

A PRELIMINARY DETERMINATION OF THE PART PLAYED BY MYELIN IN REDUCING THE WATER CONTENT OF THE MAMMALIAN NERVOUS SYSTEM (ALBINO RAT)

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ONE CHART

It is a familiar fact that there is a progressive loss of water in the brain and spinal cord with advancing age. This is illustrated for the albino rat in Chart 1.

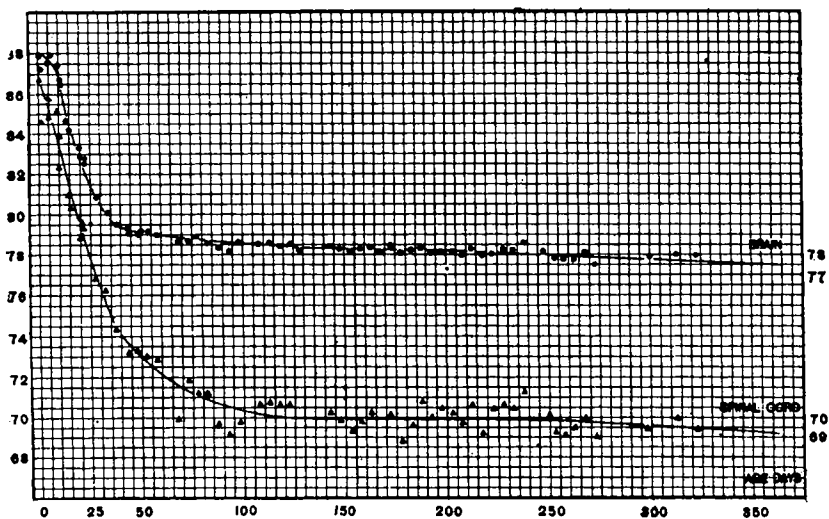


Chart 1 Showing the percentage of water on age in the central nervous system of the albino rat. The upper graph gives the values for the water in the brain as determined by the formulas (Hatai, in 'The Rat,' Donaldson, '15). The lower graph gives the corresponding values for the spinal cord, determined in the same way.

The small black dots indicate the observed values for the several age groups for the brain, and form the data for the formulas. The small black triangles have a like value in relation to the spinal cord.

It is also well known that in some mammals all the axons in the central nervous system are unmyelinated at birth, while in other species a greater or smaller number of them may already have their sheaths. In all cases, however, an active formation of myelin occurs during the period of rapid growth (Koch and Koch '13).

In a previous study on the percentage of water in the albino rat I was misled by certain graphs (Donaldson '10) to the conclusion that probably both the axons and their myelin sheaths changed in their water content so as to produce the well known reduction of water which is observed, but further study has shown that this is an incorrect view, and it is the object of this paper to present the evidence of my revised conclusion.

Since it has already been shown that the loss of water in the human brain follows the same course as in the brain of the albino rat—and has similar limits (Donaldson '10)—it is permissible to use in the argument certain observations on the human brain.

From the data for the human brain already in the literature I have selected those published by de Regibus ('84), because this author evidently examined only the outer layers of the cortex when making his determinations for the water in the gray substance and because he was able also to obtain remarkably uniform results for all of his determinations.

De Regibus tested four male brains, 25 to 76 years of age and three female brains, 30 to 60 years of age.

In Table 1 there appear also determinations of the water content in the human cortex and in the fibers at birth. These are based on the records of Weisbach ('68).

This enables us to contrast in Table 1 the conditions at birth with those at maturity.

TABLE 1
Percentage of water in the gray and white substance of the human brain at birth and at maturity

	CORTEX (GRAY)	CALLOSUM (WHITE)
	<i>per cent</i>	<i>per cent</i>
At birth (Weisbach).....	88	88
At maturity (de Regibus).....	86	70.4

According to this table the (gray) cortex has lost 2 points and the (white) callosum 17.6 points in the process of maturing.

It is never possible at maturity to obtain the cortex or any other gray mass without some admixture of myelinated fibers and I have therefore, provisionally, credited one point of the loss, noted by de Regibus in the water content of the cortex, to the presence of myelin. This implies but a small proportion of myelin since if we assume that myelin has 48 per cent of water the reduction of 1 per cent would mean that about one thirty-ninth of the mass was represented by myelin. According to this assumption the mature gray substance (cortex)—when the myelin is excluded—contains 87 per cent of water, and in the computations which follow the neurons without myelin are assumed to have 87 per cent of water, except at ten days, when they are credited with 88 per cent.

The fact that the fibers without myelin have at birth a high percentage of water (88%) while at maturity, after myelination, they have lost 17.6 points is the sort of evidence which furnishes the basis for the current, but unsupported view, that the loss of water is to be associated with the formation of the myelin. It is our object to present more precise information bearing on this point.

To obtain a notion of the approximate distribution of the water between the myelin and neurons proper, it is necessary to have data on the relative abundance of these two constituents of the brain.

In 1913 W. Koch and M. L. Koch made a study of the chemical composition of the brain of the albino rat at six ages between birth and maturity, and of the spinal cord at one age. The data thus obtained are those which will be utilized here. The authors determined seven fractions: Proteins, organic extractives and inorganic constituents, which three taken together, we shall designate protein (or non-lipoid), and phosphatides, cerebro-sides, sulphatides and cholesterol, which four taken together, we shall designate lipid.

These data give us at each age, therefore, the protein and the lipid present in the brain, or to be a little more exact, we should

say the lipid and the non-lipid fractions. The lipid (in part) represents the myelin sheaths, while the protein, with the remainder of the lipid, represents the cell bodies and their unsheathed axons.

With the exception of the one day group, the ages for which analyses were made, are given in table 2.

TABLE 2

To show, for the brain of the albino rat at five ages and for the spinal cord at one age the percentage of water in the myelin as computed according to the method described. The protein and lipid are given in percentages of the total dry substance. (Based on table 2. Koch and Koch '13)

Brain

(1) AGE IN DAYS	(2) CORRECTED PROTEIN	(3) CORRECTED LIPOID	(4) PROPORTION OF LIPOID FOR DIFFERENT AGES. LIPOID AT 20 DAYS = 1	(5) (6) PERCENTAGE OF WATER		(7) WATER. IN MYELIN = LIPOIDS (C) (COMPUTED) (See p. 447)
				Entire brain observed	In Neurons = protein (C) (assumed)	
10	<i>per cent</i> 93.80	<i>per cent</i> 6.2		86.5	88	63.8 ¹
20	88.88	11.12	1.0	82.5	87	46.5 ²
40	82.86	17.14	1.5	79.4	87	42.7
120	75.11	24.84	2.2	78.4	87	52.4
210	76.36	23.63	2.1	78.1	87	47.4
Average of 20 to 210 days.....						47.8

Spinal cord

120	52.92	47.08	4.2	70.4	87	51.0
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¹ First traces of myelin

² Myelin well shown.

At one day—or practically birth—it is found that the lipid is present to the extent of 0.31 or nearly one-third of the weight of the protein. There is, however, no visible myelin at this age, so it is concluded that this proportion of the total lipid is normally associated with the protein and is not to be included in the lipid which forms the myelin sheaths. We have treated the data for the later age groups in accordance with this relation, and in each case have taken from the total lipid found an

amount equal to 0.31 of the protein found. The remaining amount of lipid is assumed to be that used for the sheaths.

In table 2, the column (2) headed Corrected Protein gives the observed protein (non-lipoid) plus 0.31 of itself and the column (3) headed Corrected Lipoid gives the observed lipid less the amount of lipid added to the protein.

In table 2 the data are given in five age groups for the brain and in one age group for the spinal cord. It is to be noted that the 10 day brain group—which stands just at the beginning of the myelin formation—is for the moment excluded from the discussion and we begin the comparisons which are to be made, with the 20 day brain group.

In the brain series (with one exception) the corrected protein diminishes and the corrected lipid increases with advancing age. Between 20 and 210 days the proportion of the lipid doubles—column (4). We have in column (5) the observed percentage of water in the brain as a whole. It is assumed, as previously noted, that the corrected protein (neurons in the strict sense=both cell bodies and axons) have 87 per cent of water. From these several data we can compute the percentage of water to be assigned to the corrected lipid, which represents the myelin.¹

The method of computation may be illustrated by the data for the 20 day group. Reference to table 2 shows that at this age there is 1 part of lipid (11.12%) to 8 parts of protein (88.88%). This gives 9 parts, representing the entire brain and having 82.5 per cent of water. The product, $9 \times 82.5 = 742.5$. We assume that the 8 parts of protein have 87 per cent of water. The product, $8 \times 87 = 696$. The 1 part of lipid, representing the myelin, will then have a percentage of water equal to the difference of these products $= 742.5 - 696 = 46.5$ per cent.

¹It is hardly necessary to point out that a division of the brain into myelin on the one hand and neurons on the other fails to enumerate several structural elements which are also present though representing a relatively small mass. There are, in addition to the neurons, glia and ependymal cells; blood and lymph vessels; blood and lymph and a slight amount of connective tissue. Over against the neurons plus this group of elements we put the myelin, but for the purposes of the present discussion it is convenient to speak of the rest—the non-myelin portion—of the brain as if represented by the neurons alone.

As table 2 shows, similar computations give percentages of water for the myelin in the brain, which range from 42.7 per cent to 52.4 per cent and which yield a mean value of 47.8 per cent. The computation for the spinal cord gives 51 per cent. The significance of these results lies not in the particular percentage of water here determined for the myelin—as that depends somewhat on the percentage of water assumed for the protein—but on the similarity of the values found in all the five cases examined.

However, it is found on trial that one cannot depart far from the value of 87 per cent for the proteins without obtaining rather improbable percentages for the myelin, so that this value is probably nearly right.

DISCUSSION

Those familiar with the published data for the percentage of water in the cortex will at once perceive that the value given by de Regibus (86%) seems high. The various water records for the cortex run down as low as 83.5 per cent. The differences between the various determinations are, however, almost certainly due to the varying amounts of white substance included in the sample, and as has already been stated the value chosen is probably close to the true value.

In connection with the computations there are, however, two conditions which have been assumed to be constant but which in all probability, are subject to variation. I refer to the density of the myelin and to the fraction of the lipoid to be assigned to the protein. As to this last condition, it would be plausible to think of a larger fraction of lipoid in the axons than in the cell bodies. If this were true it would be necessary to increase this fraction in the case of the spinal cord or the callosum.

It is also possible that aside from this method of distribution the fraction of lipoid in the neuron may increase with age. The slight loss of water during the first ten days of (rat) life is possibly due to such an increase. Finally, the density of the myelin may change with age, as its chemical composition certainly does, and it is conceivable that it has a higher water content when first formed, as is suggested by the 63.8 per cent given for the ten day record in table 2.

If we compare the drawings of Watson ('03), showing the increase of the visible myelin in the cerebral hemispheres and in the spinal cord of the rat, with the chemical results here used, we see that the histological pictures show a more gradual appearance of myelin than the chemical results, or the water determinations, would suggest. This probably depends on the fact that it is only a fraction of the lipoids forming the sheaths, which takes the haematoxylin stain, and this stainable fraction forms at first a smaller, but later a larger portion of the entire sheath (Koch and Koch '13; Smith and Mair '08).

There is still one more modification in the formation of myelin. Tribot ('05) has contrasted in terms of dry substance the relative amounts of albuminoides and the fats in the nerve tissue of the guinea-pig at different ages. The percentage value of the fats increases from 11 days (his first observation) up to 120 days—after which it begins to fall. The fats of Tribot are the lipoids of Koch's analysis and it is of interest to note that the 120 day record for the brain in table 2, column (4), also shows the highest proportion of corrected lipoids. The observations of Dunn ('12) which show in the myelinated fibers of the second cervical nerve of the rat the highest relative areas for the myelin sheaths at 75 days and 132 days—seem to fit with these other observations and to suggest that the formation of lipoids with advancing age fluctuates in such a way as to show a maximum about the end of the active growing period of the central nervous system.

This discussion of possible factors modifying these determinations has been introduced to clear the way for further work on the main question, but so far as one can foresee the effect of taking them into consideration, it would tend to make more uniform the values thus far obtained.

CONCLUSIONS

We conclude from these results that there is no evidence that the cell bodies and their unsheathed axons suffer more than a slight loss of water between birth and maturity, and that the progressive diminution in the water content of the entire brain and spinal cord is mainly due to the accumulation of myelin,

with a water content of about 48 per cent. Moreover, the myelin must be regarded as a more or less extraneous substance, having but little significance for the characteristic activities of the neurons.

If we compare the loss of water in the case of the nervous system with that in the muscular system, which also contains a large proportion of fat (Tribot '05), we find that while the two systems lose about the same percentage of water between birth and maturity (Lowrey '13), yet in the case of the nervous system alone is this lipoid (or non-protein substance) accumulated outside of the cell. From this it is seen that the neuron is peculiarly able to maintain its early water-solids composition and that it accomplishes this by throwing out the material, which in the muscles is retained within the cells.

As the diminution in the percentage of water in the central nervous system is preëminently a function of age, and as it appears to be due almost entirely to the formation of the myelin, it follows that the myelin formation is also a function of age (Donaldson '11). A glance at the graphs in Chart 1 shows that the most active production of myelin, as indicated by the rapid loss in the percentage of water, occurs early, i.e., during the first forty days of rat life, in the brain, and during the first hundred days, in the spinal cord.

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