the BLOOD CHOLESTEROL of CHOLECYSTOSTOMY.

Group V consists of 19 CASES.

TABLE XV.

Blood Cholesterol in mgms.p.100 cc.

	BE-FORE OP.	4th DAY	8th DAY	15th DAY	22nd DAY	DRAIN-AGE	POST-OP. FALL	FALL as %age of PRE-OP. FIGURE.
34	192	134	174	159	-	+	58	30
69	224	158	168	178	-	+	66	29
52	218	170	157	174	184	+	61	28
60	184	174	134	-	-	+	50	27
38	162	128	131	160	-	++	34	21
70	242	208	203	194	-	+++	48	20
74	179	171	145	184	159	+++	34	19
36	214	222	220	183	219	+	31	14
87	166	180	143	163	-	+	23	14
64	178	157	169	163	183	++	21	12
53	170	209	162	149	-	++	21	12
46	159	182	140	154	-	++	19	12
78	184	199	167	172	-	++	17	9
67	148	159	137	138	151	++	11	7
55	176	179	171	224	-	+	+ 48	+ 27
56	158	179	177	195	-	+	+ 37	+ 23
32	139	139	138	133	167	+++	+ 28	+ 20
49	178	210	171	185	214	++	+ 32	+ 18
59	158	179	152	-	-	+	+ 21	+ 13

ANALYSIS/

ANALYSIS of TABLE XV.

In this group the rise or fall was delayed in some cases, but the net result was as follows:-

13 or 68.42%	showed	a fall of Blood Cholesterol.
1 or 5.26%	"	no change.
5 or 26.32%	"	a rise of Blood Cholesterol.

Of the 13 that fell, 5 showed a definite preliminary rise. The majority of cases showed a tendency to return to the normal level during the second and third weeks.

The average change was a fall of 17.26 mgms. per 100 cc.

OPERATIVE FINDINGS and PROGRESS NOTES.

CASE 38 - had slight rise of temperature till the 8th day.

CASE 74 - developed acute rheumatism on the 10th day.

CASE 36 - was very deeply jaundiced and this faded slowly, accounting for the delayed fall in Blood Cholesterol.

CASE 46 - had a slight degree of jaundice which was still present 4 days after the operation.

CASE 56 - had a temp. of 100° on admission.

CASE 49 - was very deeply jaundiced and continued so until the 3rd week.

DISCUSSION/

DISCUSSION.

Although the majority of cases in this group showed a fall of Blood Cholesterol, it is difficult to explain why 5 should show a definite rise. In one (CASE 56) it may have been a return to the normal level, but in the others no explanation is forthcoming.

Also, 5 of the cases which ultimately fell showed a preliminary rise, marked in 3 cases, slight in 2.

EFFECT/

EFFECT upon the BLOOD CHOLESTEROL of OTHER OPERATIONS on the GALL-BLADDER and DUCTS.

Group VI. consists of 4 CASES, and the operations were as follows:-

CASE 62 - CHOLECYSTOTOMY with Common Bile Duct Drainage.

" 72 - CHOLECYSTOSTOMY with Common Bile Duct Drainage.

" 83 - CHOLECYSTGASTROSTOMY.

" 30 - CHOLECYSTENTEROSTOMY

TABLE XVI.

Blood Cholesterol in mgms. per 100 cc.

	BEFORE OP.	4th DAY	8th DAY	15th DAY	DRAINAGE	POST-OP. FALL	FALL as %age of PRE-OP FIGURE.
62	188	187	153	142	++	46	24
72	263	262	204	156	++	107	41
83	180	168	164	167	-	16	9
30	136	110	138	138	-	26	19

ANALYSIS/

ANALYSIS of TABLE XVI.

DURING the 1st WEEK -

all 4 cases showed a fall of Blood Cholesterol.

DURING the 2nd WEEK -

1 case rose again.

1 case remained in statu quo.

2 cases fell still further.

Av. change in Blood Cholesterol was a fall of 48.75 mgms per 100 cc.

OPERATIVE FINDINGS and PROGRESS NOTES.

CASE 72 - was deeply jaundiced, and this lessened as Blood Cholesterol fell.

CASE 30 - was slightly jaundiced until the 2nd week.

DISCUSSION.

The very great fall of Blood Cholesterol in CASE 72, was to be expected because of the pre-operative jaundice and its gradual disappearance after operation.

The other cases show no special features.

DISCUSSION/

DISCUSSION of 10 CASES in which the BLOOD CHOLESTEROL ROSE after OPERATION.

1. CASE 5. The pre-operative figure was normal; during the 1st week the Blood Cholesterol rose 47 mgms., and fell again during the 2nd week to approximately the original figure. No explanation is forthcoming.
2. CASE 9. The pre-operative figure was normal; during the 1st week it rose 30 mgms. In this case and the preceding one the operation consisted of a laparotomy only. No explanation is forthcoming.
3. CASE 11. The pre-operative figure was very low, and the rise of 23 mgms. per 100 cc. was probably the beginning of a return to a normal level as the patient's general condition improved.
4. CASE 40. The pre-operative figure was low, and the patient was acutely ill, with a perforated gall-bladder. The rise of 45 mgms was probably a return to normal.
5. CASE 43. The pre-operative figure was high. The patient was just recovering from an acute attack with fever, so that it is possible that the normal figure may have been still higher.

On/

On the other hand, the patient was slightly jaundiced, she was (according to SHISKIN) at the height of the monthly variation in Blood Cholesterol, and drainage was very copious, so that a fall was to be expected. Nevertheless there was a rise of 29 mgms. No explanation is forthcoming.

6. CASE 55. The pre-operative figure was normal. Drainage was moderate and ceased after 5 days. The rise of 48 mgms took place in the 2nd week.

7. CASE 56. The pre-operative figure was normal, but the patient was just recovering from an acute attack and had a temperature of 100°. It is possible therefore that the rise of 37 mgms may have been a return to normal. Drainage was scanty.

8. CASE 32. The pre-operative figure was low. Drainage was copious, and the rise of 28 mgms. occurred in the 3rd week, following the removal of the drainage tube.

9. CASE 49. The pre-operative figure was normal. The patient was deeply jaundiced and acutely ill. It was natural therefore that the Blood Cholesterol should vary - it rose 32 mgms., fell 39 mgms., and then rose 43 mgms.

10/

10. CASE 59. The pre-operative figure was normal. The patient was very ill, with a gangrenous gall-bladder, therefore the rise of 21 mgms. can be explained by the patient's improved general condition.

Classified according to the probable cause of the rise in Blood Cholesterol, these cases are as follows:-

a) Improved General Condition - CASES, 11,40,56,59.
b) Continued Jaundice - CASE 49.
c) Cessation of Drainage - CASES 55, 32.
d) No explanation - CASES 5, 9, 43.

The explanation regarding insufficient drainage is unsatisfactory, because in other cases in which drainage has been scanty and has ceased soon after operation, there has been no corresponding rise in Blood Cholesterol.

Therefore in only 5, i.e. 50%, is there a satisfactory explanation for the anomalous result.

WILENSKY & ROTHSCHILD (29) suggest that when a post-operative rise in Blood Cholesterol takes place it may be due to swelling and occlusion of the common bile duct due to trauma. It is possible that this may be the true explanation, even in/

in cases in which other evidence of obstruction, i.e. jaundice, is not present.

It is difficult to account for the rise occurring in 2 Cases where only a laparotomy was performed; and also for the preliminary rise which occurred in several cases that ultimately fell.

It is well known that the effect upon the liver of chloroform anaesthesia varies very greatly in different individuals and it may well be that these cases were particularly susceptible to this drug - at anyrate in regard to the metabolism of cholesterol. They did not, however, show other evidence of chloroform poisoning in any marked degree.

DISCUSSION/

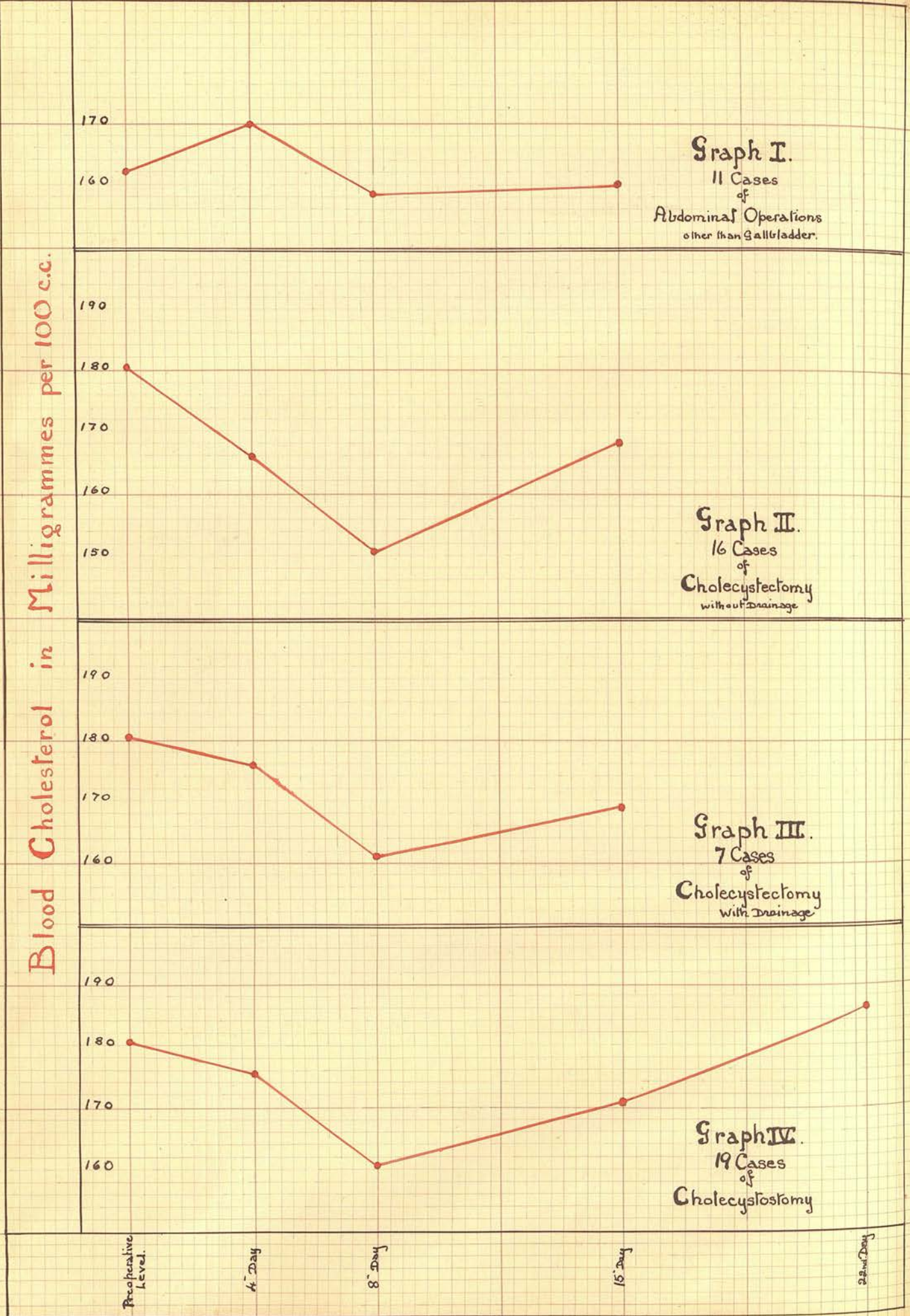
DISCUSSION of 8 CASES in which the BLOOD CHOLESTEROL did not alter after OPERATION.

A careful examination of these cases reveals nothing of note, except that the pre-operative Blood Cholesterol value was normal in 6 Cases and low in 2.

The behaviour of different groups of Cases varies very greatly in this respect, and the highest percentage of Cases showing no change is that in the group of "Abdominal Operations, other than Gall-bladder".

One must presume that in these Cases the ordinary effect of any operative procedure in causing a fall of Blood Cholesterol has been counteracted by some unknown factor, perhaps the action of the anaesthetic.

DISCUSSION/


Blood Cholesterol in Milligrammes per 100 c.c.
170
160
Graph I.
11 Cases
of
Abdominal Operations
other than Gallbladder.
190
180
170
160
150
Graph II.
16 Cases
of
Cholecystectomy
without Drainage
190
180
170
160
Graph III.
7 Cases
of
Cholecystectomy
with Drainage
190
180
170
160
Graph IV.
19 Cases
of
Cholecystostomy
Preoperative Level.
4th Day
8th Day
15th Day
22nd Day

DISCUSSION of GRAPHS.

GRAPH III. consists of 4 cases of Cholecystectomy with Cystic Duct Drainage and 3 with Common Bile Duct Drainage, as these groups were too small to be taken separately.

The group of Abdominal Operations (other than Gall-bladder) showed much less consistent post-operative changes in the Blood Cholesterol than any of the other groups. GRAPH I. therefore shows only variations which fall within the limit of experimental error.

GRAPHS, II, III, and IV. show a striking similarity to one another - in fact the averages for each day's estimation gave almost identical figures for the two Groups, III and IV.

GRAPH II differs only in that the fall in Blood Cholesterol is greater.

It is worthy of note that in all four Graphs the lowest point reached is very similar, the Blood Cholesterol values being between 150 and 160 mgms. per 100 cc.

The factor which produces this remarkable uniformity will be referred to later.

COMPARISON/

TABLE XVII.

COMPARISON of the EFFECT upon the BLOOD CHOLESTEROL of VARIOUS OPERATIVE PROCEDURE.

	NO of CASES.	PERCENTAGE SHOWING FALL	PERCENTAGE SHOWING NO CHANGE	PERCENTAGE SHOWING RISE	AVERAGE CHANGE in MGMS. PER 100 cc. FALL OF:
1. Abdominal Cases, other Than Gall-gladder.	11	45.4	27.3	27.3	10.0
2. Cholecystectomy without Drainage	16	81.25	18.75	0.0	31.37
3. " with Cystic Duct Drainage	4	75.0	0.0	25.0	27.5
4. " with Common Bile Duct Drainage	3	33.3	33.3	33.3	2.3
5. CHOLECYSTOSTOMY	19	68.42	5.26	26.32	17.26
6. CHOLECYSTOTOMY with Common Bile Duct Drainage	1	100.0	0.0	0.0	46.0
7. CHOLECYSTOSTOMY " " " " "	1	100.0	0.0	0.0	107.0
8. Cholecystgastrostomy	1	100.0	0.0	0.0	16.0
9. Cholecystenterostomy	1	100.0	0.0	0.0	26.0
All Cases Combined	57	68.43	14.03	17.54	21.94

ANALYSIS/

ANALYSIS of TABLE XVII.

In considering the comparative results shown in this table, GROUPS, 3 and 4 are too small to be taken separately, and will therefore be classed together.

The 4 types of operation in which only 1 case occurs are omitted from the comparison. The chief points which emerge from a study of this table are:-

FIRSTLY that in cases of "Cholecystectomy without Drainage" a greater percentage than in any other group showed a fall in Blood Cholesterol after operation, and that no case in this group showed a rise.

SECONDLY that in all the other groups the percentage showing a rise was similar i.e. between 25 and 30%.

THIRDLY that the group of "Abdominal Cases other than Gall-bladder" showed the least percentage of Cases showing a fall and the greatest percentage of Cases showing no change.

DISCUSSION/

DISCUSSION of TABLE XVII.

Reference has already been made to the possibility that the pre-operative Blood Cholesterol level is of importance in determining the effect of the operation.

The following figures are of interest:- Average Blood Cholesterol for different groups was as follows:-

11	Cases	of	"Abdominal Operations other than Gall-bladder	163.45
16	"	"	Cholecystectomy without Drainage	181.12
7	"	"	Cholecystectomy with Drainage	181.30
19	"	"	Cholecystostomy	180.50

This leads us to a further comparison of the pre-operative Blood Cholesterol levels and the alteration effected in them by operative procedures.

COMPARISON/

I. COMPARISON of PRE-OPERATIVE BLOOD CHOLESTEROL with the EFFECT of OPERATION OUT of 57 CASES:

A. 24 had a BLOOD CHOLESTEROL of 180 mgms or more per 100cc.

Of these 5 were jaundiced and showed an average fall of 48.8 mgms per 100 cc, or 22.2% of preoperative figure.

19 were not jaundiced and showed an average fall of 42.8 mgms per 100 cc, or 20.74% of preoperative figure.

B. 33 had a BLOOD CHOLESTEROL of less that 180 mgms per 100 cc.

Of these 9 showed a rise of Blood Cholesterol after operation:

of these 6 had a normal pre-operative figure.

" 3 had a low " ".

16 showed a fall of Blood Cholesterol after operation.

None was jaundiced and the average fall was 20.45 mgms per 100 cc. or 12.54% of the pre-operative figure.

Of the 10 Cases in the series that showed a post-operative rise of Blood Cholesterol, only one had a Blood Cholesterol value above 180 mgms, and that one is CASE 43, with a Blood Cholesterol of 210 mgms, of which details have already been given. It is in all respects the most anomalous case in the whole series.

II/

II COMPARISON of the TIME at which the BLOOD CHOLESTEROL fell after OPERATION.

A. 23 CASES with a Blood Cholesterol of 180 mgms or more per 100 cc. showed a post-operative fall of Blood Cholesterol.

Of these 15 or 65.21% showed a definite fall by the 4th day.

B. 16 Cases with a Blood Cholesterol of less than 180 mgms. per 100 cc. showed a post operative fall of Blood Cholesterol.

Of these 4 or 25% showed a definite fall by the 4th day.

III/

III. COMPARISON of the EFFECT of OPERATION upon the CHOLESTEROL in CASES grouped according to BLOOD CHOLESTEROL LEVEL.

TABLE XVIII.

BLOOD CHOLESTEROL in MGMS per 100 cc.	NO. of CASES.	NO THAT FELL	NO THAT SHOWED NO CHANGE	NO THAT ROSE
120-140	4	1	1	2
140-160	11	4	3	4
160-180	16	11	3	2
180-200	13	12	1	0
200-220	3	3	0	0
220-240	5	5	0	0

ANALYSIS of TABLE XVIII.

Cases with jaundice were omitted from TABLE XVIII. As the Blood Cholesterol rises there is a steady increase in the percentage of cases showing a post-operative fall of Blood Cholesterol.

A consideration of the comparisons given above suggests very strongly that the behaviour of the/

the Cholesterol content of the blood after any operation depends more upon the post-operative level than upon the operation performed, or any other fact.

Thus for the higher values the percentage fall was 20.74 of the pre-operative figure, whereas in the lower values it was only 12.54.

Again, of the higher values 65.21% showed a fall by the 4th day, but of the lower values only 25% had fallen by that time.

TABLE XVIII. shows how much greater the tendency is in the lower values either to rise or to remain unchanged after operation.

This apparent importance of the pre-operative level could only be confirmed by a long series of post-operative results in cases that did not involve any operation upon the gall-bladder, and in which many cases had a "high normal" or definitely high Blood Cholesterol.

IV. COMPARISON of CASES in which the GALLBLADDER was removed with THOSE in which it was left.

46 Cases of Various Gall-bladder Conditions.

1. Gall-bladder removed.23 CASES.

   73.9% showed a post-operative fall of Blood Cholesterol.

   8.7% showed a post-operative rise of Blood Cholesterol.

2. Gall-bladder not removed.23 CASES.

   73.9% showed a post-operative fall of Blood Cholesterol.

   21.9% showed a post-operative rise of Blood Cholesterol

It is most striking that the percentage of cases in these two groups showing a post-operative fall of Blood Cholesterol should be exactly the same. This point will be discussed more fully later, and these results compared with the work of other investigators.

V. COMPARISON of CASES in which there was DRAINAGE with those in which there was NO DRAINAGE.

46 CASES of Various Gall-bladder Conditions.

1. DRAINED - 28 CASES.

64.3% showed a fall of Blood Cholesterol.

25% showed " rise " " "

2. NOT DRAINED - 18 CASES.

83.3% showed a fall of Blood Cholesterol

0.0% " " rise " " ".

The result of this comparison tends rather to minimise the importance of drainage in reducing the Cholesterol content of the Blood. At the same time it must be pointed out that the main factor in producing the lower percentage in the Cases Drained is the existence of 5 Cases where a Cholecystostomy was done and the Blood Cholesterol rose after the operation. WILENSKY & ROTHSCHILD (29) point out that if drainage is being instituted with a view to counteracting some degree of hypercholesterinaemia as well as to remove infection, then cholecystostomy is quite inadequate.

Taking this comparison alone, without referring to the previous one, it would appear that removal/

removal of the gall-bladder has more effect upon the Blood Cholesterol than drainage however thorough, but the difference is more apparent than real, as will be seen from a consideration of all the comparisons detailed above.

DISCUSSION/

DISCUSSION of POST OPERATIVE RESULTS.

Until recently, very little interest appears to have been taken in the effect of removal of the Gall-bladder upon the Cholesterol content of the blood, and the majority of investigators have confined themselves mainly to a study of the microscopic anatomy of the Gall-bladder wall in health and Disease.

WILENSKY & ROTHSCHILD (29) state that the effect of cholecystectomy upon the cholesterol metabolism is nil, and that the operation merely removes a focus of infection and the end products of disturbed metabolism. MOYNIHAN (52) also states that the Blood Cholesterol falls after gall-bladder operations, as after any operative procedure, but that it soon returns to its previous level.

Experimental evidence has recently been produced by SWEET in favour of the view that the gall-bladder plays a very active part in the metabolism of Cholesterol. He studied the blood Cholesterol in dogs before and after fat ingestion; he then removed the gall-bladder, and found the following/

following interesting facts, namely, that the rise of Blood Cholesterol following fat ingestion, normally occurring at the 6th hour, was delayed until the 9th hour; and that the Blood Cholesterol rose after the operation, reaching its height about the 20th day, and not returning to normal till 40 days after the operation.

It is of course impossible to attempt to verify the first point in human beings, but this investigation shows that a rise of Blood Cholesterol does not occur after cholecystectomy in human beings. In the great majority of the cases examined the Cholesterol content of the blood fell after the operation; as a rule it started to rise again during the 2nd week. It might be argued that, as SWEET'S dogs did not show the summit of their rise till the end of the 3rd week the comparison is not close enough. Out of 23 Cholecystectomies in this series, 5 had a Blood Cholesterol estimation done at the end of the 3rd week; in 3 that had fallen it was still below the pre-operative level, 1 remained unchanged throughout, and 1 that had risen after the operation had returned to its pre-operative level. The results for 7 Cholecystostomies

at/

at the end of the 3rd week were similar and need not be detailed. SWEET does not give any details of controls, nor does he refer to the anaesthetic used and its possible effect upon Cholesterol metabolism.

BOYD, in his extremely interesting work on the "strawberry gall-bladder (34) found that the gall-bladders of normal dogs contained a lipoid deposit in the wall, similar both in character and distribution to the cholesterol found pathologically in the human gall-bladder. It would appear therefore that in the dog the metabolism of Cholesterol and the part played in it by the gall-bladder differ considerably from these processes in human beings, so that conclusions drawn from experiments on this animal must be guarded.

The same investigator found that in rabbits fed on Cholesterol, much less appeared in the blood after cholecystectomy than had been found before operation, suggesting that absorption had been interfered with by the operation. Rabbits are not accustomed to deal with Cholesterol, so that it is difficult to compare the reactions which they show to experiments with the probable process in human beings.

One/

One point which emerges very definitely from the present investigation is the fact that we are entirely in the dark in regard to the factors which influence minor variations in Blood Cholesterol - much more so than in regard to major variations. This is particularly well brought out by the occasional occurrence of a post-operative rise, where none of the known factors which produce hypercholesterinaemia can be produced to explain the anomaly.

Looking at the post-operative results in cases of Cholecystectomy - a fall of Blood Cholesterol in 73.9% - one is tempted to conclude immediately that a source of absorption has been removed. Exactly the same percentage of cases showing a fall was obtained in cases in which the gall-bladder was not removed, but it might be argued that the considerable traumatism to the gall-bladder might interfere so much with absorption for the time being, that the results were similar to those following Cholecystectomy. On the whole it seems probable that the gall-bladder does absorb some Cholesterol and that this accounts for the very consistent results obtained in cases of Cholecystectomy.

Reference has already been made to the very striking difference in post-operative blood Cholesterol/

Cholesterol levels, apparently depending on whether the pre-operative blood Cholesterol was high or low. A most interesting point in this connection is that of 27 gall-bladder Cases with a blood Cholesterol below 180 mgms per 100 cc., 16 or 59.2% showed a fall after operation; 10 of these were Cholecystectomies and of these only 6 or 60% showed a fall. The percentage showing a fall is therefore considerably less in Cases with a Blood Cholesterol below 180 mgms., than it is for the total number of Cholecystectomies.

Thus we see that not only is the fall much less in extent and slower in appearance in those cases with normal and subnormal values, but that a smaller percentage of cases show a fall compared with those cases that have a higher Blood Cholesterol.

TABLE XVIII gives a clear demonstration of this remarkable correlation between the pre-operative blood Cholesterol level and its variation after operation.

This fact would not seem so striking but rather a process to be expected, if we were dealing mainly with abnormal Blood Cholesterol values. But here we are concerned chiefly with what - for lack of/

of more exact knowledge - we refer to as "normal values". Yet this finely adjusted mechanism for the control of Blood Cholesterol seems to be working all the time towards the attainment of a mean within very much narrower limits than those that we think of as normal. It seems far from likely that the results would have the coherence and order that they exhibit, if, in connection with gall-bladder disease, we are dealing with, and sometimes removing the actual organ concerned in this adjustment.

One recalls here the phrase used by JOELSON & SHORR (22), when they suggest that some secretion from the suprarenals may control Cholesterol "as insulin controls the blood sugar". The work of disentangling the cholesterol problem is made unusually difficult by the fact that considerable variations in its metabolism, as disclosed to us by the alterations in the quantity circulating in the blood, can undoubtedly take place without producing characteristic signs or symptoms. WILENSKY & ROTHSCHILD (29) in discussing the various conditions which cause a recurrence of symptoms after operations on the gall-bladder and ducts lay great emphasis on the occurrence of a certain group of individuals in whom the diathetic factor is really the/

the main one, and in whom a recurrence of symptoms is due entirely to hypercholesterinaemia.

WILENSKY (58) has published full details of one case in which he believes this to have been not merely the most important but the only factor in produing symptoms similar to these produced by Cholecystitis and Cholelithiasis. The patient in question had at one time suffered from trigeminal neuralgia, and the author makes some very interesting speculations as to the possibility of this having been due to the hypercholesterinaemic condition. This point, however, is likely to remain in the region of speculation for some time to come. At present one is forced to conclude that greater variations in the quantity of cholesterol in the blood can occur without producing symptoms than is possible with any other substance which has been fully investigated.

It is far from easy to compute the value of efficient bile drainage in producing a normal Blood Cholesterol when the pre-operative level has been high. Although a large proportion of the cases in this series were not drained, there is no case in/

in which one can say that the effect of the operation would have been better if drainage had been instituted. No light therefore has been thrown upon the question as to whether the height of the Blood Cholesterol affords any indication as to the best operative treatment.

In concluding the discussion of the results of this investigation, I must refer once more to the part of this work concerned with the question of the diagnostic value of Blood Cholesterol estimations in Cases of Cholelithiasis. The average, though well within normal limits, was found to be distinctly above that occurring in a group of other (unselected) diseases, and also above that in a group of other Abdominal Conditions. It must however be pointed out in this connection that a patient with, for example, a gastric or duodenal ulcer, is frequently in a much poorer state of health than the individual who has recurrent attacks of gallstone colic, but enjoys reasonably good health in the intervals. This factor alone would tend to lower the average Blood Cholesterol in any group of Abdominal Conditions (other than gall-bladder disease); and if one takes into consideration the possibility that adiposity <u>per se</u> may conduce/

conduce to a hypercholesterinaemia, then we have two metabolic factors working in opposite directions which would tend to produce this difference in the average Blood Cholesterol level for the two contrasted groups.

The, point, however, which is of practical importance is whether or not in any individual case the Blood Cholesterol content is of any value in diagnosis, and the answer is most emphatically that it is not. Two cases illustrate this point very well - one, an unmarried woman of 38 had a Blood Cholesterol of 127 mgms. per 100 cc., and was found at operation to have a healthy gall-bladder full of stones; the other, a married woman of 28, had a Blood Cholesterol of 222 mgms., and the only pathological condition found at operation was a slight degree of Chronic Pancreatitis. In both cases the Cholesterol level was outside the normal limits, but for this no reason could be discovered in either case.

Because the importance of hypercholesterinaemia in the etiology of gallstones is now well-recognised, a knowledge of the Blood Cholesterol in individuals with gallstones will always be a subject of great interest, and possibly also (as WILENSKY believes) of great importance to the surgeon.

CONCLUSIONS/

C O N C L U S I O N S

1. The Blood Cholesterol in Cholelithiasis is normal in the great majority of cases, and is of no value in the diagnosis.

2. In the majority of Cases of operations upon the Gall-bladder, the Blood Cholesterol falls during the 1st and 2nd weeks, and gradually returns towards the pre-operative level.

3. When the Gall-bladder has been removed the behaviour of the Blood Cholesterol differs very little from Cases in which the Gall-bladder was left.

4. Other Abdominal Operations cause, as a rule, a fall of Blood Cholesterol, but the results are much more variable.

5. These facts are not inconsistent with the view that the Gall-bladder to some extent absorbs Cholesterol.

6./

6. The Blood Cholesterol after any operation shows a marked tendency to vary in the direction of a definite mean.

7. It is evident that the limits of the Blood Cholesterol level laid down as normal are purely empirical and the factors which govern minor variations of this substance in the blood are quite unknown.

In conclusion I wish to express my indebtedness to Professor Fraser for suggesting this investigation; to the Surgeons and Assistant Surgeons of the Royal Infirmary for access to their Wards and Case Records; and to Professor Murray Lyon for permission to carry out the estimations in the Biochemical Laboratory.

REFERENCES.

1. BLOOR W.R. Jour. Biol.Chem. 1916 XXIV 227.

2. AUTENREITH & FUNK München. med. Wochschr. 1913 IX 1243.

3. MAYER & SCHAFFER Jour. de Phys. et de Path. 1913 XV 696, and 1914 XVI 930.

4. FLINT AUSTIN Jour.Amer.Med Sci. Oct. 1862.

5. DORÉE C. & GARDNER J.A. Proc. Roy. Soc. 81B 109.

6. FRASER M.T. & GARDNER J.A. Ibid 81B 230.

7. ELLIS G.W. & GARDNER J.A. Ibid. 84B 461.

8. GARDNER J.A. & FOX F.W. Ibid. 92B 358.

9. GARDNER J.A. & FOX F.W. Ibid. 98B 76.

10 ARTOM C. Arch.Int.Physiol. 1923 XXII, 17, 32, 173.

11 GARDNER J.A. & FOX F.W. Proc. Roy. Soc. 93B 486.

12 MOYNIHAN SIR B. Brit. Jour. Surg. 1922 X 127.

13 SPERRY W.M. Jour. Biol. Chem. 1927 lxxi No 2 351

14 NAUNYN Klinik der Cholelithiasis, Leipzig, 1892

15 SCHÄFFER E.A. Text Book of Physiology, 198B.

16/

16. JANKAU — Cholelithiasis trans. by A.E. GARROD, New. Syd. Soc. 1896.

17. McADAM W. & SHISKIN C. — Quart. Jour. Med. 1922-23 XVI 193.

18. CAMPBELL J.M.H. — Ibid. 1924-25 XVIII 393.

19. GRIGAUT A. — Le Cycle de la Cholesterinémie - Thèse de Paris - 1913.

20. MCNEE J.W. — Quart. Jour. Med. 1914 vii 221.

21. MAHLER A. — Jour. Biol. Chem. 1926 lxix 653.

22. JOELSON & SHORR — Arch. Int. Med. 1924 xxiv 841.

23. ROTHSCHILD M.A. — Zeiglers Beitrage 1914-15 lx 39 and 60.

24. LEATHES J.B. — Lancet. 1925 i 803, 854, 957, and 1019.

25. ADAM N.K. — Proc. Roy. Soc. 1921. xcix 336; 1922 ci, 452 and 516, and 1923 ciii 676. A

26. BLOOR W.R. — Jour. Biol. Chem. 1921. xlix

27. CHAUFFARD, LA ROCHE and GRIGAUT — L'Obstétrique, Paris, 1911 iv 481.

28. ROUS, McMASTER & DRURY — Jour. Exp. Med. xxxix 77.

29. WILENSKY A.O. & ROTHSCHILD M. — Amer. Jour. Med. Sci. 1918 clvi. 239, 404 and 564; 1924 clxviii,66

30. MUELLER J.W. — Jour. Biol. Chem. 1916 xxvii 463.

31. ROUS P. & McMASTER P.D. — Jour. Exp. Med. 1921 xxiv 47.

32. ASCHOFF L. — Munchen. Med. Wochschr. liii 2. 1906. 1847

33/

33. POLICARD A. — Comptes Rend. de Soc. de Biol lxxvi 1914 338.

34. BOYD WILLIAM — Brit. Jour. Surg. 1923 X 337.

35. HARER, HARGIS & VAN METER — Surg. Gyn. and Obst. 1922 xxxiv 307.

36. SWEET J.E. — Int. Clinics Vol i 34th Series 1924.

37. CHAUFFARD A. — La Lithiase Biliaire - Paris 1914.

38. RANDLES & KNUDSON — Jour. Biol. Chem. 1925 xlvii 459.

39. BLOOR & KNUDSON — Jour. Biol. Chem. 1916 xvii and xxix. 7.

40. ROUZAUD & CABANIS — Presse Méd. Mars. 1913. 197

41. McADAM W. and SHISKIN C. — Brit. Jour. Surg. 1925 xii 436.

42. BLOOR W.R. — Jour. Biol; Chem 1921 xlix 201.

43. ROLLESTON SIR H. — Lancet 1925 i 1207.

44. WILKIE D.P.D. — Brit. Med Jour. 1924 ii 1199

45. MAYO — Ann. Surg. 1925 lxxxii 955.

46. ASCHOFF L. — München. med. Woch. 1913. 60

47. MANN F.C. — Jour. Amer. Med. Ass. 1924 lxxxiii 829.

48. REIMANN & MEIGON — Surg. Gyn. and Obst. 1918 282.

49. SCHNABEL T.G. — Amer. Jour. Med. Sci. 1920 clx 423.

50/

50. MYERS V.C. Jour. Lab. and Clin. Med. 1920 V 776.

51. DENIS Jour. Biol. Chem. 1917 xxix 93.

52. MOYNIHAN SIR B. Brit. Med. Jour. 1925 i, 393.

53. BELL J.R. Ibid. 1924 i, 35.

54. GRIFFITHS H.E. Lancet 1924 ii,203

55. WILENSKY A.O. & ROTHSCHILD M. Med. Jour. and Rec. New York 1924 119.

56. MANN F.C. Jour. Amer. Med. Ass. 1916 lxxii 172.

57. WILENSKY A.O. Ann. Surg. 1923 lxxviii 594.