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CONTENTS.

SAWDUST AND FISH LIFE.

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(Read 14th February, 1903.)

CONTENTS.

NOTE.—The trees mentioned in the following report are: White Pine (Pinus Strobus, L.), Red Pine (Pinus Resinosus, Ait.), British Columbia Cedar (Thuya Gigantea, Nuttall), Ontario Cedar (Thuya Occidentalis, Linn.), Hemlock (Thuya Canadensis, Carr.), Maple (Acer Saccharinum, Wang.), Elm (Ulmus Americana Linn.), Ash (Fraxinus Sambucifolia, Lam.), Oak (Quercus Rubra, Linn). Spruce (Picea Alba, Linn).

PART I.—HISTORICAL.

THE following is a continuation of my preliminary report upon the effects of polluted waters on fish life. The work was first begun at the Dominion Biological Station, St. Andrews, N.B., in 1900, and has been continued since then at the biological laboratory of Queen's University, Kingston, and along the saw dust beds of the Bonnechere River in the county of Renfrew, Ontario.

The investigation was begun at the suggestion of Professor Prince, the fish commissioner for the Dominion of Canada, and has been carried on largely through the encouragement which he has given from season to season.

The question, "Is sawdust injurious to fish life?" has been before the Canadian public for over forty years. The *Fishery Act* of 1858 for the two Canadas provided that fish ways should be erected upon dams that obstructed the passage of anadromous fish to their spawning grounds in the shallow head waters of rivers; and it forbade also throwing lime, chemicals, and other poisonous material into such rivers. It did not mention sawdust or mill rubbish, but it provided for the making of regulations by the executive, and in the exercise of this power we find that on May 16th, 1860, a by-law was passed making it illegal to throw "slabs, edgings, and mill rubbish into any river or stream which may have been leased or reserved by the Crown for propagation, or where fish ways have been erected."

This by-law was embodied in the amended Act of 1865, the clause relating to sawdust reading as follows:—

"Lime, chemical substances, or drugs, poisonous matter (liquid or solid), dead or decaying fish, or any other deleterious substance shall not be thrown into, or allowed to pass into, be left, or remain in any water frequented by any of the kinds of fish mentioned in this Act, and sawdust and mill rubbish shall not be drifted or thrown into any stream frequented by salmon, trout, pickerel, or bass under a penalty not exceeding a hundred dollars."

Immediately after confederation the Act was further amended, and a very important proviso was attached to the foregoing clause, viz.:—

"Provided always that the Minister shall have power to exempt from the operation of this sub-section, wholly, or from any portion of the same, any stream or streams in which he considers that its enforcement is not requisite for the public interests."

Evidently the promoters of this legislation either did not feel sure that sawdust was poisonous, or they thought it just, in the interests of the lumber industry, to exempt from the operations of the Act certain large rivers in the maritime provinces, Quebec and Ontario. Exemptions were continued by the minister from year to year down to 1894, when they ceased by Act of Parliament. Parliament itself, however, extended these exemptions down to 1899

In 1873 an Act was passed making it illegal to throw mill refuse into navigable rivers, on the ground that in some parts of the Dominion rivers once navigable had ceased to be so on account of the accumulation of mill rubbish. The Otonabee River in Ontario, and the La Have in N.S., were two rivers which were obstructed in this way.

Most of the Eastern United States have legislated against throwing sawdust into streams containing protected fish; but so far as I have been able to discover, the promoters of the legislation have never been able to prove conclusively the poisonous action of sawdust. At any rate, the scientists of the United States Fish Commission have not been unanimous in their opinions regarding the matter.

For example, in the Fish Commissioner's report for 1872-3, part i., "Inquiry into the Decrease of Food Fishes," Mr. Milner, one of the investigators, says (page 49): "In a number of rivers entering into Green Bay, the white fish was formerly taken in abundance in the spawning season. Saw mills are numerous on all these streams at the present day, and the great quantity of sawdust in the streams is offensive to the fish, and has caused them to abandon them. In one or two rivers of the north shore (Michigan) they are still found in autumn."

In this same report another scientist, Mr. Atkins, referring to the Penobscot River, says (page 303): "The extensive deposits (of sawdust) have in some instances so altered the configuration of the bottom as to interfere with the success of certain *fishing stations;* but beyond that I see no evidence that the discharge of the mill refuse into the river has had any injurious effect on the salmon. It does not appear to deter them from ascending, and being thrown in below all the spawning grounds it cannot affect the latter."

In the Fish Commissioner's report for 1872-3 and 1873-4, vol. I., we meet with another confident statement, but no proof. Mr. Watson, in an article on "The Salmon of Lake Champlain and its Tributaries" (page 536), says: "The sawdust stained and polluted the water, and the sediment and debris of the mills settled largely on the gravelly bottoms, which had been so alluring to the salmon, changed their character, and revolted the cleanly habits of the fish."

Four years after this the Commissioner inserts in his report (1878) a translation of an article by Professor Rasch, of Norway, on "The Propagation of Food Fishes": "That the rivers on which there is considerable cutting of timber gradually become more and more destitute of salmon is an undeniable fact; but while it is asserted that the sawdust introduced into the river from the saw mills causes the salmon coming from the sea either to forsake the foster stream because of meeting the sawdust, to seek another river not polluted, or else when the fish attempts to pass through the areas quite filled with sawdust then this by fixing itself in the gill openings, or between the gills causes its death, yet later experience seems to entitle us to the assumption that sawdust neither causes the salmon to forsake its native stream, nor produces any great mortality among the ascending fishes. The hurtfulness of the sawdust to the reproduction of the salmon is not so direct, but is exceedingly great in this, that it partly limits and partly destroys the spawning grounds of the river."

In his report for 1879, the Commissioner gives a translation from another Norse writer, W. Landmark, on "The Propagation of Food Fishes." This scientist mentions four objections to sawdust:—

1. "Sawdust gradually sinks to the bottom, and thus fills the very place where the fish eggs are to develop, with impure and injurious matter."

2. "When eggs are brought into contact with sawdust or any other rotting wooden matter for any length of time, the eggs are overgrown with a species of fungus, which invariably kills the germ."

3. "When the water rises and causes the masses of sawdust which have gathered in the river to move, a large number of young fish are carried away with it, and are gradually buried in the newly-formed piles of sawdust." In a foot-note he says: "It has been said that sawdust will drive the salmon entirely away from a river, but I think that

this is very improbable, and could only be possible in cases where a river has been completely filled with it."

4. "The refuse from the saw mills, in many places, interferes with the fisheries."

For the next eight years we find little or nothing in the reports of the United States Fish Commissioner regarding the ill-effects of sawdust. In an appendix to his report for 1887, entitled "Fisheries of the Great Lakes in 1885," we find the following expression of opinion from Hugh M. Smith and Merwin Marie Snell: "The fishermen appear to be considerably hampered in their operations by the presence of great quantities of drift wood and sawdust from the mills. At times this debris covers the lake (Michigan) for miles around, and very seriously interferes with the seining and netting. The most disastrous effects, however, are seen on the fish themselves, especially during the spawning season. Spawning grounds formerly existed in this vicinity, but they have been deserted for some years owing to the deposit of sawdust thereon."

On November 29th, 1888, there was started in *Forest and Stream* a very remarkable correspondence, which lasted nearly a year. The general topic was the effect of sawdust upon trout. The writers lived in Canada, the New England States, and some in the west as far as California. Both sides of the question were presented with great vigor. Most of the correspondents were evidently keen sportsmen and close observers of nature, and the only regret one feels in reading through these letters is that some of the men did not test their observations and conclusions by experimenting with sawdust. The following is a typical letter:—

A CENTURY OF SAWDUST.

Editor FOREST AND STREAM.

I was delighted with the intelligent way in which your correspondent "Piscator" handled the sawdust question in your issue of December 27th. It is a comfort to listen when a well-informed person speaks, but in these days of callow pretension experience is usually elbowed back from the front.

In my opinion the famous Mill Brook, of Plainfield, Mass., which has a record of a century as the finest trout water in the Hampshire hills, supplies those very conditions and corroborative data which "Piscator" declares are essential to determine what pernicious effect the presence of sawdust has upon the denizens of mill streams. Here is a water power which carried no less than thirteen manufactories fifty years ago. These included a tannery, a sawmill and factories for making brush and broom handles, whipstocks and cheese and butter boxes, all of which discharged, more or less, sawdust and shavings into the streams, to say nothing of three satinet factories and a felt hat factory, whose waste must have been deleterious to fish life.

Most of the buildings have since been destroyed by fire or tumbled into pieces by decay, but the old foundation, walls, and dams remain, and untold tons of tanbark and sawdust still cover the beds of the abandoned mill ponds knee deep, all of it in a perfect state of preservation, as I happen to know from wading the stream last summer. Nevertheless, the brook continues fairly stocked with small trout, despite the supplementary fact that it has been unmercifully fished ever since the memorial days of the "Mountain Miller," fifty fingerlings per rod being not unusual now for a days' catch. Besides, at no time within my recollection have there been less than three sawdust-producing mills on this stream at once, so that it may be asserted that its waters have not been normally clear for a century. Where the current is rapid and the water broken by ledges and boulders, the presence of the sawdust is scarcely perceptible, but at mill-tails, and in the basins above the dams, it accumulates in quantity and remains, becoming water soaked and sinking to the bottom.

Obviously, in localities where the entire bottom is imbedded by sawdust, fish can neither spawn nor feed; but it happens that such deposits do not form on their breeding places, nor is the area of their foraging ground appreciably diminished by their presence. Even in the half-emptied and now useless ponds, the current constantly scours out a central channel through the sawdust, leaving the bottom clear and pebbly: so that, in fact, these local beds are of no more detriment to the fish than so many submerged logs. The trout can range far and wide without encountering them at all. Yet, strange to say—that is, it must seem strange to those persons who take it for granted that sawdust kills fish—the most likely places for the larger trout are these self-same pebbly channels in the old ponds, along whose edges, despite a hundred freshets and ice-shoves, the persistent sawdust and tanbark lie in wind-rows so deep that the wader feels as if he were going to sink out of sight whenever he puts his foot into the yielding mass, every movement of which stirs up a broadening efflorescence which spreads for rods away, distributing itself throughout the stream.

From these sawdust beds I can always fish out three or four good trout with a cautious fly, and at certain times the surface is fairly dimpled with breaking fish, which presumably are after larvæ and insects which the sawdust has harboured, though careful investigation might discover other inducements for their congregating there.

In passing I would remark that this Mill Brook is fed by seven lateral brooklets, which tumble into it from the adjacent hillsides at intervals between dams, and are so effectually protected by overgrowth that they must always serve as prolific breeding places, secure from predatory birds and small boys, as well as places of refuge to trout which wish to escape the sawdust of the main stream. I have seen trout streams, especially in the pine barrens of Northern Wisconsin and Michigan, which were by no means as favoured as this Mill Brook, the current being comparatively sluggish, and not so capable of purging itself of sawdust; yet I know of few trout streams in any lumber region where its denizens cannot avoid the sawdust if they will, by withdrawing to the headquarters or lateral tributaries, provided fishways are supplied to enable them to surmount the dams where the accumulations chiefly occur. What I remark as most singular in the Mill Brook is, that the trout gather most where the sawdust is thickest, both on old mill sites and on sites where mills are running now. I take my best trout right from under the flume of a whipstock factory and sawmill, where the refuse is dumped as fast as it forms.

But I recall to mind a still more striking example of the innocuousness of sawdust. There are in Hampshire county, Massachusetts, a series of three large natural reservoirs, varying from half a mile to two miles in length, which for fifty years have abounded in pickerel, perch, eels, and bullheads.

It is said that they originally contained trout, but the water is dark and discolored by the drainage of spruce and cedar swamps. At the outlet of the lowest pond once stood a village called Hallockville, which operated a grist mill, sundry sawmills, and what was then the largest tannery in Massachusetts. It was burned in 1846 and never rebuilt, and the dams and foundation walls are now almost destroyed and buried by a new growth of forest. But the sluice and flood stream below are still clogged with the sawdust and tan bark deposited a half century ago, and the water is black and forbidding, though much broken into swirls and rapids by boulders and ledges. But for the colour of the water, it is a most likely 'ooking place for trout, though it has been tested time and time again without successful results. It has always been maintained, from the date of the building of the tannery, that there were no trout in it. I used to fish it myself when I was a boy. Last summer I took therefrom five small trout with a worm. They had doubtless worked their way up from the Buckland streams below, for they never came through the dam from the pickerel ponds above. Nevertheless, the lower streams are occupied by many sawmills, and carry their proportion of sawdust, that substance which some of your correspondents maintain is fatal to fish life. I leave your readers to draw their inferences, and trust that Mr. Fred. Mather will feel himself sustained by this testimony of the streams. That gentleman is not apt to make mistakes. He is grey with the experience of years, and that is better than guess work.

WASHINGTON, *December 29th.* CHARLES HALLOCK.

In this same year (1889) a very remarkable report on this subject was sent to the Hon. C. H. Tupper, the Minister of Marine and Fisheries, Ottawa, by W. H. Rogers, late Inspector of Fisheries for Nova Scotia. The report did not appear among the State papers, and it was consequently published in Halifax under the title of "*The Suppressed Sawdust Report.*" No one can read this pamphlet without being staggered with the mass of information which is supplied to prove the harmlessness of sawdust, and the marvel is that the Minister did not order a thorough investigation to be made into the whole subject.

Of course, diametrically opposite views were expressed by other fishery officers, in whose judgment, no doubt, the Minister had perfect confidence. For example, Mr. S. Wilmot, the Superintendent of the Dominion Fish Hatcheries, wrote a very vigorous report denouncing the deadly effects of sawdust, and his opinions were certainly entitled to some weight. But there was this marked difference between the reports of the two officers: Mr. Rogers' was bristling with facts and observations based evidently upon first hand knowledge of the subject, whereas Mr. Wilmots' report showed no close acquaintance with it.

Turning again to the reports of the United States Fish Commissioner, we do not find any further reference to sawdust until 1892, when Mr. Hugh M. Smith again reports upon "The fisheries of the Great Lakes." At page 404 he says:—"At first white fish and trout were both abundant. . . . Since 1881 or 1882 they have been comparatively scarce. . . . The gill-net fishermen lay the blame on the small meshed pound-nets. The pound-net fishermen, on the other hand threw the responsibility on the saw mills and the gill-net men. The saw mills, they say, pollute the waters with sawdust and vegetable refuse, and the gill-net men lose a great many nets, which with the fish in them soon decay and become a putrid mass, which contaminates the fishing grounds, and causes the fish to leave for other places."

Comparing this with his report for 1887 it will be seen that Mr. Smith refrains from asserting any ill effects from sawdust, and places the responsibility for such statements upon the fishermen. A similar remark applies to the International Fish Commissioner's report for 1893, *and to the report of Mr. Richard Rathbun in 1899 on the "Fisheries in* the Contiguous Waters of the State of Washington and British Columbia." "Attention," he says, "has been especially called to the Skagit river, on whose banks there are numerous shingle mills, from which a very large amount of refuse is allowed to enter the water. According to the statements of the fishermen in that region this practice has caused a great deal of damage to the spawning grounds of the salmon and has affected the fishery in other ways."

Coming to 1899 we find a very important report from the Dominion Fish Commissioner, Professor Prince, and one from the Deputy Commissioner for the Province of Ontario, Mr. Bastedo. Both reports command attention from the fact that they take opposite sides upon the sawdust question. Professor Prince says: "So far as our present knowledge goes, sawdust pollution, *if it does not affect* the upper waters, the shallow spawning and hatching grounds, appears to do little harm to the adult fish in their passage up from the sea." . . . "There is no case on record of salmon, or shad, or any other healthy adult fish being found choked with sawdust or in any way fatally injured by the floating particles."

Again, in summing up his conclusions upon all forms of pollutions: "In the first place it is evident that circumstances modify the effects of all forms of pollutions, so that waste matters which would be deadly in one river will pass away and prove of little harm in another, where the conditions are different. In the second place it shows how varied are the effects of various waste products under the same conditions upon different species of fish. Salmon will survive unharmed where shad and gasperaux would be *killed off. Further, these notes indicate* how little is actually known of the effects upon fish life of these various pollutions from accurate and thoroughly scientific experiments."

Contrast with this Mr. Bastedo's opinion as published in his *report* for the same year: "There can be nothing more destructive of fish life than the depositing of sawdust in the rivers and lakes. It is said to absolutely kill all vegetation, and it is well known that in waters where there is no vegetation fish life is noticeably absent. Minute crustacea of various kinds feed upon the juices of the plants which are to be found at the bottom. These afford food for the smaller fish, and again these furnish food for others of larger size."

Such was the state of our knowledge in 1900, when at the suggestion of Professor Prince, I undertook some experiments at St. Andrews, N.B., for the purpose of ascertaining whether or not sawdust was injurious to fish life.

PART II.—EXPERIMENTAL.

The results of these experiments were published in the report of the Minister of Marine and Fisheries, Ottawa, in 1901, and went to show that brook trout were not injured by living for two weeks in a water tank largely filled with sawdust, so long as a copious supply of water was allowed to run into and out of the tank. These results were abundantly corroborated this summer (1902) in a series of experiments carried on for several weeks in the biological laboratory of Queen's University, Kingston. Perch, rock bass and black bass fry were all used. In fact, the tests this season were, if anything, more exacting than they were in 1900. The volume of pine and of cedar sawdust used was 20 per cent. of the whole volume of the tank, and both adult fish and black bass fry (these latter only about six weeks old and an inch long) were kept for four or five days in the mixture, without any apparent injury.

When, however, sawdust was allowed to lie in still water, or in very slowly running water, entirely different results were obtained. Then, the most disastrous effects followed the immersion of different animals in the poisonous mixture. Not merely did adult fish die in it, but fish eggs, fry, aquatic worms, small arthropods, animalcules and water plants. Nor was the cause of death due to suffocation from lack of oxygen, because when air was made to bubble rapidly through the solution the final results were the same, the only difference being that death was somewhat delayed. No one could paint too vividly the deadly effects of strong solutions of pine or cedar sawdust when soaked in standing water. Adult fish died in two or three minutes; fish eggs in a few hours; fry and minnows in from ten to fifteen minutes; aquatic worms and insects, eight to twenty-four hours; aquatic plants, a few days. Every living thing died in it, and if one were to judge of its effects by laboratory experiments alone, then the prohibitory legislation needs no better defence.

Without anticipating further the results of these experiments, I shall proceed to describe them, so that the reader may be in a position to draw his own conclusions, if he differs from mine.

THE SINKING OF SAWDUST.

As regards the sinking of sawdust, the following experiment was typical of a large number which were carried out, in order to determine how much and how quickly sawdust sank after being thrown into the water at the tail end of a mill.

A litre measure was filled up to 900 c.c. with tap water, and then 100 c.c. of moderately packed pine sawdust was poured upon the water. The moment the sawdust touched the surface, particles began falling to the bottom, and continued to fall for nearly twenty minutes. During this time the water had penetrated 100 c.c. of the floating sawdust, and this volume of it began to sink very slowly *en masse*. Figure 1 represents the conditions in the experiment at the end of the 20 minutes. No less than 70 c.c. of the sawdust lay at the bottom; 100 c.c. were between the 700 and 800 marks, and about 20 c.c. only were floating. The 100 c.c of sawdust at the beginning of the experiment had swollen to nearly 200 c.c. On giving the vessel a slight tap, the 100 c.c. of water-logged sawdust, lying between the 700 and 800 c.c. marks, suddenly upset and most of it sank to the bottom. The large particles, however, rose again to the top, so that in less than three minutes more, only 30 c.c. were floating, and the rest, swollen to 170 c.c., were lying at the bottom.

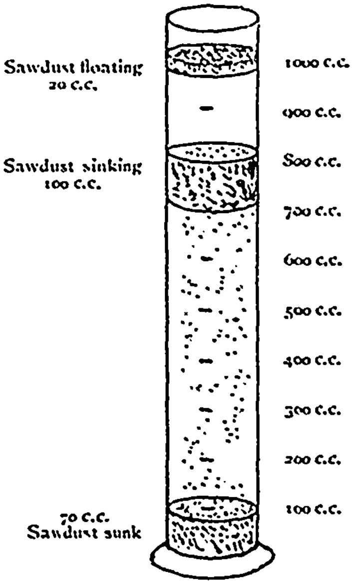


FIG. 1.
Litre measure at end of 20 minutes.

The following conclusions are based upon the results of many similar experiments: From 50 per cent. to 80 per cent. of white pine sawdust sinks in standing water, in from two to three minutes. The variations in quantity and time depend upon, (1) the size of the particles (2) upon the manner in which they are made, (3) upon whether the water is perfectly still or agitated, and (4) upon whether the particles are dry or moist.

Large particles sink much more slowly than small ones, because the latter are more easily penetrated through and through by the water.

Dust made with a hand-saw sinks more slowly than sawdust made with a large mill saw. The difference seems to be due to the difference in the force with which each is made. A large upright or circular lumber saw strikes the log with great force, squeezes out the imprisoned air from the wood fibres, renders them denser, and as a consequence they sink more quickly than particles of a similar or smaller kind which have been made by a hand-saw.

When water is slightly agitated, sawdust thrown upon it sinks more quickly than when the water is perfectly still. Consequently, in the swells of a steamer, in the waves made by wind, and in the ripple of a slight rapids, all the sawdust excepting the largest particles would sink to the bottom in a few minutes.

If thrown into a rapidly flowing stream, sawdust is carried downwards until it reaches comparatively still water, and then the finer particles sink; the coarser may be carried for miles and miles down a river and out into the bays of a lake or sea.

In laboratory experiments the coarser particles would float for days, because the water is unable to penetrate the fibre and displace the imprisoned air, which gives to wood its buoyancy. Wood fibre is, of course, heavier than water, and therefore sinks; and pine logs would sink much more quickly than they do only that the water cannot penetrate their interstices and drive out the air. Yet they do sink in considerable numbers, as every lumberman knows.

Hardwood logs cannot be floated to market at all, because the water of the cell-sap permeates them, rendering them heavier than water and they sink. A very simple experiment illustrates how pine logs sink after being in the water some time. Throw a piece of blackboard crayon into a dish of water. At first it floats, but soon bubbles of air escape from the chalk, and in a few moments it sinks to the bottom. So is it with sawdust and logs.

Sawdust from cedar takes a longer time to sink than that from pine. In fifteen minutes 66 per cent. only had sunk, probably because it contains more resin and consequently water-logs more slowly. Maple sawdust ranged half way between pine and cedar—66 per cent. sinking in eight minutes. Elm sawdust differed from pine, maple, or cedar in that only about 30 per cent. sank in twenty minutes; 75 per cent. of oak sawdust sank in six minutes. So that as far as my experiments went the different kinds ranged as follows: oak sank most quickly, then white pine, maple, cedar, elm. But it must be remembered that the particles in my experiments differed from each other in size and in the moisture they contained, and consequently different results might easily be obtained. The important point is that all kinds sink in a few minutes, especially in agitated water, but not, of course, in a stream with anything like a rapid current.

EXTRACTS FROM SAWDUST.

The first experiments of the season were performed for the purpose of determining the effects of sawdust upon fish eggs. The St. Andrew's experiment had shown that adult trout were not injured by sawdust in rapidly running water; but two other points remained to be determined: (1) Whether sawdust killed fish eggs, and (2) whether it destroyed the food of young, or full grown fish.

Perch eggs were collected along the shallows of Collins Bay, just west of Kingston, and brought to the laboratory on May 12th. They were placed in a clean aquarium with a stream of tap water (from Lake Ontario) running into and out of the vessel. On the same day a bag made of bleached cheese cloth, and filled with a peck of white pine sawdust was placed in an aquarium, 40½in. x 15in. x 16½in. It was weighted with stones to keep it on the bottom. Water entered the aquarium very slowly, so that the conditions of the experiment approximated somewhat to those in the pools of a sluggish stream.

Next morning it was noted that as a result of the bag of sawdust being in the aquarium all night, the water had dissolved out a sufficient amount of material from the sawdust to turn the bottom layer of water a yellowish brown color. This layer measured 1¾in. in a total depth of 16½ inches. Above the yellowish brown layer, and separated from it by a well-defined surface, the water was as clear as that of Lake Ontario. Only about $\frac{4}{5}$ths of the bottom of the aquarium

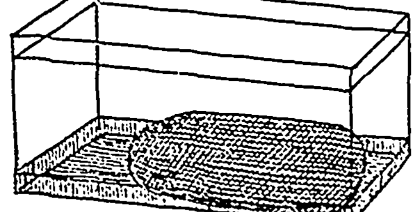


FIG. 2.

was covered by the bag; its upper surface stood about half an inch above the brownish liquid. These conditions are represented in figure 2. Four batches of eggs were placed in the aquarium at 10 a.m. of the 13th of May, viz.: two batches on the very bottom of the aquarium in the brownish water, and two on the surface of the bag of sawdust, well within the clear water.

Next morning at 9.00 a.m. every egg in the yellowish brown water was dead; and every egg in the clear water was alive.

Assuming that the brownish water was a saturated solution of material extracted from sawdust, two other solutions were made from it,—one of 25 per cent., and one of 50 per cent. strength, in tap water. Fresh batches of eggs were placed in each of them. In twenty-four hours the eggs in the 25 per cent. solution were all alive; half of those in the 50 per cent. solution were dead. In twenty-four hours more some of the fry had hatched out, but eggs and fry in both solutions were all dead.

In order to ascertain whether the death of both larvæ and fry was not due to lack of oxygen, rather than to poisonous extracts dissolved from the wood, air was made to bubble rapidly through some of the brown water. This experiment was begun at 12.30 p.m., and 800 c.c. of air per minute were passed through 230 c.c. of the discoloured water. At 5.30 p.m. of the same day, a batch of 60 eggs was placed in this aërated water, and air was passed continuously through it all night at the rate of 400 c.c. per minute. Next morning at 10 a.m. every egg in the batch was dead. The conclusion, therefore, is quite clear. The eggs were killed, not by lack of oxygen in the water, but by the poison contained in the water and evidently dissolved out of the sawdust.

The water had changed during the night to a much darker shade of brown. This marked change in colour will be discussed in a subsequent report.

SOURCE OF POISON.

The source of the poison given off by sawdust is undoubtedly to be found in the contents of the wood cells. Sugar, starch, oil, resin, gum, jelly, alkaloids, and acids are all examples of material stored in different parts of plants.

In the older parts of trees the protoplasm and sap disappear completely from the cells, and they may then contain nothing but the stored material. In the pine family there is stored in the wood and bark cells an abundance of crude turpentine and resin. The Norway spruce of Europe furnishes in this way turpentine and Burgundy pitch. The yellow pine of the Southern United States yields spirits of turpentine by distillation of the crude turpentine which runs away from the tree by cutting into it. The residue after the distillation is resin.

Now the poisonous material in sawdust must be either the cell wall or the stored material. It cannot be the cell wall, for this is just the wood fibre or material used in making paper, and pure paper is certainly not harmful to fish life. The poison can scarcely be anything else than the turpentine and other substances stored in the cells.

Different trees, such as tamarack, pine, cedar, spruce, etc., generate and store different kinds of reserve material. When a log from one of these trees is cut into boards, the sawdust gives off proportionately much more poisonous matter than the slabs, edgings and bark. The reason of this is easily understood. As each cell or vessel is microscopic, and contains only a very small quantity of poison, and as the cell wall must be broken open in order to let out the contents, it follows that the greater the number of cells that are opened, the greater will be the quantity of turpentine, resin, etc., poured out. Hence, a saw log converted into sawdust, or ground into shreds, as in a pulp mill, gives out the maximum of poison; whereas a similar log sawn into boards, edgings and slabs, will give out a much less quantity. The minimum will be given out by a saw log floating in the water.

The total waste in manufacturing saw logs into boards is sometimes stated as equal to the lumber obtained for market; but this is a gross exaggeration. Prominent manufacturers like the Rathbun Co., W. C. Edwards, M.P., and J. R. Booth estimate the waste as varying between 25 per cent. and 35 per cent. of the whole log. The proportion of refuse varies with the size of the logs, with the kind of lumber into which the log is cut, and with the kind of saw used in the mill. The old-fashioned gang saw and the large circular saw produce a higher percentage of waste than the more modern band saw. There is more waste in cutting a log into inch boards than into 3-inch deal, and small logs produce proportionately more waste in bark, slabs and edgings than large logs. The waste in sawdust alone varies from 10 per cent. to 20 per cent.

PULP INDUSTRY.

There are other industries in Canada, which in preparing their products for market grind up plants and trees, and thus let out their cell contents. One of these is the pulp industry—likely to become very extensive in the near future. Two processes are in vogue in this industry. In one, the logs are macerated with chemicals, the mills being known as sulphite mills. In the other process, the logs are ground into shreds in what are known as mechanical mills. Both processes liberate the greatest possible quantity of stored material from the wood cells, and if this material is equally poisonous with that liberated from sawdust, then the waste water discharged from a pulp mill should be much more poisonous than from a sawmill. The St. Andrew's experiments determined the percentage of poison from a sulphite mill which is fatal to fish life, but, so far as I know, the percentage of poison from a mechanical mill has never been determined. A provisional conclusion, however, may be based upon some of the experiments to be described later in this paper.

BEET SUGAR INDUSTRY.

The manufacture of sugar from the maple and from the beet depends upon the fact that sugar is one of the reserve materials stored in the cells of these plants. In order to liberate the sugar from the beet roots they must be thoroughly ground into a mash, so as to rupture the cell walls. The more effectively this is done, the higher is the percentage of sugar obtained from the beet. It is easily conceivable that the water that escapes from beet sugar factories may contain matter that is poisonous to fish life.

Professor Prince called attention to both these sources of pollution in his report for 1899, and they are referred to now merely for the purpose of emphasizing the fact that other industries may pollute the streams of Canada to even a greater extent than lumbering. In all three industries the source of pollution is the contents of the wood or plant cell.

There is a similar action going on in nature all the time. Leaves, branches, and trunks of dead trees are decomposing continuously; their cell contents are being dissolved in rain and melting snow, and are in part carried away in streams and rivers. The only difference is that in this latter case the poisons come away so slowly that air (oxygen), sunlight, and bacteria have ample time in which to change their poisonous character; whereas in the saw mill, the pulp mill, and the beet sugar factory, the poisons are quickly discharged into running water, and tend at once to produce their effects upon fish and other life.

STRENGTH OF SAWDUST EXTRACTS.

As already explained, the first experiments were made with solutions obtained by soaking white pine sawdust for at least twenty-four hours in tap water from Lake Ontario. When the sawdust was soaked for four days in tap water, 1,000 c.c. of the yellowish-brown solution already described as oozing out from the bag of sawdust, and lying at the bottom of the aquarium, yielded 1,240 milligrams of solid matter after evaporation in a platinum crucible. The ash from this weighed 80 m.gs., which was found to be exactly the same as that from tap water. Deducting this from 1,240, leaves 1,160 m.gs. as the weight of the material stored in the pine cells of the sawdust, and dissolved out in 1,000 c.c. of water in four days.

After filtering off the first water, and adding fresh water to the same sawdust, and allowing the mixture to stand five days longer, it was found that 1,000 c.c. of this second solution yielded a total of 360 m.gs. of solid, or allowing for the ash in tap water, a net residue of 260 m.gs. of reserve material was dissolved out the second time.

The corresponding figures for cedar (Ontario) sawdust were as follows:—

1. Solid from 1,000 c.c. soaking four days...............	1,300 m.gs.
2. Same sawdust with first water filtered off, fresh water added and allowed to stand five days	=550 m.gs.
3. Same operations repeated, soaking five days	=350 m.gs.

No allowance is made in these figures for the ash from tap water, viz., 80 m.gs.

These figures indicate clearly enough that the reserve material stored in the wood cells comes away in diminishing quantities every time fresh water is added to the sawdust.

The next point sought to be determined was the number of times that fresh water could be added to a fixed weight of sawdust and continue to produce solutions which would be poisonous to fish life. For the purpose of getting information on this point two series of experiments were carried on, one with cedar sawdust and one with white pine.

EXTRACTS FROM CEDAR (ONTARIO).

On the second of June 400 grams of cedar sawdust were placed in a cheese-cloth bag and sunk to the bottom of a small glass aquarium (12 in. x 8 in. x 6 in.) containing 7,000 c.c. of tap water. Next day there had formed at the bottom, to a depth of two inches, a dark yellowish brown solution. The uppermost four inches were tinged a light yellow by diffusion, but there was a perfectly distinct surface of a greyish colour separating the upper from the underlying dark water. These characters became still more marked during the week, at the end of which time 1,400 c.c. of the lower liquid were siphoned off into a shallow circular dish and a perch weighing seventy grams immersed in the solution. In thirteen minutes it was lying on its back moribund, but revived when returned to fresh water. The control animal was kept twenty-four hours in 1,400 c.c. tap water in a similar vessel and then returned to the aquarium.

A perch weighing twenty-five grams was placed in 400 c.c. of this extract and air bubbled rapidly through it all the time. In twenty-eight minutes it was dead. The control animal in tap water under similar conditions was alive at the end of seventy hours.

Three *Daphniæ* in this extract died within two hours.

Fresh water Hydra died almost instantly in it. Paramœcia were unaffected by either cedar or pine extracts. These scavengers were often observed apparently feeding upon the dead bodies of Hydra that had died in the poisonous extracts.

On the 13th, all the water was poured off and fresh water added. On the 14th, a perch weighing seventy grams was placed in 1,400 c.c. of the solution formed during the preceding twenty-four hours. It was moribund in six minutes. With air bubbling rapidly through some more of this solution, another perch placed in it was moribund in seven minutes.

Pond silk appeared to be unaffected by an immersion of four days in this water.

June 15th. A perch moribund in fourteen minutes in a third solution—all the water being poured off and fresh added.

June 16th. A perch moribund in eight minutes in a fourth solution from the same sawdust.

June 16th. A perch moribund in ten minutes in a fifth solution formed by soaking this same sawdust in fresh water for seven hours.

June 17th. A perch moribund in ten minutes in a sixth solution. With air bubbling through more of this solution, another perch moribund in twenty-one minutes.

June 18th. A perch moribund in twelve minutes in a seventh solution. The water was poured off twice to-day, making an eighth solution,

June 19th. A perch moribund in eight minutes in a ninth solution. At 11 a.m. a pond leech was placed in this solution. It had eighteen young ones, five large and thirteen small, attached to its back. Three of these young at once detached themselves from the mother's back. The adult showed every symptom of discomfort by swimming rapidly round the vessel, then pausing and rolling itself up into a wheel as if to escape the effects of the water. It tried to leave the vessel, but was put back again. Gradually the smaller young detached themselves from the mother until only two or three of the smallest (about ¼ inch in length) remained on her back. The larger young ones were about an inch long, but when extended they were an inch and a half. Finally the smallest dropped off and wriggled about with the others on the bottom. The mother came to rest in about three-quarters of an hour. The young were all dead at 4.30 p.m.; the mother was moribund at 6 p.m., and died during the evening.

Two pond snails lived just twenty-four hours in this solution. The larva of an aquatic insect lived five and a half hours in it.

At 11.30 a.m. three bunches of vorticellæ were placed in the solution. Their cilia at once stopped moving in all the individuals, and they assumed the spherical form. By 5 p.m. most of these animals had dropped from their stalks and lay quite motionless at the bottom of the glass. Apparently they were dead. Returned them to fresh water, but found them all apparently dead next morning.

A bunch of embryos of the pond snail placed in this extract at 5.30 p.m. to-day were all found dead the next morning.

June 20th. Placed a perch weighing seventy grams in 600 c.c. cedar water drawn off for the eleventh time; air bubbling through it very rapidly. Moribund in ten minutes.

Placed in this extract about a dozen worms gathered from the mud in water about three feet deep. These animals were massed together and lying among the roots of aquatic plants. The moment the solution touched them they separated all over the bottom of the watch glass, wriggling in all directions and voiding their faeces. In three hours they were all dead. So were two small phyllapod crustaceans which happened to be along with the worms.

June 24th. Placed a batch of about fifty aquatic worms in cedar extract drawn off for the twelfth time. At first great wriggling ensues with evacuation of faeces; then constrictions occur in each segment of the body, making the animal look somewhat like a string of beads; then the hinder end appears to disintigrate but leaves the front end living and moving; finally the head dies. In two hours most of them were dead, but in a few the head was still alive.

June 25th. At 9.15 a.m. placed a tadpole one inch long in cedar extract drawn off for the twelfth time. Apparentty dead in fifteen minutes, but revived in about an hour when returned to fresh water.

Up to this time all experiments with cedar extract had been conducted with what might be considered as saturated solutions; that is, the solution used was siphoned off from the bottom of the aquarium where the sawdust was lying and where the dark colour showed the extract to be the strongest. From this date the experiment was varied by throwing out all the water, filling up the aquarium with fresh water and allowing the bag of sawdust to float at the top of the water. In this way the solution was uniform in colour and strength throughout the aquarium. The animals were then as a rule placed in the aquarium and usually swam about below the floating sawdust bag.

June 27th. Mr. Halkett, an officer of the Department of Marine and Fisheries, arrived to-day from Belleville, bringing with him about 100 black bass fry. The weight of one of these of medium size was found to be 135 milligrams; its length one inch.

Placed two of these fry in cedar extract drawn off fourteen times. Both appeared to be dead in two minutes. Placed in fresh water they did not revive.

June 28th. Two black bass fry in cedar extract drawn off sixteen times from the same sawdust, died in two hours.

June 30th. Changed all the water on the cedar sawdust to-day no less than five times. Immediately after adding the fresh water black bass fry were placed each time in the aquarium, and in each case the animal was dead in from half an hour to forty minutes.

July 7th. The last experiment with this sawdust was performed to-day. The water was changed this morning at 9 a.m. for the thirty-first time, and immediately afterwards a black bass fry was immersed in it. It swam about below the floating bag which contained the sawdust. The odour of the cedar was scarcely perceptible in the water. The strength of the solution was, of course, increasing all the time. At 11 a.m. the fry was dead.

Some of the water that was drained off from the sawdust at 9 a.m. was found to contain 235 m.gs of solid matter per 1,000 c.c. Allowing for the residue after ignition, there would still remain 155 parts per million of poisonous extract dissolved out of the cedar cells in the thirtieth withdrawal. This is quite remarkable when it is remembered that the sawdust had been soaking continuously for five weeks, and the water on it changed thirty times.

Comparing the solid in this solution with that in a saturated solution already given, viz., 1,240 per 1,000 c.c., we conclude that there has been a continuous withdrawal of poisonous extracts from the cedar. The question, therefore, of whether a river is polluted with sawdust or not, simply becomes a question of determining the quantity of sawdust poured into a known volume and flow of water, and the further question of determining whether the resulting solution is poisonous enough to kill fish eggs, fry, adult fish or fish food.

Warm water was found to extract the poison from wood cells much more quickly than cold water.

EXTRACTS FROM WHITE PINE.

The general effect of pine extracts upon fish eggs has already been described. It only remains to point out some special effects under varying conditions. One of these is that eggs live longer in aërated sawdust water than in unaërated. This is quite clear from the following experiment: At 9.45 a.m. of May 18th, two batches of eggs were placed in pine water at the bottom of the aquarium. At 5.30 p.m. every egg but two was dead.

At 11.15 a.m. of May 17th two batches were placed in pine water through which air was bubbling at the rate of 400 c.c. per minute. At

9.45 a.m. of the 18th, twenty out of one batch of thirty-three were dead; and in the other, thirteen out of seventy-three were dead. At 5.30 p.m. of the same day all of the first batch of thirty-three were dead, and only seven were alive in the other.

The effect of aërating the pine water was made apparent in another way. At 10.25 a.m. of the 18th, 120 eggs were placed in pine water in a shallow dish so that the water was only three-eighths of an inch deep. At 5.20 p.m. only a few were dead; all the rest were very quiet. At 9 a.m. the next morning forty-seven were dead; at 6 p.m. all were dead except five. At 10.30 a.m. of the 20th four of these five had hatched out and were quite lively. This experiment shows that the large surface exposed to the air absorbs oxygen, and therefore tends to prolong the life of both larvæ and fry. In contrast with this it is interesting to note that the same quantity of poisonous water put into a tall jar at 6 p.m. of the 19th had killed every egg in a batch of nineteen by 10.30 a.m. the next morning. In this case, the depth of the water and the small surface exposed to the air prevented the diffusion of the oxygen downwards to the eggs, lying at the bottom of the vessel.

There can be no doubt that fish instantly perceive the poisonous character of pine or cedar extracts. A minnow was placed in the large marble aquarium already described, and being driven to one end of the vessel, it sank through the clear water and into the yellowish brown extract lying at the bottom. The moment his head touched it he started towards the surface. I drove him back several times, and each time he sank into the coloured water he made frantic efforts to escape from it. He refused finally to be driven into it. Immersed in the pine extract in a separate vessel the minnow was moribund in three minutes and could not be resuscitated in fresh water.

A perch placed in 900 c.c. of pine water, in a shallow dish, was moribund in three minutes.

Another perch in pine water with air bubbling rapidly through it lived three and a half hours.

Two limicolous worms died in thirty minutes; two rotifers lived only ten minutes.

One tadpole half an inch long lived two hours. Another tadpole of the same size died in half an hour in a weak solution. A similar animal in strong cedar extract lived only six minutes.

A copepod placed in it at 11 a.m. was alive at 3.30 p.m., but died during the early evening.

Daphnia and the larva of an aquatic insect lived three days.

One hydra immersed at 10.40 a.m. was dead at 5 p.m. Its body was partly disintegrated and many paramœcia appeared to be feeding on it. Another hydra on being placed in the extract contracted its tentacles, detached itself from its support, contracted the lower half of its body, voided the intestinal contents, and appeared to be dead in two hours. It revived in fresh water.

A colony of vorticellæ at first showed no signs of discomfort; the cilia kept on moving, and the stalks contracting spirally. Soon the stalks ceased their movements; a little later the cilia stopped; the animals took the spherical form and within one and a half hours all were apparently dead.

A rock bass weighing seventy grams when placed in 1,300 c.c. of the extract became moribund in twelve minutes. All of the fish revived in from five to twenty minutes when returned to fresh water.

A perch weighing thirty grams when placed in a jar containing 400 c.c. of the pine water with air bubbling rapidly through it was moribund in ten minutes.

June 16th. Up to this time my experiments with this extract had been made with strong solutions. To-day a series of experiments were begun for the purpose of ascertaining, if possible, how long the same sawdust would continue to give off poisonous solutions when the saturated water was drained off and fresh water added from time to time.

With this end in view 360 grams of white pine sawdust were placed in a cheese cloth bag at 9 a.m. and sunk to the bottom of a small glass aquarium, 12 in. x 8 in. x 6 in., containing 7,000 c.c. of tap water. Two hours after, 800 c.c. were siphoned off from the extract at the bottom of the vessel. This was found to be very slightly poisonous to adult fish. Next morning at 10 a.m. 800 c.c. more were siphoned off. A rock bass lived in this one hour and twenty minutes. Another fish lived six hours in it when air was made to bubble rapidly through it. The third and fourth withdrawals of 800 c.c. each were thrown away.

June 21st. The larva of an aquatic insect lived twelve hours in the extract drawn off for the fifth time: vorticellae lived twenty hours, limicolous worms twenty hours, a pond snail seventy hours. The sixth and seventh withdrawals were thrown away.

June 27th. The eighth withdrawal of 800 c.c. made by soaking the pine eighteen hours, killed a perch in three hours, and three black bass fry in half an hour.

June 30th. A slight modification was made in this experiment. In place of siphoning off the strong extract at the bottom of the aquarium, the whole 7,000 c.c. of water were drained off, and the aquarium was filled up with fresh water. The weights were removed from the bag, which at once rose to the top of the water. Consequently the extract coming off from the sawdust, being heavier than the fresh water, fell towards the bottom and became uniformly diffused throughout the vessel. This was the twelfth withdrawal. Black bass fry lived five hours in this water, which was, of course, becoming more poisonous all the time.

July 7th. The last experiment with this sawdust was made to-day. The bag is still floating. The water was changed for the twentieth time at 9 p.m. last evening. At 9 a.m. to-day a black bass fry was immersed in this solution. In two hours it was dead. Some of this solution was evaporated and was found to contain 160 m.gs., or, allowing for the residue after ignition, eighty parts per litre. That is, pine sawdust soaking continuously since June 16th, with the water on it changed twenty times furnished in twelve hours eighty parts per million of poisonous extracts from its wood cells.

Comparing these figures with those for a saturated solution already given, viz., 1,160 parts for 1,000 c.c., we see that there has been a continuous withdrawal of poisonous material from the sawdust. The question, therefore, of determining whether any stream is polluted with pine sawdust or not is largely the question of determining the minimum amount of sawdust extracts which will kill fish eggs, fry, adult fish, and fish food. Needless to say, such determinations would have to be made for every sawmill stream in Canada, and for each separate kind of fish.

OTHER WOOD EXTRACTS.

A number of experiments were made with extracts from other woods besides pine and cedar. Norway, or red pine, British Columbia cedar, maple, hemlock, oak, ash, elm were all used, but it was soon discovered that the most poisonous extracts were obtained from the pines and cedars. Consequently experiments with the hard woods were soon discontinued.

From all hard woods, however, the saturated yellowish-brown extract was found to be very poisonous to both adult fish and fish eggs.

The following experiments give typical results in the case of each of these woods.

MAPLE SAWDUST.

A dark orange liquid oozed out from maple, and lay at the bottom of the aquarium. This was separated from the clear liquid above by a perfectly well-defined greyish surface. At the top of the water (16½ inches deep) intake and outflow pipes allowed tap water to flow into and out of the aquarium at the rate of 600 c.c. per minute. A perch having sunk into this extract once or twice could not afterwards be driven into it. The animal soon found where the fresh water inlet was, and when driven to other parts of the aquarium would always come back to the fresh water.

Aquatic plants in maple extract lost their chlorophyl in three days. Returned to fresh water they regained their colour, but the tips of their leaves had died.

HEMLOCK SAWDUST.

Hemlock has always had a bad reputation, but does not deserve it.

On July 27th, six black bass fry were placed in a mixture of five volumes of water to one volume of hemlock sawdust. The vessel was covered with four layers of cheese cloth, and a copious stream of water was made to fall upon it from a tap about a foot above it. The fry were all alive and well at the end of three days, when they were returned to the aquarium.

As a control experiment, five black bass fry were kept for the same length of time in the same volume of water, viz., 600 c.c., with air bubbling through it all the time. These animals also were quite lively and well at the end of the experiment.

BRITISH COLUMBIA CEDAR SAWDUST.

This sawdust sank rapidly, 75 per cent. falling to the bottom of perfectly still water in two minutes. It gave off a very poisonous extract. Two black bass fry lived only one minute in a solution made by standing five and a-half hours. The colour was a beautiful amber with a strong smell of cedar. A solution made by one gram of sawdust standing in 500 c.c. water for three hours rendered a black bass fry moribund in two hours. A solution from one gram in 750 c.c. water for twenty-seven hours, killed another fry in two and a-half hours. Even as homeopathic a solution as one gram in 1,500 c.c. killed fry in less than eighteen hours.

If much of this sawdust is poured into British Columbia streams, the stockholders of British Columbia Fish Canning Co's will need to look closely into the future prospects of their industry.

NORWAY, OR RED PINE.

Within three minutes, 90 per cent. of the sawdust from this wood had sunk. A strong solution made in eighteen hours rendered a black bass fry moribund in one hour. This water when aërated, but not filtered, rendered another fry moribund in exactly the same time. In both cases the gills of the animals seemed to be affected by fine particles of the wood fibre clinging to the filaments and preventing respiration. This was not observed to be the case with any other kind of sawdust.

A solution made by soaking one gram of this sawdust for nine hours in 250 c.c. of water killed a fry in less than an hour.

Another fry lived fifteen hours in a solution made by soaking one gram in 850 c.c. water for six hours.

OAK.

Contrary to expectations, oak sawdust was not so poisonous as pine and cedar. It communicated an orange colour to the water just as other woods did. A tadpole lived three days in a strong solution, and was quite lively at the end of that time.

ELM.

A few experiments were made with elm sawdust. Here again a dense yellowish-brown layer forms at the bottom of the aquarium. This kills adult fish in from half an hour to two hours. A tadpole lived over an hour in it. When this water was thoroughly aërated a perch lived twenty hours in it, and was then active and apparently well.

EXTRACTS QUICKLY SOLUBLE.

The experiments hitherto described would seem to indicate that some considerable time was required for the water to dissolve out the poisonous extracts from white pine sawdust, but such is certainly not the case. This was clearly shown in the following experiment, Fig. 3. Two minnows were confined in a bottle containing 600 c.c. water and eighteen grams of white pine sawdust. Fresh water was made to enter and leave at the rate of 100 c.c. per minute. The inlet tube passed straight to the bottom of the vessel, and its lower end was therefore buried in about an inch of sawdust. One animal lived forty minutes, the other fifty. When the incoming water was reduced to 80 c.c. per minute three

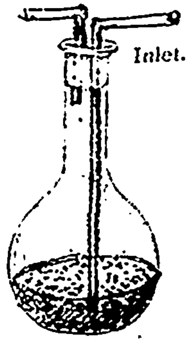


FIG. 3.

minnows lived only from three to five minutes. When the fresh water entered at the rate of 125 c.c. per minute, minnows lived from twenty to ninety minutes. The control animals were kept for a week in a similar bottle, without sawdust, of course, and with water coming in at the rate of 110 c.c. per minute. In these experiments the poisonous extracts must have been coming away all the time. The moment the bottle was full of water the minnows were slipped into it. Consequently, when the fish were killed in five minutes, the 600 c.c. at first in the bottle. and 400 c.c. additional water were poisoned. When they were killed in ninety minutes, no less than 11,250 c.c. were poisoned. That is, the percentage weight of sawdust to poisoned water was .16 per cent. This determination is important, as we shall see later, when we come to compare it with the percentage of sawdust thrown into the Bonnechere River.

FISH AT MILL-ENDS.

Millmen and anglers alike testify that many kinds of fish are taken by hook and line at mill-ends, no matter how excessive the sawdust may be. The sawdust does not kill the fish so long as there is a rapid and abundant flow of water. Why do fish thus congregate at mill-ends? To answer this question we must remember two things : first, rapidly running water is better aërated than sluggish water ; and secondly, some fish, such as trout and salmon, ascend streams until they reach suitable spawning grounds, or are stopped in their ascent by high falls or mill-dams. In ascending a river these fish are but obeying a law of their nature ; in congregating at mill-ends they are equally obeying a law of their nature, and are instinctively seeking water which furnishes their blood with a plentiful supply of oxygen. This instinct is well illustrated in the experiment just described in Fig. 3. The experiment was repeated a number of times, and in every instance the fish discovered where the fresh water came in. In one instance, in order to get close to the incoming water, a minnow stood on its head for fifteen minutes with more than half of its body buried beneath the sawdust. It was thus acting under the impulse of two fundamental instincts, viz., the instinct to avoid poisoned water on the one hand, and to seek fresh water on the other. The experiment seems to throw light upon the experience of anglers who have found that trout desert the main stream when saw mills are running, and betake themselves to the unpolluted branch streams lower down.

A STAGNANT ARTIFICIAL POOL.

Reference has already been made to the fact that black bass fry, minnows and perch, when placed in an aquarium, invariably avoided the poisonous sawdust water at the bottom. Having sunk into it once or twice, it was found almost impossible to drive them into it again. Here was a conflict between two fundamental instincts. On the one hand was the natural instinct to hide in deep water; on the other hand, the equally natural instinct to avoid the poisonous solution at the bottom. Which instinct would the fish obey if compelled to make a choice?

The following experiment was designed for the purpose of seeing which instinct was the more powerful, and for the further purpose of imitating what might possibly occur in a stagnant pool along the course of a sawdust polluted stream.

A glass aquarium 12 in. x 8 in. x 6 in. was placed in a much larger vessel and a mixture of ice and salt packed in the latter so as to surround the aquarium. The aquarium was then half-filled with white pine extract which had been forming for three weeks, and which killed adult fish in from one to three minutes. After the extract had been cooled down to 8° c., tap water at the temperature of 13° c. was slowly admitted to the aquarium so as not to disturb the underlying poisonous water The tap water, being warmer, floated clear and transparent on the dark purplish extract below. The clear water entered and left the aquarium at the rate of 150 c.c. per minute. The arrangement of apparatus is represented in Fig. 4.

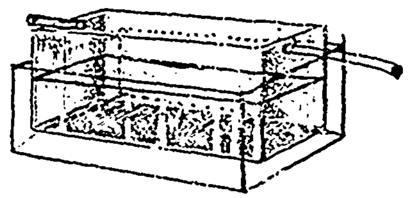


Fig. 4.

At first two minnows were placed in the aquarium. They at once dove to the bottom, encountered the poisonous water, immediately came up again, repeated the operation a few times, and finally remained swimming about in the clear water. Three black bass fry, liberated one after the other, went to the bottom and never came up—suffocated and poisoned in the dark stagnant water at the bottom. Of two other minnows dropped into the aquarium, one large one never came to the surface; the other joined its fellows in the clear water above. All three soon found the end at which the fresh water was entering and remained there facing the stream.

This experiment shows what might possibly happen in pools partially filled with sawdust. Wood extracts would form, and being cooler and heavier than the clear water, would lie at the bottom of the pool. Of course, fish already in the pool would be driven away, but those coming up or down stream through shallow stretches, and trying to hide in the deeper waters of the pool, might be suffocated or poisoned.

COMPARATIVE RESULTS.

After obtaining the general results detailed in the preceding part of this paper, it seemed desirable to plan a series of experiments that would show comparative results at a glance. With this end in view, two grams each of different kinds of sawdust were placed in shallow circular dishes containing respectively, 300, 400, 500, 600, 700, 800, 900, 1,000, 1,200, 1,500, and 1,700 c.c. of fresh water. After soaking for about five hours in each case, a minnow was placed in each of the dishes. The length of time each animal lived was carefully noted, except in those cases where death occurred during the night. The results are given in the following tables:—

WHITE PINE SAWDUST.

Weight of Sawdust.	Volume Water c.c.	Time Soaking.	Time at which minnow was immersed.	Results.
2 grams.	300	From 10 a.m.	2.43 p.m.	Lived about 9 minutes.
"	400	"	"	" "
"	500	"	"	" "
"	600	"	"	" "
"	700	"	"	" "
"	800	"	"	" 10 minutes.
"	900	"	"	" 13 "
"	1000	"	"	" 15 "
"	1200	"	"	" 20 "
"	1500	"	"	" 29 "
"	1700	"	"	" 29 "

ONTARIO RED PINE.

2 grams.	300	10 a.m.	2.47 p.m.	Lived 47 minutes.
"	400	"	"	" 50 "
"	500	"	"	" 50 "
"	600	"	"	" 1 hour and 28 minutes.
"	700	"	"	" 1 " 14 "
"	800	"	"	" 1 " 14 "
"	900	"	"	" 1 " 53 "
"	1000	"	"	" 2 hours and 20 "
"	1200	"	"	" 2 " 50 "
"	1500	"	"	" 3 " 45 "
"	1700	"	"	" 3 " 45 "

ONTARIO CEDAR.

Weight of Sawdust.	Volume Water c.c.	Time Soaking.	Time at which minnow was immersed.	Results.
2 grams.	300	From 10 a.m.	2.33 p.m.	Lived 8 minutes.
"	400	"	"	" 9 "
"	500	"	"	" 19 "
"	600	"	"	" 20 "
"	700	"	"	" 21 "
"	800	"	"	" 22 "
"	900	"	"	" 27 "
"	1000	"	"	" 27 "
"	1200	"	"	" 1 hour.
"	1500	"	"	" 1 " and 48 minutes.
"	1700	"	"	" 1 " " 55 "

BRITISH COLUMBIA CEDAR.

2 grams.	300	10.15 a.m.	2.51 p.m.	Lived 6 minutes.
"	400	"	"	" 6 "
"	500	"	"	" 15 "
"	600	"	"	" 53 "
"	700	"	"	" 43 "
"	800	"	"	" 1 hour and 9 minutes.
"	900	"	"	Jumped out of dish unnoticed.
"	1000	"	"	Lived 1 hour and 32 minutes.
"	1200	"	"	" 1 " 36 "
"	1500	"	"	" 3 " 50 "
"	1700	"	"	" 3 " 29 "

HEMLOCK BARK.

Bark. 2 grams.	300	10.10 a.m.	2.36 p.m.	Lived 55 minutes.
"	400	"	"	" 1 hour and 32 minutes.
"	500	"	"	" 1 " 43 "
"	600	"	"	" 1 " 49 "
"	700	"	"	" 2 hours.
"	800	"	"	" 1 hour and 32 minutes.
"	900	"	"	Jumped out of dish unnoticed.
"	1000	"	"	Lived 2 hours and 18 minutes.
"	1200	"	"	" 3 " 24 "
"	1500	"	"	" 4 "
"	1700	"	"	" 4 " 15 "

HARD MAPLE SAWDUST.

Weight of Sawdust.	Volume Water c.c.	Time Soaking.	Time at which minnow was immersed.	Results.
2 grams.	300	From 10.38 a.m. July 15th.	July 15th. 3.30 p.m.	Lived 2 hours and twenty minutes.
"	400	"	"	July 21st, 10 a.m. Still alive.
"	500	"	"	" 16th. Died last night.
"	600	"	"	" 21st, 10 a.m. Still alive.
"	700	"	"	" 16th. Died last night.
"	800	"	"	" 21st, 10 a.m. Still alive.
"	900	"	"	Lived only 2 hours.
"	1000	"	"	July 18th. Died between 4 p.m. and 8 p.m.
"	1200	"	"	Lived 3 hours and 30 minutes.
"	1500	"	"	July 18th. Died between 4 p.m. and 8 p.m.
"	1700	"	"	" 20th. Died 3 p.m.

This experiment was discontinued July 21st, 10 a.m.

ONTARIO CEDAR BARK.

2 grams.	300	10.20 a.m.	2.41 p.m.	Lived 37 minutes.
"	400	"	"	" 1 hour and 20 minutes.
"	500	"	"	" 50 minutes.
"	600	"	"	" 50 minutes.
"	700	"	"	" 1 hour and 20 minutes.
"	800	"	"	" 1 " 31 "
"	900	"	"	" 1 " 40 "
"	1000	"	"	" 1 " 57 "
"	1200	"	"	" 2 hours 10 "
"	1500	"	"	" 4 "
"	1700	"	"	" 4 " 20 "

ELM SAWDUST.

2 grams.	300	10.44 a.m. July 15th.	3.30 p.m.	Lived 4 hours and 30 minutes.
"	400	"	"	Died 10 a.m. July 16th.
"	500	"	"	Lived 1 hour and 30 minutes.
"	600	"	"	" 2 hours and 30 "
"	700	"	"	" 1 hour and 30 "
"	800	"	"	July 21st, 10 a.m. Still alive.
"	900	"	"	" 18th. Died last night.
"	1000	"	"	" 21st. Died last night.
"	1200	"	"	Lived 1 hour and 30 minutes.
"	1500	"	"	" 4 hours and 30 "
"	1700	"	"	" 1 hour and 30 "

This experiment was discontinued July 21st, 10 a.m.

OAK SAWDUST.

Weight of Sawdust.	Volume Water c.c.	Time Soaking.	Time at which minnow was immersed.	Results.
2 grams.	300	Since 10.15 a.m. of 23rd.	July 23rd. 2.30 p.m.	Lived 2 hours and 30 minutes.
"	400	"	"	" 2 " 30 "
"	500	"	"	" 3 " 30 "
"	600	"	"	" 7 " 30 "
"	700	"	"	" 2 " 20 "
"	800	"	2 animals	{ One lived 2 hours and 20 minutes. July 24th. Died last night.
"	900	"	2 animals	{ One lived 7 hours and 30 minutes. July 24th. Died last night.
"	1000	"	"	July 25th. Jumped out unnoticed.
"	1200	"	"	" 30th, 9 p.m. Still alive. Released.
"	1500	"	"	Lived 3 hours and 30 minutes.
"	1700	"	"	July 25th, 3 p.m. Dead.

ASH SAWDUST.

2 grams.	300	10.48 a.m. of July 15.	3.30 p.m. July 15th.	July 21st, 10 a.m. Still alive.
"	400	"	"	Lived 1 hour and 30 minutes.
"	500	"	"	July 21st, 10 a.m. Still alive.
"	600	"	"	Lived 1 hour and 40 minutes.
"	700	"	"	" 2 hours and 10 "
"	800	"	"	July 21st. Died last night.
"	900	"	"	Lived 1 hour.
"	1000	"	"	July 21st, 10 a.m. Still alive.
"	1200	"	"	" 21st. Died last night.
"	1500	"	"	" 21st, 10 a.m. Still alive.
"	1700	"	"	" 19th. Died to-day.

This experiment was discontinued July 21st, 10 a.m.

HEMLOCK SAWDUST.

2 grams.	300	10.15 a.m. of 23rd.	2.30 p.m. July 23rd.	July 26th, 9.30 a.m. Dead.
"	400	"	"	" " "
"	500	"	"	July 30th, 9 a.m. Released.
"	600	"	"	" " "
"	700	"	"	" " "
"	800	"	"	July 26th. 9.30 a.m. Found dead.
"	900	"	"	Lived 45 minutes.
"	1000	"	"	July 26th, 11 a.m. Dying.
"	1200	"	"	" 28th, 3.00. Dead.
"	1500	"	"	Lived 1 hour and 45 minutes.
"	1700	"	"	July 26th, 9.30 a.m. Dead.

SPRUCE SAWDUST.

Weight of Sawdust.	Volume Water c.c.	Time Soaking.	Time at which minnow was immersed.	Results.
2 grams.	300	10.30 a.m. of 23rd.	2.40 p.m. July 23rd.	Lived 3 hours and 30 minutes.
"	400	"	"	July 24th, 9.30 a.m. Found dead.
"	500	"	"	" " "
"	600	"	"	" 26th, " "
"	700	"	"	" 24th, " "
"	800	"	2 animals	July 24th, 9.00. Dying. " 25th, " Found dead.
"	900	"	"	July 26th, 9.30 a.m. Found dead,
"	1000	"	"	" 30th, 9.00 a.m. Released.
"	1200	"	"	" " " Dying.
"	1500	"	"	" 27th, 7.30 p.m. Dying.
"	1700	"	"	" 26th, 9.30 a.m. Found dead.

The reader will, of course, understand that in all experiments due. allowance must be made for constitutional differences in individual fish. Some men survive the effects of cold, hunger or poisonous drugs longer than others. In the same way some species of fish, and some individuals in each species, are naturally more hardy than others, and can survive in poisoned water a longer time. Some are more delicately organized and are, therefore, more easily killed. For example, black bass fry lived longer than minnows in some of my experiments. Consequently too much importance must not be attached to the exact number of minutes or hours that a fish will live in any given strength of sawdust solution. When we are dealing with vital phenomena, all we can consider is the general average of a number of experiments. Keeping this in view, some conclusions may fairly be drawn from the foregoing results.

1. White pine sawdust is by all odds the most poisonous substance.

2. Next comes Ontario cedar.

3. Then British Columbia cedar.

4. Red pine, cedar bark and hemlock bark are moderately poisonous.

5. Maple, oak, ash, elm, hemlock and spruce may all be grouped together as only slightly poisonous.

EXPERIMENTS WITH BARK.

From the frequent references to the pernicious effects of bark which may be found in the literature of sawdust pollution, one would naturally expect to find that bark solutions were very destructive to fish life and fish food. The very opposite was found to be the case. Compared with the wood extracts, the bark solutions were comparatively harmless. Even tan bark, much execrated by fishermen and anglers alike, was not so poisonous as one might expect, but the experiments must speak for themselves.

WHITE PINE BARK.

Only 11 per cent. of sawdust from this bark sank in ten minutes. A black bass fry seemed perfectly unharmed after being three hours in a solution of this bark that had been forming for twenty-two hours (one gram in seventy-five c.c. tap water). The animal was then returned to the fresh water aquarium.

This same bark, after soaking two weeks, gave a solution that killed solely by suffocation. This was quite apparent from the fact that two minnows when placed in this water (freely aërated) lived for fwenty-four hours and were then liberated. When the solution was unaërated the minnows died in an hour or two. Pouring the solution several times from one vessel to another arëated it sufficiently to enable two minnows to live three days in it without apparent harm.

After standing six weeks a scum formed on the surface. This was removed and the solution aërated by pouring it several times from one vessel to another. A minnow now lived in it for two days and was liberated, apparently as well as ever.

HEMLOCK BARK.

A solution made by soaking one gram of this sawdust bark for fifteen hours in 100 c.c. of tap water killed a minnow in six minutes. After soaking for two weeks this water killed a minnow in one hour, even when thoroughly aërated. But these were very strong solutions compared with the ones obtained from wood.

CEDAR BARK.

Only 5 per cent. sank in fifteen hours. In two days it had all sunk excepting about 1 per cent. A 1 per cent. solution (one gram in 100 c.c.) made in fifteen hours, rendered a minnow moribund in fourteen minutes. Here again the solution was a very strong one compared with those obtained from wood sawdust and used in the experiments previously described.

As bark extracts, therefore, are not more poisonous than those from pine and cedar woods, it seemed useless to conduct separate experiments upon their effects.

DECAYING SAWDUST.

One objection frequently urged against the practice of throwing sawdust into streams and rivers is that the decaying sawdust imparts such a disagreeable odour to the water that sensitive fish are driven away to other waters not so polluted. It seemed to me, therefore, that some progress might be made towards a definite conclusion in this matter, if sawdust were allowed to stand for several weeks in an aquarium and tested from time to time as to the changes going on in it, and the influence of these upon fish.

With this end in view about 1,000 grams of white pine sawdust were placed in an aquarium three feet four inches long, fifteen inches wide, and filled up to sixteen and a half inches deep with fresh water. This was done June 24th. No water was allowed to enter or leave the vessel. No direct sunlight fell upon it.

The usual results followed, viz., a well defined layer of pale, yellow water one and three-quarter inches deep formed in a few hours and lay at the bottom. On top of this was the perfectly clear layer about fifteen inches deep.

After soaking for two days, bubbles of gas began to rise to the surface of the water, but no attempt was made to analyze it. The bottom yellowish layer had become so dense that no object could be seen across it—a thickness of fifteen inches. Its upper surface was sharply marked off from the overlying transparent water by a thin greyish layer. Microscopic examination of this layer showed it to be swarming with bacteria.

At the end of a week, only about an inch at the bottom had retained the original yellow colour; the next inch had changed to a yellowish brown; then came a greyish layer about one-sixteenth of an inch thick; above this, what had at first been fourteen inches of perfectly clear water had turned to a dark grey, though still quite transparent. Black bass fry placed in the aquarium at this time at first sank to the bottom, but after meeting the poisonous extract once or twice could not subsequently be driven into it. On the contrary they swam along the top with their nose just touching the surface of the water, and behaved as if suffering from lack of air. They lived only about two hours.

Four days after this, black bass fry placed in the upper fourteen inches lived only about one hour. They also swam along the surface and appeared to be gasping for air. That they were suffocating in both cases was proved by the fact that when fry were placed in a wash bottle of this water with air bubbling through it, they lived on for twenty-four hours, and were then apparently well and exceedingly active. On being transferred from the wash bottle to the aquarium the animals at first plunged downwards to the bottom, paused there a moment, but soon came towards the surface breathing very rapidly. Evidently they were suffering from lack of oxygen. They swim along the top with noses upwards and body inclined at an angle of about thirty degrees with the surface. Gradually they tire; sink towards the bottom; rise again; swim convulsively towards the surface; jump clear out of the water with gaping mouth; become exhausted by their convulsive efforts and finally sink to rise no more. Of all the fish killed in this extract not one ever rose to the surface after death.

It would be difficult to say whether this experiment throws any light upon a point much discussed in the literature of sawdust. The point is this: if sawdust kills fish, why are they not found dead in considerable numbers along the course of the stream? In my experiments the dead bodies of the fish never rose out of the poisonous liquid.

AROMATIC COMPOUND.

The foregoing experiments show that the oxygen naturally dissolved in the upper fourteen inches of water had, at the end of a week, all disappeared. It was used up either in supporting the life of the bacteria, or in oxidizing the wood extracts through the agency of the bacteria. Bacteria were abundant in every part of the aquarium, but especially in the underlying solution. Moreover, either by their action on the pine extracts, or by the chemical decomposition of these extracts, an aromatic compound of a sweetish pleasant smell had begun to form. At the surface the smell was faint; but in the water siphoned off from the bottom the perfume was strong and agreeable. The production of this compound is possibly due to micro-organisms, and if the special bacterium could only be isolated and used upon the extracts without admixture with other forms, it might be possible to manufacture a perfume from pine which many people would find agreeable. Alcohol, lactic acid, acetic acid, etc., are all formed by the action of bacteria upon vegetable substances in solution; the quality, too, of butter and cheese is determined by the action of bacteria on the constituents of milk; and it would, therefore, be only in accordance with well known facts to find that aromatic compounds, some pleasant, some unpleasant, could be formed from pine extracts by the action of different kinds of micro-organisms.

Some of the bottom water was distilled for the purpose of seeing whether this aromatic compound could be thus separated from the water, but the attempt failed. The distillate had the aromatic odour of the original water, but mixed with it was a disagreeable burnt smell. This distilled water killed minnows in half an hour, both when aërated or unaërated.

At the end of three weeks the uppermost fourteen inches of water had gradually become a steel grey or slaty colour and was quite opaque. The outlines of a window sash ten feet away could not be seen through it. The extract at the bottom still killed by its vegetable poison; the slate coloured water above still killed by suffocation.

At the end of five weeks these conditions were but slightly changed. In place, however, of the pleasant aromatic odour previously arising from the surface, a musty, disagreeable smell had taken its place. As the laboratory windows were always open, mosquito larvæ became numerous and appeared to be feeding upon the bacteria. These larvæ died in sawdust solutions only when prevented from coming to the surface to breathe.

The water at the very bottom was still of a yellowish tinge; the uppermost was smoky or slate coloured, as already explained. About 6,000 c.c. of this slate coloured water was siphoned off from the middle, on July 31st, and placed outside of the laboratory in direct sunlight. The object of this was to compare changes taking place in the slaty water placed in sunlight and breeze, with changes taking place in the slaty water which remained in the aquarium.

Dr. W. T. Connell, Professor of Bacteriology, made cultures from these two waters and compared them on three different occasions. His report which will be found in the appendix to this paper, shows that while the number of colonies from water in the shade increased from 3,435 per cubic centimetre to 7,870 per cubic centimetre; the number of colonies from water in sunshine increased from 3,435 per cubic centimetre to 37,070 per cubic centimetre. These latter were different bacteria from the former. Sunlight and air had killed off those kinds of bacteria which flourish in shade and in absence of oxygen, and had stimulated the growth of other kinds of bacteria which flourish in sunshine and moving water. As a result of sunlight, warmth and breeze, what had been exceedingly disgusting water was changed in a fortnight to water brownish in colour, without any odour, and perfectly transparent. A heavy precipitate lay at the bottom. Minnows were able to live in it, and soon made havoc with the mosquito larvæ. In short the water had, within the fortnight, changed to normal water, while that in the shade still retained all its disagreeable and poisonous characters. The decaying mass of sawdust and water was kept for three months, and up to the very last showed no improvement. Slimy, a dark slate colour, foul smelling, teeming with anaerobic bacteria and mosquito larvæ, it was utterly unfit to support any kind of fish life.

NUTRITIVE RELATIONS.

However, the connection between a few links in the chain of animal life was apparent enough, viz., wood extracts supported bacteria, bacteria supported mosquito larvæ, and these again (after aëration of the water such as would occur in running water) supported fish life. These observations dispose to some extent of the oft repeated charge against sawdust that it destroys the food of young or newly hatched fish. When minnows relished mosquito larvæ as food, and I frequently saw them eating the larvæ, it requires no great stretch of the scientific imagination to understand how fish fry of different kinds, such as trout and salmon, might subsist upon the larvæ of mosquitoes and other aquatic insects, these latter in turn subsisting upon bacteria, and the bacteria subsisting upon the organic matter derived from the decaying vegetation of the forest.

Another thought comes up in connection with the presence of organic matter in streams and rivers. The organic matter which passed into a river when Canada was covered with forest must have been quite different in character from that which this same stream receives to-day from the vegetation of the farms along its valley. The surface drainage from a forest must differ in kind from the surface drainage of a farm, and the bacterial life in each must differ also. Moreover, the waters of our smaller streams were, years ago, shaded by trees, and the varieties of their bacterial life must thus have been quite different from the bacterial life in sunlit streams of to-day. Consequently, it may fairly be argued that the insect life, in and along the streams of an agricultural district, differs both in kind and number from what characterized these same streams 100 or 200 years ago. And if larval and adult insect life has dwindled or disappeared, so must the fish life which subsisted upon it.

The Anglo-Saxon has always been a disturbing factor in the balance of life. Forests, game and fish all disappear with his arrival. To get good fishing or good hunting now-a-days one must travel back to unsettled districts. No one expects game to be plentiful along the shores of Lake Ontario, but many people are amazed that fish are not abundant in it. They still hug the pleasing delusion that if brooks have been overfished, the fish hatchery can restock them. But with the disappearance of our forests it is exceedingly doubtful whether we can ever again, by all the help of hatchery, overseers and fish commissioners, re-people the streams which have been depleted by man through overfishing and deforestation. He has upset the balance of life; it can only be fully restored by a return to primitive conditions. When game, therefore, becomes plentiful on the streets of Ottawa city, fish will be equally abundant below the saw mills of the Chaudiere Falls.

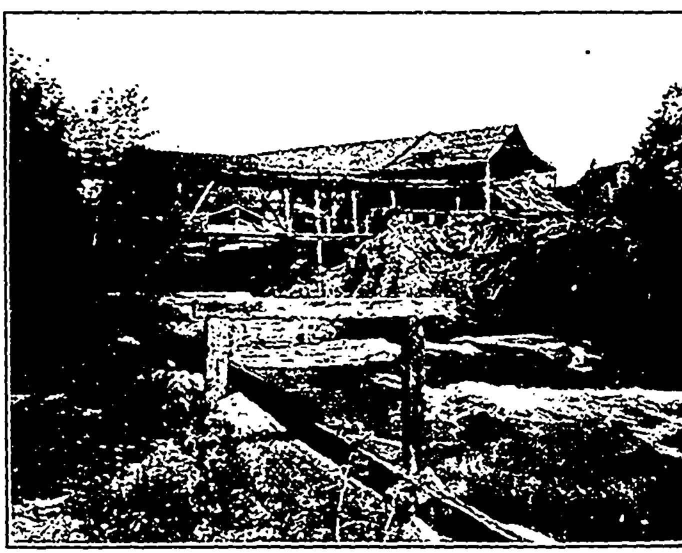
Such, at least, is the conclusion to which my experiments point, notwithstanding the indisputably poisonous effects of strong solutions from sawdust near the source of pollution. As I have already pointed out the question of whether any particular stream is sufficiently polluted with sawdust to kill fish life is simply the question of determining whether enough sawdust is passed into the stream to poison its waters. The forestry engineer will soon be trained to determine the strength of sawdust solutions, and will then be able to settle this question of pollution beyond the possibility of doubt.

ON THE BONNECHERE RIVER.

At present, however, a final judgment cannot be pronounced upon the poisonous effects of sawdust. These effects must be studied near the mills and along the sawdust beds of our rivers. A three weeks' study of the Bonnechere river, a tributary of the Ottawa much polluted with mill rubbish, led me to modify very considerably the conclusions which I had based upon my laboratory experiments. I visited the mill represented in two of the illustrations of this report fully expecting that not one fish could survive in such surroundings. But pike were abundant for miles below the mill, and fish (chub) could be caught any day along the side of the submerged driftwood. Stranger still, the fish so caught lived for three hours in a pailful of sawdust water drawn from the very centre of a sawdust bed. A few brook trout had been caught earlier in the season just below the mill when it was running. At the date of my visit, August 20th, 1902, the mill had been closed for seven weeks and no sawdust was then passing into the river.

The owner of the mill furnished the following information: The water passing over the dam is a stream nineteen and a half feet wide, by one and one-half feet deep, and moving two feet per second. This would mean that about sixty cubic feet of water were passing over the falls every second. Add to this, leakage through the dam, mill, and timber slide, estimated as equal to what passes over the dam, or sixty cubic feet more, a total of 120 cubic feet per second. The total water, therefore, passing down the river in July, August and September, would average 10,368,000 cubic feet per day, and weigh 642,816,000 pounds.

The mill cut an average of 375 logs per day. The logs averaged twelve inches in diameter and were chiefly sixteen feet long, but many

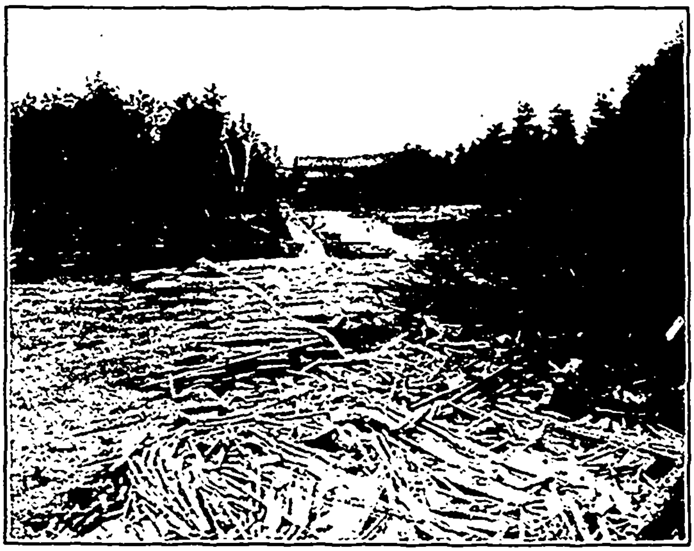


Sawmill on the Bonnechere river, a branch of the Ottawa. Sawdust and edgings pass into the river from the end of the mill.

were thirteen feet. Taking the specific weight of wet pine as .75, each log would weigh about 560 pounds. Of this weight about 13 per cent. would pass into the river as sawdust. This 13 per cent. was obtained as the average of five estimates furnished by such lumbermen as E. W. Rathburn, Esq., J. R. Booth, Esq., and W. C. Edwards, Esq. Consequently about seventy-two pounds of sawdust would pass into the river from every log cut into inch boards, or a total of 27,000 pounds of sawdust per day. Expressing this as percentage of water (642,816,000 pounds) we get .004 as the percentage strength of sawdust in this water.

During the high water of April, May and June the strength of the solution would be considerably less than .004 per cent., and as chub and brook trout were caught on and off all summer below the mill, this strength of sawdust solution was certainly not strong enough to kill off all the fish, though it is quite conceivable that it might drive fish down the river into tributary streams where there could be no sawdust pollution.

Comparing this percentage with that in two of the laboratory experiments described on pages 450 and 452 we find that in one case two grams of white pine sawdust in 1,700 c.c. of fresh water, *i.e.*, .12 per cent. strength, soaking for five hours, killed a minnow in twenty-nine minutes; and in the other case a percentage of .16 killed in ninety minutes.



Slabs, edgings and sawdust, half-a-mile below the mill.

Of course, these figures are mere approximations, but they point unmistakably to the conclusion that the sawdust poured into the Bonnechere river is not destroying its fish life. Moreover, in Golden Lake, an expansion of this same river, and ten miles above any saw mill, lake trout used to be very abundant. Every October large numbers were caught in nets along their spawning beds. Now these spawning grounds are reported to be deserted by the fish; and certainly sawdust cannot be blamed for their disappearance. Higher up the river, in Round Lake, the October fishing is still good, solely because there are fewer settlers and less fishing.

CONCLUSIONS.

1. Strong sawdust solutions, such as occur at the bottom of an aquarium, poison adult fish and fish fry, through the agency of compounds dissolved out of the wood cells.

2. The overlying water in such an aquarium does not at first kill fish. After about a week it does kill, but solely through suffocation, the dissolved oxygen having all been used up.

3. Bacteria multiply enormously throughout all parts of such an aquarium, and through oxidation change the poisonous extracts to harmless compounds. Mosquito larvæ live on the bacteria. No doubt, in natural pools, other aquatic insect larvæ live on bacteria also.

4. Subsequent aëration and sedimentation of sawdust water purify it, so that fish can live in it without injury.

5. Since adult fish and black bass fry both refused to be driven into pine extracts in the bottom of an aquarium after they had experienced its poisonous effects, we may infer that fish would desert a river much polluted with sawdust, going down stream and into tributaries to escape from the disagreeable influence of sawdust extracts.

6. No stream can be pronounced off hand as poisoned by sawdust. Each stream must be studied by itself and the varying conditions must be thoroughly understood before a judgment can be pronounced. The chief things to be considered are (1) the quantity of sawdust, and (2) the volume of water into which the sawdust is discharged. Subordinate conditions are the rapidity or sluggishness of the stream, the amount of sunlight or shade, and the character of the water, whether from agricultural lands or from primitive forests.

7. Further observations and studies along sawdust polluted streams and rivers of Canada are urgently needed before more definite conclusions can be reached.

ACKNOWLEDGMENTS.

Acknowledgment is due to Toronto University, the Public Library, Toronto, and the Canadian Institute, for the privilege of consulting their libraries in order to write the historical part of this report.

I am under special obligations to my colleague, Prof. J. C. Connell, M.A., M.D., for the large number of minnows which he procured for me, and which were so indispensable for the laboratory experiments.

Dr. John Waddell and Mr. C. W. Dickson, M.A., both of the School of Mining, Kingston, rendered valuable aid in determining the amount of solid matter in sawdust water.

The Ontario Fisheries Department facilitated my task on the Bonnechere by instructing their overseers to assist me in every way possible.

APPENDIX TO DR. KNIGHT'S REPORT ON SAWDUST AND FISH LIFE.

BACTERIOLOGICAL EXAMINATION OF SAWDUST WATER IN SHADE AND IN SUNSHINE.

Examination of sawdust water in aquarium made July 31st, 1902.

Two agar plates made. The *first* averaged 3,300 colonies of bacteria per cubic centimetre. None of the colonies were spirilla which were present in large numbers in direct microscopic ex. mination of the water. The chief colonies were those of a spore bearing bacillus, a variety evidently of B. Subtilis; also a few sarcinae, particularly one like Sarcina Lutea. The second plate averaged 3,570 colonies per cubic centimetre. In general characters they were the same as in the first plate.

AUGUST 4TH, 1902. *Water in aquarium.* Agar plates averaged 3,570 colonies per cubic centimetre. These were in all respects like those of July 31st.

Same water in sunlight since July 31st. Agar plates average 4,200 colonies per cubic centimetre. These colonies contain the same bacteria as in the aquarium water, but in fewer numbers. Further, there is present a fluorescent bacillus, making up half the number of colonies present.

AUGUST 8TH, 1902. *Water in aquarium.* Agar plates develop 7,870 colonies per cubic centimetre. These colonies are of the same type as those found on previous plates with the addition of about 1,000 colonies of B. Mesentericus Vulgatus per cubic centimetre.

Water in sunlight. Agar plates develop 37,070 colonies per cubic centimetre. These consist mainly of B. Fluorescens Liquescens; also of Sarcina Lutea, and an occasional colony of B. Subtilis.

W. T. CONNELL,
Prof. of Bacteriology.

THE BACTERIAL CONTAMINATION OF MILK AND ITS CONTROL.

BY F. C. HARRISON, PROFESSOR OF BACTERIOLOGY, ONTARIO AGRICULTURAL COLLEGE.

(*Read 28th February, 1903.*)

MILK as sold in cities, towns or villages contains a varying number of bacteria according to its age, the amount of sediment in it, and the temperature at which it has been kept. Soxhlet,[1] Uhl,[2] Backhaus[3] and others have shown that the more dirt or sediment, the more bacteria there will be, and Renk[4] has given us some interesting experimental data on the amount and kind of filth present in ordinary market milk. This filth is largely made up of excrementitious matter, vegetable fibres, epithelial debris, hairs of the cow, dust particles, etc., and the amount of filth contained in a litre of milk furnishes a positive index of the degree of cleanliness observed in the dairy stable. Renk found the following amount of dried impurities in the milk supply of the following German cities: Leipzig, 3.8 milligrams; Munich, 9 milligrams; Berlin, 10.3 milligrams; Halle, 12.2 milligrams per litre; and Hird[5] found in the Washington, D.C., milk supply, 5.30 milligrams of filth per quart.

Backhaus[3] has also shown that 50 per cent. of fresh manure dissolves in milk and does not appear as sediment; and therefore the weight of undried filth in all these samples would have been more than doubled. This investigator has also determined by actual tests that the daily milk supply of Berlin, Germany, contains about 300 pounds of dirt and filth. Further, many of the bacteria derived from such sources are very harmful, for not only are such fecal bacteria concerned in the intestinal troubles of infants, but they also give rise to abnormal fermentations in butter and cheese, producing taints, off-flavours, and decomposition products in these foods.

For the guidance of the dairyman who buys milk for sale, and for the housewife, Renk[4] suggests the following rule: If a sample of milk shows any evidence of impurity settling on a transparent bottom within two hours, it is to be regarded as containing too much solid impurities.

When we examine the results relative to the number of bacteria in European market milk, we are at once struck by the enormous numbers that are frequently present.

Thus, Clauss[6] found that the number of germs per c.c. of Wurzburg milk ranged from 222,000 to 2,300,000. The average was between one and two millions per c.c. Knopf[7] found from 200,000 to 6,000,000 per c.c. in the milk of Munich. Bujwid[8] examined the milk of Warsaw, where there was an average of 4,000,000 per c.c. In the milk immediately after it was drawn from the cow he found 10,000 to 20,000 per c.c. In Amsterdam, Geuns[9] found 2,500,000 per c.c. in fresh milk. Renk[10] examined the market milk of Halle and found from 6,000,000 to 30,700,000 per c.c. Uhl[11] in 30 tests of Giessen milk found from 83,000 to 169,600,000 per c.c. In the month of June he found an average of 2,900,000 per c.c. The average in May was 22,900,000. Uhl explains this difference by the supposition that the cows and stables were kept clean during this latter month, and there was less night's milk mixed with the morning's.

Knochenstiern[12] examined more than 100 samples of the milk of Dorpat. He divided the samples into four classes, according to their sources. The averages of the numbers in the several classes ranged from 10,000,000 to 30,000,000 per c.c.

The milk supply of Helsingfors was studied by Hellens,[13] who found in samples taken in the summer from 20,000 to 34,300 bacteria per c.c., while in the winter the bacterial content ranged from 70,000 to 18,630,000 and averaged 2,111,000 per c.c. About 60 per cent. of the summer samples contained over 1,000,000 bacteria per c.c., against 35 per cent. in the winter samples.

Rowland[14] found in twenty-five samples of London, England, milk an average of 500,000 bacteria per c.c.

Sacharbekoff[15] examined more than eighty samples of St. Petersburg milk. The number of bacteria ranged from 400,000 to 115,300,000 per c.c., with an average of 16,596,000.

Conn[16] has already pointed out the fact that the market milk of American towns and cities contains fewer bacteria than are to be found in European supplies; and he has explained as the reason for this difference, the free use of ice in North America.

Sedgwick and Batchelder[17] examined a number of specimens of milk from Boston. They found, as an average of several tests that milk obtained in a clean stable from a well-kept cow, milked into a sterilized bottle, contained 530 bacteria per c.c.; but when the milking was done under the ordinary conditions of farm practice, the number of bacteria reached on the average, 30,500. From fifteen samples of milk obtained from the houses of people in the suburbs of Boston, the average was 69,000 germs; from fifty-seven samples from milkmen the average was 2,350,000, and from sixteen samples secured at groceries, the average was 4,577,000 bacteria per c.c.

In 1901, Park[18] reported on the milk supply of New York. He found that during the coldest weather the average was about 250,000 bacteria per c.c., during cool weather about 2,000,000, and during hot weather about 5,000,000. Regarding the harmfulness of these bacteria the writer cites the universal clinical experience "that a great many children in cities sicken on the milk supplied in summer, that those who are put on milk that is sterile, or that contains few bacteria, as a rule, mend rapidly, while those kept on the impure milk continue ill or die."

Leighton[19] determined the number of bacteria in the milk supply of seventeen dairies at Montclair, N.J., the investigation extending over a period of three years. In dairies of the most approved type the average number of bacteria per c.c. was below 15,000. Poorly equipped dairies, in which the owners had endeavoured to do their utmost to produce a pure product with the crude means at hand, gave an average of between 40,000 and 70,000 per c.c.; and in those dairies in which neither good equipment nor good intentions prevailed, the average number of bacteria was over 180,000.

McDonnell[20] sampled 352 lots from eleven American cities. The worst samples were found in restaurants, and with small retail dealers. Twenty-eight per cent. of all samples contained less than 100,000 bacteria per c.c., while 34 per cent. had less than 500,000 per c.c.

Loveland and Watson[21] found in the supply of Middletown, Conn., from 11,000 to 85,500,000 per c.c.; and milk as delivered by milkmen to their private customers in the city of Madison, Wis.,[22] ranged from 15,000 to 2,000,000 organisms per c.c., varying mainly with the seasons of the year.

The writer[23] examined about twenty samples of Guelph market milk, a few years ago, and found an average of 650,000 bacteria per c.c.

Eckles[24] has made a bacteriological study of the milk supply of a creamery. He found from 1,000,000 to 5,000,000 organisms per c.c. in winter, and in summer from 10,000,000 to 80,000,000 per c.c.

During the summer of 1901,[25] whilst investigating an affection known as bitter milk in a large cheese factory, the writer had the opportunity of analysing the milk of ninety-six patrons who delivered milk to the factory. The results of this examination showed that the mixed milk of each patron contained from 4,000,000 to 30,000,000 of micro-organisms per c.c. Astonishingly large numbers (from 100,000 to 5,000,000) of the Colon bacillus were found in many samples.

Having seen from this review of the bacterial content of European and American milk supplies, let us now examine the sources of this contamination.

The bacteria which find their way into milk come from:—(1) The fore-milk; (2) The animal and milker; (3) Dusty air; (4) Unclean utensils. And, as Russell remarks, "the relative importance of these various factors fluctuates in each individual instance."

I.—CONTAMINATION FROM THE FORE-MILK. (*See Fig. 1*).

The constant presence of bacteria in freshly-drawn milk is a matter of considerable importance, and this fact helps to explain the ineffectual attempts to obtain milk in commercial quantities uncontaminated by bacteria. At the same time it has been but very recently that investigations as to the number and nature of the organisms that gain access to the milk through their localization and multiplication in the milk ducts, have been made. The first recorded experiments are those of Leopold Schultz[26] in 1892. He examined milk bacteriologically at the first of the milking, in the middle of the milking and at its close. This examination consisted merely in counting the number of bacteria present, and as a result, the following figures were determined:—The first milk contained from 55,000 to 97,200 germs per c.c.; the middle milk from 2,000 to 9,000 germs per c.c.; and the last milk was in some cases sterile, and sometimes contained about 500 germs per c.c. The number of germs in the last milk, he says, depended upon the quickness with which the milking was done. When done quickly, all the germs were washed out, so that "the last milk was often, but not always, sterile."

Gernhardt[27] investigating the same subject found a larger number in samples from the middle of the milking than at the beginning. To explain this result, as well as to explain irregularities in the numbers, he suggested that the bacteria made their way up through the milk-ducts of the teats, through the cistern, and into the smaller ramifications of the ducts which connect the cistern with the ultimate follicles. As many of the colonies so formed are not easily removed, they are not found in the first milk, but appear later when they have become broken up by the persistent movements of milking.

Von Freudenreich,[23] on the other hand, states that when in the udder milk is free from bacteria, except when the milk glands are in a diseased condition.

H. L. Bolley and C. M. Hall,[29] in their studies of the bacterial flora of the milk of ten healthy cows, isolated sixteen distinct species of bacteria, some of which were common to both the first and last milk, and others to only one of these. All the micro-organisms found were bacteria, and none were found which produced gas.

Russell[30] in his text-book on Dairy Bacteriology, published in 1894, states that he has found an average of 2,800 germs per c.c. in the fore-milk, while the average of the remainder of the milk only had 330 germs per c.c. In characterizing this, he says that " the number of species is usually small, one or two kinds usually predominating to a large degree. Those that are commonly found are those that produce lactic acid, as these microbes find in milk the best medium for their growth."

Gosta Grotenfelt,[31] however, in his text-book on the " Principles of Modern Dairy Practice," reasserts the statement of Von Freudenreich that when the milk is drawn from the udder of a healthy cow, it is germ-free or sterile.

Rotch[32] concludes, from an examination of the bacteria found in four cows' milk, that the bacteria do not necessarily come from external sources, but that they may also come from some part of the milk-tract between the udder and the end of the teat. The few colonies, however, obtained in the plates from the latter half of the milkings, are considered as possible contaminations between the " cow " and the " plates."

Moore[33] states that in investigations made upon this subject, he found that, in addition to the bacteria in the fore-milk, the last milk from at least one-quarter of the udder in every case contained bacteria.

Conn,[34] reviewing this subject, says that the different results of many of these early experiments are due to the small quantities of milk taken, while in the latter experiments large quantities have been taken. He adds, " Undoubtedly the milk-gland of the healthy cow produces milk which is uncontaminated with bacteria, but the large calibre of the milk ducts makes it possible for bacteria to grow in the duct to a considerable extent, so that it becomes a matter of extreme difficulty to obtain milk from the cow, even with the greatest precautions, which will not be contaminated."

Harrison,[35] in 1897, in a report of investigations upon this subject stated. "When milking is done there remains in the teat of the cow a little milk that affords nourishment to any bacteria that may come in contact with it through the opening at the end of the teat." The average of a number of analyses made by him shows the presence of 18,000 to 54,000 germs in the fore-milk, and 1,000 to 3,000 in the after-milk.

Experiments have also been conducted on human milk by Palleske,[36] Honigmann,[37] Knochenstiern,[38] and Ringel,[39] but all of these have independently found it impossible to get human milk from the mammary gland in such a way as to be sterile.

The most recent work upon this subject has been done by Moore and Ward,[40] of Cornell University. They investigated the source of a gas and taint producing bacterium in cheese curd for a certain factory that was troubled with "gassy curd." They easily located the trouble in the herd of a particular patron. On inquiring into the history of the herd it was ascertained that at the time of parturition, the placentae had been retained by a number of cows, and these had been allowed to decompose in the uterus. It was soon after this that the "gassy curd" began to appear. A thorough bacteriological examination located the bacillus which was the cause of the "gassy curd" in the udders of the cows of the herd; and it seemed very probable, though, of course, not demonstrable, that it had gained access to the udders from the decaying placentae.

Subsequent to this, Ward conducted further experiments, and in an article on "The Persistence of Bacteria in the Milk-Ducts of the Cow's Udder,"[41] he concludes, (1) "certain species of bacteria are normally persistent in particular quarters of the udder for considerable periods of time, and (2) it is possible for bacteria to remain in the normal udder and not be ejected along with the milk." These conclusions controvert the statement previously made by Von Freudenreich and Grotenfelt that the milk-ducts are always sterile at the close of milking, becoming tenanted from the outside alone by organisms which chance to come into contact with the end of the duct.

The results of still later investigations by the same author are published in a bulletin on the "Invasion of the Udder by Bacteria."[42] In these investigations a bacteriological examination was made of the udders of milch cows slaughtered after reacting to the tuberculin test. In all cases the udders were perfectly normal. Just before slaughtering the animals were milked as thoroughly as possible and samples of the milk taken, and a bacteriological examination made. After slaughtering, a similar examination was made of the tissues of the udder. In all cases, even in the upper third of the udder, bacteria were found, and they were identical with those found in the milk. He concludes that "milk, when secreted by the glands of the healthy udder, is sterile. It may, however, immediately become contaminated by the bacteria which are normally present in the smaller ducts of the udder." However, "the bacteria so far found in the interior of the udder do not affect milk seriously. This, however, does not preclude the possibility that forms more injurious to milk may invade the udder."

From the above resumé it is apparent that widely different results have been obtained by different investigators, and it has been a very interesting study to see whether the experiments conducted at Guelph would throw any light upon these divergencies in results.

The plan of experiment has been as follows: For a number of days samples were taken from the fore and after milk of a number of cows on the College Farm. The samples were collected in sterile test-tubes, and previous to taking the milk, the flank, udder, and teats of the cows were thoroughly washed with a 1-1000 solution of mercuric chloride. Gelatine plates were then made from these samples, and afterwards the number of colonies counted and the different species isolated and cultivated on the various media. It soon became apparent that while several species were more or less constant in the udders of all the cows, yet there were many variable species present in the milk of some cows that were not present in that of others, and not even in the same udder on two successive days. Therefore, in making a systematic study, it was deemed best to confine our attention to those species that were more or less constantly present in the milk of all the cows, and to make a complete study of those existing in the udder of one particular cow.

The number of bacteria present in both the fore and after milk of the various cows, and of the same cow, and even in the different quarters of the udder of the same cow, were so widely different that little stress can be laid upon an exact enumeration.

The following samples, which are typical of many others, will illustrate the point:

Cow No. 1.	*Determination 1.*
Fore-milk, right front teat.................	86,400 per c.c.
" " hind "	120,000 "
Strippings, " front "	40,800 "
" " hind "	57,000 "

				Determination 2.
Fore-milk,	right	front	teat	48,000 per c.c.
"	"	hind	"	24,080 "
"	left	front	"	22,400 "
"	"	hind	"	35,100 "

Cow No. 2.	*Determination 1.*
Fore-milk	200-500 per c.c.
After-milk	0-100 "

The results of a large number of determinations, of which the above are typical, showed on an average 25-50,000 germs per c.c. in the fore-milk. The numbers in the strippings or after-milk varied greatly with the manner of taking. For example, when the milking was done quickly, but very few and sometimes no colonies were found in the "strippings," whereas, when the milking was done slowly and some time lost before the samples from the last milk were taken; the number of bacteria was very variable, being in one case as high as 57,000.

The important point, therefore, is not the exact number, but the fact that bacteria were found in large numbers, not only in the fore, but in the middle and last milk of nearly all the cows tested.

The number of species present in the udders of cows is very small. Of this number some are more or less constantly present, whereas others are very variable in their presence. Of those species which are present, the characters are in many cases so slightly marked that their identification proved a very difficult matter. In fact, with the exception of *Bacillus acidi lactici*, not a single species discovered was strongly characterized. A number had a very little or no effect upon milk, and even the digestors were in every case very slow digestors.

B. acidi lactici (Conn No. 206), *B. acidi lactici* (Conn 202), and *B. lactis aerobans* (Conn 197) are the only ones that have been found constantly present in the samples, and in every case they have composed at least 95 per cent. of the germs present. The following species have been only more or less variably present, and in no cases have been found in large numbers.

B. halofaciens (N. Sp.). This bacterium approaches in characteristics *Bact. annulatum* (Wright), but differs from it in several details, so that we do not hesitate to call it a new species. The name refers to the characteristic halo found in gelatin cultures. As this bacterium was of quite frequent occurrence, we made butter from cream ripened with a culture of it, and found that the flavour of the butter, while not strong was quite disagreeable. At the same time, its presence in relatively small numbers, and the fact that the flavour was not strongly marked, make this an inconsiderable item so far as the "natural ripening" of such milk is concerned.

Micrococcus varians lactis (Conn 113 and 104). Conn, in speaking of this coccus, considers it one of the most important of dairy species, and suggests that it likely exists in the milk-ducts.

Bacillus No. 18, Conn. This species appeared very frequently in all samples examined, but never in very large numbers. In old gelatin and agar cultures, spores appeared at the ends of the bacilli. Two cultures from these heated for ten or fifteen minutes at a temperature of 85° to 90° germinated in one day. The species is very similar to *Bacillus No. 18 Conn*, but differs in that growth on potato is not spreading, and no spores are found in potato culture.

Bacillus No. 7. This bacillus was quite constantly present for some weeks, but afterwards disappeared. It seems to resemble *B. cremoris* (43) (*B. lactis No. 9 Flugge*), but differs in its effect on milk, so that it would appear to be an allied species.

Bacterium No. 8. The gelatine plate colony appears very similar to No. 7, but the organisms otherwise differ, both morphologically and culturally in many particulars. Like No. 7 it appeared quite constantly for some weeks and then disappeared.

B. exiguum (Wright).[44] This bacterium was found almost constantly present in the milk of one of the cows tested, but was never found in that of any other. There were never more than from one to four colonies per plate present, so that its effect on the milk was very inconsiderable. Its similarity to *Bact. exiguum* (Wright) is most marked. Wright isolated this bacterium from water, but the fact that he found its optimum temperature to be 36° and that it is a facultative anaerobe does not make it at all surprising that it should be found in the udder of a cow.

Micrococcus No. 10. This coccus comes in the same class as Conn's 167, and may be identical with it. The most marked variations are the gelatin colony, and the fact that in no culture of this germ was there the slightest indication of a yellow colour. At the same time it agrees in morphological characters, and especially in the fact that, although a liquefying coccus, it fails to curdle milk.

Like several of the forms previously described, this coccus was found to be present for some time in the samples taken, but afterwards, in a few weeks, completely disappeared. In no cases was it present in large numbers.

Comment has already been made upon the fact that by far the largest number of species determined in all the samples tested, were lactic acid species, and that other species, although more or less constantly present, were not invariably so, and never in very large quantities. This is a most important, practical consideration, for it means that, although by the most scrupulous care, it may not be possible to procure milk free from germ-life because of those that are present in the udder of the cow, yet the species that gain access to the milk through this source are, for the most part, beneficial ones. In Bulletin No. 21, 1900, of Storr's Agricultural Experimental Station, Conn speaks of a method, now widely adopted in American dairies, for procuring what is known as a "natural starter." The method consists in drawing milk, just as has been done in all the examinations we have made, into sterilized flasks, and using cultures from these as starters. Conn says, "there can be no question that the use of natural starters thus made has been a very decided advantage to the buttermaker," for the reason that "the bacteria which are within the cleanly cow's udder and thence get into the milk, are most commonly of the desired character." There is, no doubt, some uncertainty about this method, but so far as all examinations conducted by us are concerned, the cultures so obtained would be good ones, being largely composed of lactic acid species.

While the large per cent. of lactic acid species present is the paramount characteristic of the bacterial flora of freshly drawn milk, yet there are other peculiarities of considerable, if not equal, importance.

By reviewing the description of the species determined, it will be noted in every case that, although each species would grow at room temperature, yet the optimum temperature was in the neighbourhood of 37°. This fact was well demonstrated in comparing gelatin plates, made from the general milk supply, with those made from the aseptically drawn milk. These plates cannot be kept at a temperature higher than 22°, and it was most marked that when plates from the former were quite covered with bacterial growth, those from the latter were still clear. On the other hand, when agar plates were used and kept at a temperature of 37°, the order of growth was slightly reversed. This explains facts that were noted in reference to the keeping quality of the aseptically drawn milk, as compared with that of the general milk supply; for, when kept at room temperatures, the former remained good considerably longer than the latter; whereas, when kept at 37°, both became curdled in less than twenty-four hours.

Another marked characteristic of all the species was that they were facultative anaerobes. The anaerobic faculty was especially marked in the two species, Nos. 1 and 2, that were found to be so uniformly present in all the milk tested. This is just what one would naturally expect, for the conditions in the udder must be largely anaerobic conditions. In virtue of these conditions, and possibly other undetermined conditions, the udder, so to speak, exerts a selective action upon the bacteria which may be temporarily present in it. In this they are, of course, aided by the mechanical expulsion of bacteria in the process of milking.

In order to throw a little light upon this problem, experiments were conducted in inoculating udders with well-marked but harmless bacteria, which could be easily recognized by their cultural characteristics. *Bacillus prodigiosus* and *B. exiguum*, both of which were marked by their pigment production, were the ones experimented with. Cultures of these were smeared upon the ends of the teats, so that the bacteria might work their way up into the udder, just as any other germ might which comes in contact with the ducts of the teat. In the case of *B. prodigiosus*, about 20,000 per c.c. were present in the fore-milk at the first milking eight hours after inoculation. By the third milking only a few were present, and after that it disappeared completely. The experiment was repeated with *B. exiguum* with similar results, although a smaller number—240 per c.c.—were present in the first milking, and by the fourth milking it had disappeared. No doubt the small number was due to the fact that this germ grows more slowly than *B. prodigiosus.*

In view of Ward's discovery of *B. fluorescens liquefaciens* in the udders of certain cows, it seemed advisable to attempt to colonize this germ in the udder, and a bouillon culture was smeared upon the ends of the teats of a cow in the manner already described. This bacillus was discovered in the fore-milk six hours after the teats were smeared, but was not found in the fore-milk of the second and third milkings.

It does not seem probable that an aerobic bacterium of this character is able to live and compete with facultative anaerobic bacilli. Further, the optimum temperature for the fluorescing bacterium is not 37°.

Possibly, by continuous experimentation, we might have finally discovered a species which would persist in the udder, but at the same time, the bacteria chosen have evidently fared much the same as other more or less injurious forms which may occasionally find temporary lodgment in the udder. Exception, however, may occur, as we have the gas and taint producing bacillus located by Ward and Moore in the udders of the cows of a particular herd.

Another fact that may have a bearing on this problem is that normal healthy organs, taken from the body immediately after death, may contain bacteria which are capable of development. Thus, Ford[45] has shown that 80 per cent. of healthy organs, removed from killed guinea pigs, rabbits, dogs and cats, contained living bacteria. No udder tissues were examined by him, and in order to ascertain if bacteria existed in the udder of healthy animals a few experiments were made along these lines, but they are open to criticism because it is impossible to say, with any degree of certainty, that the bacteria found came from the animal's glands or blood, or from infection through the teat. However, by selection of cows which had been dry for several weeks before slaughter, the latter objection is to some extent overcome. The liver was examined at the same time, and its bacterial content, if any, noted.

The methods employed in this work were essentially those used by Ford—a large piece of the tissue to be examined was excised with a sterilized knife, placed in a sterile jar, and immediately taken from the slaughter house to the laboratory. Small pieces of tissues were then cut from *the inside* of the large piece with sterilized knives, and then held in the flame of a Bunsen burner with sterilized forceps, until the whole of the outside of the piece was well scorched. The piece was then transferred to beef bouillon or peptone whey bouillon, and the preparations placed in the incubator at 37°. On the fourth day gelatine plates were made from the different pieces of tissue, and the bacteria, if any, were isolated.

I. An aged cow, dried up five weeks before slaughtering, udder small, with considerable fatty tissue. All organs perfectly healthy.

	Bouillon.	*Peptone Whey.*
Liver	+	+

Subsequent plating and sub-cultures gave :—

B. mesentericus vulgatus.

B. subtilis, and a Micrococcus, identity not established.

	Bouillon.	*Whey Peptone.*
Udder	+	+

Plates gave colonies of:—

B. subtilis.

Micrococcus (sp ?).

II. An aged cow, dry for some time, the butcher not knowing the exact length of time. Udder of a fair size, and well formed. All organs apparently healthy and normal.

	Bouillon.	Whey Peptone.
Liver	+	+

Plates gave colonies of:—

B. lactis aerogenes.

Proteus.

	Peptone.	Whey Peptone.
Udder	—	—

III. An aged cow, dry for four weeks previous to slaughter. Udder fair size. Organs normal and apparently healthy.

	Bouillon.	Whey Peptone.
Liver	+	+

Gelatine plates gave colonies of:—

B. subtilis.

A spore-bearing bacillus, which produces no effect in milk.

	Bouillon.	Whey Peptone.
Udder	+	+

Gelatine plates gave cultures of:—

Micrococcus varians lactis.

These .results, whilst agreeing with Ford's, are not sufficiently authoritative to allow us to assert positively that the bacteria found in the udders of the two cows came from the blood or lymph stream, rather than through the teat, but in conjunction with the results obtained by Ford, they threw doubt on the supposition that all udder infection comes originally through the orifice of the teat. It is also noteworthy that a spore-bearing bacillus belonging to the *subtilis* group, and several micrococci were isolated by Ward from udder tissue. Another fact, which is difficult to explain and which may possibly have some influence on the bacterial content of the udder in its normal condition, is the strong germicidal power of freshly drawn milk. This property was first noticed by Fokker,[46] and subsequently confirmed by Freudenreich,[47] and quantitative studies of freshly drawn milk, inoculated with various bacteria, show that an actual destruction of bacteria took place. This germicidal property has been shown to exist in the milk-serum, and is evidently allied to the similar bactericidal property of blood, for Brieger and Ehrlich[48] and Wassermann[49] have found that the milk of immune animals can confer immunity.

If then, this germicidal power exists in fresh drawn milk, it is certain to be present whilst the milk is still in the udder, and may inhibit or prevent the rapid multiplication of adventitious bacteria, which penetrate up the opening of the teat. Although we have frequently found large numbers of lactic acid bacteria in freshly drawn milk, yet the reaction of this milk is never acid.

De Freudenreich[47] has also shown that the bactericidal power is not the same for all species, that whilst the cholera vibrio, the typhoid bacillus, and even *B. Shafferi* (a colon bacillus), are destroyed in large numbers, the bactericidal action is less pronounced on lactic acid bacteria. These facts may possibly explain why the germs of the colon type are so seldom found in the healthy udder, for we know that the teats and udder of the cow are constantly brought in actual contact with particles of manure, and even the hands of the average milker are soiled with stable filth, which undoubtedly contains colon bacteria.

It might be reasonably asked if the advice, commonly given to those who wish to procure milk as near sterile as possible, to milk the first few streams in a separate utensil, is good. And in reply we would say, decidedly yes, for not only is the number of bacteria in the fore-milk much in the excess of the bacteria found in the rest of the milk, but frequently the number of species found in the fore-milk is considerably larger than that in the after-milk. (*See Fig. 2*).

In reviewing the subject there can be no doubt that the number of bacteria present in the milk as it exists before being drawn from the udder is somewhat startling, and were nothing more than an enumeration of the germs given, there might be some occasion for alarm.

However, a systematic study of the germs proves that, with the possible exception of rare cases, this source of bacterial life is much more beneficial than baneful to the average consumer of milk and its products.

CONTAMINATION FROM ANIMAL AND MILKER.

A prolific source of contamination is from the animal and milker, and the following realistic statement,[50] describes a condition which unhappily is only too common on many farms:—

"The day has gone by when a pretty milkmaid went, in clean white apron and with shining milk pail, to milk the cow with the crumpled horn, out among the buttercups on a dewy morning. Instead, some old fellow stumbles out of the house and to the barn, with the stump of a clay pipe in his mouth, and wearing overalls and boots saturated and covered with the filth acquired by a winter's use. When he reaches the barn he selects some recumbent cow, kicks her until she stands up dripping and slimy, and as he is a little late and the milk will hardly have time to cool before the man who carries it to the city will come along, he does not stop to clean up behind the cow, but sitting down on a stool, proceeds to gather the milk, and whatever else may fall, into a pail which perhaps is clean and perhaps is not. Of such refinements as washing the udder of the cow or wiping her flanks he has never heard. If he has, it is only to scoff. Then he stands the milk behind the cows. That is bad enough, but it is not all the story. Everyone knows that in straining the milk the strainer becomes obstructed, more or less, with dirt and filth, and when the milk does not run fast enough, he would be a rare milker who hesitated to scrape away a place with his fingers so that the milk might run more freely. Those who have seen certain fingers, as I have, know what that means."

This description seems hardly credible, but when one visits an ordinary cattle stable, he is prepared to believe almost anything under this head. The hair of cows, even those that are kept very clean, swarms with bacteria. (*See Fig. 3*). Many hundreds may be isolated from a few particles of hair, and this fact alone shows the importance of keeping cows clean, well carded and well brushed. When in this condition, they are not so liable to lose hairs, nor are the hairs so easily dislodged during the movements of milking. The particles of manure and filth which cling to the sides, flank, udder and tail of animals are laden with germ-life. Wüthrich and Freudenreich[51] have found far more bacteria in manure when the animals are given dry food than when kept upon grass, and the most numerous species present were the colon bacillus, the hay bacillus, and other species able to liquefy gelatine, and peptonise casein. Great care should be taken in the construction of cow stalls. If they are too long the hind-quarters of the animal are apt to be plastered with manure when she lies down; and when too short, the hind-quarters and tail find their resting place in the gutter. (*See Fig. 4*).

The milker, too, is not always above reproach. Clothed in dust-laden garments, used for all kinds of farm and stable work, without even washing his hands, he does the milking as he would do any other job on the farm. Too often the milker has the filthy practice of moistening his hands and the cow's teats with milk. Freudenreich[23] reports some experiments in which the germ-content of milk was reduced from several thousands to 200 where the hands were well rubbed with vaseline before milking, and as pointed out by Russell,[52] a pinch of vaseline not only helps the milker to obtain a firmer grasp, but also prevents scales or dirt from being rubbed from the teats, and its effect on sore or chapped teats is healing.

METHODS OF PREVENTION.

Contamination from the milker can, however, to a very large extent, be prevented by moistening thoroughly the flanks and udder of the cow before milking. Germs cannot leave a moist surface, and the dust-like particles are thus held in place. (*See Fig. 5*). The following instructive experiment is cited by Russell[52]:—

"When the animal was milked without any special precautions being taken, there were 3,250 bacteria per minute deposited on an area equal to the exposed top of a ten-inch pail.. When the cow received the precautionary treatment, as suggested above, there were only 115 *bacteria per minute deposited on the same area. This indicates that* a large number of organisms from the dry coat of the animal can be kept out of the milk if such simple precautions are carried out." It has been frequently found that the germ-content of the milk in the pail is increased from 20,000 to 40,000 bacteria per minute during the milking period by the dislodgment of organisms from the animal.

By diminishing the exposed surface of the milk-pail, (*see Fig. 6*), a considerable amount of dirt may be excluded, as it is obvious that less dirt will fall in a pail with a small opening. A number of different types of such pails are now in use, and Eckles[53] and other investigators have given us experimental data on the subject. Thus 43,200 bacteria per c.c. were found in the milk drawn in a common pail, as against 3,200 per c.c. in the covered pail. The milk soured in forty-three hours in the first case; sixty-four hours in the latter case.

The milker should put on a clean, loose, cotton or linen smock over his clothes, and invariably wash his hands immediately before milking. The milking smock should be washed frequently and should be kept, not in the open barn or stable, but in some place as far removed as possible from all kinds of dust and dirt. The practice of moistening the hands or cow's teats with milk should be scrupulously avoided.

MILKING MACHINES.

Of late years several milking machines have been introduced to obviate the difficulty in obtaining milkers, and to lessen the time taken in milking. Such machines, in order to be a success, must do the milking naturally, quickly, thoroughly and without any annoyance to the cow. Further, milk drawn by such a machine must be of good keeping quality, and the machine must be adaptable to the requirements and arrangements of an ordinary dairy farm.

When first introduced many dairymen expressed the opinion that these machines would guard against the admittance of all dirt, but unfortunately this requirement has not been fulfilled by a machine installed in the Dairy Stable of the Ontario Agricultural College.[54] This machine, called the "Thistle Milking Machine," (*see Fig. 7*), was in more or less constant use in our stable during the summer of 1899, and we took advantage of this opportunity to make bacteriological analyses of the milk milked with the machine as compared with milk drawn by hand milking. We found that the machine milk had a far larger germ content than that milked in the usual manner, and after making a direct comparison between the number of bacteria in machine-milk and the number in hand milk, we found that the proportion varied greatly, from three to twenty times as many bacteria being found in the machine milk as in the hand drawn milk. The averages were as follows :—

Machine Milk.		*Hand Milk.*	
No. of Analyses.	No. of Bacteria in c.cm. of *morning's milk.*	No. of Analyses.	No. of Bacteria in c.cm. of *morning's milk.*
161	141,600	78	10,600
	Evening's Milk.		*Evening's Milk.*
74	165,000	16	12,900

These results were greatly in favour of hand-milking, and the large number of bacteria found in machine milk was attributed to three causes:

1. When the rubber teat-cups are fastened on the cow, a small portion of the hairy coat of the udder is included in the cup, and no matter how clean the animal is, germs are sure to be present on this coat in considerable numbers, depending upon the cleanness of the udder.

When the suction of the machine is applied, the force exerted naturally draws any loose or dry particles that may be on the eats and that portion of the udder within the cups, down into the mi.k. In this way, many germs on these particles gain access to the milk, and find in it suitable conditions for their growth and multiplication.

2. The teat-cups and connecting tubes to the milk pail are made of rubber, and consequently cannot be scalded or steamed, as scalding water or steam would crack and spoil the rubber ; hence it is impossible to cleanse them thoroughly from germ life. They may look clean after being rinsed in warm water and kept in cold water, but they are certainly not bacteriologically clean, *i.e.*, free from germs; and in the process of milking many of the germs on the inside of the rubber and in the crevices of the tubing are washed into the milk. Conclusive evidence on this is afforded by the fact that, time and again, germs that were constantly present in water in which the rubber tubing was kept between milkings, were also found in the milk.

3. In detaching the cups from one cow and putting them on another, attendants sometimes let them fall upon the floor of the stable, and in this way germ-loaded particles of dust and dirt get into the teat-cups and find their way into the pail as soon as the milking of the next cow begins. Of course, this may be put down to carelessness on the part of the attendants ; but in our experience, no matter how careful the transfer was made from one cow to another, instances of the cups falling occurred from time to time, and each time undoubtedly made a large addition to the germ content of the milk.

In 1898, the Highland and Agricultural Society of Scotland[55] offered a prize of £50 for the best milking machine. Only two makers entered their machines for competition, viz., Mr. W. Murchland of Kilmarnock, (the Murchland Milking Machine Company), (*see Fig 8*), and the Thistle Mechanical Milking Machine Company. The judges, after an exhaustive trial, awarded the prize to the Murchland Milking Machine, it having in every respect most effectually filled the conditions which they originally agreed should guide them in making their awards. In every instance the samples of milk drawn by this machine were found to keep satisfactory. After a lapse of forty-eight hours, they were found in no respect inferior to the samples of milk drawn by hand, in fact, if anything, rather superior in point of flavour. The judges regarded the Murchland machine as a practical success. On the other hand, the chief defect in the Thistle Milking Machine was the effect it had upon the keeping qualities of the milk. Most of the samples from it developed sourness in twelve to twenty-four hours, while samples drawn by hand from the same cows at the same time, and kept under precisely the same conditions, remained perfectly sweet for from thirty-six to fifty hours.

The Murchland Machine was also placed in competition for a prize of £50 at the York meeting of the Royal Agricultural Society[56] at York. In the opinion of the judges it presented such difficulties for efficient cleaning, that they were unable to report that it adequately fulfilled the requirements set forth in the regulations for these trials. No award was made in the competition.

CLEANING MILK BY THE USE OF A GRAVEL FILTER.

The gravel filter in most general use is the model used by the Copenhagen Milk Supply Company, (*see Figs. 9 and 10*). It consists of two enamelled iron tanks placed at different levels; a pipe in the form of a siphon has its long limb connected with the bottom of the upper tank, and its short limb with the bottom of the lower tank, so that the milk poured into the upper tank comes up as a kind of spring at the bottom of the lower one. On the bottom tank there are three layers of gravel, that in the lower layer being about the size of a pea; in the middle layer somewhat smaller, and in the third or top layer, a little larger than a pin's head. The layers are separated from each other by perforated tin trays. On the top of the uppermost layer of gravel are five thicknesses of fine cloth. The whole is kept in position by a pyramidal frame-work which presses down the tin trays. As the milk rises to the top of the tank, it passes into a large storage or mixing receptacle. These filters require the most careful management, and are generally taken to pieces immediately after use, when the gravel is washed in hot water until the water comes off clean. It is then steamed at a temperature of 202° F., after which it is spread out in shallow trays, and baked at a high heat. For the concluding operation, the gravel is placed in a winnowing machine, which drives off all particles of fine dust.

Schuppan[57] seems to have first called attention to the use of gravel filters as a means of reducing the number of bacteria in milk. He reported in 1893 that the bacterial content of milk was reduced 48 per cent. by sand filtration; and at the meeting of the German Dairy Association in 1893, he strongly recommended the use of sand filters for removing dirt and germs from milk. The use of sand filters was, however, questioned by other members.

Backhaus,[58] whilst giving no numerical data, reports that these filters have no effect in reducing the number of bacteria in the filtered milk. The mechanical separation is good, all coarse particles, such as hair, straw, manure, etc., are arrested; but the bacteria are washed out of the manure, and the milk contains more bacteria than before filtration.

In 1899, Dunbar and Kister[59] made an exhaustive study of the working of this class of filter. In twenty-two analyses of raw and filtered milk there were in seventeen cases more bacteria present after filtration, and in four cases fewer bacteria. A few examples of their results will suffice:

Raw Milk.	*Filtered Milk.*
80,000 per c.c.	60,000 per c.c.
793,000 "	44,100 "
95,000 "	49,400 "
819,000 "	94,000 "
Average 446,700 "	Average 61,800 "

In these cases filtration diminished the bacterial content of the milk.

Of the seventeen other samples, the bacterial content after filtration was increased, thus:—

Raw Milk.	*Filtered Milk.*
350,000 per c.c.	600,000 per c.c.
650,000 "	950,000 "
650,000 "	1,260,000 "
320,000 "	620,000 "
3,900,000 "	14,300,000 "

The average of the seventeen analyses was:—

Raw Milk.	*After Filtration through Gravel.*
1,300,000 per c.c.	5,567,000 per c.c.

CLEANING MILK BY CENTRIFUGAL FORCE.

Clarified milk, (*see Fig. 11*), or milk that has been passed through a separator, has been recently quite extensively advertised. The effect of this method of cleaning milk is similar to that of the gravel filters, and according to Backhaus,[58] 95 per cent. of the mechanical impurities (hairs, manure particles, etc.), are eliminated. The separator divides the milk into three parts, the slime which adheres to the bowl of the machine, the skim-milk and the cream. Several investigators have given us data of the number of bacteria which are found in these three products. Thus Popp and Becker[60] found the germ content per c.c. of the whole milk, to be 72,954; of the cream of this milk, 58,275; the separator skim-milk, 21,735; and the separator slime, 43,891.

Scheurlen[61] found in one litre of milk 2,050,000,000 of bacteria, and after separation 1,700 in the 200 c.c. of cream, 560,000,000 in the 800 c.c. skim-milk, and 18,000,000 in the 6 c.c. of slime.

Other investigators have also shown that centrifugation does not decrease the number of bacteria in milk. Thus, Fjord and Fleischmann[62] claim that centrifugal separation has little value as a means of purification, and Conn[63] states that " milk after passing through a centrifuge, although it contains less gross impurities, shows more bacteria than before. This is explained by the fact that masses are broken up, and large numbers of bacteria liberated, "and again," the same writer says, "centrifugal purification does not materially affect the bacteria, for there seem to be about as many after treatment as before."

Niederstadt[64] obtained similar results, for he found that by the centrifugal treatment of 300 litres of milk, about 130 grams of sediment were obtained. The cream was richer in bacteria than the sediment. The separator effected no purification of milk from bacteria, and 75 per cent. of the bacteria went into the cream.

Dunbar and Kister,[59] in an exhaustive series of experiments, found in four instances fewer bacteria after separation, the average of these four instances being as follows:—

Raw Milk.	*Centrifuged Milk.*
446,000 per c.c.	146,000 per c.c.

But in the remainder of the experiments, twenty-four in number, more bacteria were found in the separated milk, the averages in this case being:

Raw Milk.	*Centrifuged Milk.*
1,400,000 per c.c.	2,200,000 per c.c.

It would seem from these figures that the smaller the number of bacteria present in the whole milk, the more efficient was the separator in reducing their numbers.

Eckles and Barnes[24] have also investigated the purification of milk by the centrifugal separator. They found a large proportion of the bacteria removed by centrifuging, but no enhancement in keeping quality.

Russell in a private communication to the writer expresses his opinion thus :—" I do not think clarification is worth the trouble, unless the milk is exceptionally dirty.'

At the suggestion of the Ontario Department of Agriculture, we (my assistant, Dr. Streit and myself) have reinvestigated this subject.

A power belt separator was used, run at the speed indicated by the manufacturers. The milk came from farms in the vicinity, and was of average quality, similar to the ordinary factory supply. About 150 pounds of this milk were thoroughly mixed in a sterilized can with a sterilized stirrer. A half-pint sample of the milk was taken in a sterilized jar, the rest of the milk being put through a separator. The cream and skim-milk were caught together in a sterilized can, and were again thoroughly mixed with a sterilized stirrer, and another half-pint sample of the clarified sample was taken. Both samples were immediately carried to the laboratory, where suitable dilutions were made and plates poured.

The culture medium used was whey gelatine, with one per cent. of peptone. The plates were kept at 20° C. and counted at the end of forty-eight or seventy-two hours, depending on the size of the colonies. In most cases the plates were counted by each of us independently, so as to reduce the personal equation.

Each result given in the table is the average of four plates, and thus the gross average represents the numerical results obtained from 240 plates or analyses.

THE BACTERIAL CONTENT OF MILK BEFORE AND AFTER SEPARATION.

DATE.	BEFORE SEPARATION.		AFTER SEPARATION.		
	Total No. of Colonies.	Liquefying Colonies.	Total No. of Colonies.	Liquefying Colonies.	More bacteria after Separation + or less −
April 8	447,000	25,000	775,000	64,000	+
" 8	391,000	23,300	1,000,000	196,000	+
" 10	491,000	6,500	529,000	18,700	+
" 10	442,000	7,500	469,000	16,000	+
" 12	1,351,000	88,500	2,495,000	271,000	+
" 12	1,990,000	67,500	2,070,000	110,000	+
" 17	1,958,000	......	4,250,000	21,600	+
" 17	3,000,000	3,800	3,750,000	9,000	+
" 19	1,850,000	6,600	2,700,000	30,700	+
" 19	2,500,000	6,000	2,800,000	25,700	+
" 22	1,100,000	4,200	1,160,000	10,850	+
" 22	1,200,000	10,850	1,200,000	18,750	+
" 24	2,000,000	15,000	2,000,000	10,000	−
" 24	2,000,000	11,000	2,250,000	13,000	+
" 26	996,000	6,000	1,100,000	12,600	+
" 26	1,100,000	11,000	994,000	8,600	−
" 24	2,700,000	4,800	2,900,000	12,000	+
" 24	3,000,000	13,000	2,700,000	7,600	−
May 1	714,000	22,800	790,000	56,000	+
" 1	646,000	30,000	730,000	32,000	+
" 3	950,000	38,000	908,000	36,000	−
" 3	832,000	26,000	964,000	38,000	+
" 7	530,000	30,000	710,000	40,000	+
" 7	480,000	13,000	805,000	22,000	+
" 17	2,250,000	31,000	2,470,000	61,000	+
" 17	2,060,000	6,000	3,000,000	61,000	+
" 20	2,300,000	......	2,750,000		+
" 20	2,800,000	.....	2,300,000	.....	−
" 22	16,000,000	20,000	15,000,000	19,000	−
" 22	12,000,000	26,000	17,000,000	26,000	+
Average	2,359,000	19,800	2,759,000	44,540	+

400,000 bacteria per c.c. more in centrifuged milk.
24,740 liquefying bacteria per c.c. more in centrifuged milk.

A perusal of the table will show that on six occasions there were fewer bacteria after separation than before, and on twenty-four occasions more bacteria present after clarification than in the raw milk.

Another striking fact brought out by this investigation is the large increase of liquefying colonies in the separated milk. The bacteria which liquefy gelatine are usually harmful, some are spore-producing germs and they give rise to off flavours in both cheese and butter. Many of this class are present in manure, on particles of fodder, etc., and our results seem to show that these bacteria exist in clumps or masses in such material and the centrifugal process breaks these up and distributes them through the milk.

These results obtained at Guelph are identical with those obtained by Dunbar and Kister, and go to show that centrifugal purification, as far as bacteria are concerned, is ineffectual.

CONTAMINATION OF MILK FROM THE STABLE AIR.

Although it is difficult to separate contaminations from animal and milker and that from the air, it is better to consider the latter source of infection separately, as the number of germs floating in the air depends to a great extent upon the amount of dry fodder and straw that may be used in the stable. If manure is not frequently and thoroughly cleaned out, it gets dry and small particles from it help to swell the number of microbes in the air. The greater disturbance of these dusty fodders at any time, the greater will be the germ content of the air at that time. The following data show the number of bacteria per minute deposited in a 12-inch pail. In series A. (*see Fig. 12*), the exposure was made during bedding; in B. (*see Fig. 13*), one hour after this operation.

Series A.	16,000	13,536	12,216	12,890
	15,340	19,200	23,400	27,342
	42,750	27,820	18,730	12,210
Average...	20,100			
Series B.	483	610	820	715
	1,880	1,987	2,112	1,650
	990	1,342	2,370	1,750
Average....	1,400			

These results indicate that many bacteria are attached to particles of considerable weight, as they soon settle on the floor.

Cows are frequently bedded with dusty straw at the very time when milking is going on, a forkful of straw in some instances that have come under my observation, having been thrust under the cow that was being milked. *Dusty fodders are often thrown down from the loft when the* milking is in progress, filling the stable with dust, every particle of which carries spores of moulds and bacteria. It must be remembered also that very undesirable spores which are very difficult to kill, even by long-continued steam heat, abound in straw and hay.

Much benefit would ensue either from moistening the fodder or from feeding and bedding an hour or so before milking commences, to allow the dust, etc., of the air time to settle. In many of the more modern dairy farms, the stables are thoroughly cleaned and ventilated, the floors sprinkled, and the manure removed from the building before milking commences, or a milking room is provided, into which the cows, one, two or three at a time, are brought for milking. This room is supplied with water, conveniently located, and kept in an absolutely clean condition.

CONTAMINATION OF MILK FROM DAIRY UTENSILS.

Probably more trouble is caused to butter and cheese makers by the use of dirty utensils than any other way. Every article that is brought into contact with milk is at once infected with bacteria. When milk is left in storage cans for some time, a tremendous number of microbes develop, and a vast number of spores or latent forms of bacteria are produced. In this way, vessels are infected, and the bacteria find lodgment in all the cracks and crevices of pails, cans, dippers, strainers, etc. Take any milk-can and run the point of a pen-knife along the seam of the can, and you will find a stinking, cheesy mass, composed very largely of bacteria, all ready to grow and re-produce when fresh milk is poured in the can. Nothing is more difficult to clean than these dairy utensils, with the facilities at hand on the average farm. Scalding with hot water is often insufficient to kill the bacteria on the inner surface of the can, and in the cracks and crevices which are usually present. The following experiments will suffice to show the importance of utensils as a factor in milk contamination. Thus Russell[32] took two cans, one of which had been cleaned in the ordinary way, while the other was sterilized by steaming. Before milking the udder of the cow was thoroughly cleaned and special precautions taken to avoid the raising of dust; and the fore-milk was rejected. Milk drawn into these cans showed the following germ-content:

	Number of Bacteria.	*Hours before Souring.*
Steamed pail........	165 per c.c.	28½
Ordinary pail........	4,265 "	23

The writer[35] has also shown the great differences in the bacterial content of cans by a bacterial analysis of the can washings. Cans were rinsed with 100 c.c. of sterile water, and numerical determination of this rinsing water was made.

The following data are from cans poorly cleaned, (*see Fig. 14*), cans washed in tepid water and then scalded—the best farm practice—and cans washed in tepid water and then steamed for five minutes (*see Fig. 15*).

BACTERIAL CONTENTS OF CANS CLEANED IN VARIOUS WAYS.

	Number of Bacteria per c.c. of Can Washings.		
Poorly Cleaned.......	238,500	342,800	215,400
	618,000	806,000	510,000
	230,000	600,000	418,000
Average..442,000.			
Ordinary Method......	89,000	84,000	26,000
	24,000	38,000	76,000
	15,000	44,000	93,000
Average..54,300.			
Approved Method.....	1,100	1,800	890
	355	416	725
Average....880.			

All cans should be constructed so as to facilitate cleaning. Stamped pails, without seams, may now be purchased, but if seams are present they should be examined to see that they are well flushed with solder The bottoms of all cans should be concave, and not convex, to expedite cleaning.

After thorough rinsing to remove organic matter, cans should be washed in hot water, to which borax or soda may be added. After washing, rinse with boiling water, and if available place the can over a steam-jet for a few minutes.

THE EFFECT OF TEMPERATURE.

After the milk is infected with bacteria, the temperature at which it is kept exerts an important influence on the rate of growth or multiplication of the bacteria in the milk. Freudenreich[65] obtained some milk, which when delivered at his laboratory two and a-half hours after the milking, had 9,300 germs per c.c. Samples were stored at 15°, 25° and 36° C. (or 59°, 77° and 95° F.), and the results were as follows per c.c.:

	15°	25°	35°
3 hours......	16,000	18,000	30,000
6 "	25,000	172,000	12,000,000
9 "	46,500	1,000,000	35,280,000
24 "	5,700,000	577,000,000	50,000,000

If we analyze this table we find that at 15° the increase during the first half hour was 700, or 7 per cent., which would indicate that the average duration of a generation is thirteen hours. In three hours more, the increase is 15,000, or 150 per cent., which gives the average duration of a generation as two hours. In the next three hours, the increase is 21,500, and the average duration of a generation about 3½ hours. From the ninth to the twenty-fourth hour, the average is about 2½ hours. At 25°, the times required for a doubling of the number are for the successive periods, about half an hour, about an hour, and about 7½ hours. At 35° the time occupied in a generation was about twenty minutes at first, then about forty-five minutes, and at last thirty-seven hours. These results are curious, but could only be explained by a fuller knowledge of the species concerned, and of the cause influencing the changes.

The most important point brought out in this experiment is the tremendous rate of increase at the higher temperatures; therefore, much may be done to restrain this rapid multiplication by cooling the milk as rapidly as possible. Milk allowed to cool naturally takes some time before it reaches the temperature of the air. Hence, measures should be promptly taken to reduce the temperature quickly.

CERTIFIED MILK.

Of late years a number of sanitary, model dairies have been established in the vicinity of large cities in various parts of the United States and Canada, (*see Figs. 16 and 17*), which have placed on the market, milk with a relatively low bacterial content. Such milk is known as "hygienic," "sanitary," or "certified." It is interesting to note that these establishments prosper, an indication that the discriminating public appreciate the honest endeavour of these dairies to produce milk which will fulfil the requirements of the most exacting sanitarian.

These establishments put into practice the suggestions made by various experimenters and investigators, as the result of their experimental inquiries, and these have been more or less briefly outlined in this paper. The freedom from bacteria obtained in these dairies depends on the thoroughness with which all details are carried out. Russell[52] has shown that when samples of milk are secured under as nearly aseptic conditions as possible, the germ content was 330 organisms per c.c.; but when drawn under ordinary conditions, the bacterial content was 15,500 organisms per c.c. Marshall[66] gives similar results, for example, milk drawn under aseptic conditions averaged 295 bacteria per c.c.; under ordinary conditions, 786,000 per c.c.

In certain cases milk coming from sanitary dairies is endorsed by a board of examining physicians and experts. Thus, the Milk Commission[67] of the Medical Society of the County of New York endorses milk from various dairies when the acidity of the milk is below 0.2 per cent., and when the milk contains less than 30,000 bacteria per c.c. The Milk Commission of the Philadelphia Pediatric Society give their endorsement for milk free from pus and injurious germs, and having not more than 10,000 germs per c.c.

Such milk naturally has enhanced keeping qualities, and milk and cream from several such hygenic dairies in the United States were shipped to the Paris Exposition in 1900, arriving in good condition after 15 to 18 days in transit.

The adoption of a numerical standard seems a very necessary step. Bitter suggests that 50,000 organisms per c.c. should be a maximum limit in milk intended as human food. Park thinks that any intelligent farmer, with sufficient cleanliness and a low temperature, can supply milk averaging not over 100,000 bacteria per c.c., when twenty-four hours old, and suggests that the sale of milk should be so regulated that that containing more than this number per c.c. should be excluded from the market. Rochester, N.Y., has already tried the enforcement of this standard, with good results. In the opinion of Russell, "the practical difficulties to contend with in establishing a milk standard based upon a quantitive bacterial determination are such as to render its general adoption extremely problematical." On the other hand, this investigator advocates the employment of the acid test, and postulates that milk should not contain more than 0.2 per cent. figured as lactic acid, and if possible the acidity should be brought down to 0.15 per cent.

To conclude, from what has been brought up before you, it is undoubtedly easy to see the reason for cleanliness in all operations connected with the dairy business. "All the results of scientific investigation," says Fleischmann, "which have found such great practical application in the treatment of disease, in disinfection, and in the preservation of various products, are almost entirely ignored in milking," and the only remedy for this state of affairs is "a campaign of education among the farmers who produce milk, concerning, first, the simple protection of a readily putrescible fluid from pollution with dirt or other elements of decay; and, second, the sanitary protection of milk from infection."[68]

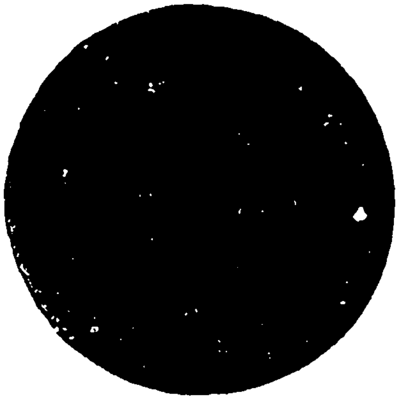
REFERENCES.

1. SOXHLET. Deut. Viertjschr, f. off. Gesundhtspflg, Bd. 24 (1892), pp. 8-75.
2. UHL. Zeitsch. f. Hyg. Bd. 12, (1892) p. 475.
3. BACKHAUS. Milch Zeitung, 26, (1897), p. 357.
4. RENK. Cent. f. Bakt., Bd. 10, (1891), p. 193.
5. HIRD. Report Health Officer, District of Columbia, 1895.
6. CLAUSS. Inaug. Diss., Wurzburg, 1899.
7. KNOPF. Cent. f. Bakt., Bd. 6, (1888), p. 553.
8. BUJWID. Quoted from 7th Report Storrs' Experimental Station, Conn., (1894), p. 70.
9. GEUNS. Neder. Tydschr. v. Geneesk., 2 R., XXI., (1885), Arch. f. Hyg. 3.
10. RENK. Cent. f. Bakt., Bd. 10 (1891), p. 193.
11. UHL. Loc. cit.
12. KNOCHENSTIERN. Inaug. Dissert. (1893), Dorpat.
13. HELLENS. Inaug. Dissert. Helsingfors (1899), Cent. f. B. II., VI., 261.
14. ROWLAND. British Medical Journal, II. (1895), p. 321.
15. SACHARBEKOFF. Cent. f. Bakt. 2 Abt., Bd. 2 (1896), p. 545.
16. CONN. Report Secretary Conn. Board of Agriculture, 1895.
17. SEDGWICK AND BATCHELDER. Boston Medical Surgical Journal, Jan. 14, 1892.
18. PARK. Science, n. ser., 13 (1901), p. 322.
19. LEIGHTON. Science, n. ser. 11, (1900), p. 461.
20. MCDONNELL. Penn. Department Agriculture Report (1897), p. 561.
21. LOVELAND AND WATSON. Report Experimental Station, Storrs, Conn. (1894), p. 72.
22. RUSSELL. Dairy Bacteriology, 3rd Edition, p. 59.
23. HARRISON. Ontario Agricultural College and Experimental Farm Report 22, (1896), p. 107.
24. ECKLES. Iowa Experimental Station, Bulletin 59, 1901.
25. HARRISON. Ontario Agricultural College and Experimental Farm Bulletin 120, May, 1902.
26. SCHULTZ. Archiv f. Hyg., Bd. 14, (1892), p. 260.
27. GERNHARDT. Inaug. Dissert. Dorpat, 1893.
28. FREUDENREICH, E. VON. Dairy Bacteriology, London, 1895.
29. BOLLEY & HALL. Cent. f. Bakt., II Abt., (1895), pp. 1-795.
30. RUSSELL. Loc. cit.
31. GROTENFELT. The Principles of Modern Dairy Practice, N.Y.
32. ROTCH. Transactions Association American Physicians, IX., (1894), p. 185.
33. MOORE. Bureau of Animal Industry, 12th and 13th Annual Reports, p. 261.
34. CONN. United States Department of Agriculture, Office of Experiment Stations, Bulletin, 25, p. 9.
35. HARRISON. Ontario Agricultural College, Report 22nd (1896), p. 207.
36. PALLESKE. Virchow's Archiv, C XXX., (1894), p 185.
37. HONIGMANN. Zeitsch. f. Hyg. XIV. (1893), p. 207.
38. KNOCHENSTIERN, H. Inaug. Dissert., Dorpat, 1893.

39. RINGEL. Munch. Med. Wochenschr., (1893), No. 27.
40. MOORE AND WARD. Cornell University Agricultural Experimental Station, Bulletin 158.
41. WARD. Journal App. Micr. I., p. 205.
42. WARD. Cornell University Agricultural Experimental Station, Bulletin 178.
43. CHESTER. Determinative Bacteriology. N.Y., 1901.
44. WRIGHT. Memoirs Nat. Acad. Science, VII. (1895), 246.
45. FORD. Transactions Association American Physicians, XV. (1900), p. 389. Journal of Hygiene, 1901.
46. FOKKER. Fortschr. d. Medecin. VIII. (1890), p. 7.
47. FREUDENREICH, VON. Ann. de Micrographie, III. (1891), p. 118.
48. BRIEGER AND EHRLICH. Zeitschr. f. Hyg., XIII., (1893), p. 336.
49. WASSERMANN. Zeitsch. f. Hyg. 18 (1894), p. 235.
50. SMITH. Journal Mass. Association Boards of Health, II. (1892), p. 23. (Quoted by Sedgwick (68).
51. WÜTHRICH AND FREUDENREICH. Cent. F. Bakt., II. Abt. 2., (1895), p. 873.
52. RUSSELL. Dairy Bacteriology, 5th Edition (1902), p. 36.
53. ECKLES. Quoted from Russell's Dairy Bact., p. 38.
54. HARRISON. Cent. f. Bakt., II. Abt., 5 (1899), p. 183.
55. DRYSDALE. Transactions Highland and Agricultural Society, Scotland, 5 Ser., 10 (1898), p. 166.
56. Journal Royal Agricultural Society, Eng., (1900), p. 468.
57. SCHUPPAN. Molk. Zeit. 7, (1893), p. 241. Cent. f. Bakt., XIII., (1893) p. 155.
58. BACKHAUS. Milch Zeit. (1897), p. 357.
59. DUNBAR AND KISTER. Milch Zeit. (1899), p. 753. 787.
60. POPP AND BECKER. Hyg. Rundsch., 3 (1893), pp. 530-534.
61. SCHEURLEN. Cent f. Bakt., XI., (1892), p. 54.
62. FLEISCHMANN. The Book of the Dairy, p. 98.
63. CONN. The Dairy (1902), p. 21, London, Eng. Agricultural Bacteriology (1901), p. 189, Phila.
64. NIEDERSTADT. Chem. Centbl. I (1893), p. 396.
65. FREUDENREICH. Ann. de Microg., 2 (1890), p. 115.
66. MARSHALL. Michigan Agricultural Experimental Station, Bulletin 182, 1900.
67. PEARSON. Seventeenth Report Bur. An. Ind., Washington, D.C. (1900), p. 191.
68. SEDGWICK. Principles of Sanitary Science, N.Y., 1902.

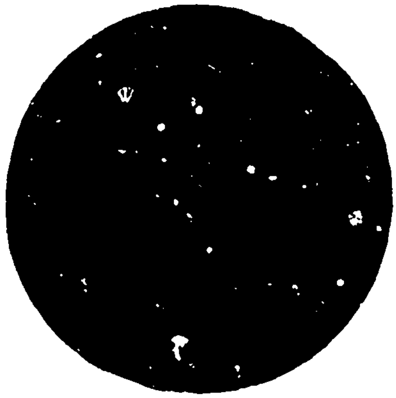
NOTE.—I desire to acknowledge the courtesy of Mrs. Massey, Dentonia, for the use of Figs. 16 and 17; Major Alvord, of the United States Department of Agriculture, for Fig. 6; The City Dairy Co. for Fig. 11; The Kjobenhavn Maelkforsyning for Figs. 9 and 10.

Fig. 1—Contamination from the Fore Milk.



Gelatine Plate shewing colonies of bacteria in $\frac{1}{15}$ c. c. (about 2 drops) of the fore milk. (First few streams from all four teats).

Fig. 2—Bacterial Content of Milk, the Fore Milk being Rejected.

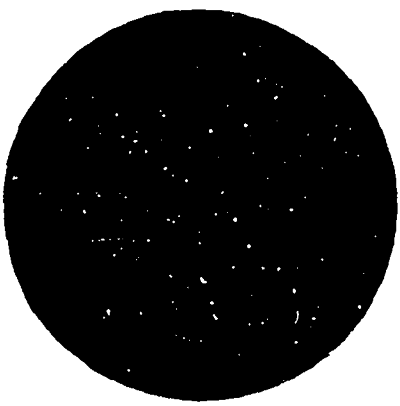


Gelatine Plate shewing colonies of bacteria in $\frac{1}{15}$ c. c. (about 2 drops) of milk taken from the middle of the milking.

Fig. 3.

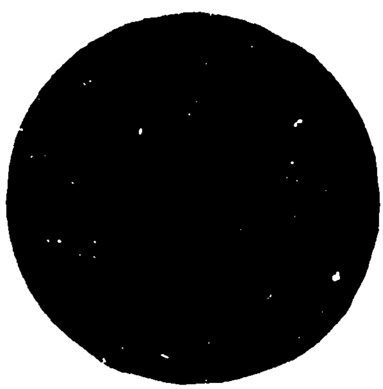
Shewing the bacterial contamination from hairs.

Fig. 4—Contamination from Animal and Milker.



Gelatine plate exposed under the udder of a cow for one minute, while milking under ordinary conditions.

Fig. 5—Contamination from Animal and Milker.



Gelatine plate held under cow for one minute while milking. The udder and flanks well moistened with water.

FIG. 6—SANITARY MILK PAIL AND MILKER PROPERLY CLOTHED.

FIG. 7—THISTLE MILKING MACHINE.

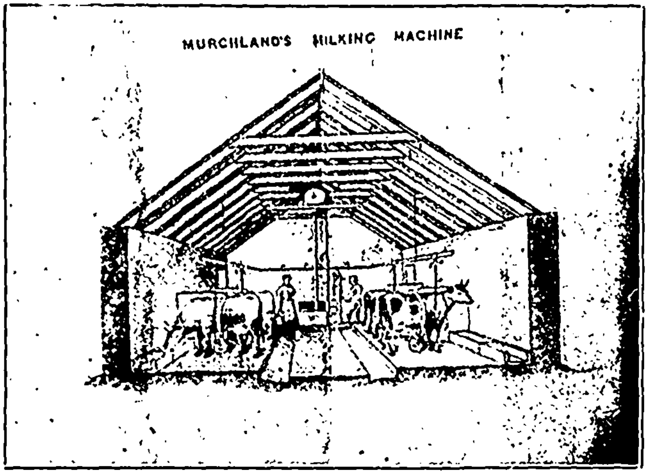


Fig. 8—Murchland Milking Machine.

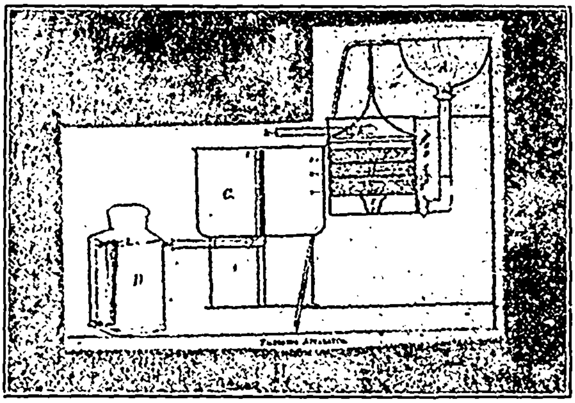


Fig 9—Section of Copenhagen Gravel Filter.

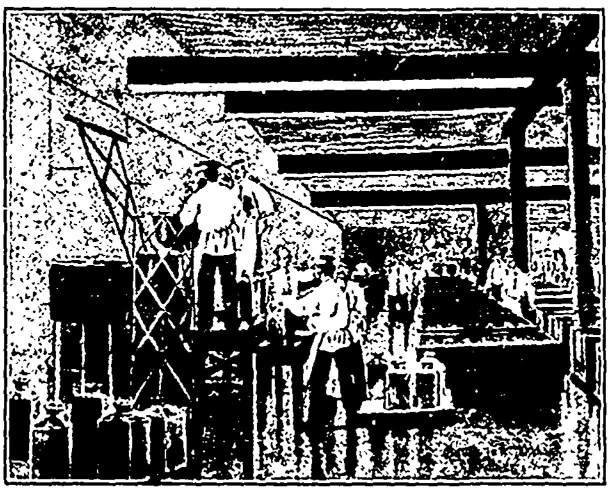


Fig. 10—Filtering Milk Through Gravel.

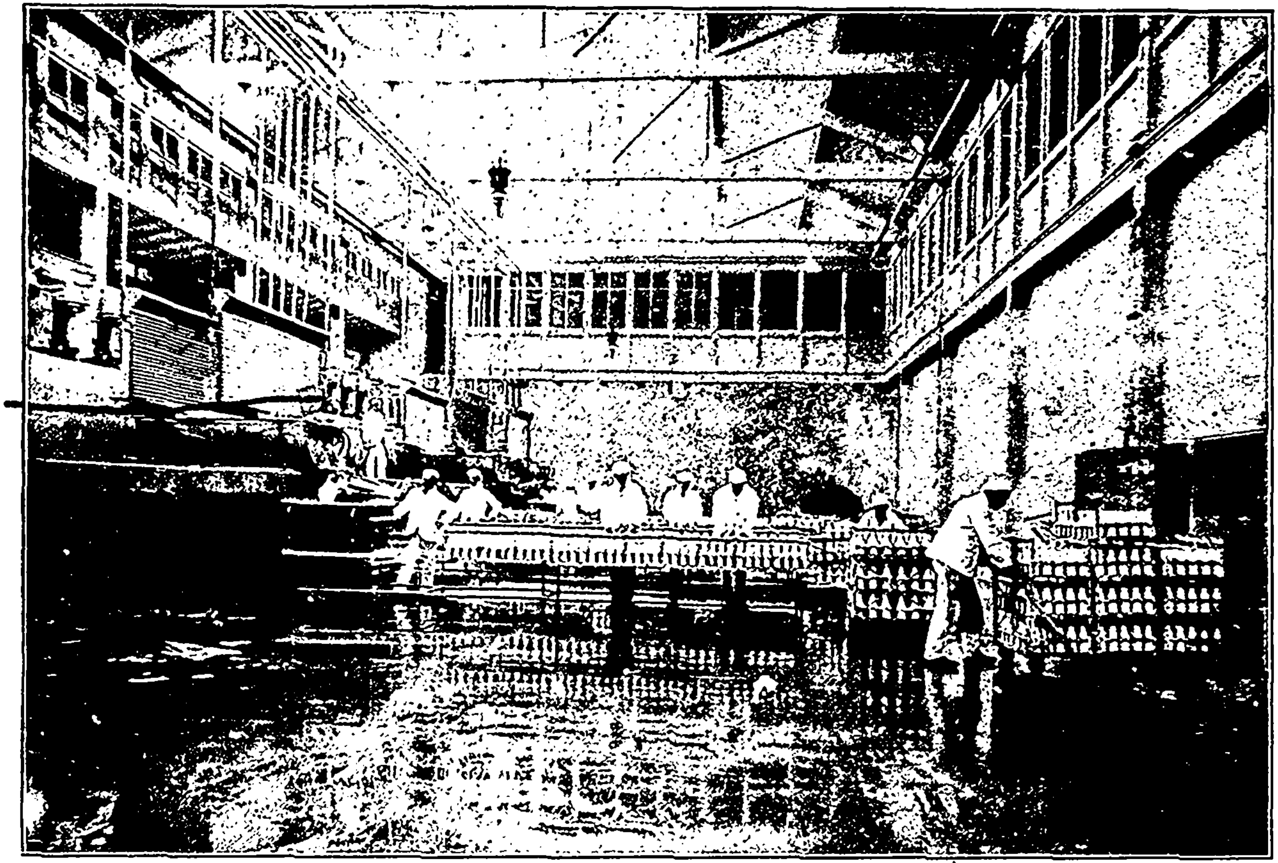


FIG. 11—CLEANING MILK BY CENTRIFUGAL FORCE. S. SEPARATORS.

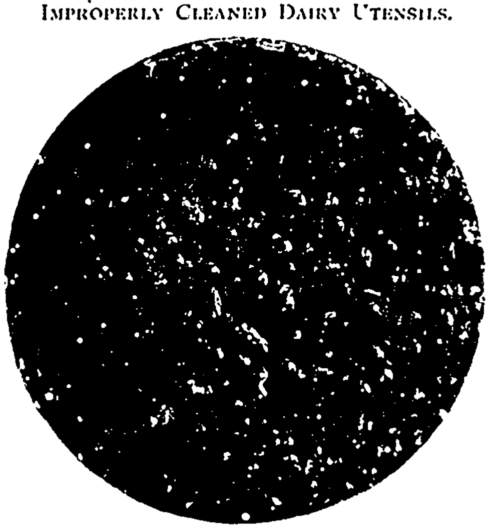
Fig. 12—Germ Content of Barn Air During Bedding, Cleaning Up, Feeding, Etc.

Gelatine plate exposed to the deposition of germs for one minute in a stable when some of the above operations were in progress.

Fig. 13—Germ Content of Barn Air During Bedding, Cleaning Up, Feeding, Etc.

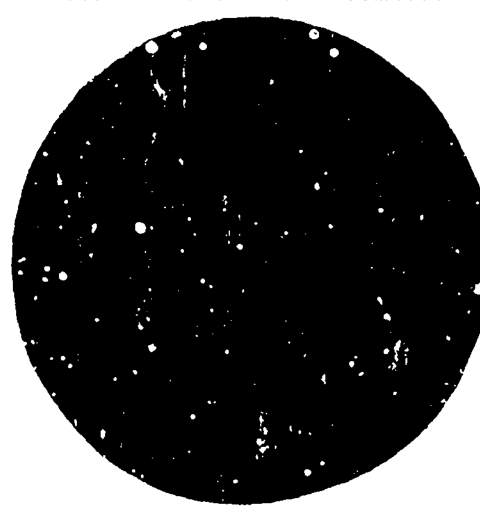
Gelatine plate exposed to the deposition of germs for one minute in a stable when all the above operations had been completed.

Fig. 14—Contamination from the Use of Improperly Cleaned Dairy Utensils.



After a can had been washed with warm water, and thoroughly drained, a quantity of sterile water was added and the can well rinsed. This gelatine plate was made from $\frac{1}{100}$ c. c. (a very tiny drop) of this water.

Fig. 15—Contamination from the Use of Properly Cleaned Dairy Utensils.



After a can had been washed, scalded and steamed for five minutes, and thoroughly drained, a quantity of sterile water was poured in and the can well rinsed. This gelatine plate was then made from $\frac{1}{3}$ c. c. (about six drops) of this water.

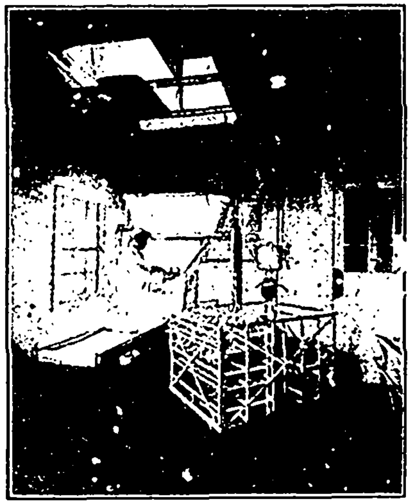


FIG. 16—A SANITARY DAIRY. FILLING BOTTLES.

FIG. 17—A SANITARY DAIRY. BOTTLE WASHING AND BOTTLE STEAM STERILIZER.

THE CHEMISTRY OF WHEAT GLUTEN.

BY GEO. G. NASMITH, B.A.

(Read 26th April, 1902.)

I.—HISTORICAL.

THE first preparation of gluten from wheat flour by washing away the starch from dough seems to have been made by Becari,[1] but Einhof[2] was the first to give special attention to its composition. He extracted wheat gluten with dilute alcohol, and he found that the substance which precipitated on cooling, diluting or concentrating the solution was practically identical with gluten itself.

Taddei[3] named the portion soluble in alcohol gliadin, the residue zymom.

Berzelius[4] thought that he found a second constituent in the part of the gluten soluble in alcohol, which he called mucin, and which was precipitated by acetic acid. He[5] regarded Taddei's gliadin as identical with the substance obtained by Einhof from wheat, barley and rye. The insoluble residue Berzelius called plant albumin, from its great similarity to animal albumin.

De Saussure[6] found that wheat gluten contained about 20 per cent plant gelatin, or glutin, as he proposed to call it, 72 per cent. insoluble plant albumin, and 1 per cent. mucin; the latter, although differently prepared, he considered to be similar to the mucin of Berzelius, and it had, as he thought. the power of transforming starch into sugar.

Boussingault,[7] like Einhof, considered that part of the gluten soluble in alcohol to be identical with the entire gluten proteid.

Liebig[8] named the portion of the gluten insoluble in alcohol plant fibrin; he rejected the term zymom given by Taddei, and also that of plant albumin of Berzelius, in the latter case because solubility in water is a characteristic of albumins. The portion soluble in alcohol he called plant gelatin, and considered it to be a casein-like compound of a proteid with an undetermined organic acid.

Bouchardat[9] found in gluten a substance soluble in extremely dilute acid, which he named albumin, since he regarded it as forming the chief constituent of egg albumin, blood fibrin, casein and gluten.

Dumas and Cahours[10] found four proteids in flour, namely, an albumin which was obtained from the water used in washing out the gluten; plant fibrin left as a residue on extracting gluten with alcohol; a proteid from this alcohol which separated on cooling, and finally a second proteid which precipitates from the same alcohol on concentration and cooling. This latter he called glutin.

Mulder[11] prepared plant gelatin by extracting gluten with alcohol, filtering hot, allowing to cool and redissolving the white precipitate which settled out twice. This he considered to be a compound of sulphur with protein, and he found that it did not contain phosphorus.

Von Bibra[12] stated that on exhausting gluten with hot alcohol insoluble plant fibrin remained behind, while plant gelatin and plant casein dissolved; the plant casein separated on cooling. These bodies he thought had the same elementary composition, and were in fact isomers.

Günsberg[13] held that gluten was composed of three proteids, gliadin being a mixture of two. These were, (a) gluten fibrin, soluble neither in alcohol nor warm water; (b) gluten casein, insoluble in hot water but soluble in alcohol; (c) gluten gelatin, soluble in alcohol and hot water.

Ritthausen[14] found four proteids in gluten, namely, gluten casein, gluten fibrin, plant gelatin or gliadin, and mucedin, of which the last three are soluble in dilute alcohol. His casein was prepared by extracting gluten with boiling alcohol, cooling, exhausting the casein which settled out with absolute alcohol, then with acetic acid, and finally neutralizing the clear filtrate from this with ammonia. The decanted alcoholic fluid from the casein contained the gelatin, which separated on evaporation.

Scherer[15] digested gluten with artificial gastric juice and observed that the greater part went into solution in about fourteen hours.

Martin[16] found that only one proteid was extracted from gluten by dilute alcohol or hot water, which gave the reddish violet reaction of proteoses and peptones. Because of this reaction and its comparative insolubility he called it insoluble phytalbumose. The residue was coagulated by boiling water, and was soluble only in acids and alkalies. He claimed that dilute alcohol extracted only fat from dry flour, and came to the conclusion that insoluble phytalbumose was produced from a soluble albumose, and gluten fibrin from a globulin by pre-existing ferments.

Chittenden and Smith[17] made preparations of gluten casein according to Ritthausen's method, which averaged 15.86 per cent. of nitrogen.

Osborne and Voorhees[18] in an exhaustive research brought many opposing views into harmony. Like Martin they found only one proteid in gluten that was soluble in alcohol, and considered that the various proteids claimed by previous investigators to have been soluble in alcohol were impure preparations, perhaps mixtures with fat. Martin's gluten fibrin they termed glutenin, and found its composition to be practically identical with that of gliadin, a conclusion that had not hitherto been suggested. The high percentage of nitrogen they thought due to their improved method of preparation by which all starch, etc., had been removed. Contrary to Martin's experience they found that dilute alcohol extracted gliadin directly from flour.

Osborne and Voorhees further arrived at the conclusion that gluten is made up of two forms of the same proteid, one being soluble in cold dilute alcohol and the other not. They found that flour exhausted with sodium chloride solution yielded the same amount of gliadin as was obtained from the gluten made from an equal quantity of flour, or by direct extraction of the flour with 70 per cent. alcohol. They, therefore, held that gliadin exists as such in the seed.

Teller[19] noted again the fact that gliadin possessed proteose-like characters, as previously stated by Martin. Gliadin he found to be slightly soluble in dilute salt solution, and he regarded it as identical with that body classified by Osborne and Voorhees as proteose.

O'Brien[20] found himself in agreement with Osborne and Voorhees in considering that gluten pre-existed as such in flour in the same proportions as in gluten, and that there was but one mother substance in flour which gave rise by a process of hydration to gluten. His conclusions were, (*a*) that the differently described derivatives of gluten soluble in alcohol merge into one another; (*b*) that the portion soluble in alcohol may be made to pass into the insoluble stage; (*c*) that a proteose is readily formed as a secondary product from gluten.

Fleurent,[21] whose work, like that of O'Brien, was unfinished when Osborne and Voorhees' results were published, found himself also in agreement with the latter in concluding that only one body soluble in alcohol was present in gluten. Besides gliadin and glutenin he found a very small quantity of a third body which he called conglutin.

Morishima[22] found only one proteid in gluten (that he named artolin) which he prepared by kneading gluten with dilute alkali and treating the decanted fluid with hydrochloric acid until it contained one per cent. of the acid in excess. The precipitate washed with one per cent. hydrochloric acid and made up with alcohol to a solution containing 70-80 per cent. of alcohol, was filtered and saturated with absolute alcohol and ether.

Its composition he found to be:

ARTOLIN (MORISHIMA).

C	52.29
H	7.02
N	16.51
S	.74
O	....
Cl	1.57

Mayer[23] held that the old name of plant gelatin was inappropriate, since it was chemically unlike gelatin: the latter does not contain sulphur, and is insoluble in dilute alcohol.

Ritthausen[24] commented on Fleurent's and Osborne and Voorhees' careful work, but, nevertheless, clung to the views propounded in his early paper. In criticizing the work of Morishima, he pointed out the fact that proteids are soluble in dilute acids, and concluded that artolin was glutenin linked with hydrochloric acid.

The subjoined analyses, taken from the paper of Osborne and Voorhees were found by reference to the original papers to be accurate:

	BOUSSINGAULT. Gliadin.	JONES. Plant. Gelatin.	DUMAS and CAHOURS. Casein.	DUMAS and CAHOURS. Glutin.	MULDER. Plant. Gelatin.	VON BIBRA. Plant. Gelatin.
C	52.30	54.44	53.46	53.27	54.85	53.57
H	6.50	7.42	7.13	7.17	7.05	7.12
N	18.90	15.98	16.04	15.94	15.71	15.57
S	....	....	....	....	.60	.88
O	22.30	22.16	23.37	23.62	21.79	22.86
	100.00	100.00	100.00	100.00	100.00	100.00

	Gessberg. Plant. Gelatin.	Ritthausen. Gluten fibrin.	Plant. Gelatin.	Mucedin.	Osborne and Voorhees. Gliadin.
C..........	52.68—52.65	54.31	52.76	54.11	52.72
H..........	6.77 — 6.88	7.18	7.10	6.90	6.86
S..........	....	1.01	.85	.88	1.14
N..........	17.76—17.45	16.89	18.01	16.83	17.66
O	22.79—23.02	20.61	21.08	21.48	21.62
	100.00-100.00	100.00	100.00	100.00	100.00

GLUTENIN.

	Jones.	Scherer.	Dumas and Cahours.	Von Bibra.	Ritthausen.
C.......	52.79	54.60—52.34	53.37—53.23	55.57	52.94
H.......	7.02	7.45— 7.13	7.02— 7.01	6.95	7.04
N.......	15.59	15.81—15.36	16.00—16.41	15.70	17.14
S.......	....	....	.. ....	1.02	.96
O.......	24.62	22.14 -25.17	23.64—23.35	22.76	21.92
	100.00	100.00-100.00	100.00-100.00	100.00	100.00

	Chittenden and Smith.	Osborne and Voorhees.
C	52.87	52.34
H....	6.99	6.83
N...........	15.86	17.49
S....	1.17	1.08
O	23.11	22.26
	100.00	100.00

II.—OBSERVATIONS.

While working at the composition of wheat flour, Professor Macallum suggested that I should trace to its source the phosphorus which he found to be present in the cellular elements of the wheat grain. It was sought for and found in gluten, no matter how carefully prepared or how long it had been washed in tap or distilled water. Gluten was prepared in the usual manner by kneading dough in a stream of water until free from starch, dried at 110 C. until the weight was constant and the phosphorus estimated according to Neumann's[25] method, which was found by experiment on known solutions of phosphoric acid to be perfectly accurate. Two quantities of gluten yielded 0.11 and 0.12 per cent. of phosphorus respectively.

In order to determine next which of the constituents of gluten (*i.e.*, gliadin or glutenin) contained phosphorus, gliadin was prepared by extracting starch-free gluten with 70 per cent. alcohol, filtering the solution *repeatedly, and afterwards evaporating completely to dryness.*

Average of five estimations 0.83 per cent. ash.
" two " 0.29 " phosphorus.

Gliadin was prepared by extracting gluten with 70 per cent. alcohol, filtering and diluting with twice its volume of 1 per cent. sodium chloride solution; the white precipitate, separating out, was collected, washed with distilled water, till free from chlorine, and dried at 110 C. The analyses gave :—

	I.	II.	III.	IV.
Phosphorus........	0.19	0.19	0.18	......
Ash..............	0.205	0.201	......	......
Nitrogen	17.705	17.435	17.64	17.555

The ash from these was dissolved with hydrochloric acid; the solution evaporated almost to dryness in a platinum crucible, was diluted with distilled water, and treated with a quantity of dilute hydrochloric acid containing also potassium ferrocyanide. A blue colouration immediately indicated the presence of iron; repeated trials invariably yielded the same result.

In order to determine whether the iron was organic or inorganic, a solution of gliadin in ammonia-free distilled water was added to a solution of hæmatoxylin. No darkening whatever occurred, showing that the iron must be organically combined. Inorganic iron salts with hæmatoxylin give an intense dark blue colour. Pieces of freshly-prepared gliadin, suspended in hæmatoxylin, gave no reaction in thirty hours. *The iron, like the phosphorus, must be in organic combination.*

Previously to this I had found that on digesting gluten with artificial gastric juice, and repeatedly renewing the fluid, a part remained insoluble even after two months. This residue, after extracting with absolute alcohol and ether, was dissolved in 0.2 per cent. sodium hydrate, and precipitated by 0.2 per cent. hydrochloric acid, the precipitate being insoluble *in excess of the acid. Evidently this was a nuclein, and must* have come from the gliadin or glutenin of the gluten.

A gram of gliadin, purified by precipitating, dissolving, reprecipitating, and extracting with absolute alcohol and ether, was digested with artificial gastric juice at 38°C. A residue remained which gave all the reactions for nuclein, and undoubted reactions also for organic iron and phosphorus.

A large amount of gliadin was now prepared by extracting gluten with 70 per cent. alcohol, filtering, concentrating to a small quantity, precipitating with 95 per cent. alcohol, extracting in the Soxhlet apparatus for sixteen hours with absolute alcohol to remove fat and lecithin, and finally drying for three hours at 110°C.

Analyses gave the following:—

GLIADIN.

C.............	52.39	Av. 6.
H.............	6.84	Av. 6.
N.............	17.46	Av. 2.
S.............	1.12	Av. 2.
O.............	21.89	
P.............	0.267	Av. 2.
Fe	0.034	Av 2.
	100.00	

The iron was determined gravimetrically since the amount was so small that only a few drops of 1 40 normal solution of potassium permanganate were necessary by the volumetric method, and the exact end point was consequently difficult to determine. Taking all necessary precautions to eliminate aluminium and calcium, results were obtained by extracting the iron from the ash, which were concordant with those obtained from the filtrate after precipitating the phosphorus as ammonium phospho-molybdate. The weight of ferric oxide seldom exceeded 0.6 milligram. The analyses in other respects agree very well with those of Osborne and Voorhees, except that the carbon and nitrogen contents are slightly lower, and that they obtained no phosphorus.

A large quantity of gliadin was prepared and digested at 38°C. with artificial gastric juice in litre flasks. Digestion was continued for three weeks, the flasks being frequently shaken, and the clear supernatant fluid renewed several times. The considerable residue was collected on filters, washed free from proteoses and peptones with water, then with 70-95 per cent. alcohol which removed some fat. The residue dissolved in 0.2 per cent. sodium hydrate solution, was filtered, and the solution precipitated with excess of dilute hydrochloric acid, the process of solution and precipitation being repeated several times; the precipitate was then collected on "hardened" filters and washed with distilled water till free from chlorides. Extracted with absolute alcohol in the Soxhlet apparatus for sixteen hours, dried at 110 C., and analyzed, the residue yielded the following results:—

GLIADIN NUCLEIN.

C	49.47	per cent.	Av. 2.
H	6.98	"	Av. 2.
N	16.60	"	Av. 2.
S	0.80	"	One.
P	0.29	"	Av. 2.
Fe	0.04	"	Av. 2.
O	25.82		
	100.00		

Ash .... 0.24 per cent. Av. 2.

The amount of phosphorus was very small, practically the same, in fact, as the gliadin from which it was prepared. The chemicals used were carefully tested in blank experiments, but no trace of phosphorus was found in any of them. Possibly the prolonged digestion with frequent renewals of hydrochloric acid solution had removed some of the phosphorus. The result was, however, quite unsatisfactory, since it was to be expected that the amount of phosphorus and iron would be much greater than in the substance from which it was derived. The analyses of the gliadin and the nuclein, derived from it, may be compared side by side :—

	Gliadin.	Gliadin Nuclein.
C............	52.39	49.47
H	6.84	6.98
N	17.47	16.60
S	1.12	0.80
O	21.89	25.82
P..........	0.267	0.29
Fe	0.034	0.04
	100.00	100.00

From this it may be gathered that the two compounds are quite distinct chemically as well as physically.

Glutenin was prepared, as recommended by Osborne and Voorhees[18] by extracting all the gliadin from gluten by dilute alcohol, dissolving the residue in 0.2 per cent. potassic hydrate, and precipitating by exactly neutralizing with 0.2 per cent. hydrochloric acid; the precipitate washed with 70-95 per cent. alcohol, was again dissolved in 0.2 per cent. potassic hydrate and filtered perfectly clear through heavy filter paper in an ice chest. Precipitated from the solution by exact neutralization with 0.2 per cent. hydrochloric acid, washed with distilled water till free from chlorides, then with 70-95 per cent. alcohol, extracted in the Soxhlet apparatus for ten hours with absolute alcohol, and dried for three hours at 110 °C., the glutenin so prepared gave on analysis the following :—

GLUTENIN.

	NASMITH.	OSBORNE.
C	52.75	52.34
H	7.22	6.83
N	16.15	17.49
S	1.06	1.08
O	22.58	22.26
P	0.215	....
Fe	0.026	....
	100.00	100.00

Ash 0.188 per cent.

Another preparation by Fleurent's method[21] which was also carefully filtered, yielded 16.55 per cent. nitrogen. The figures are not at all in agreement with those of Osborne and Voorhees for this compound. Mine are considerably higher in carbon and hydrogen, and much lower in nitrogen, a result which might be accounted for by carbohydrate impurity. Since, however, it was prepared exactly as described by him this seems unlikely. The fact that the amount of iron and phosphorus is practically the same as in gliadin at once suggested the possibility of these elements being derived from a certain amount of nuclein mechanically carried along with these compounds in the attempted purification process.

Impure glutenin was digested with pepsin and hydrochloric acid, but the insoluble residue was so difficult to separate from soluble starch, and was so evidently impure that the complete analysis was not made, though the presence of iron and phosphorus in it was demonstrated.

In repeating the work of Morishima[22] a copious precipitate as usual occurred at the neutral point, but when more acid was added nearly all went into solution; after twenty-four hours only a trace of precipitate settled out. Glutenin has again and again been shown to be soluble in dilute acids. Artolin, as I found, is derived from another source than Morishima supposed. A 0.4 per cent. hydrochloric acid extract of flour was made, filtered perfectly clear and potassic hydrate added until neutral, when a precipitate was thrown down, which proved to be nearly all gliadin. If to this 0.4 per cent. hydrochloric acid extract more acid was added, a precipitate began to appear which increased with the acidity. This in large part separated on heating, and it proved entirely soluble in 70-80 per cent. alcohol, the result showing it to be gliadin.

This property of gliadin, of being precipitated with excess of acid, has not, I think, been hitherto noted. Since the compound of Morishima was prepared in practically the same way artolin is evidently gliadin in acid combination. Glutenin remains in solution. The body obtained under these circumstances by Morishima would perhaps correspond to a proteid salt,[35] *e.g.*, a chloride of gliadin. I obtained the substance called conglutin by Fleurent[21] but in quantity insufficient for analysis.

In order to decide whether the iron and phosphorus in gliadin and glutenin were actually in molecular combination in these compounds, resource was had to the microscope. Grains of Manitoba hard wheat were imbedded in celloidin and sectioned. Macallum's methods for determination of iron[26,27] and phosphorus[28] were used.

For iron the celloidin was removed by equal parts of alcohol and ether, the sections passed through absolute alcohol and inorganic iron salts removed by 2.5 per cent. hydrochloric acid in 95 per cent. alcohol. Sections so treated showed no trace of colour with pure hæmatoxylin in aqueous solution (0.5 per cent.) after the lapse of thirty minutes. The sections now placed in sulphuric acid alcohol (4 vols. acid, 100 alcohol) at 40° C. were removed at intervals of half hours; on washing out the acid, and placing in hæmatoxylin, the sections gave a marked reaction for iron, the organic iron combination having been broken up and the inorganic iron salt formed retained in sitû.

Sections unextracted by hydrochloric acid showed much inorganic iron in the aleuron layer and germ. When this had been removed by hydrochloric acid no colour whatever appeared after standing for twenty minutes in hæmatoxylin solution. After treatment with sulphuric acid alcohol the nuclei of the aleuron and large parenchymatous endosperm cells were stained with hæmatoxylin purplish blue-black. The aleuron cell contents gave no reaction, nor did the proteid matter of the endosperm, which constitutes gluten. Gliadin and glutenin, therefore, do not contain iron in their molecules, and that present must have been derived from the nuclei of the cells of the endosperm and aleuron layer, and possibly in small amounts from embryo cells.

The distribution of iron in the embryo, or germ, is a point of interest. The closely packed cells of the embryo each contained a large nucleus coloured with hæmatoxylin almost black. In the rapidly dividing cells *of the radicle and plumule a diffused purplish blue-black reaction* occurred, which under the highest power could not be identified with any definite granules or structures. Some of the cells, other than those in a rapid state of division, gave a faint purplish reaction, perhaps from iron derived by diffusion from the nucleus.

In order to show the distribution of organic phosphorus the inorganic phosphates were first removed by soaking for half an hour in acetic acid alcohol. Sections removed at the end of this time, placed for a few minutes in the nitric-molybdate solution, and then in one per cent. solution of phenylhydrazine hydrochloride showed no trace of green colouration, this fact indicating that all inorganic phosphates had been removed.

Such extracted sections were now placed in nitric-molybdate solution at 35° C. and removed in series at intervals of half an hour. When placed in a solution of phenylhydrazine hydrochloride for a few minutes, they showed a green colour, which increased in depth with the times during which the section remained in the molybdate solution. In twenty hours the aleuron layer and embryo were stained a bright green. Sections which had had the celloidin removed by alcohol and ether, and which were subsequently extracted with absolute alcohol in the Soxhlet apparatus for several hours, gave exactly the same reactions as those unextracted. Consequently lecithin could not have been present. The aleuron cells in such preparations showed a large nucleus of a much deeper green than the rest of the cells, and under the high power the colour was seen to be confined to the spaces between the aleuron grains, the coloured parts appearing in the form of a network. The network had a more or less punctated appearance, the grains themselves were perfectly colourless.

In the endosperm of such preparations the nuclei alone were coloured, though sometimes, after twenty-four hours, the proteid matter packed between the starch grains, and even the cellulose gave the phosphorus reaction. Possibly phosphorus had diffused from the nuclei. The manner in which the phosphorus is distributed in the different types of embryo cells is quite varied. The palisade-like absorption cells between the endosperm and embryo appeared finely granular and of a uniformly dark green tint. The cytoplasm of the radicle and plumule cells were of a finely granular character, and gave the phosphorus reaction. Around these tightly packed cells of the radicle and plumule were other cells much more loosely connected, whose contents appeared vesiculated. The intercellular material gave a faint phosphorus reaction, while the large granular nucleus was much darker and very prominent.

Between these vesiculated cells and the absorption tissue of the embryo were large cells loosely bound together. These cells, even under the low power, were very different from the others, containing large, well-separated granules, coloured a bright green. Under the high power these granules appeared round, angular, or often crescent-shaped. In very thin sections they were quite separated from one another and very brilliantly coloured; in thin sections the nucleus often was not apparent. In thicker sections these granules were seen to be connected, forming a loose kind of meshwork, the spaces between being filled with a finely granular substance, giving a faint but distinct phosphorus reaction. When treated for the iron reaction a very faint violet tinge appears in these cells, but only between the bodies which stain so brightly for phosphorus.

From this it seems that, with the exception of the rapidly dividing cells such as those of the radicle and plumule, iron is found in the nuclei only of the various cells of the wheat grain.

Phosphorus is more widely distributed, appearing between the aleuron grains; in fine grains in the radicle and plumule cells; in the foam-like mesh work of another type of embryo cell; in the very distinct large granules just described, and in the nuclei of all these cells. From the various ways in which these different cells stain, and the several methods of phosphorus distribution in them, one may conclude that there are probably several nucleins present.

Osborne and Campbell[29] extracted wheat germ with petroleum naphtha, ground the residue to a fine flour, extracted this with water, saturated the clear filtrate with sodium chloride, and subjected the resulting precipitate to a vigorous peptic digestion. The nuclein so prepared, they conclude, " is not an original constituent of the extract nor of the cells of the embryo, but results through several molecules of nucleic acid with one of Protein." To this nuclein, washed with water and dissolved in dilute potassic hydrate solution, was added hydrochloric acid until a precipitate formed, which readily separated. When this was filtered off a considerable excess of hydrochloric acid was further added to the filtrate, whereupon a precipitate of nucleic acid separated out which became so dense and brittle that it could be ground under water.

This operation, as described, I repeated, but a small quantity only of nucleic acid was obtained, which, however, did not become brittle under water. As I expected, the ash of this nucleic acid and of the nuclein, also prepared, gave distinct reactions for iron, even after standing for several weeks under dilute hydrochloric acid, a fact unnoticed by Osborne, and showing that part at least of his nuclein had come from the nuclei of the cells. If this nuclein had been derived from the nuclei of the embryo cells, it must have contained iron, since, as above demonstrated, its presence is invariable in the nucleus. Probably his nuclein was derived both from nuclei and ground substance of the cells.

It may then probably be admitted that the phosphorus and iron invariably found in gliadin and glutenin, no matter how carefully they have been prepared, are present in the form of nuclein or nucleic acid, which have been derived from the nuclei of the parenchymatous endosperm cells chiefly, and carried with them in the purification process. Perhaps aleuron and embryo cells imperfectly separated in the milling process contribute part of them.

III.—PROPERTIES OF GLIADIN.

Gliadin extracted directly from raw flour by dilute alcohol is always contaminated with fat, which gives to its solution a yellow tinge. On diluting this solution with an equal volume of sodium chloride solution, a snow-white precipitate separates, which, if the dilution is sufficient, collects into brownish flocculent masses, and either rises or sinks, according to the strength of the salt solutions. Prepared in this way gliadin is exceedingly viscid, adhering to everything with which it comes in contact. When precipitated by water alone, gliadin will not readily separate. Evaporation of the alcoholic solution and cooling cause a considerable gummy mass of gliadin to separate, while a few drops of sulphuric acid to the supernatant fluid throw down almost all of the gliadin left in solution.

A solution of gliadin evaporated to dryness forms a glue-like brittle, opalescent, yellow mass; hydrated gliadin, exhausted with absolute alcohol and ether, and dried over sulphuric acid, forms a pure white friable mass. Either variety will almost wholly go into solution on warming in dilute alcohol. Gliadin is slightly soluble in distilled water, and then gives the pink biuret reaction; it is not entirely insoluble in dilute salt solutions, as stated by Osborne and Voorhees. In dilute alkalis it readily dissolves, and the greater part of that dissolved separates on neutralizing. Its action with hydrochloric acid is peculiar; it may be extracted directly from flour by dilute acids, filtered perfectly clear, and yet an additional drop of acid throws down a cloudy precipitate which increases in quantity with further addition of acid, but separates completely only on heating; as it cools, however, more or less of the precipitate goes back into solution. A drop of alkali to the acid solution only produces a faint opalescence, which does not increase with additional alkali until the neutral point is reached, when a sudden clouding occurs, and a precipitate settles out on heating.

A cold alcoholic solution of gliadin filtered clear, clouds slightly in twenty-four hours, depositing a small precipitate which increases in quantity with the length of time under alcohol. It is much more soluble in boiling than in cold alcohol, a saturated solution of the former depositing a heavy precipitate on cooling. Heating to 130° C. in the autoclave renders gliadin insoluble in alcohol. In artificial gastric juice at 38° C. it rapidly dissolves, depositing a small amount of nuclein, and yielding a considerable amount of true peptone, as evidenced by the deep red colouration with potassic hydrate and cupric sulphate in the filtrate after removal of proteoses by saturation with ammonium sulphate. It is a unique proteid, in that it gives this red biuret reaction before as well as after digestion. In this particular the name "insoluble phytalbumose" applied to it by Martin[16] does not appear appropriate. The proteid is entirely insoluble in absolute alcohol, and is precipitated by strong alcohol from solutions in weak. Addition of salt to a solution of gliadin in 70 per cent alcohol does not produce precipitation until water is added. Millon's reagent, and nitric acid give the usual proteid reactions.

Gliadin is distributed throughout the endosperm, especially toward the periphery, where the small proteid granules are much thicker and the starch granules they enclose smaller. It is also contained in bran, and probably in aleuron cells as part of the packing between the aleuron grains, for both bran and shorts yield gliadin to dilute alcohol.

IV.—PROPERTIES OF GLUTENIN.

Glutenin is almost completely insoluble in salt solutions, water, and alcohol; readily soluble in dilute acids and alkalis, from which solution the proteid is precipitated unaltered when the solution is rendered neutral to litmus. It has a definite coagulation temperature which lies about 70° C. Gluten dehydrated with absolute alcohol and ether, is very slowly soluble in dilute acids and alkalis, more or less remaining undissolved. Experimental evidence seems to show that glutenin exists as such in the wheat grain. Its composition, according to Osborne, is practically identical with that of gliadin, results differing greatly from those of previous investigators, who had only in one instance obtained from glutenin as much as 17 per cent. of nitrogen.

Osborne considered it an altered form of gliadin, but the fact that it has a definite coagulating point, while gliadin has none, would indicate that it is improbable. No one has yet succeeded in making gliadin assume a form at all resembling glutenin. In my opinion the two proteids are entirely distinct in origin as well as in properties. Osborne states that glutenin is slightly soluble in cold but much more in hot dilute alcohol, the dissolved proteid separating on cooling. Since glutenin is coagulated at about 70° C. the proteid dissolved must have either been due to gliadin imperfectly separated from the glutenin, or to part of the latter split off by heat. The trace soluble in cold alcohol, as Osborne himself hints, may have been gliadin, which is exceedingly difficult to separate from glutenin.

V.—THE FERMENT THEORY OF GLUTEN FORMATION.

The question whether gluten exists as such in flour, or whether it results by the activity of a ferment, is one on which there are considerable differences of opinion. Weyl and Bischoff[30] considered gluten to be formed from pre-existing globulins by a pre-existing ferment in flour. They held that flour extracted by 15 per cent. solution of sodium chloride, and heated to the coagulation point of globulin, gave no gluten. They were, however, unable to isolate the ferment.

Martin[6] thought that gluten did not pre-exist in flour as such, but that his gluten fibrin was derived from a precursor globulin, and his insoluble phytalbumose or gliadin, from a soluble albumose. He stated that gliadin was not extracted directly from flour by 70 per cent. alcohol.

Johannsen[31] advanced arguments against the ferment theory, and thought gluten existed as such in a finely divided state in the wheat grain. He stated that a temperature of 60°C. did not injure the gluten-forming power of flour, and that flour made by mixing dry starch and finely-powdered gluten behaved like ordinary flour.

Ballard[32] maintained that gluten pre-existed as such in flour. Osborne[23] arrived at the same conclusion. O'Brien[20] found that flour heated to 100° C. for thirteen hours gave practically the usual amounts of gluten; also that a paste made with boiling water yielded gluten in apparently normal quantities: that flour left twenty-four hours under absolute alcohol and ether, yielded gluten when these evaporated. He concluded that there is but one compound soluble in alcohol, that the portion soluble in alcohol may be made to pass over into the insoluble stage, and that there exists but one mother substance of gluten in flour.

None of the proofs as to the existence or non-existence of a ferment appear at all conclusive. Dry heat at 100° C. or even 110° C. for several hours does not kill ferments, neither does alcohol for a short period. To prove the non-existence of a ferment presents in this case peculiar and apparently unsurmountable difficulties, but a few facts bearing on the point may be given here.

Seventy per cent. alcohol, cold or hot, applied directly, extracts gliadin from dry flour; warm 95 per cent. alcohol does the same; flour moistened with 95 per cent. alcohol and heated to 80° C. yields abundant gliadin, as does flour stirred into boiling water and then extracted with alcohol. When flour, however, is slowly sifted into boiling water, so that every particle comes into instant contact with water or steam at 100° C. it yields no gliadin to dilute alcohol.

Dough made from flour and boiling water does yield gluten on washing, as stated by O'Brien, but it is smaller in amount and is of irregular consistency. The temperature of the dough when mixed was found to be only 52.5° C. Now glutenin has a definite coagulation point. Martin[16] stated that the residue after extracting gluten with dilute alcohol was coagulated by boiling water. Before noticing his work I had found the coagulation point of glutenin to be about 70° C. When, therefore, a dough was made with boiling water, and only reached the temperature of 52° C. only a comparatively small amount of the flour must have been heated to 70° C., a temperature which coagulates glutenin. Consequently a quantity of gluten would be formed from the portion of the flour not heated to that point. A dough made in this way and gradually heated till it reached a temperature of 80° C. yielded no gluten, proving that its formation depended upon the glutenin not being coagulated.

A dry heat of 110° C. for ten hours does not coagulate proteid, and flour heated to this point still yields gluten; but if flour is heated to 120° C., or even 100° C., for half an hour in the autoclave a dough of little coherence results, and no gluten is obtainable on washing even over silk. The glutenin had been coagulated. In other words any temperature or manipulation that would kill a ferment which might be present would coagulate the glutenin and therefore gluten could not be obtained. The fact that gluten has a definite coagulation point would seem to indicate that it is not derived from the same substance as gliadin. I have never been able to transform one of these compounds into anything at all like the other. With the idea of finding out whether gluten changed into gliadin, I extracted all the latter from flour, let one half stand over night under water and the other under alcohol for twenty-four hours, but neither yielded anything to dilute alcohol.

The fact that ground, dried glutin mixed with starch yielded dough of normal properties, as stated by Johannsen[31] is no proof as to the non-existence of ferment action, since if ferment action were present the dried gluten itself would have been the resultant product of the ferment action.

Flour was slightly moistened with absolute alcohol and heated on a warm bath to 70° C., being stirred all the while with a stout thermometer in order to heat the mixture evenly throughout. Alcohol was used to prevent any possibility of ferment action. After drying in the air, one half was taken and made into a dough, from which, as I expected, gluten could not be obtained. A small quantity of raw flour was intimately mixed with the other half and this was also made into a dough. In this case also no gluten could be obtained. This proved that the formation of gluten depended altogether on whether glutenin was coagulated or not, since the ferment if existing should have been present in the added raw flour.

Now ground air-dried gluten mixed with starch and made into dough yields gluten of normal properties. Such a dough of ground gluten and starch warmed above 70° C. does not yield gluten since the glutenin has been coagulated. Therefore when glutenin which had been already made, as in the the second case, or glutenin, or even its predecessor in the raw flour in the first case, was coagulated, a similar result obtained. The probability, therefore, seems to be strong that glutenin is present in flour as such. And since gliadin is extracted directly from flour or bran with 70 95 per cent. alcohol, cold or boiling, and also by dilute acids or alkalis, it also apparently is present as such in flour, and not derived, as O'Brien[20] holds, from the same parent substance as gluten.

VI.—THE ALEURON LAYER OF WHEAT.

The outer endosperm layer of wheat was stated by Sachs[33] in 1862 to be rich in oil and nitrogenous compounds. Ten years later Pfeffer[34] pointed out the fact that gluten was not derived from the aleuron layer as was commonly believed. He maintained that the high nitrogenous value of the latter was due to substance not proteid in nature, and to adhering endosperm rich in gluten.

Johannsen[31] in 1888 again emphasized the fact that aleuron cells do not contain gluten; he stated that these cells contained nitrogenous granules imbedded in a soft protoplasmic mass, rich in fatty matter.

According to O'Brien[20] the protoplasm of an aleuron cell is continuous with that of adjacent cells, aleuron as well as endosperm. He found oil present in considerable quantities. The individual aleuron grains on addition of water appeared to consist of a central core which was more or less soluble in water, salt solutions, dilute acids and alkalis, and not readily stainable. The layer surrounding this core he found to stain readily with iodine, hæmatoxylin and aniline stains, and to be insoluble in any of the above mentioned reagents.

From an aqueous extract of bran he obtained a coagulable proteid, probably a globulin, and proteose which, when evaporated to dryness, yielded a gelatinous semi-transparent substance, partly separating in small round spherules, regarded by him as artificial aleuron grains, since they gave all the reactions of those imbedded in cell protoplasm.

He also extracted from bran by means of dilute alcohol a proteid which corresponded to gliadin.

Dilute alcohol, I found, extracted gliadin from both bran and shorts. Aqueous extracts of bran gave a globulin coagulable by heat, and also a proteose-like body which was not gliadin. On evaporation of this proteose extract no granule corresponding to O'Brien's artificial aleuron grains could be obtained, although a granular material did separate; the solution at the same time exerted a very strongly reducing action upon Fehling's fluid. I was unable to make out a double coat to the aleuron grains. The substance between the aleuron grains seems to be chiefly gliadin, and contains inorganic iron, calcium salts and phosphorus-holding compounds.

VII.—CONCLUSIONS.

Gliadin and glutenin do not come from the same parent substance, nor are they of the same composition. Gliadin has not a definite coagulation point, while glutenin has. Gliadin is obtained from rye, barley, and maize, and from the bran and shorts of wheat, while glutenin cannot be obtained from these. By chemical or other means one has as yet not been transformed into anything at all resembling the other.

Both gliadin and glutenin invariably give the reactions for organic iron and phosphorus, but are not nucleo-proteids. Under the microscope the gluten matrix in thin sections of wheat does not show any indication of iron or phosphorus, and it must, therefore, be concluded that the organic iron and phosphorus found in gluten are due to nucleins or nucleic acid derived from the nuclei of the large endosperm cells. Probably part is derived from nuclei of the aleuron cells, or of the embryo cells, or from the nucleins present in the cytoplasm of the embryo cells.

Gliadin exists as such in the wheat grain, and the theory of its formation by means of ferment action is not justifiable. Strong alcohol mixed with flour and then diluted with water to a 70 per cent. solution extracts gliadin from it ; boiling alcohol also extracts gliadin from flour or bran.

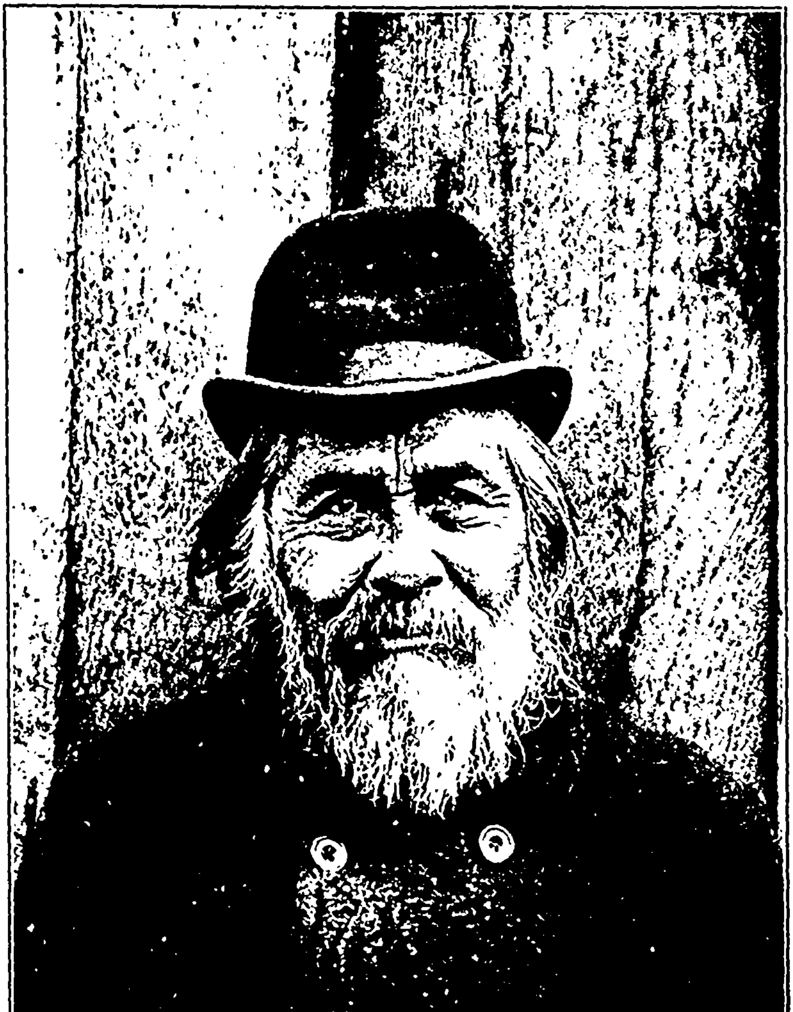
Glutenin exists as such in the wheat grain ; any manipulation that will destroy the hypothetical ferment will coagulate glutenin, thus making gluten formation impossible.

Gluten formation is not merely a mechanical mixture of gliadin with glutenin, but a definite physical state of the two mixing substances is necessary. Coagulated glutenin with gliadin does not form gluten.

There are probably several nucleins or nucleo-proteids in wheat, as shown in the various ways phosphorus is distributed in the different types of embryo cells. Organic iron is found only in the nuclei of the endosperm, aleuron, and embryo cells, and in the cytoplasm of the absorption layer, plumule and radicle cells. The proteid between the aleuron grains shows the presence of organic phosphorus only.

VIII.—BIBLIOGRAPHY.

1. Common Bonon I, 1, p. 122.
2. Journ. d. Chemie, von Gehlen, V, p. 131, 1805.
3. Abstr. Schweiger's Journ. f. Chem. u. Physik, XXIX, 514.
4. Lehrbuch d. Chemie, 3te Aufl., VI, p. 453.
5. Berzelius, Jahresb., VII, p. 231, 1826.
6. Schweiger's Journ., LXIX, p. 188, 1833.
7. Ann. de Chem. et de Phys., LXV, p. 30; Abst., Berzelius Jahresb., XVIII, 327, 1837.
8. Ann. der Chem. u. Pharm., XXXIX, p. 129, 1841.
9. Ann. der Chem. u. Pharm., XLIII, p. 124, 1842.
10. Journ. f. prakt. Chem., XXVIII, p. 398, 1843.
11. Ann. d. Chem. u. Pharm., LII, 419, 1844.
12. Die Getreidearten und das Brot, 1860.
13. Journ. für prakt. Chem., LXXXV, p. 213, 1862.
14. Journ. für prakt. Chem., LXXXV, p. 193.
15. Annalen der Chemie, XXXIX–XL, 1841.
16. Br. Med. Journ., II, p. 104, 1886.
17. Journ. Physiol., XI, p. 419, 1890.
18. Am. Chem. Journ., XV, 392, 1893.
19. Am. Chem. Journ., XIX, p. 59, 1897.
20. Annals of Bot., 1895, p. 182.
21. Comptes Rendus, 1896, p. 327.
22. Arch. für exp. Pathol. u. Pharm., XLI, p. 345; Abst. Chem. Central-blatt, II, 1898.
23. Chem. Central-blatt, I, p. 465, 1898.
24. Journ. f. Prakt. Chem., p. 474, 1899.
25. Archiv. f. Anat. u. Physiol., Physiol. Abth., 1900, p. 163.
26. Proc. Roy. Soc., L, p. 277, 1891-2.
27. Journ. of Physiol., XXII, p. 95, 1897-8.
28. Proc. Roy. Soc., LXIII, p. 471, 1898.
29. Journ. Am. Chem. Soc., XXII, p. 379, 1900.
30. Berichte d. D. Chem. Gesel., 1880.
31. Ann. Agronom., XIV, p. 420; Abst. Journ. Chem. Soc., March, 1889.
32. Journ. de Pharm. et de Chem., 1883-84, Ser. 3, T. VII.
33. Bot. Zeitung, 1862.
34. Pringsheim's Jahrb., VIII, 1872.
35. Journ. Am. Chem. Soc., 1902.



A Nah'ane Medicine Man in Modern Costume.

THE NAH·ANE AND THEIR LANGUAGE.

BY THE REV. FATHER A. G. MORICE, O.M.I.

(Read 4th April, 1903.)

OF the twenty odd tribes which compose the great Déné family, few, if any, are so little known as the Nah·ane.

Many are the travellers who had passing references to them in the course of their writings, but exceedingly few are those who had as much as seen one of them. In fact, Dr. G. M. Dawson is the only author who can be said to have introduced them to us, and his information, fragmentary, and at times inexact as it is, is confined to the limits of a few pages.

Writers are not even agreed as to their very name as a tribe. Thus while Pilling in his valuable Bibliography of the Athapaskan Languages has adopted the spelling Nehawni, Kenticott calls them Nahawney; Ross writes their name Nehawney; Richardson changes this into Noh'hanne; MacKenzie dubs those he met Nathannas; Campbell and Dawson alternate between Nahanie and Nahaunie; others prefer Nahawnie, and Petitot himself never speaks of them but as the Na"anne, his " being the equivalent of my upper dot, which stands for the hiatus.

He derives that appellation from Nari'an-o'tine, "people of the West," but does not state from which dialect the word is borrowed. All the western Déné who know of that tribe, as well as its members themselves, pronounce it Nah·ane, and there can be no doubt that Petitot is correct in the meaning he ascribes to that term, whatever may be said of its derivation. For sunset or occident, the Tsiikotin say *nare·iN*, the Carriers *naanai*, the Tsé'kéhne *naren·oN*, and the Nah·ane themselves *naean*. The final e is expressive of personality and sometimes of plurality or collectivity.

On the other hand, Mr. J. W. MacKay[1] repeatedly calls the tribe Ku-na-na, the name given it by the Tlhinket, its neighbours in the south-west. But that he is somewhat mixed as to the ethnographical status of those Indians is shown by his remark that "the Ku-na-nas of the Stickine valley are closely allied to the Tlinkeets of that section,

1 B.A.A.S. Tenth Report on the North-Western Tribes of Canada, p.p., 38-39.

i.e., the Skat-kwan."[1] As a matter of fact, the latter are just as pure non-Déné as the former are undoubted Déné.

In common with all the Déné and many other aboriginal families, the Nah·ane recognize as their property no other vocable than Déné, "men," though the branch of that tribe best known to me, the Thalhthan, will occasionally call themselves Tcitco'tinneh or "stick-people," whereby they simply translate the name given them by outsiders, since, according to Dawson, and as I have myself ascertained, "the interior Indians are collectively known on the coast as 'stick Indians.'"[2]

So much for the name of the tribe. Now as to its ethnographical status. This seems even more of a mystery to the few writers who have ever referred to it.

It is now over nine years since I stated myself that the Nah·ane "hunting grounds lie to the north of those of the Tsé'kéhne. But I am not familiar enough with their tribal divisions to state them with any degree of certainty, nor do I sufficiently possess their technology to speak authoritatively of it."[3]

I am glad to be now in a position to say that, in the course of the present year, I have taken a trip to their chief village Thalhthan,[4] in order to add as much as possible to my knowledge of that tribe and its language. I have succeeded in gathering besides the material for a grammatical compendium, quite a goodly little dictionary, and not a few texts in its dialect which I intend shortly to publish. Yet I must confess that we must still fall back, for the details of their frontiers and some other particulars, on what the late Dr. G. M. Dawson wrote of them in 1887—Notes on the Indian tribes of the . . . northern portion of British Columbia.[5] Inaccurate as it is from a philological standpoint, his is the only account of the Western Nah·ane worth referring to.

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1 Notes on the Indian Tribes of the Yukon District, etc., p. 2.

2 Tenth Report, p. 39, note.

3 Notes on the Western Dénés, Transactions Canadian Institute, Vol. IV., p. 31.

4 Most writers spell this word Tahltan, when they do not have it simply Taltan, and Dr. Boas corrects them by changing it into Ta'tltan. All sin through ignorance of the Déné phonetics and of the meaning of that word, which is a contraction of *Tha-sælhthan*, *tha*, the usual alteration of *thü*, water n compounds, and *sælhthan*, a verb which has reference to some heavy object lying therein.

5 Annual Report of Geological Survey of Canada.

I.

Broadly speaking the tribe consists of four main divisions. To my certain knowledge, its principal seat in the west is Thalhthan, a salmon fishery at the confluence with the Stickeen of a river of the same name, by about 58° 2′ of latitude north. From the new village in the immediate vicinity of that place, these aborigines radiate as far south as the Iskoot River, taking in all its tributaries and some of the northern sources of the Nass, and in the east to Dease Lake and part of the Dease River, extending also to all the northern tributaries of the Stickeen. Further north, we meet the Taku branch of the tribe, which claims "the whole drainage basin of the Taku River, together with the upper portions of the streams which flow northward to the Lewes, while on the east their hunting grounds extend to the Upper Liard River and include the valleys of the tributary streams which join that river from the westward."[1]

The third division of the Nah'ane is the so-called Kaska, about whom much misapprehension seems to exist among the whites I met in the course of my journey, a misapprehension of which Dr. Dawson constituted himself the echo when he wrote: "The name Kaska is applied collectively to two tribes or bands occupying the country to the eastward of the Tahl-tan. I was unable to learn that this name is recognized by these Indians themselves, and it may be, as is often the case with names adopted by the whites, merely that by which they are known to some adjacent tribe. It is, however, a convenient designation for the group having a common dialect. This dialect is different from that of the Tahl-tan, but the two peoples are mutually intelligible, and to some extent intermarried."[2]

In the first place I must remark that Kaska is the name of no tribe or sub-tribe, but McDane Creek is called by the Nah'ane Kasηa—the η representing a peculiar gutturalo-sibilant aspiration—and this is the real word which, corrupted into Cassiar by the whites, has, since a score of years or more, served to designate the whole mining region from the Coast Range to the Rocky Mountains, along, and particularly to the north of the Stickeen River.

All the whites who mentioned the subject to me concurred in Dawson's opinion that the so-called Kaskas form quite a different tribe, and in a footnote to the latter's essay, a Mr. Campbell goes even so far

1 Notes on the . . . northern portion of British Columbia, p. 3.

2 *Ibid.*, p. 9.

as to state that the "Nahanies of the mountains (who correspond to a subdivision of the Kaskas), are quite a different race from the Nahanies of the Stickeen (Tahl-tan)"[1] Now the Thalhthan Indians I questioned on the subject unanimously declared that those pretended foreigners spoke exactly the same language as themselves, with, of course, some local peculiarities. From a Kaska boy, with whom I travelled for a number of days, I ascertained that even such non-Déné words as *'käk*, paper, *khukh*, box, *'kunts*, potatoes, which I thought proper to the Thalhthan Indians, who borrowed them from the coast, were the only ones current among his people to designate those objects.

The physique of the Kaska is somewhat different from that of the Thalhthan aborigines, inasmuch as I recognized in the former the thin lips and small, deeply sunk eyes of the Tsé'kéhne, while the latter resemble more the Carriers of the Coast Tlhinket, with whom from time immemorial they have more or less intermarried.

The sociology of the two divisions of the Nah'ane is as widely different, and their respective mode of life aud social organization confirm my previous assertion in former papers that, to all practical purposes, the western Nah'ane are Carriers, while their eastern brethren are Tsekehne.

Another circumstance which has contributed not a little to the estrangement of the two tribal divisions, is the long-standing feuds arising out of difficulties concerning the hunting grounds, the making of slaves, and other causes. Even to this day the Kaska resent the Thalhthan's assumed or real superiority, and will not be confounded with them as co-members of the same tribe. Hence their declarations to the whites and the travellers' and traders' printed statements.

According to Dr. Dawson, the so-called Kaskas are sub-divided into the "Saze-oo-ti-na" and the "Ti-tsho-ti-na" and their habitat is in the neighbourhood of the Dease, Upper Liard and Black Rivers. His "Saz-oo-ti-na" may be Sas-otine or "Bear-People," while his Ti-tsho-ti-na's real name is no doubt Tihtco'tinne, or Grouse-People, an appellation which would seem to leave it open to discussion whether we have not in them rather the names of two different phratries or gentes than those of two genuine ethnical subdivisions of a tribe.

"Eastward they claim the country down the Liard to the site of old Fort Halkett, and northward roam to the head of a long river (probably

[1] Notes, etc., p. 1[illegible]

Smith River) which falls into the Liard near this place, also up the Upper Liard as far as Francess Lake."[1]

This statement would seem to dispose of Petitot's Bad-People or *Mauvais-Monde*, a "very little known tribe," he says, "which used to trade at the now abandoned Fort Halkett to the number of 300 or 400 souls."[2]

Father Petitot furnishes us with our fourth division of the Nah'ane when he states that "a little band of 300 Na"annes (Déné) roam over the mountains of the MacKenzie. They are the Nathannas of Sir A. Mackenzie. We can add thereto the Etaottines of the Good Hope mountains, and the Espa-t'a-ottines of Fort des Liards in equal number."[3]

To the above certain divisions of the Nah'ane tribe, we should perhaps add the Ts'Ets'aut, an offshoot of some inland Déné, whom Dr. F. Boas discovered some years ago on Portland Inlet, on the Pacific Coast, somewhat to the southwest of the Nah'ane proper. That Dr. Boas would himself connect them with the Nah'ane tribe is apparent from the statement that "Levi (his informant) named three closely related tribes whose languages are different, though mutually intelligible; the Tahltan (Ta-tltan) of Stickeen and Iskoot Rivers, the Laq'uyip or Naqkyina, of the headwaters of the Stickeen, and the Ts'Ets'aut."[4]

This surmise is fully confirmed by Mr. MacKay, his annotator, who states that those Indians "belong to the Kunâna, a tribe which inhabits the lower Stickine valley and whose headquarters are at Tahltan."[5]

But here *scinduntur doctores*. According to Dr. Boas this handful of natives, which now consists of a mere dozen individuals, would have numbered about 500 souls sixty years ago, while Mr. MacKay has quite a different story to account for their separate existence as a tribe. He relates that, not more than forty years ago,[6] three or four families hailing from Thalhthan in the course of their wanderings made for Chunah, on the sea coast, but took a wrong direction and struck on the west shore of Portland Channel, where they were practically forced to remain in a

1 Notes, etc., p. 10.

2 *Mémoire abrégé sur la Géogrphie de l' Athabaskaw-MacKenzie*, p. 46.

3 *Ibid. ibid.*

4 Tenth Report, B.A.A.S., p. 34.

5 *Ibid.* p. 38.

6 It is now eight years since both statements were published.

subject condition by the Tsimpsians, among whom they had unwittingly tumbled.[1]

Be this as it may, the language of the Ts'Ets'aut such as recorded by Dr. Boas himself, while it shows here and there undeniable traces of a Déné origin, has become so corrupt by the admixture of foreign terms and the alteration of its original lexicon, that the propriety of their being classified as Nah'ane is now quite problematical.

The population of the whole Nah'ane tribe must remain little more than a matter of guess. From the Iskoot, close to the Pacific, to the Mackenzie, across the Rocky Mountains, is indeed a broad stretch of land, and the very fact that it is so sparsely peopled renders it so much the more difficult to obtain anything like an exact computation of the tribesmen. I myself took some years ago a census of the Thalhthan village, and my figures were in the close vicinity of 190 souls. The population has since decreased, so let us call it 175.

From native sources I ascertained that the "Kaska were more numerous, perhaps 200. Petitot puts at 600 the number of the transmontane Nah'ane and allied subtribes. Allowing for the probable decrease and possible exaggeration, let us say 500. There remain the Taku, of whom I have no means of ascertaining the exact numbers. Probably 150 would be a conservative figure.

We thus obtain a total of 1,025, or in round numbers, 1,000 souls for the whole tribe, and I believe this is as fair an estimate of its population as could possibly be had at the present time.

As already stated, the eastern Nah'ane somewhat differ in physique from their western congeners, the only portion of the tribe with which I am familiar enough to describe it *de visu*. Their stature would be rather below than above the average, the maximum height being five feet eight inches. Their feet and hands are small and well shaped, and their head is round and not so large as that of the neighbouring

1 In the course of his account of that adventure and the circumstances which lead to his getting acquainted therewith. Mr. McKay takes occasion to speak of an invasion by the Tsimpsian of the territory which is now the Tsimpsian peninsula, whereupon Dr. Boas remarks that "there is no traditional evidence of the invasion of the Tsimshian tribe to which Mr. McKay refers," adding that "it is probable that the Tsimshian were originally an inland people." two statements which, apparently difficult to reconcile as they at first appear, nevertheless are in no way conflicting. There may be no tradition of such an invasion among the Tsimpsian, but their very name betrays their origin. The Skeena River is known to them as the *K'sièn*, and they call themselves *T'sœm-sièn*, people from the Skeena, or the river. To this day, anybody can see, two miles from Hazelton, on the Upper Skeena, a prairie or ancient townsite, where one can distinguish the cavities over which were built their winter subterranean houses. Now the name of the locality is *Tamlarh-am*, the beautiful place, in Tsimpsian, and those two words are still used in that connection by the inland Kitkson to the exclusion of any name in their own dialect.

Tlhinket. With them the nose, without being of the regular aquiline type, is not so squatty as among the Tsilhkoh'tin and other tribes. The lips are full, the eyes dark and not quite as large as is common with the Carriers. The forehead is low, broad and bulging immediately above the eyes. The hair is invariably black, coarse and straight.

Their beard is scanty, though a few, especially such as have taken to shaving—they are very progressive and great imitators—disport a fair quantity of dark, bristly facial hair.

As to their complexion, it varies considerably according to the individuals. Contrary to what I have noticed in other tribes, some of the eastern Nah'ane women have cheeks of a tinge which might almost be characterized as rosy, though the facies of others is quite swarthy.

All the adults above forty have the septum pendent and pierced through with a hole which held formerly a large silver ring, perhaps two inches in diameter. The leading men or notables wear likewise silver rings hanging from the lobes of the ear, and these are the only present remnants of the many ornaments which the helix was originally made to support.

Neither in blood, customs nor language are the western Nah'ane pure Déné. They are indebted to no small extent to the Tlhinket of Fort Wrangell for their present make-up. To them also they undoubtedly owe that lack of moral strength and force of character which has left them such an easy prey to the vices of unscrupulous white men. Very few are to-day the western Nah'ane who can be represented as bodily sane. Syphilis, a disease hardly known among the other Déné, is but too prevalent among them. Liquor is also slowly but surely killing them out.

I am bound to add, however, that adverse circumstances are a great deal to blame for the development of such pitiful results. Had missionaries established themselves among them before the rush of strangers to the Cassiar mines, the natives would not, in all probability, be the degraded beings they have become. Since the last few years, a representative of the Anglican Church has struck his tent on the arid hill of Thalhthan. But I am sure he could not well himself take exception to my statement that his influence has not been in the interest of temperance.

Though no other Déné that I know of have had to undergo the test of being left alone to wage their war against such a degraded foe as a majority of the Cassiar miners have shown themselves to be, it is difficult for me to imagine for a moment, for instance, the Tsè'kéhne tribe sunk to the low moral level of the present Nah·ane whom I have met or have been told about.

While the eastern. Nah·ane lead the simple patriarchal life of the Tsè'kéhne, with hardly any sign of a social organization, their western congeners, with the remarkable adaptiveness proper to the Déné race, have adopted practically all the customs and some of the mythology of their heterogeneous neighbours on the sea coast. Thus it is that matriarchate or mother-right is their fundamental law governing and regulating all inheritances to rank or property.

Though they have no totem poles, they know of the gentes, which at Thalthan are those of the Birds and of the Bears. Each of these have several headmen or *téné-thîe* (the equivalent of the Carrier *tœnœza*), who alone own the hunting grounds, and on festival occasions, such as dances or potlatching, are granted special consideration. These ceremonial banquets are much in vogue, and as a result, almost every house in Thalhthan is now crowded with a quantity of trunks containing goods publicly received or to be likewise given away.

Those houses are now of rough unhewn logs, with stoves instead of fire-places. But the tribe's residences were originally much less elaborate, and consisted of brush shelters, sometimes with low walls made of long, slender poles. Therein they dwell, generally several related families together.

Marriage was never accompanied with any ceremony or formality. It seems to have been based principally on the bestowing of furs or other goods on the parents of the prospective bride.

Polygamy was known everywhere, but it is now practically abolished, the only exceptions being a very few cases among the present Kaska. As to divorce, it is obtained without any formality, and is often enough resorted to.

Shamanism was originally the only form of worship common to the whole tribe, and in the east witchcraft, and the social disturbances it entails seem even now quite prevalent. The Kaska boy I have already mentioned as a companion on part of my trip from Thalhthan was just being taken away from revengeful fellow-tribesmen who had already done to death two of his brothers under the plea that their parents were responsible for the sickness and ultimate death of some Indian or

Indians against whom they were believed to have exercised their black art.

As among the other Déné, such deaths were the cause of family feuds of long duration and bitter hatred, when they did not lead to reprisals and a series of murders. Thus would originate their internecine wars, which consisted merely in ambuscades, surprises and massacres, accompanied sometimes with the enslaving of the women and the children.

But their "wars" were more frequently directed against foreigners, such as the Tsimpsian of the upper Skeena, or against the Tlhinket of the coast. They had no war chiefs, or indeed any chief at all in our sense of the word.

In times of peace, their special avocation and means of subsistence are hunting and fishing, to which a few of the younger men add packing for the miners and the Hudson's Bay Company. As their territory is so extensive, it still abounds in fur-bearing animals and game of almost all descriptions. I found moose especially plentiful all over the country. The mountains are also rich in sheep and goats.

No wonder then, if the Nah'ane are well-to-do. In fact I consider that the western part of the tribe is at present dying on a golden bed. In the house of my hosts at the time of my visit were to be seen, besides gilt bronze bedsteads and laces of all kinds, two sewing machines, two large accordeons, and, will the reader believe it?—a phonograph! All this in the forests of British Columbia, north of the 58th degree of latitude!

Since I have mentioned death, I may remark that cremation was, until recently, the mode adopted by the western Nah'ane to dispose of their dead. And, in this connection, we have a ludicrous admixture of the new order of things with the olden ways, in the small travelling trunks bought from the whites, which are to be seen planted on two posts, in several places along the trails, and which contain some of the bones of the dead picked up from among the ashes of the funeral pile.

II.

As to the language of the Nah'ane, much might be said. I shall point out in the following pages only those particularities which are its exclusive property, and leave out most of the general features which are common to all the Déné dialects, and which the reader will find detailed in my paper on "the Déné Languages,"[1] and in my forthcoming complete grammar of the Carrier language. Furthermore, all the following remarks shall apply more particularly to the idiom of the western Nah'ane. the only one I have ever studied.

Neglected by the ethnographers as the Nah'ane have remained to this day, their dialect has still been more of a *terra incognita* to the philologists. With not even the least grammatical note has it been honoured so far in all the linguistic literature at my command, and the only vocabulary by which it has ever been represented in scientific publications consists of the four columns of Thalhthan words printed by the late Dr. Dawson.[2]

And here I may be allowed to state that, after a careful study of their language, I have had the satisfaction of ascertaining that of all the corrections in the latter's vocabulary which I lately declared[3] were demanded by the general rules of Déné phonetics and suggested by my knowledge of the other related dialects, not one have I found to be unwarranted.

Before going further I must also correct the one statement Dr. Dawson makes concerning their language. Speaking of the Thalhthan and Taku Nah'ane, he writes: "These Indians speak a language very similar to that of the Al-ta-tin, if not nearly identical with it, and so far as I have been able to learn, might almost be regarded as forming an extension of the same division. They appear to be less closely allied by language to the Kaska, with which people they are contiguous to the eastward."[4]

I have already done justice to the latter assertion. By Al-ta-tin, Dr. Dawson means the Lh'ta'tin, or "People of the beaver dams," as the Tsé'kéhne are called by the Carriers. His notion about the similarity of the two dialects I have found prevalent in other quarters. To prove its utter groundlessness, I need but reproduce here the Nah'ane and the Tsé'kéhne versions, for instance, of the doxology. Was the Chippewayan version available, I have no doubt that it would be found more alike to that of the Tsé'kéhne than to that of the Nah'ane. Grammatically speaking, there is more affinity between the Tsé'kéhne and the Chippewayan—two very distinct tribes—than between the Tsé'kéhne and the Nah'ane.

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1 Transactions Canadian Institute, Vol. 1.

2 Notes on the northern portion of British Columbia, p. 19, *et seq.*

3 Transactions Canadian Institute, Vol. VI., pp. 99-103.

4 Notes, etc., p. 2.

THE DOXOLOGY.

IN NAH'ANE.

Séesôga Œtha· 'ka'tcéh, Œtcimé· ka'tcéh, Ahtige-Ti 'ka'tcéh hut'sihkaihtin.

Lhanu kastséh tûda ahih'té la, tû'gu 'ka'tcéh, ue'té 'katcéh, ét'tha ta'da œtû wotôzite a'téh éyéne 'ka'tcéh hu'karo'té ni.

IN TSÉ'KÉHNE.

Utqon Œtha· qûh, Œtcwinh qûh, Yétqire-Inqi qûh ut'sœrhautœz.

Sé rhasséh tarhît'qé ille a, qû qûh, awuz'on qûh int'lhon qé ta ussé utœtûzit e'tah éyétœ qûh hahut'qé.

To start with the sounds as such, I will remark that the following desinential letters or groups of letters are never found in Carrier or Chilcotin, but are quite common in Nah'ane: *c, ts, tc, tlh, klh,* to which we must add the medial *-slh-*, as in *aslhé*, I make, and *-srh-*, as in *ctisrhuh*, I snore. Final *ts* occurs often enough in Babine, and final *tc* is as frequent in Tsé'kéhne, but the other compounds are never found even in those idioms.

On the other hand the letter *m*, which sometimes terminates a word in Carrier, never occupies that position in Nah'ane. We should not forget either to notice that the double letter *tj* or *dj*, which is so frequent in Kut'chin appears also in Nah'ane to the exclusion of all the other Déné dialects.

Some Carrier letters have their fixed equivalents in Nah'ane. Thus the Carrier initial *n* is often replaced by *t* in Nah'ane. Ex.: *ni*, mind, Nah'ane, *ti*: *na*, eye, Nah'ane, *ta*: *aunilh*, purposedly, Nah'ane, *atilh*; *dûni*, he will say, Nah'ane, *dûti*. The initial *p* of many Carrier words becomes *m* in Nah'ane (as well as in Tsé'kéhne), and we have *pœn*, lake, in Carrier, but *men'* in Nah'ane; *thapa*, shore, in Carrier, *thama* in Nah'ane; *pœ-*, his, in Carrier and *me-* in Nahane.

A Nah'ane sound, which I have found in no other Déné idiom is that which I render by H. It is a kind of a guttural aspiration, much more pronounced than that of the common *h*. Its equivalent in the other dialects is *rh*, or the Greek *rho*, and in the possessive case, it is inflected into a soft *r*. Ex.: *His*, pus; possessive, *me-rize*, his pus.

The first particularity which strikes a Déné scholar in his study of the Western Nah'ane, is the presence therein of a regular accent, something quite unknown in all the northern Déné dialects. I have no doubt that the intercourse of that subtribe with the Tlhinket of Fort Wrangell is responsible for that feature of its language. This accent has for effect, not only to lengthen the syllable it affects, but even to raise the pitch of the voice when the accented syllable is pronounced. Thus it often falls on monosyllables. Gun is *tl'na* (a Tlhinket word) in Nah'ane; *kussa'* means, "I do not know" in the same dialect. Much stress must be laid on the *u* of the first word and on the *sa* of the last, otherwise neither would be understood.

On the other hand the voice must also be raised with a sort of constrained effort when one pronounces the words *'khon'*, fire, *nehn'*, land, *tsé*, gum, etc., though many other monosyllables lack this distinguishing feature.

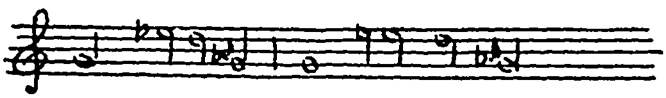
In this connection I must not fail to record what, to a student of the Carrier idiom, seems something of an anomaly. In my "Notes on the Western Dénés,"[1] I wrote some years ago: "In these nouns there is generally one syllable which is more important and contains, as it were, the quintessence of the word. Thus it is with the *ne* of *tœne*.... In composite words such syllables only are retained.

Now it happens that in Nah·ane the accent falls precisely on the first syllable of that word (which means "man" in all the dialects), and not on the second, which is hardly audible when pronounced by a native. In the same way, instead of using only the second half of the word, as is usual with the Carriers and the Chilcotin when they refer to the human body or to any part thereof, as in *ne-yœs'te*, human body; *ne-na*, human eyes; *ne-t'sĭltcœn*, human neck, etc., a Nah·ane will always utter the whole word, giving particular prominence to its first part, and say, for the same objects, *tèn'e-ri*, *tèn'e-ta*, *tèn'e-'kwos*, which the careless listener will most probably take for *tèn'ri*, *tèn'ta*, etc.

Beside their accent, the Nah·ane have, when speaking, a particularly marked intonation. This is so pronounced that it could almost be compared to a song. In fact, I have noticed the following modulation as being of very frequent occurrence. Its finale especially is hardly ever omitted.

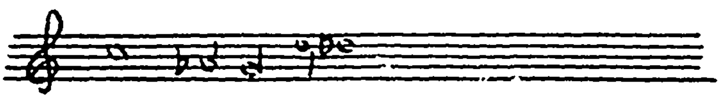
Students of native languages must have noticed that most tribes or portions of tribes have their own peculiar way of singing out, as it were, the sentences of their respective idioms. When there is nothing in their elocution which can be compared to a song, the finale, at least, is almost certain to stray out of the *recto tono*. So the ending of each Shushwap sentence is infallibly from G to upper C, while the Coast Salish, or at least the Sicalh, content themselves with raising the voice from G to A, or one full tone.

The intonation of the Carriers varies too much according to the different groups of villages to be recorded here. I will choose but one, which is characteristic of the Hwozahne, or people of Stony Creek.



Ntœn lhecœtni, au t'sœtœst'sœk ;
i.e., What does he say, we don't understand him.

The elocution of the Chilcotin and of the Tsé'kéhne is more uniform. Any member of the former tribe would, I think, easily recognize the following sentence, which they are ever ready to utter when anything is asked of them which they are not disposed to grant.



Tla kan'te kule ; *i.e.*, There is none.

But to return to our Nah'ane dialect. From a terminological point of view, it has all the appearances of an eclectic language. Indeed, I would fain compare it to English, as it occupies to some extent, with regard to the other Déné dialects, the position held by that language relatively to the European idioms. Its vocabulary furnishes us, besides fully forty nouns,[1] which are more or less Tlhinket, several terms which duly belong respectively to the Kutchin, the Hare and the Chippewayan dialects. Here are a few examples. Kutchin: *djugu*, now, Nah'ane, *tugu* ; *lhaon*, quite, Nah'ane, *lhan* ; *œkwœt*, knee, Nah'ane, *ekwœt* ; Hare *due*, no, Nah'ane, *tueh* ; *guntie*, elder brother, Nah'ane, *etiye* ; Chippewayan: *sorha*, well ; Nahane, *sôga* ; *esdan*, I drink, for which the Nah'ane have an absolutely homonymous synonym.

Even the Tsimpsian has lent them one word, *lelk*, to designate the snake, a reptile which is not found within Nah'ane territory. A few words of English have also crept into the Nah'ane vocabulary, and it is worthy of remark that whenever an *l* occurs in them, the Nah'ane have

1 I may here draw the reader's attention to the fact that a people of a low mental standard, a nation of uncultivated intellects, may borrow many unchangeable words from the vocabulary of its heterogeneous neighbours, but will never attempt to appropriate verbs. The former they will leave in their original form, or allow them through neglect or ignorance to slowly degenerate into more or less different terms; but when it is a question of verbs, the low type intellect is not up to the task of adapting them to the exigencies of its grammar. In other words it cannot digest and assimilate them.

altered it into an *n*. Thus for gold, they say *gon*; for silk, *sink*; for dollar, *dana*. The word *kas* for barrel they owe to the Tlhinket, who had themselves borrowed it from the English speaking skippers and traders (*kas* = cask).

Chinook has contributed *masmas* (a corruption of *musmus*), cattle, and probably *kimdan* (for *kiutan*), horse. Following the example of the coast Indians, the Nah·ane have likewise changed the Chinook for cat, *pus*, into *tuc*.

At times this propensity for appropriating foreign terms leads to curiously hybrid compound words. For instance, the Nah·ane equivalent of organ is half Tlhinket and half Déné. All the Déné call that instrument a "paper that sings." As the Nah·ane had already borrowed the Tlhinket work *'kŭk* for paper, and on the other hand, as they did not know or could not use the Tlhinket synonym for "sings," they unscrupulously retained the first vocable to which they added their own equivalent for the verb and said *'kŭk-etqine*.

The dictionary may be regarded as a thermometer which faithfully registers a people's status and chief avocation. Its readings are seldom at variance with fact, and when it records, for instance, a multitude of fish names or, still better, when it possesses several names for the same fish according to its age or condition, it will inevitably denote a nation of fishermen. In like manner the sociological status of our Nah·ane is betrayed by their vocabulary, which abounds in fine distinctions for the names of the larger animals on which they mainly subsist.

I will take but one example to illustrate my meaning. With them the generic name of the marmot is *tœtiyé*, and the female is called *hosthelh*, while the name is known as *oet'qetha*. A little marmot in general is named *oe'kane*, or *usthe-tsetle*. But if it is only one year old it goes as *usaze*; the next year it will be known as *oekhutze*, and when in its third year, it will be called *tœtiyé-tucitze*. And note that all of those eight words apply to only one kind of animal, since there is another term to denote the smaller variety of marmot (*arctomys monax*).

We have therefore our Nah·ane stamped by their very vocabulary as a people of trappers and huntsmen, and the abundance of their terms for a mountain animal furthermore sheds a good ray of light on the topography of their country.

Another reliable indicator of a primitive people's main occupations, to which it adds a valuable hint at the nature and climate of its land, is the calendar. Subjoined is that in use among the western Nah'ane, and the careful student of Americana will perhaps find it worth his while to compare it with those of the Carriers and of the Tsé'kéhne published in my "Notes on the Western Déné."[1] Of course, all the months therein recorded are lunar months, and coincide but imperfectly with our own artificial divisions of the year.

January, *sa-t'séslhie,* moon of the middle (of the year).
February, *tœnon-thene,* the snow is a little frozen over.
March, *ihťsi-sa,* moon of the wind.
April, *tlhi-pœnetsé-e,* the dog uses to bark.
May, *ih'aze-sa,* moon when all the animals leave their winter retreats.
June, *œyaz-e-sa,* moon of the little ones (when animals have their young.
July, *œtcitc-e-sa,* moon when they moult.
August, *ti'ka-e-sa,* moon when they fatten.
September, *hosthelh-e-sa,* moon of the female marmot.
October, *mœn-then-tsetle,* moon of the small ice.
November, *mœn-then-tco,* moon of the big ice.
December, *kœrh-urwoesse,* the rabbit gnaws.

We have tarried so long over the sounds and substantives of the Nah'ane language that our remarks on the other parts of speech must necessarily be brief.

In its numerals we find a confirmation of what I said some time ago when I wrote, speaking of the roots of languages in general: "The numerals and the pronouns . . . generally have a kind of family air in cognate dialects. As to the pronouns, I think that hardly any qualificative reservation is necessary, but it is not so with all the numerals."[2] Of the ten Nah'ane numbers, only three (one, *lhige,* Carrier, *ilho*; three, *thadé'téh,* Carrier, *tha*; and five, *lholla,* Carrier, *kwollai*) have any affinity with the Carrier, Babine, Chilcotin or even Tsé'kéhne numerals. The other seven have not the faintest resemblance thereto.

A peculiarity worth recording in this connection is the fact that the numbers two, three and four are in Nah'ane perfectly regular verbs which are conjugated with persons—plural, of course—and tenses. Let us take, for instance, the number three, *thadé'téh.* We have at our disposal any of the words of the following conjugation:

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1 Transactions Canadian Institute, Vol. IV., p. 106.

2 The Use and Abuse of Philology, Transactions Canadian Institute, Vol. VI., p. 92.

PRESENT.	PROXIMATE FUTURE.
tha-desi'téh, we are three	*tha-di'tilh*, we are going to be three
tha-dah'téh, you are three	*tha-dah'tilh*, you are going to be three
tha-hidé'téh, they are three	*tha-hædi'tilh*, they are going to be three
PAST.	EVENTUAL FUTURE.
tha-desi'tée, we were three	*tha-dû'tée sa*, we will be three
tha-dah'tée, you were three	*tha-dah'tée sa*, you will be three
tha-hidé'tée, they were three	*tha-hædû'tée sa*, they will be three

In all these words the main root for three is, of course, *tha*. Yet *thadesi'téh*, etc., are single words whose neither first nor last component parts can be used separately.

The only approach to these conjugable numerals I know of is to be found in the speech of a small portion of the Carrier tribe. It is restricted to the number two, *natne*, which becomes *nat'soetne*, we are two (persons), *nahtne*, you are two, etc. I should not forget, however, a peculiar set of numerals for which I find no more appropriate qualificative than the epithet "inclusive." These not only have in Carrier all the persons and tenses of the above, but they are even modified so as to form a separate class of adverbal numerals. Here are a few examples: *na-t'sæl'torh*, both of us;[1] *na-nel'torh*, both of us; *na-nælh'torh*, both of you; *na-rhæl'torh*, both of them.

The following are impersonal verbs: *na-hwul'torh*, both times; *na-hwothil'torh*, it is going to be both times, etc.; *tha-hwul'torh*, all of the three times; *ti-hwul'torh*, all of the four times, etc.

All these forms, tenses or persons can be applied in Carrier to all the numerals of that class, except the first, the ninth and the tenth, and in this respect, as in so many others, that language surpasses in richness all the other Déné dialects.

The Nah'ane lacks an equivalent for the personal plural particle *ne*, which the Carriers suffix to the verb when in English we make use of the demonstrative and relative pronouns "those who," as in *hwot'sit-ne*, "those who lie," the liars. Instead of this, the Nah'ane will say, by a curiously abnormal commingling of a plural pronoun with the corresponding singular verb: "he-lies they," *tset'sit ockhune*. This renders speech unnecessarily long and rather unwieldy.

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1 With an idea of impersonality, which it is impossible to express in English, and which is absent in *nanel'torh*.

Nah'ane Women in Dancing Costume.

A feature of the possessive pronouns which the Nah'ane shares with some related idioms is the absence of a term for the second person of the plural. Most of the eastern Déné dialects even lack altogether the same person of the personal pronouns, but the Nah'ane are not so verbally destitute. In their minds, however, there lurks some vague confusion about the difference between the first and the second person plural of those pronouns which, at times, does not seem to be fully grasped.

In common with those of the other Déné dialects, the Nah'ane verbs are rich in persons, some, like the verbs of station and the verbs of locomotion, having as many as eighteen for each tense, as against the twenty-one their Carrier equivalents boast of. In the face of that relative richness it is somewhat of a surprise to find that the regular or common verbs have not even a single person representing the dual, which is rendered, as with us, by the plural, while even the Carrier, which is rather deficient in that respect, possesses, at least, the first person dual for all the verbs.

A point of resemblance with the eastern dialects is the plural of some Nah'ane verbs, which is formed by the incorporation of the particle *da,* without any alteration of the desinential syllable. Thus, until we come to the plural, the conjugation of the verb *t'sé-mészit,* I wake up, is practically that of its Carrier equivalent. But after this, the similarity is confined to the main or initial root, which, through all tenses and with any person, remains invariable in all the dialects. The following partial conjugation of the present of the above mentioned verb will illustrate my remark:

CARRIER.	NAH'ANE.
Dual.—*t'se-nîtzit,* we wake up, both of us.	Dual.—*t'se-nîtzit.*
t'sé-nœhzit, you wake up, both of you.	*t'se-nahzit.*
t'sé-rhœnzit, they wake up, both of them.	*t'se-hœnîzit.*
Plural.—*t'se-t'sœntilh,* we wake up.	Plural.—*t'sé-dasîtzit.*
t'sé-nœhtilh, you wake up.	*t'sé-dahzit.*
t'sé-rhœntilh, they wake up.	*t'sé-dahezit.*

Another most important point of resemblance of the Nah'ane with the eastern Déné dialects, is the utter absence in the former of any special negative form. This particularity may be said to constitute its fundamental difference from the Carrier, Babine and Chilcotin idioms, the verbs of which are distinguished by at least one, and frequently two or even three syllabic inflections in addition to the negative particle.

Instead of this the Nah·ane set that particle before the verb, which remains under its affirmative or normal form.

To sum up. The Nah·ane language is much less complicated and verbally poorer than the Carrier. It is also less pure in its lexicon, more embarrassed in its phraseology, and owing to its accent, even more delicate in its phonetics.

THE PALÆOCHEMISTRY OF THE OCEAN IN RELATION TO ANIMAL AND VEGETABLE PROTOPLASM.

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CONTENTS.

I.—INTRODUCTION.

THE history of the composition of ocean water is a question of very great interest to the geologist, the physiographer and the biologist. To the geologist and physiographer its importance lies chiefly in the fact that it is associated with the history, on the one hand, of erosion and denudation of land surfaces of the globe, and, on the other, of the formation of all the sedimentary strata. The ocean, ever since the first condensation of water on the rockcrust of the earth, has acted as a gigantic solvent, and the salts it now holds in solution represent what it has retained after its action for millions of years as a leaching and filtering agent. The sedimentary rocks are thus but a vast precipitate from the ocean of what had been partly suspended and partly dissolved matter in it during all the geological periods. The history of the composition of the ocean is, on this view, the complement of the history of all the terrigenous changes necessary to fill out all the pages of the record of events that have transformed the surface of the earth.

To the biologist the value of the question obtains from a different point of view. The sea is the original home of all life on the globe, and it was in the sea that the differentiation between animal and vegetable life, as well as the evolution of the great divisions of the animal kingdom were effected. Indeed the great events in the evolution of animal forms have been rendered possible by changes which have taken place in the composition of ocean water. These changes have modified organisms, and have created conditions which have served as factors in directing the course of development. This may be specially illustrated by reference to the case of the calcium salts in sea water. That the earlier Archæan seas contained comparatively small quantities of calcium compounds seems to be clearly indicated by the fact that in pre-Cambrian strata the limestone deposits are very limited, not more than two per cent. of the thickness of the beds, the Huronian portions of which, now generally recognized as of sedimentary origin, are, according to Lawson,* over 50,000 feet in thickness. The small amount of limestone deposits could not have been due to the absence of living organisms, for the oldest Cambrian beds contain Trilobites and Brachiopods, and such highly specialized forms postulate a long course of pre-Cambrian life. The very fact that the Brachiopods of the early Cambrian were largely those provided with a horny or chitinous shell, indicates that all the animal forms of the preceding period had imperfectly acquired the lime "habit," which, one may reasonably believe, would have earlier made its appearance had calcium salts been present in considerable quantities in ocean water from the first. It is perhaps due to the absence of this lime "habit" that fossils do not obtain in pre-Cambrian strata.

Once, however, the lime "habit" was acquired, through adaptation of the animal cell to its environment, the course of development became accelerated, and the evolution of the higher types of Invertebrate life, as well as all the forms of Vertebrata, became possible. The Vertebrate skeleton, and all that it implies in evolution, is, therefore, a result of the gradual increase in the quantity of calcium in the oceans of the pre-Cambrian period.

To both the geologist and the biologist the history of the chemistry of the ocean has recently acquired an additional interest from the attempt made by Joly† to determine the age of the earth, who uses for that purpose as factors the amount of sodium now in the ocean, and that

* Geol. Survey of Canada, 1887, pp. 101 and 102, F.

† An Estimate of the Geological Age of the Earth. Trans. Roy. Dublin Soc., Vol. 7. (Ser. 2), 1899, p. 23.

estimated to be in the annual river discharge of the globe. Joly took for these the results of Murray,* who, basing his calculations on the discharge of nineteen of the principal rivers of the world, estimated the total amount of the sodium and other salts annually put into the sea by river water. Joly finds from Murray's tables that the sodium annually discharged is 157,270,000 tons, and the quantity in the sea is 14,151,-000,000,000,000 tons. Dividing the latter by the former he gets as quotient, approximately, 90,000,000, which, expressed as years, would be the age of the earth, or, rather, the period of time which has elapsed since the first condensation of water vapour took place on the globe. Joly admits that the ocean at first contained a considerable quantity of sodium as sodium chloride, and this he puts at about 14 per cent. of the present amount in the sea. This would make the amount discharged into the sea by river water less than that stated above, but, on the other hand, the volume of the ocean may, as a result of more recent estimations, be given a higher value, and in consequence the mass of sodium in it would be 15,627,000,000,000,000 tons. Further, of the sodium annually put into the ocean, Joly allows as much as 10 per cent. for that which is taken from the ocean by the rain and returned again in river water, and this estimate would make the amount of river sodium, which is annually leached out of the rocks and strata, as 97,800,000 tons. With these values Joly finds that the corrected figures for the age of the earth is 89,300,000 years.

In support of his contention Joly shows that as compared with the igneous rocks there is in the sedimentary rocks, which are derived from them, a deficiency of sodium, and that the sodium now in the sea would approximately account for the difference. The bearing of this fact is that all the sodium now in the ocean was derived from the original rock crust by processes which to-day are in operation in decomposing rock material and removing the sodium therefrom. In other words, the discharge of sodium into the sea has been in the past a uniform one, or at least subject to no great variations that would constitute a factor against determining the age of the earth by this method.

This estimate has been ably criticized by the eminent geologist, the Rev. Osmond Fisher,† who points out that the sodium which is derived from the decomposition of crystalline or igneous rocks is in the form of carbonate rather than chloride; and he asks whether it is not possible that the chloride of river water is derived, not from crystalline,

* On the Total Annual Rainfall on the Land of the Globe, and the Relation of Rainfall to the Annual Discharge of Rivers. The Scottish Geogr. Mag., Vol. 3, 1887, p. 65.

† Geol. Mag., New Ser., Vol. 7, p. 124, 1900.

but from sedimentary rocks, or from what Sterry Hunt calls "fossil sea water, still to be found imprisoned in the pores of the older stratified rocks, and presumably in the younger as well." To answer this affirmatively would be of necessity to assert that the sodium which now goes to the sea as sodium chloride comes from the supply derived from and deposited by the sea in ancient geological strata—that is, what was at one time in the sea is being returned to it again. Fisher also points out that the strata which are now in the process of formation, imprison sodium chloride in their mass, taking it from the sea. There would thus be a constant circulation of sodium chloride from the ocean to the stratified rocks and back again to the ocean. That would also postulate that the sea was almost as rich in sodium chloride in Silurian times as it is now, and it would go far to support the view that "the sea was salt from the first;" but if we assume that the sodium of the sea is derived from those sodium compounds supplied by rivers other than the chloride, the estimate of the age of the earth, as given by Joly, would have to be multiplied several times in order to get the approximate length of the period which has elapsed since the oceans of the globe were first formed.

Another criticism of Joly's view, made along the lines followed by Fisher, is that advanced by Dubois,* who, from a comparison of the amounts of sodium and chlorine supplied to the sea by a large number of rivers, concluded that only a small portion, if any at all, of the sodium derived from denudation appears in river water as sodium chloride; that the sodium chloride discharged into the sea annually is derived from the rainfall, and the salt deposited in the older strata by the sea.

As Fisher has already pointed out, it is the sodium compounds other than the chloride that ought to be considered as being primarily derived from the disintegration of rock mass, and, therefore, primarily added to the sea. What the total amount of this sodium is cannot be determined with approximate certainty, but Dubois is inclined to regard it as about one quarter of the total discharge of sodium into the sea as given in Murray's tables, and, consequently, Joly's estimate of the length of the period which has elapsed since water first condensed on the earth's surface would have to be multiplied by four, the product being approximately 400 million years.

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* *On the Supply of Sodium and Chlorine by the Rivers to the Sea.* Kon. Akad. v. Wetensch., Amsterdam, Proceedings of the Section of Sciences, Vol. 4, p. 388, 1902.

II.—THE ORIGIN OF THE PHYSIOLOGICAL RELATION OF THE CHEMICAL ELEMENT IN BLOOD PLASMA.

I have thus dealt at some length upon the importance of the history of sea water, and with Joly's views and those of his critics, because all this leads up to a question which is of very great importance to the physiologist. The life of the globe in the earlier geological ages, so far as the strata reveal to us the past history of the earth, as already pointed out, was closely associated with the sea. It is indeed almost universally assumed that life began in the ocean and continued in association with it alone till the close of the Cambrian period, although the presence of graphite in Cambrian and older rocks seems to indicate that vegetable organisms were accommodating themselves to a land life. Even this may not be an exception, for these rocks must have been laid down under water, and therefore their organic remains would be those of the sea. If accordingly we could know what the composition of the sea water in the Cambrian and pre-Cambrian periods was, we would, in all probability, be able to determine some of the chemical and physical forces to which living matter was then subjected and thus explain the relations which obtain to-day in living matter between it and its salts. In a recent paper* I have pointed out that the relative proportions of the elements, sodium, potassium, and calcium in the plasma of the blood are surprisingly very like those which are found in the ocean water of to-day, and that the differences which obtain between the two series of proportions of these elements may be explained on the ground that such proportions in the blood plasma are those that obtained in ancient sea water when the ancestral form of Vertebrates, in which sea water was the circulatory fluid, as it is in many marine forms to-day, acquired a closed circulatory system. That the ancient proportions are reproduced to-day in all forms, which have a closed circulation, I attribute to the influence of heredity, the cells of the organisms having for ages been associated with the sodium, potassium, and calcium in certain proportions, and having been accommodated to them, the relations ultimately became so fixed that living matter reproduces the ancient proportions in the fluids which bathe itself. There is one point in which the proportions in the circulatory fluid and those in sea water differ, and that is in respect to the magnesium. In the sea water of to-day there are 11.99 parts of magnesium for every 100 of sodium, while in plasma there are 0.8 parts of magnesium to 100 of sodium. This is

* On the Inorganic Composition of the Medusæ, Aurelia flavidula and Cyanea Arctica. Journ. of Physiol., Vol. 29, p. 213, 1903.

a striking difference but it is easy of explanation. The proportion of magnesium in sea water is now slowly growing. In the pre-Cambrian oceans it must, therefore, have been very small, not perhaps as low as it is in blood plasma, for in the latter the magnesium would only represent the proportion of an earlier period than that in which the circulation became closed, as the tissues would only reproduce the proportion which had by long accommodation become fixed in them. Even the organisms which live in the sea to-day, whose ancestral forms have lived in the sea since the Cambrian, do not take up the magnesium from the sea water in the full proportion which it has in the latter.

III.—THE ORIGIN OF THE RELATION OF THE CHEMICAL ELEMENTS WITHIN PROTOPLASM ITSELF.

There is, therefore, so far as the circulatory fluid of Vertebrates is concerned, a reproduction of the proportions of the sodium, calcium, and potassium of the pre-Cambrian oceans. The problem which now arises is one whose solution involves greater difficulties. If organisms should reproduce in their own circulatory fluids the proportions of the elements in the early geological periods, what contributed to those remarkable proportions which obtain, not in the circulatory fluids, but in the living matter itself? These proportions are widely different from those found in the circulatory fluids, and one cannot bring oneself to regard the former as derived from the latter. In vegetable organisms the potassium and the calcium much exceed the sodium, and even the magnesium may be greater in amount than that of the latter. In animal organisms the proportions are difficult to ascertain owing to the presence of skeletal and other structures in which the calcium and sodium greatly preponderate, but even in these the potassium is nearly equal to the sodium, and in muscle it is greatly in excess, while the calcium and the magnesium are much less than the sodium. Thus, in the muscle of the dog the relative values for each are* :—

Na.	*K.*	*Ca.*	*Mg.*
100	354	7.26	25.1

These proportions may or may not represent approximately those found in unicellular organisms like an Amœba, or even a white blood corpuscle, but do they represent to any degree the proportions which obtained in the early pre-Cambrian seas when life was represented by unicellular organisms only, which accommodated themselves to the sodium, potassium, calcium, and magnesium in their habitat, just as the

* Julius Katz, Pflüger's Arch., Vol. 53, p. 1, 1896.

marine unicellular organisms of to-day have accommodated themselves to these elements in the sea water? If the blood plasma of Vertebrates, because of the forces of heredity, reproduce the proportions which obtained in pre-Cambrian oceans, why should not the cells of the tissues because of the same forces, reproduce in themselves the proportions which obtained in sea water of a much earlier geological period? In other words, if the proportions in the plasma are inherited, why should not those found in the living matter be considered as inherited also? An affirmative answer to this question would postulate that the proportions of the four elements in early pre-Cambrian seas were very greatly different from what they are now in the ocean—as different almost as the proportions of the four elements in muscle are from those found in the blood plasma.

The question is one of great importance in physiology, and, though its solution presents great difficulties, its very interest compels a consideration of it. We know that the unit of living matter, the cell, whether of animal or vegetable kingdom, presents, on the whole, the same type of structure, and it goes through the same morphological changes. Some of these are grouped under the process of division, and its characteristic details are the same in both animal and vegetable forms. Now, the animal and vegetable cells are derived from a single type which must have existed at the very dawn of life on the globe. The whole process of division, with its peculiar morphological features, was elaborated in this single-celled organism, which transmitted it to its descendants. Since, as already stated, the process of division is the same in both kingdoms, it is obvious that it has continued almost unchanged through an infinity of generations, animal and vegetable, and for many millions of years, and that this preservation of the original type is due to heredity. If, now, heredity is so powerful in regard to structure, is it a negligible force in regard to chemical composition? Is living matter fixed in structure almost beyond change, however widely the conditions under which it lives may vary, but unfixed and changeable in its relations to the chemical elements? As structure depends so largely on composition, it would be difficult to explain how living matter could so widely vary its relations to the elements and at the same time retain its structure.

We are, therefore, forced to a choice of hypotheses of which one postulates that all of the relations of living matter to sodium, potassium, calcium and magnesium are a result of inherited forces, while the other concedes that in regard to the circulatory fluids the proportions are determined by heredity, but the relations of these elements in living matter itself are due to quite different forces in which heredity is a small factor or no factor at all. The acceptance or rejection of either hypothesis depends on the evidence which we can bring as to the composition of the ocean in the very earliest geological periods.

The conclusions which we can formulate on this point depend on what we accept as the composition of the original crust of the lithosphere, and in our knowledge of the character and composition of the sedimentary rocks, and they must also be based on the changes which are admitted to have taken place in the composition of the ocean during all the periods. These conclusions I propose to deal with here in a general way only, for a full consideration of all the facts which have a bearing on them would demand a detailed treatment which would far exceed the limits set for this paper.

IV.—THE COMPOSITION OF THE PRIMEVAL OCEAN.

The original condition of the earth was a molten mass in which the temperature was so high that many of the elements now in the rock crust were in a gaseous condition, and dissociated, just as they are at present, in the solar atmosphere. As the dissipation of heat went on some of these must have condensed at degrees of temperature which approximated their present respective volatilization points, while the remainder, oxygen, hydrogen, chlorine, sulphur and carbon would combine to form water, hydrochloric, sulphuric and carbonic acids. The elements, sodium, potassium, calcium, magnesium, and aluminum would also before condensation take out of the original atmosphere chlorine, sulphuric acid, oxygen, and perhaps, carbonic acid, to form the chlorides, sulphates, oxides, and carbonates of these elements, but whether these compounds obtained after condensation depended on whether the temperature of the heated rock surface was still as high as their respective dissociation points. When the molten magma had cooled down to a degree below the lowest dissociation point, all the compounds referred to would be either deposited on the hot rock surface or in the form of vapour in the then atmosphere. When the temperature of the latter had fallen to about 1000°C, all these compounds were removed by condensation, for although, under the atmospheric pressure which now obtains, the temperature of condensation is for nearly all these compounds about 200° lower, the very great atmospheric pressure of the pre-oceanic period must have rendered the combination of the dissociated elements and the condensation of the compounds formed from them possible at a much higher temperature.*

At such a temperature the previously molten rock had become rigid, and of course the condensed compounds would be deposited on its surface, and when refusion of the rockcrust occurred, as it must have done over large areas, large quantities of the deposited compounds would be diffused through the superficial crust. When the cooling of the atmosphere and globe progressed until the temperature of the former was 370°C, the first condensation of water took place on the rock surface. The atmospheric pressure, according to Joly,† must have been about 270 times what it is now. According to Clarke's ‡ estimate of the relative values of water and carbon dioxide to that of the solid portion of the globe, the atmospheric pressure before the first condensation took place, was about 247 times what it is at present. Joly affirms that at 370°C a pressure of 190 atmospheres would produce a condensation of water, and, as the pressure was much higher, condensation would go on till the pressure fell below 190 atmospheres. This would entail rapid evaporation, for at many points the temperature of the rock surface would be so high that the water would condense only to boil away immediately. This would collect the salts deposited on the surface in masses, and it would, as in the case of the chlorides of magnesium, iron and aluminium, convert these into oxides of these metals and free chlorine, which, uniting with hydrogen, would form free hydrochloric acid. The other chlorides, namely, those of sodium, potassium and calcium would be unaffected. The ferric chloride would in some cases be volatilized but to be recondensed.

This condensation of the water vapour, and the re-evaporation would occur a countless number of times before there would obtain a permanent body of water on the globe. Where such first occurred there would be a lower temperature than elsewhere, and in consequence further condensation of water vapour would occur there also. The result would be the first ocean basin, the weight of the body of water acting on the

* The volatilization points of potassium, sodium and magesium are 667°C, 742°C, and 1100°C respectively. The melting points of calcium and aluminium are unknown. The melting points of certain sodium and potassium compounds are, according to V. Meyer & Riddle (Ber. d. d. Chem. Gesell. Vol. 27, p. 2,443.) as follows:

Na Cl	851°C.	K Cl	766°C.
Na Br	727°C.	K Br	715°C.
Na I	650°C.	K I	623°C.
Na_2 CO_3	1098°C.	K_2 CO_3	1045°C.
Na_2 SO_4	843°C.	K_2SO_4	1073°C.

† *Op. cit.*

‡ F. W. Clarke, The Relative Abundance of the Chemical Elements. Bulletin U. S. Geol. Survey No. 78, 1891.

thin crust and easily affecting the depression. These phenomena would be repeated at other points as the temperature of the crust and the atmosphere gradually lowered, until at a point below 100°C. nearly all of the water originally present in the atmosphere had condensed to form the oceans of the globe.

The composition of the ocean would follow from the occurrence of the soluble chlorides, sulphates and carbonates of the metals which came in contact with the first condensations. As pointed out, the condensation of superheated water would convert the chlorides of magnesium, iron and aluminium into magnesia (Mg O) oxide of iron ($Fe_2 O_3$) and alumina ($Al_2 O_3$), the first of which is soluble only in 55368 parts of hot or cold water,* while the two latter are practically insoluble, even in dilute acids. The magnesia, of course, would dissolve in water which contained either hydrochloric or carbonic acids, but the amount dissolved would, on account of the slight quantity of these acids in the water, be very small. The other chlorides, namely, those of sodium, potassium and calcium, although equally abundant, would not be leached out of the rock surface in equal amounts. The solubilities of these salts differ. For example, 100 parts of water dissolve at 99°C 154 parts of calcium chloride, 56.3 parts of potassium chloride, but only 39.7 parts of sodium chloride. In consequence there would be different quantities of each chloride dissolved, and the calcium chloride would by far predominate, while the potassium chloride would be more abundant than the corresponding sodium compound. There would, as already pointed out, be very little ferric chloride and what would be dissolved would gradually all be converted, first into the colloidal ferric hydrate, and eventually into the insoluble oxide of iron.

It does not follow that the ocean would contain, even after a long period of action on the rockcrust, the whole of the chlorides of calcium, potassium and sodium originally disposed over and diffused through the now more or less rigid rockcrust. The constant washing out of the land areas would no doubt tend to remove these salts from the rocks until there would be little left in the latter and at the same time they would become correspondingly more abundant in the sea water. But other salts would begin to appear there also. The magnesia derived from the chloride of magnesium, through the action of superheated water, would, under the action of carbonic acid in the rain water, go into solution as carbonate, but the amount so dissolved, would, on account of its low degree of solubility, be very small and it would only

*Fresenius, Liebig's Annalen, Vol. 59, p. 123.

after a long period of time become appreciable in the ocean. The carbonic acid in the rain water must have acted, as it does now, on the silicates of sodium, potassium and calcium in the rocks and produced free silica and carbonates of these elements, these latter going into solution and thus reaching the ocean, where, acting on the chloride of calcium, carbonate of lime and chloride of sodium and potassium would be formed. The calcium carbonates would be removed by deposition and thus constitute the origin of the limestone beds of the pre-Cambrian age, but the chlorides remaining in solution, thus contributed to an increase in the amount of sodium and potassium in the sea water.*

The sulphates in the rock crust disintegrated or affected would also be carried to the sea, but, as these would be small in quantity, they need not be specially considered here.

Thus the history of the sea must have begun and continued for a period of unknown length. The only change came from the discharge into the sea of the carbonates, the consequent removal of the lime and the slow increase in amount of magnesium, sulphuric acid, and of potassium and sodium. The two latter elements were not removed from the sea except through the rainfall. As I shall presently point out, the potassium compounds are to-day removed from the ocean apparently as rapidly as they are added by river water, and, in consequence, the amount in sea water now appears to be stationary. In the earliest geological period the conditions which now contribute to this result did not exist, and the ocean retained all the potassium it held or received through river discharge. In all probability the potassium equalled, and even exceeded, the sodium in amount.† When sediments began to form, and, when soils made their appearance, then, and then only began the elimination of the potassium from the ocean. It has been long established that potassium manifests a marked capacity to unite with silicates of alumina to form firm compounds, and these obtain whenever potassium salts in solution come in contact with argillaceous material, sedimentary or otherwise,‡ while the sodium, magnesium, and calcium are unaffected.

* Sterry Hunt (Chemical and Geological Essays, Boston, 1875) held the view that the most abundant constituent in primeval sea water was calcium chloride, and that with the gradual addition of sodium carbonate calcium was removed as carbonate and sodium chloride consequently took its place.

† Joly (*loc. cit.*) assumes that the greater part of the chlorine now in the ocean was originally united with the iron, calcium, magnesium, potassium, and sodium, these elements entering into combination in proportion parallel to the proportions in the rockcrust as determined by F. W. Clarke (*loc. cit.*) This postulates that 14 per cent. of the chlorine now in the ocean was united with sodium, and consequently the ocean originally contained about one-seventh of the sodium it now holds. As the proportion of sodium to potassium in the rock crust is 100 to 95, on Joly's hypothesis the potassium in the primæval ocean must have really equalled in amount the sodium therein. Joly, however, is in error in supposing that the chlorides of magnesium and iron could have existed, and he should consequently have made a greater allowance for the amounts of chlorine combined with the sodium, potassium, and calcium.

‡ Sterry Hunt (*op. cit.* p. 95.)

The capacity to abstract the potassium is increased if the silicates are mixed with organic matter. Consequently the potassium which rain water may contain is in great part removed when the latter filters through soils, and, therefore, the water discharge from alluvial areas is always richer in sodium than potassium. This capacity of soils to abstract potassium is a matter of direct demonstration, and it "explains the presence of so small an amount of potassium salts in the waters of rivers, lakes, streams, and oceans where the lime and soda have accumulated."* This cause of deficiency acts not only in the case of the potassium leached out of disintegrating rock by rain water, but also on the potassium carried from the sea to the land areas by rain water. The potassium thus carried is not inconsiderable, for, according to M. J. Pierre,† the rain water in the neighbourhood of Caen (France) annually carries to each hectare of land, about 7.9 kilograms of this element, or about 1.23 tons per square mile.

This mode of elimination also operates in the ocean, where, however, the organic matter responsible for the removal, is derived from plankton organisms, which, on dying, fall to the sea bottom and their remains decomposing, the potassium they hold reacts with the argillaceous material on which the deposits rest and forms the mineral known as glauconite, containing as low as 0.95 per cent. of oxide of potassium, but other estimates range from 2.52 to 4.21 per cent. The sodium present is very much less in quantity.‡ This mineral is now being formed, as it has been formed in the past, on the ocean bottom over the areas which fringe the continental coasts and it constitutes as much as, or more than, half of the deposits in shallower waters. Considering the extent of these areas as well as the fact that they cover the sea bottom of those localities into which river discharge takes place, it will be recognized what a very important factor the constant formation of glauconite is in eliminating potassium from sea water and thus preventing an increase in the amount of that element in the ocean. This formation has been going on in the past geological periods, for it is to be found§ in the primary formations of Russia and Sweden, in the sands

* Mendeleef's Chemistry, Vol. 1, p. 547, 1897.

† The reference is given in Dr. Angus Smith's "Air and Rain," which is quoted by Joly (*loc. cit.*)

‡ The analysis of five specimens as given by Murray & Renaud (Challenger Report, Deep Sea Deposits, p. 389) gave:

	I.	*II.*	*III.*	*IV.*	*V.*
CaO	1.69	1.26	1.27	1.34	1.19
MgO	2.49	3.13	3.04	2.83	4.62
K_2O	2.52	4.21	3.86	3.36	0.95
Na_2O	0.90	0.25	0.25	0.27	0.62

Other analyses quoted by Roth. (Allgemeine and Chemische Geologie, Vol. 1, p. 559, 1879) gave a percentage of potassium (not K_2O) varying from 2.8 to 7.3.

§ Murray & Renaud, op. *cit.*, p. 384.

and gravels of the Cambrian sandstone of North America, in the Quebec group of Canada and in the coarse Silurian sands of Bohemia. In the Mesozoic period it was more abundantly formed and its deposits are very marked in the strata of the Cretaceous division. It is also found in the Tertiary from the lowest strata to the highest of the series. It is thus shown that the formation of glauconite occurred in all the geological periods from the commencement of the Palæozoic Age to the present time and that thus a very large proportion of the potassium which the ocean would now contain, were it not for the formation of glauconite, has been removed from it.*

In the formation of glauconite, organisms appear to play a very distinct part and amongst these the Foraminifera are the most important. The decomposing organic matter of the dead forms liberates sulphur which combines with the iron in deposits to form sulphide.† This latter is converted into sulphuric acid which, acting on the fine clay sets free colloidal silica and ferric hydrate in a condition which promotes their union and the silicate so formed combines with potassium to form glauconite. It is obvious that organic matter is a very important factor in the process and that in the absence of animal organisms no glauconite would be formed, a view which explains the almost complete absence of this mineral from the deep sea areas, but it also postulates as decidedly, that before the appearance of living forms in the primeval ocean, there was little or no potassium eliminated from it, and this, taken in conjunction with the fact that in earlier pre-Cambrian times there could not have been much or any soil to affect the potassium in the waters discharged from the land areas, makes it quite clear that there was a period during which the potassium content of the ocean must have increased absolutely and that this was succeeded by a period in which the amount of the potassium ceased to increase or remained practically stationary, while decreasing relatively to other constituent elements. The beginning of this latter period coincided with the appearance of living forms in large numbers in the sea.

The history of sodium in the ocean has been one of uniform increase through all the geological ages. The addition that is to-day being made by river discharge is large and must have obtained as abundantly in the past. There have, on the other hand, been no important agencies which have served to eliminate it from the ocean. The great salt deposits, some of which are as old as the Cambrian, are,

* Forchhammer was the first to point out that potassium is being removed from the ocean, (British Association Report, 1844, p. 153.) From his analysis of Fucoids and of the metamorphosed Fucoid schists of Scandinavia he came to the conclusion that Fucoids constitute a very important factor in the process.

† Murray and Renaud, *op. cit.*, p. 389.

as is certainly the case with the Stassfurt beds, the result of the evaporation of land-locked arms of the sea.* and they are constituted of but an infinitesimal fraction of what is contained in the ocean. Sodium chloride, like other constituents of sea water, is carried landward with evaporation and rain clouds, but it appears to be returned to the ocean, without any perceptible loss, through the river discharge. The only method of elimination which at all possibly counts is that in which it is imprisoned mechanically in the sedimentary deposits during their formation. That sodium chloride is removed in this way has been pointed out and emphasized by Osmond Fisher, but there are no data which serve to indicate that this is a considerable factor in diminishing the sodium content of the ocean. All the known facts point in the contrary direction. There is no mineral in the course of formation, which is extensive or abundant in its distribution and which also requires considerable quantities of sodium for its production, and there are, further, no agencies acting in the soils which serve to remove sodium compounds from the percolating water.

In these considerations we find a full explanation for the relative proportions (100: 3.613) of the sodium and potassium which now obtain in sea water, and also for those which obtain in the river discharge of the globe. According to Murray's estimate for nineteen principal rivers, the proportions would be 100: 38.6. We may postulate from this that in the early geological periods of the pre-Cambrian period, when soils did not exist, the quantities of each element discharged by rivers or bodies of water derived from the land surface, were nearly equal. Since the primeval ocean, as pointed out above, contained these elements in almost equal quantities, this condition must have continued until long after soils holding organisms and organic matter had appeared, and even for an indeterminable period after organisms had made the ocean their habitat. The change in the relative proportions once begun must have gone on with extreme slowness, and oceanic organisms, at first wholly of the unicellular kind, must have, after acquiring a relation to these elements, just as slowly responded to the changes in the proportions of their medium.

The river discharge of the globe has been from primeval times adding also magnesium and calcium to the sea.. According to calculations based on Murray's data, the proportions relative to the sodium shown in these are 134 and 591 respectively to every 100 of the latter. This is, of course, based on approximate estimations, and they may be incorrect,

* See G. P. Merrill's "Treatise on Rock and Rock Weathering and Soils," p. 120, 1897.

as they seem to be, if one scrutinizes the proportions that are found in rivers whose waters have been carefully analysed. There are only two rivers, the Amazon and the St. Lawrence, which give nearly the proportion of magnesium called for by Murray's estimates, while the Ottawa, the Mississippi, and the Nile give quantities much below that of the sodium, and the quantities of the calcium are found to vary very much for the different rivers. If we disregard Murray's estimates and base our observations on the analyses of the various rivers, we can safely conclude that, while the quantity of calcium added, except in the case of the Nile, is always, and sometimes very much, greater than the sodium addition, the latter does not probably exceed the amount of the magnesium discharged. In the ocean, however, the sodium, calcium, and magnesium have the proportions of 100, 3.91 and 12.0.

The comparatively low proportion of magnesium in sea water is explainable. In the first place, as pointed out above, there must have been in the primeval ocean but very little magnesium, owing to the conversion of all the chloride of magnesium into magnesia which is, except in minute quantities, insoluble. The conditions which so affected magnesium chloride left the chlorides of calcium, sodium and potassium unchanged, and in conseqence these went into solution in primeval sea water, and were, therefore, as compared with magnesium, very abundant. Further, the ocean at first must have contained only traces of the latter element and the subsequent addition of it through river discharge would increase the amount in sea water, but not to such an extent as to make it overtake the sodium.

There is another factor which operated in limiting the amount of the magnesium. This is the tendency shown by the chloride to interact with the carbonate of lime when the latter undergoes deposition to form limestone, and, in consequence, this always contains carbonate of magnesia. When the latter exceeds 10 per cent. the mixture of the carbonates is given the conventional name of dolomite, and in some formations of this kind the magnesia is found greatly to exceed the lime. Dolomites are found in all the periods down to and including the Cambrian and even in the pre-Cambrian, it is associated with the crystalline schists.* An exact estimation of the magnesium so localized is impossible, but on the average it cannot be more than 10 per cent. of the quantity of the calcium due to deposition, so that the amount of magnesium removed annually from sea water must fall far behind that of the calcium. It follows from this that whatever were

* Zirkel, Lehrbuch der Petrographie, 1894, Bd. 4, p. 499.

the proportions of these two elements in primeval sea water. the proportions must have slowly changed, and as a consequence the magnesium must have gradually increased while the calcium practically remained stationary.

It must of course be admitted that magnesium is withdrawn from the ocean by organisms, but the amount thus removed is very small, and in no case is it an important method of eliminating the element from sea water. In the hard part of corals it is as a rule under one* per cent. and in the coralreefs it is less than that in amount, while the calcium constitutes nearly 40 per cent. Forchhammer's† analyses of the ash of sea weeds reveal a quantity of magnesium which he regarded as important, and he held that the Fucoids thus remove quantities of this element and deposit them in the beds which contain the solid substances of sea weeds as far as they are insoluble in water.‡ According to the analyses of Gödechens,§ the ash of Fucoids contains from 4 to 7 per cent. of magnesium. That the element is eliminated from sea water by these forms may be conceded, but it is doubtful if the quantity removed in this way is sufficient to affect materially in time the total amount retained in the ocean.

We may conclude, therefore, that in the formation of dolomites, of magnesia-holding limestones and chalk deposits, and, to a minor degree, in the activities of animals and plants, elimination of magnesium from sea water has always obtained; and, further, that the amount eliminated annually does not equal the amount of magnesium added to the sea by river discharge. This postulates a constant increase in the amount of magnesium in the sea; and in this respect it must be ranged with sodium, which increases in amount at a greater rate, since, so far as is known, there are for it no agencies of elimination in operation which compare with those affecting the potassium, the calcium, and even the magnesium. The sodium, therefore, though it is not added in greater amount than in the case of the latter, is increasing at a greater rate, and thus the proportion of sodium to magnesium in sea water is slowly altering. As pointed out above, the primeval ocean must have contained but an exceeding small quantity of magnesium, and the amount of the latter now in it is practically wholly derived from the leaching out of the land surfaces during the intervening ages.

As regards the calcium in sea water there is less uncertainty. The

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* According to Forchhammer the corals, *Isis nobilis* and *Corallium nobile*, contain 6.36 and 2.1 per cent. respectively of magnesium carbonate.

† Roth, *op. cit.*, p. 616, where the results of analyses of a number of forms are given.

‡ *Op. cit.*, p. 159.

§ Ann. d Chem. und Pharm., Vol. 54, p. 351, 1854.

calcium of river discharge greatly exceeds in amount that of the three other elements, and yet it is less abundant than either in the ocean. If there were no elimination of calcium from sea water, the salts of the latter element would long have reached the point of saturation in the ocean. The present condition is easy of explanation. On the one hand, calcium separates from sea water through the formation of sulphate and carbonate of lime, which are to a high degree insoluble. This constitutes in part the origin of the gypsum beds and of the limestones of sedimentary origin. On the other hand the myriads of organisms that have their habitat in the sea have the lime "habit," and they consequently remove from solution enormous quantities of calcium. This is the case not only with all forms provided with exoskeleta and endoskeleta, into the composition of which lime largely enters, but also with those which exercise the precipitating effect on the calcium salts they absorb from sea water, the precipitation rarely going so far as to form a distinct deposit in the cells or tissues of the organism. This power to precipitate is universal, as shown by the fact that the capacity to form calcareous skeleta is almost universal, and this capacity is merely an enhancement of the power to precipitate. The latter, therefore, operating so largely, separates calcium from sea water, and on the death and disintegration of the organisms, the element is deposited on the sea bottom either as phosphate or carbonate of calcium.* These deposits, owing to the fact that they contain few calciferous fossils, are regarded as due to chemical reactions alone; but if they are, sedimentary limestones should be of a more uniform distribution, whereas we find them more or less localized. The explanation that they are due to protoplasmic "secretion" and not to either chemical reaction or skeletal deposition in living forms accounts for much, and indicates what a factor living protoplasm, animal and vegetable, is in the separation of calcium from sea water.

Sterry Hunt† advanced the view that in the primeval ocean the chief salts were chlorides of calcium and magnesium, and that the constant, large output by river water of carbonates of sodium and potassium, and particularly of the former, affected a conversion of these into carbonate of lime and chlorides of sodium and potassium which were retained in solution, while the carbonate was deposited. The objection to this view is that, if it is correct, the conversion ought to have taken place in the pre-Cambrian period, and, therefore, there ought to be extensive limestone deposits in the rocks attributed to that period.

* Sterry Hunt, *op. cit.*, pp. 82 and 311.

† *Op. cit.*, pp. 2 and 41.

There is, indeed, in these rocks only a small amount of crystalline or other limestone, and there appears to be still less in the divisions of the Huronian, which, as pointed out, have a thickness in the Lake of the Woods and Rainy Lake districts, according to Lawson,* aggregating 50,000 feet, all representing sedimentary formations. What limestone and gypsum are present in pre-Cambrian rocks can very well be attributed to the occurrence of calcium salts in the oceans of the Archæan in quantities, however, which could not have very greatly exceeded those which obtain to-day in sea water.

V.—THE RELATION OF THE SALTS IN THE OCEAN TO PROTOPLASM.

From the considerations advanced in the preceding section of this paper, it follows that the ocean has been, and is now, slowly changing, not in its composition, but in the proportions in it of the various elements to each other, and that, as a consequence, it is now in this respect greatly different from what the primeval ocean was in the period following the first condensations of water vapour on the rock crust of the globe. It may again be noted that in all these changes there are two distinct periods. In the first, or older, life was not represented except towards its close, and therefore, the only factors engaged in eliminating any of the elements from the sea were purely chemical ones such as are illustrated in the precipitation of lime as carbonate and sulphate and of magnesia as carbonate. In this period the elements must have differed in amounts from each other less markedly than they do to-day, and the constant addition to these from the discharge from the land surfaces did not tend to alter, even after a very long interval, the proportions which first obtained. This period must have terminated some considerable time after the appearance of living forms on the globe, and especially only after the adaptation of vegetable forms to a land life, and the consequent production of soils. The second period could not have begun at once after the appearance of living forms, for these must first have acquired a relation to the elements and then have developed the habit of disposing of the various salts which they took out of the sea water. This period may well be supposed to have begun when there had developed not only a considerable diversity of forms in the sea, but also the organisms which contribute to the production of organic matter in soils. In this period the removal of potassium from the land surfaces decreased,

* *Loc cit.*

and the combination of this element with argillaceous matter at the bottom of shallow portions of the sea began. As a result the amount of potassium in sea water became stationary. At the same time the removal of calcium on a larger scale than obtained in the preceding period commenced and this checked the increase of calcium salts.

The first forms of life in the primeval ocean were undoubtedly unicellular, and they were probably also organisms which presented features intermediate between those of the vegetable kingdom on the one hand, and those of the animal kingdom on the other. These forms must have persisted for a period of unknown but very great duration, for in them developed not only a nucleus but also the capacity on the part of the latter to divide in the remarkable and complicated manner illustrative of karyokinesis, and which is characteristic now of the cells of both kingdoms. This process of division, so alike in its main features in animal and vegetable cells, must have become fixed before specialization had gone so far as to evolve both animal and vegetable types, for, had it been otherwise, there would have been greater differences in the process in animal and vegetable forms. That the process has continued practically unchanged in all the intervening millions of years shows how deeply fixed in the organism this morphological habit has become, and, therefore, the act of fixation must have taken an incredibly long period of time during which the ocean was changing, not in the relative proportions to each of the elements it contained, but in the absolute amounts of these.

During this long period, these organisms, neither distinctly animal nor distinctly vegetable, exposed as they were to action of these same elements, must have acquired a relation to them as fixed as the karyokinetic process was becoming. Their protoplasm had established all its normal processes in the presence of potassium, sodium, calcium, and magnesium in certain proportions in sea water, and, after the lapse of the long period of time required for the elaboration of the karyokinetic method of division, these processes became unalterably dependent on the presence of the elements in the proportions which then prevailed. Without this fixed relation life could not continue, and when specialization into animal and vegetable forms occurred this fixed relation was transmitted to the forms of both kingdoms. How long these latter forms remained unicellular cannot, of course, be surmised, for there are no means of determining the length in time of this or any part of the pre-Cambrian age, but that it was of very great duration can hardly be questioned, and it must have strengthened the relation which obtained between protoplasm on the one hand and the elements in certain proportions, on the other.* In consequence, their descendant forms inherited this relation, and transmitted it to the forms and species which arose through variation and other causes. When multicellular forms arose these were endowed with the same relation.

The proportions of the elements in the early pre-Cambrian ocean with their long action on protoplasm must then have conferred a more or less fixed property on the latter and, in consequence, living matter, whether animal or vegetable, now shows in its ash proportions of the elements greatly different from those found in the media in which it lives or in the circulatory fluid which bathes it. This relation or property resists change even after exposure to altered conditions for a very long period of time. Before the circulatory fluid (blood plasma) was established in multicellular animals, a great change must have occurred in the proportions of the elements in the ocean, a change which would account for the wide differences between the proportions in the protoplasm or tissue on the one hand, and those in the blood on the other.

The proportions of the elements in living matter are due then to conditions which obtained in the ocean far back in the pre-Cambrian age, while those in the blood or plasma are due to conditions which occurred in the ocean long after this and yet before the beginning of the Cambrian period. The proportion of potassium to sodium in blood plasma is nearly† double what it is in the ocean and therefore that difference must have resulted in the period that has elapsed since the rudiments of a circulatory system were developed in those Metazoan animals which gave rise to Vertebrates.

As pointed out above, it is difficult to obtain the exact proportions of the sodium, potassium, calcium and magnesium in living matter, for, except in muscle fibre, protoplasmic structures cannot in sufficient quantities be freed from adherent material which carries these elements in very different proportions. Calcium exists in tissues apart from the protoplasm and as precipitates or deposits, and according to recent observations which I have made, this is true in a very large degree of

* Geologists concede a very long time to the pre-Cambrian, a duration which, according to the different estimates, ranges from one-third to four-fifths, and even nine-tenths, of the whole geological period. The very fact that all the chief types of animal life, and perhaps also of vegetable life as well, appeared before the close of the pre-Cambrian age, indicated that the latter was of inconceivably long duration.

† Amongst the oldest and highly specialized forms are *Olenellus* and the Brachiopods of the Cambrian. The oldest Vertebrate remains are in the Trenton division of the Silurian, more recent than the Cambrian, but these are "ganoid" in character and this fact postulates a long preceding period of development out of Protovertebrate forms which therefore could not have first appeared much later than the beginning of the Cambrian. The circulatory system of Vertebrates accordingly has a history which began in the pre-Cambrian age.

potassium. With regard to this element it may be said that active living matter has the power of absorbing it in large quantities and disposing of it in an inert form by precipitating it at the peripheries of cells or in inert organic masses within them, and, as a consequence, the ash of animal and vegetable cells shows a larger quantity of potassium than the protoplasm of the cells required. This illustrates how difficult it is to determine the primitive and fixed proportions of potassium and calcium, and further, how little we should depend, even in the case of muscle, on the analyses of the ash of organs or organisms, for this purpose. If a sufficient quantity of Amœbæ* could be obtained for analysis it might yield results of value but until that is done, the exact proportions of sodium, potassium, calcium and magnesium must be a matter for conjecture. It can scarcely be that the proportions found in muscle represent even approximately those which should obtain in undifferentiated propoplasm or cells.†

VI.—EVIDENCE FROM THE LAKES AND RIVERS OF THE PRESENT PERIOD.

It may be pointed out that in the composition of the rivers, large lakes, and seas of the world, there is evidence confirmatory of the view that potassium and calcium predominated in the pre-Cambrian seas. The conditions, of course, which contribute to the composition of the lakes of to-day are not the same as those which existed when the oceans of the globe were formed. There are but infinitesimal traces of the chlorides of calcium and potassium in the rock crust or sedimentary strata, and, further, there are, apart from the deposits of salt, and that amount of it due to rainfall, but small quantities of sodium chloride which can to-day come under the leaching action of water. There are also soils to alter the proportions of the chemical elements derived from them.

Nevertheless it happens that in lakes surrounded, either wholly or

* Fresh water, unicellular animal organisms are apparently free from excess of the elements. They are, as a rule, free from potassium, at least in such quantities as are found in other organisms.

† According to J. Katz (Pfluger's Arch., vol. 63, p. 1), the proportions in muscle from different animals are:

	Na.	*K.*	*Ca.*	*Mg.*
Man	100	400	9.3	26.4
Dog	100	354	7.26	25.1
Rabbit	100	870	40.0	60.5
Pike	100	1415	145.0	105.0

These and other results of the same observer are open to the objection that no effort was made to get muscle fibre free from all adherent tissues. Visible blood vessels, tendons, nerves and fat were indeed removed but these constitute only a part of the non-muscle portions of the tissue and they may be the cause of the variations shown in the results.

partially, by regions in which pre-Cambrian formations occur, or are the only apparent rocks, the potassium predominates over the sodium. For example, in the water from Reindeer Lake, which is situated 400 miles directly north of Lake Winnipeg, Professor Adams found the potassium to exceed very greatly the sodium. In the water from the Churchill River, as well as in the water from the Saskatchewan River above the junction of the Big Stone River, the potassium is much richer than the sodium.* These rivers drain rocky areas chiefly of the pre-Cambrian type. Rocks of the primitive kind, therefore, contrary to the prevailing opinion,† supply to the water which comes in contact with them more potassium than sodium.

Even in the case of Lake Superior which draws its supply not only from the primitive rock region on its northern side, but also from the areas covered with soils of alluvial and drift origin on the south, the potassium is about equivalent to the sodium. In the lakes of the Bavarian Highlands, Rachel See, Würm See and Ronig See, the potassium is twice in amount that of the sodium. In Lake Zurich the potassium exceeds the sodium. In Lake Geneva, in Pyrenean and Vosgean Lakes and in those of Russia, Armenia and Central Asia the potassium is approximately two-thirds of the sodium. It is probable that if proper methods for estimating potassium had been current in his day, C. Schmidt would have found for the lakes of Russia, Armenia and Central Asia a higher potassium value than he obtained, for the methods then in vogue for the determination of the element in the presence of sodium were very faulty and gave very low results. It is probable also that this may explain the low value found by Sterry Hunt for the potassium of the Ottawa River, whose waters, as well known, are derived largely from Archæan regions.

The Tables A and B show further that, in nearly all cases, the calcium is very abundant. In the Nile only, amongst the rivers, is it less than the sodium, while it very greatly exceeds it in the rest. In the lakes it is very abundant relatively, with the exception of the Rachel See and Lake Onega. In the Bavarian lakes, Lake Geneva, Lake Zurich, and some others, it is exceedingly abundant relatively.

The magnesium is always less than the calcium, and the relative difference is sometimes very great. It may fall below the sodium, but, as a rule, it is greater in amount.

These proportions, one can readily understand, must have been

* F. D. Adams Geo. and Nat. Hist. Survey of Canada, 1880-2, p. 6, 4.

† This opinion is based largely on the fact that the potash feldspars are difficult to decompose while the soda feldspars readily undergo decomposition.

uniformly maintained for an indefinitely long period. It may even be claimed that, in the case of Lake Baikal, of Reindeer Lake, and other lakes supplied from Archæan areas the proportions have obtained from pre-Cambrian times, and further, that the river discharge of that period, coming as it did from pre-Cambrian rock areas wholly, would contain the four elements in these or similar proportions. That would postulate that the primeval ocean was merely a gigantic body of fresh water, in which the sodium, potassium, calcium, and magnesium obtained in quantities and proportions as they now obtain in a lake situated in Archæan area. As already pointed out, these proportions gave place to others, and to-day, as in the past, the relative amounts of each element are changing, so that in a few million years hence the composition of ocean water will be appreciably different from what it is now.

One can, indeed, illustrate what changes have taken place in the ocean by reference to such a large body of fresh water as Lake Superior. If the latter were to lose its outlet no doubt its area would be larger than it is now, but when that had attained a certain extent the evaporation would balance the inflow as in the case of the Caspian Sea, and in consequence the salts held in solution would constantly increase in amount, but each at different rates up to a certain point, when the proportions would begin to approximate those in ocean water. One cannot of course say that this is what has happened in the case of either the Caspian or the Sea of Aral, for these bodies of water were connected with the ocean as late as the beginning of the Tertiary age, but it may be pointed out that if their composition was, to start with, the same as that of the ocean in Tertiary times, their present composition is strong evidence of the effect that the salts derived from leaching of the land areas have in modifying the proportions, for in that respect either is markedly different from the other and from the ocean.

The Great Salt Lake of Utah may be adduced as an instance of the change of a body of fresh water into one which presents a high degree of salinity, and which in the proportions of its salts is remarkably not unlike the ocean. This lake, which is in part of the area covered by the glacial Lake Bonneville, is considered by G. K. Gilbert to have been a body of fresh water about 25,000 years ago. He arrived at this result by determining the discharge of chlorine into the lake by river water and comparing it with the quantity at present obtaining in the lake. Lake Bonneville had* an outlet delivering its waters into a tributary of the Columbia River and thus the lake was kept fresh. When, however, this outlet was lost, changes climatic and physical operated to reduce

* Monographs of the U. S. Geol. Survey, Vol. 1, Lake Bonneville, 1890, p. 254.

the volume of water, and, evaporation keeping pace with the inflow, a concentration of the salts held in solution took place. An examination of the present sources of inflow shows that these do not contain the sodium, potassium, calcium and magnesium in the relative proportions which are found in the lake. Gilbert estimates that it would take only eighteen years to give the lake through its fresh water inflow, all the calcium it now contains and that 850 years would to this end be required for magnesium. He does not deal with the case of the potassium of which the analyses he reports show only traces in the inflow water, but this also may have been due to faulty methods of determining that element. These latter seem to be the only explanation for the great discrepancy between the amounts of potassium found by Talmage* in 1889 and Bassett† in 1873‡.

Short as is the extreme period required by Gilbert's calculations to affect all the changes in the composition, it has epitomized the history of the ocean. Even if we postulate that the primitive rock crust of the globe in pre-Cambrian times contained more sodium chloride than what is found now in Archæan formations, there is also more of this salt in the strata of later geological periods which cover the drainage area of Utah Salt Lake. Of course there is not a complete parallel between the latter and the ocean, for the relative proportions are not exactly the same, but their approximate similarity is striking, and, it may be added, very convincing as to the extreme probability of the thesis maintained above.§

TABLE A.

RIVERS.

	Na.	*K.*	*Ca.*	*Mg.*	*SO₃.*	*Cl.*	*Si.*	*Fe.*
1. St. Lawrence ...	100	22.9	638.0	143.4	136.0	223.0	343.0	....
2. Ottawa	100	64.2	416.7	82.5	67.3	224.3	402.0	....
3. Mississippi	100	35.5	462.0	82.0	17.1	8.4	86.4	17.0
4. Amazons.........	100	72.6	1,089.0	135.6	36.0	90.0	....	....
5. Nile	100	22.2	75.1	41.5	18.5	16.0	44.4	....
6. Assinaboine.....	100	10.5	122.0	69.4	127.9	50.0	....	....
7. Red River	100	12.3	133.3	83.2	190.0	91.4	....	....
8. Nineteen Rivers (Murray)	100	38.6	590.9	134.2	197.6	53.5	145.3	37.9

*Science, Vol. 14, 1889, p. 445.

†Chemical News, Vol. 28, 1873, p. 236.

‡See Table B, Utah Salt Lake, 26 and 27.

§ In Lake Shirwa, according to J. E. S. Moore, ("The Tanganyika Problem," 1902, p. 22.) we have a lake which was once fresh, but has become salt through the loss of its outlet. So far as I know no analyses have been made of its waters.

TABLE B.

LAKES AND SEAS.

	Na.	*K.*	*Ca.*	*Mg.*	*SO3.*	*Cl.*	*Si.*	*Fe.*
9. Superior........	100	97.2	1,015.0	208.0	27.5	103.2	19.3	....
10. Rachel See	100	199.4	13.0	....	....	17.5	....	1.6
11. Würm See	100	223.9	2,445.0	809.0	....	....	....	....
12. Walchen See....	100	117.7	2,557.0	808.5	....	....	....	....
13. Ronig See	100	207.9	5,812.0	588.8	....	....	....	....
14. Schlier See......	100	96.4	3,141.0	661.0	....	....	....	....
15. Lac Gaube (Pyrenean).....	100	66.3	421.0	17.9	....	....	....	....
16. Lac Gerardmer (Vosges).......	100	70.4	117.6	29.8	....	....	....	....
17. Lake Geneva....	100	74.5	2,345.0	341.3	....	55.0	1,806.0	....
18. Lake Zurich.....	100	111.7	1,843.9	272.0	441.0	36.0	60.8	....
19. Lake Peipus.....	100	75.2	929.5	159.9	18.5	134.0	13.3	3.4
20. Lake Onega	100	71.4	67.3	51.4	33.4	104.7	36.9	2.2
21. Lake Tschaldyr (Armenian Highlands)	100	59.3	219.3	45.7	88.2	65.3	172.1	5.5
22. Lake Baikal.....	100	58.9	399.6	60.6	98.5	41.7	16.1	17.3
23. Sea of Aral	100	2.38	18.6	24.2	113.0	156.0	....	....
24. Caspian Sea.....	100	3.35	9.4	22.7	80.0	163.7	....	....
25. Dead Sea	100	28.4	40.3	1.27	2.0	556.1	....	....
26. Utah Salt Lake..	100	3.22	1.22	7.81	14.9	169.2	....	....
27. " " ..	100	25.8	1.56	7.82	19.1	192.2	....	....
28. Ocean	100	3.613	3.91	12.0	20.9	180.9	.. .	....

REFERENCES FOR ANALYSES.

1 and 2. Sterry Hunt, Phil. Mag., 4th series, Vol. 13, p. 239, 1857.

3. Warren Upham, The Glacial Lake Agassiz, United States Geol. Survey, 1895, p. 543.

4. Mellard Reade, Am. Journal of Science, Vol. 29, p. 295, 1885.

5. O. Popp, Annalen d. Chem. und Pharm., Vol. 155, p. 344, 1870.

6 and 7. Warren Upham, *Op. Cit.*, p. 542.

8. John Murray, Scottish Geogr. Mag., Vol. 3, p. 76.

9. Warren Upham, *Op. Cit.*, p, 542.

10. Johnson und Lindtner, Liebig's Ann. der Chem., Vol. 95, p. 230, 1855.

11-14. Adolf Schwager's Analyses in Willi Ule's Der Würm See (Starnbergersee) in Oberbayern, Leipzig, 1901.

15-17. Delebecque, Les Lacs Français, Paris, 1898, pp. 197-205.

18. F. Moldenhauer, Schweizer Polytechn. Zeitschr., 1857, II., p. 52. (Reference in Kopp and Will's Jahresber. der Chem., 1857, p. 724).

19-22. C. Schmidt, Bulletin's Acad. Imp. de St. Petersburg, Vol. 28, 1883, pp. 245-6.

23 and 24. C. Schmidt, Bull. Acad. Imp. de St. Petersburg, Vol. 20, pp. 134 and 143, 1875.

25. R. F. Marchand, Journ. für Prakt. Chem., Vol. 47, p. 365, 1849.

26. J. E. Talmage, Science, Vol. 14, 1889, p. 444-6.

27. H. Bassett's Analysis, reported by G. K. Gilbert, Lake Bonneville, U. S. Geol. Survey Monographs, p. 253, 1890.

28. — Dittmar, Challenger Report, Physics and Chemistry, Vol. 1, p. 203.

SUMMARY.

The points discussed in the preceding pages may be summarized as follows :—

1. The composition of the ocean represents the result, on the one hand, of the leaching action of water on the land surfaces of the globe continued throughout all the geological periods, and, on the other, of the chemical and other agencies modifying or enhancing the power of sea water to retain in solution the mineral constituents derived from the land surfaces through river water since the beginning of the primeval period.

2. The relative proportions of the elements, and especially of sodium, potassium, calcium, and magnesium, in river discharge are not parallel to those of the same elements found in the sea. In river water, the calcium is always more, and the potassium less, abundant than the sodium, while the magnesium appears to approximate in amount the latter. In the sea, on the other hand, the sodium is much more abundant than the other three elements, and this is due to the continuous precipitation of a very great portion of the calcium added by rivers as carbonate, to the subsequent fixation in the limestone so formed of the magnesium as carbonate, and to the removal, continually taking place, of potassium, which is affected through animal and vegetable forms, and its consequent fixation in submarine deposits as glauconite and other potassium-holding minerals. The calcium and potassium appear to be stationary in amount, while the magnesium added by river water appears to exceed in amount that removed from the sea, and, in consequence, is slowly on the increase in the ocean, but its rate of increase is far behind that of the sodium.

3. The relative proportions of the elements in the ocean have, therefore, always been changing, and these proportions must have been, in the earlier geological periods, very different from what they are now. In the ocean of the earliest period the relative proportions of the elements approximated those found in river discharge, or rather those found in fresh water shed from areas covered with Archæan rocks. In this the potassium approaches the sodium in amount while the magnesium exceeds the latter, and the calcium is relatively very abundant.

4. This condition must have continued until living forms made

their appearance in the ocean when the gradual elimination of the magnesium, and particularly of the potassium and calcium, began. The forms were in all probability unicellular, and as the period must have been of great duration, the organisms and their protoplasm acquired a fixed relation to the four elements.

5. With the appearance of vegetable land forms and the formation of soils the removal of potassium from the land to the sea by river water diminished, and this, in conjunction with the elimination of the element from sea water by organisms, made the amount in the sea stationary. Through the action of living forms the calcium also in sea water has been kept stationary since that remote period.

6. In the transition from the ocean of the more ancient composition to that of the present, the unicellular forms became multicellular, and developed circulatory systems, the vascular fluids of which were at first simply modified sea water. In the blood plasma of Vertebrates, the three elements, sodium, potassium, and calcium are in relative proportions strikingly like those which now obtain in sea water. The magnesium only is considerably less than it is in sea water. The whole is due to heredity, the proportions of the saline constituents of the plasma being a reproduction of the proportions which obtained in sea water when circulatory plasmata were developed.

7. The proportions of the four elements which obtain in living protoplasm are as yet unknown, for the latter has the power of precipitating the potassium, calcium and probably the sodium and magnesium as inert compounds in itself or in its adventitious structures, and thus analyses would comprehend the inert material as well as the quantities of these elements which are actively participating in the processes of the living substance. If we could determine the latter quantities alone we could regard them as a representation of the proportions obtaining in primeval sea water to which the protoplasm of unicellular organisms had established a fixed relation.

8. That such a relation could be inherited may be inferred from the fact that the karyokinetic process, being practically the same in the animal and vegetable cell, has continued unchanged in both from the primeval period when the karyokinetic process first developed in a parent unicellular organism neither distinctly animal nor distinctly vegetable. This indicates how marked an influence heredity wields.

9. Briefly, animal as well as vegetable protoplasm owes its relations

to the elements sodium, potassium, calcium and magnesium to the composition of sea water which obtained when all forms were unicellular, just as the blood plasma owes its relations to the same four elements to the composition of sea water which prevailed when circulatory fluids were established. In other words the relation of protoplasm to salts is due to the action for ages of sea water, for incalculably long periods of time, on the living matter of unicellular organisms.

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