## THE CHANGES DURING DESICCATION AND REHYDRA-TION IN THE BODY AND ORGANS OF THE LEOPARD FROG (RANA PIPIENS)

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On account of the fundamental importance of water in the living organism, it is of interest to know how much water the body can lose from its storage depots and yet survive. The further question arises as to whether there is, after recovery from desiccation, a normal redistribution of the water in the body as shown by the water content of the various organs. Dehydration is difficult to produce in the mammalian organism, but is easily accomplished in the frog. This animal was therefore used in the present experiments.

## MATERIAL AND METHODS

Forty common leopard frogs (*Rana pipiens*) were used in the experiments. They were all males of similar weight and obtained from the same location in southern Minnesota. They were caught in October, and the experiments ran from October through February. The frogs were kept in large concrete tanks through which ran fresh, cold tap water at a temperature ranging from 8° to 11° C. The water was kept at a depth of six inches. Bricks were put into the tanks in such a manner that when the frogs rested on them, only their heads would be exposed. The frogs had reached a constant body weight before being used.

The forty animals were divided into four groups of ten frogs each. These were designated as control group A (controls for normal fresh organ weights); control group B (controls for normal total water content); test group C (frogs desiccated for organ weights), and test group D (rehydrated for organ weights). The frogs in control group A were anesthetized with ether and autopsied at once. Their fresh and dry organ weights were determined. The control group B were killed by the ether and their fresh and dry weights were determined for the body as a whole. The test group C were desiccated so as to lose forty per cent or more of their total body weight, then anesthetized and autopsied to obtain the fresh and dry organ weights. The test group D were desiccated to the same extent as group C, and then

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allowed to recover for ten days in water at room temperature of  $25^{\circ}$  C. (range  $20-28^{\circ}$  C.). They were then anesthetized and autopsied to secure the fresh and dry organ weights. The body weights of test groups C and D were taken at frequent intervals (see Fig. 1) to determine the rate of dehydration, and, in the case of group D, also the rate of rehydration.

The frogs were desiccated by exposing them to evaporation, each in a cylindrical glass jar 8 inches in height and width. These jars were covered by a metal screen with three meshes to the inch. They were placed in a room, the temperature of which was kept fairly constant by means of a thermostat. The average temperature was 25° C. (range 23–26° C.). The average relative humidity was 25 (range 20– 29), measured with a Tycos hygrometer.

Before weighing in all groups, the excess of moisture was removed from the frogs by carefully blotting with paper towels. No allowance was made for the contents of the urinary bladder, as the frogs usually voided urine during the manipulation incident to drying them.

In all cases, ether was used to anesthetize the frogs. With the exception of control group B, the (anesthetized) animals were killed by cutting open the tip of the cardiac ventricle and suspending the animals by the head and feet so that all the blood possible might drain out into a bottle. The weight and the water content of the escaped blood were determined.

The procedure at autopsy was as follows: The head was severed from the body at the atlanto-occipital articulation. The limbs were separated at the hip and shoulder joints, respectively. All parts were placed in a moist chamber until dissected. The dissection was made upon dampened blotting paper. The lungs were removed at the junction with the trachea, and were opened to remove any parasitic flukes found there. The heart was removed by cutting the large veins at their termination, and severing the aorta at its origin from the bulbus aortae. The chambers of the heart were opened and their contents (if any) removed. The contents of the stomach and intestines were removed before weighing these organs. The ano-cloacal junction was taken as the lower border of the intestines. The suprarenal glands were not removed from the kidneys. The liver was removed and weighed together with the gall bladder. The bile in all cases was allowed to drain out before the weighing. The tongue was separated by cutting the muscles at their attachment to the hyoid bone. When the integument was taken, the subjacent lymph was merely allowed to drain off. The term "remainder" includes the various items such as large blood vessels, nerves, connective tissue and other structures not included under the other headings. The remainder does *not* include the water in the subcutaneous lymph spaces.

All organs were weighed on a Wilkens-Anderson analytical balance in previously weighed weighing bottles with ground glass stoppers. The organs, and likewise the entire frogs of control group B, were dried in a Thelco electric oven at about 95° C. until constant weights were obtained.

## Results

## General Observations

The frogs of all groups were healthy and normal in all respects. The average and range of initial body weights in grams were as follows: control group A, 43.59 (35.0 - 53.2); control group B, 40.64 (34.5 - 41.6); test group C, 43.57 (37.0 - 53.1); test group D, 44.80 (42.7 - 49.0). It will be noted that in average body weight groups A and C were nearly identical, and group D differed from these by about only two per cent. In the calculations, it is assumed that the initial organ weights were equal in these three groups.

It required 28.6 hours, on the average, to desiccate the test group C to an average weight of 24.98 grams, with loss of 42.7 per cent of their initial body weight, or 51.9 per cent of their total water content (Fig. 1).<sup>1</sup> These frogs at first were quite active, but after a few hours of desiccation they remained motionless, apparently almost lifeless. The skin became very dry, and the animals appeared emaciated. The frogs maintained the sitting position, and the joints of the limbs became stiff. In addition to the ten surviving frogs in this group, four others died during the test and were excluded from the experiment.

A period of 31.8 hours was required to desiccate the test group D to an average weight of 25.40 grams, with loss of 43.3 per cent of their average initial body weight or 52.7 per cent of their total water content.<sup>1</sup> Up to this stage, the general appearances and reactions were the same as in the test group C. Upon being put back into water at room temperature ( $25^{\circ}$  C.) for rehydration, the test group D at first remained inactive. After two or three hours, they gradually began to move about, and soon became normal to all outward appearances. After 26.2 hours these frogs had returned to a nearly constant body weight, averaging 36.74 grams (Fig. 1). This weight they maintained, with slight fluctuations, for ten days before being autopsied. This nearly constant body weight averaged about 8.1 grams below the original body weight. Figure 1 shows the general weight curve fol-

<sup>&</sup>lt;sup>1</sup> These percentages for the estimated losses of water content are slightly too high, since they include (as shown later) a loss of about 0.51 gram in solids during the experiment. The corrected loss of total water content is 50.5 per cent for group C and 51.3 per cent for group D.

lowed by this group during desiccation and rehydration. In addition to the ten surviving frogs in this group, five others died during the test and were excluded from the experiment. These five frogs died during the dehydration part of the experiment. No deaths occurred during rehydration.

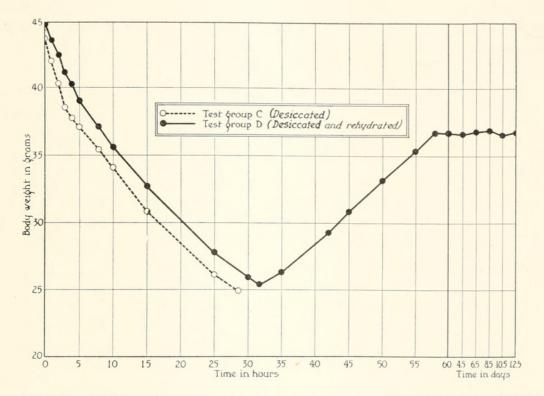


FIG. 1. Curves of average weight during desiccation and rehydration. Test group C was desiccated 28.6 hours, decreasing in average body weight from 43.6 grams to 25.0 grams. This corresponds to a loss of about 43 per cent in body weight. Test group D was similarly desiccated 31.8 hours, decreasing from 44.8 grams to 25.4 grams, with loss of about 43 per cent in body weight. This group was then replaced in water and increased to 36.7 grams in 26.2 hours (end of 58th hour of experiment). Thereafter the weight remained nearly stationary during a period of 10 days. Failure to reach the original weight is ascribed to the increase in room temperature.

#### Observations on Individual Organs and Parts

Observations at Autopsy.—At autopsy the control group A appeared normal in every respect with the exception of an occasional lung fluke. The test groups C and D also occasionally contained one to three flukes. In the desiccated test group C the blood was so thick that it would barely run into the container. There seemed to be practically no water in the tissue spaces and lymph sacs. The intestinal contents were very slight. Most of the organs appeared very dry.

The rehydrated test group D at autopsy appeared quite normal with the exception that the epidermis was shedding in large pieces quite generally over the body. This process resembled the normal ecdysis, although in this case it was caused by the desiccation. The amount of water in the tissue spaces and lymph sacs appeared somewhat below that in the normal control group A. The blood seemed normal in color and consistency.

Fresh Weights of Organs.—(Tables I and II.)—The average fresh organ weights of control group A were taken as norms, and the average fresh organ weights of test groups C and D were compared with these.

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	Control group A (normal)	Test group C (desiccated)	Test group D (rehydrated)
Eyeballs	0.3980	0.3958	0.3987
Lungs	0.2146	0.1992	0.2134
Skeleton	5.2250	5.0073	5.1246
Brain	0.1013	0.0934	0.1093
Kidneys	0.1381	0.1151	0.1442
Heart	0.1112	0.0922	0.1057
Spinal cord	0.0636	0.0498	0.0591
Γestes	0.0424	0.0314	0.0456
Stomach-intestines	0.6182	0.4326	0.5812
Muscles	16.5533	11.4997	16.0331
Skin	5.7809	3.3976	4.7196
Fongue	0.3993	0.1892	0.3800
Spleen	0.0381	0.0170	0.0296
Liver	1.6586	0.7019	1.0137
Blood	1.4274	0.2739	1.3468
Fat bodies	0.0950	0.0251	0.0463
Remainder	0.7194	0.4448	0.7808
Fotal	33.5844	22.9660	31.1317

Average Fresh Organ Weights in Grams

As shown by Tables I and II, in the test group C, the eyeballs and lungs remained nearly unchanged in weight, and the skeleton and brain lost less than ten per cent through desiccation. The kidneys, heart, spinal cord, testes, stomach-intestines and muscles form a group losing moderately (16–30 per cent), but relatively less than the body as a whole. Finally the skin, tongue, spleen, liver and (escaped) blood lost 43–81 per cent, which exceeds the relative loss of the body as a whole.

In the rehydrated test group D, all the fresh organs, with the exception of the skin, spleen and liver, had regained practically normal weight (within 8 per cent). The skin remained about eighteen per cent below the norm, the spleen 22 per cent below, and the liver nearly 39 per cent below. The significance of these changes will be considered later.

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## TABLE II

Average Percentage Difference in Fresh Organ Weights of Test Groups C and D, Compared with Control Group A

Test group C	Test group D
(desiccated)	(rehydrated)
1. Organs with slight loss during desiccation	

 Eyeballs
 -0.6  $\pm 0.0$  

 Lungs
 -0.6 -7.2 

 Skeleton
 -4.2 -1.9 

2. Organs with moderate loss during desiccation

Brain..... -7.8

Kidneys	-16.7	+4.4
Heart		-5.0
Spinal cord	-21.7	-7.1
Testes		+7.6
Stomach-intestines	-30.0	-6.0
Muscles	-30.5	-3.1

3. Organs with great loss during desiccation

Skin	-42.8	-18.4
Tongue	-52.6	-4.8
Spleen	-55.4	-22.3
Liver	-57.7	-38.9
Blood	-80.8	-5.6

Water Content.—(Tables III and IV.)—The average percentage water content found in the whole body of control group B was 82.14 per cent, ranging from 80.21 to 84.28 per cent. This average is taken as the norm for the body as a whole.

The average percentages of water found in the organs of control group A, and of test groups C and D, are given in Table III. For the normal control group A, the organs are arranged in order of increasing relative water content, from the skeleton (58.13) to the escaped blood (88.97 per cent). The muscles in this group contain almost exactly the same percentage of water as the average for the whole body. The skeleton, liver, spinal cord, skin and spleen contain relatively less, and the testes, "remainder," brain, heart, kidneys, stomach-intestines, tongue, lungs, eyeballs and blood contain relatively more water than the body as a whole.

In the desiccated test group C, the organs listed (Table III) have all lost part of their water and hence appear lower in relative water content than the corresponding organs of control group A. Assuming that all the loss of weight in this desiccated group C (Table I) is due to loss of water, we may estimate the theoretical water content of the organs in group C. This theoretical water content should agree

+7.9

fairly with that actually found in the desiccated test group C (Table III). The calculations of the theoretical water content were made in this way for every organ, but the figures in detail are omitted for economy of space. The muscles on this basis show the closest agreement with a discrepancy of only 0.2 per cent.<sup>2</sup> Agreement (within 3.5

## TABLE III

## Average Percentage of Water in Organs

	Control group A (normal)	Test group C (desiccated)	Test group D (rehydrated)
Skeleton	58.13	57.94	60.52
Liver	76.08	72.10	76.41
Spinal cord	80.75	76.73	81.67
Skin	81.05	70.84	80.15
Spleen	82.13	77.84	81.39
Muscles	82.15	74.52	82.17
Testes	82.57	75.09	81.40
Remainder	83.12	77.19	81.85
Brain	83.90	81.28	85.09
Heart	84.07	82.24	87.17
Kidneys	84.42	78.09	84.06
Stomach-intestines	85.00	75.21	83.36
Tongue	86.13	76.83	87.87
Lungs	87.11	82.52	86.15
Eyeballs	87.83	85.38	88.16
Blood	88.97	77.33	88.79

per cent) is also found in the case of all the other organs except the liver, spleen, tongue, lungs and skin. The percentage of water in the liver of the desiccated group averages 28.7 per cent greater than that to be expected from its loss of weight. The corresponding percentages in the spleen, tongue, lungs and skin are, respectively, 17.9, 6.8, 4.5 and 4.0 per cent greater than those theoretically calculated from the change in organ weight. The significance of these discrepancies will be discussed later.

The absolute amount of water given up by each of the various organs or parts in test group C is shown in Table IV. The amounts

<sup>2</sup> The method of calculation is as follows: As shown in Table III, every 100 grams of fresh muscle in the normal control group A contains 82.15 grams of water and (accordingly) 17.85 grams of solids. Table II shows that in the desiccated test group C each 100 grams of muscle had lost 30.5 grams in weight, which is assumed to be due to loss of water. Subtracting 30.5 from 82.15 gives 51.65 grams of water remaining in the 69.50 grams of muscle in the desiccated frog. This gives a theoretical water content of 74.32 per cent. The actual water content, as found in the oven-dried muscle of the desiccated test group C (Table III), was 74.52 per cent, which differs from the theoretical value by only 0.2 per cent.

are estimated from the differences in relative water content between groups A and C (Table III), assuming that the initial organ weights were equal to those in control group A. The estimate for the lymph spaces is explained later. The chief storage depots of water in the frog's body are apparently the lymph spaces, yielding 8.51 grams (or

## TABLE IV

Organs and Parts of Test Group C, arranged according to the Absolute Amount of Water in Grams Lost During Desiccation. Estimated on basis of change in percentage of water content shown in Table III and normal fresh organ weights in Table I.

Lymph spaces 8.5100	Fat bodies 0.0294
Muscles 5.0289	Lungs
Skin 2.2794	Kidneys 0.0210
Blood 1.0582	Spleen 0.0181
Liver 0.7559	Heart 0.0177
Remainder 0.2547	Eyeballs 0.0134
Stomach-intestines 0.2001	Spinal cord 0.0132
Tongue 0.1985	Testes
Skeleton 0.1363	Brain 0.0091
	Total

45.8 per cent of the total 18.5776 grams of water lost by the body during desiccation); and the muscles, yielding 5.03 grams (or 27.1 per cent). Somewhat smaller contributions are made by the skin (2.28 grams or 12.3 per cent), and the blood (1.06 grams or 5.7 per cent). All the other organs together give up only 1.70 grams, or 9.2 per cent.

In the frogs of the rehydrated test group D, the organs in general have regained nearly normal water content (Table III). All the organs average within 2 per cent of the normal water content found in control group A, excepting the skeleton, which is about 2.4 per cent above normal, and the heart, which averages 3.1 per cent above.

To determine the amount of water in the lymph spaces of control group A, the total average fresh organ weight (33.58 grams, Table I) was subtracted from the average original body weight (43.59 grams). This gives a difference of 10.01 grams, due chiefly to the contents of the lymph spaces. During desiccation, the test group C frogs lost 18.59 grams in average body weight. The organs of this group (excluding the lymph spaces) apparently gave up 10.07 grams of water, as shown by the estimates in Table IV. The remainder of the loss (8.52 grams) was assumed to come from the lymph spaces, although a small part of this loss is in solids, as will be shown later. It is assumed that the lymph spaces of test groups C and D originally contained as much water as the lymph spaces of control group A (10.01 grams). There-

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fore, at the time of autopsy the lymph spaces of test group C should theoretically contain 1.49 grams (10.01 minus 8.52 grams). If we subtract the average fresh organ weight of this group (22.97 grams, Table I) from their average final body weight (24.98 grams), we find that the actual amount of water in the lymph spaces was 2.01 grams. The apparent loss of solids in the various organs can be calculated by comparing the theoretical losses of water (Table IV) with the decreases in fresh weight (Table I). All the organs excepting the stomachintestines, lungs, eyeballs and testes show variable (usually small) losses in weight beyond those due to apparent decrease in water content. The net decrease amounts to about 0.58 gram, or apparently slightly more than sufficient to account for the difference between the theoretical and the actual amount of water in the lymph spaces (0.52 gram).

The total theoretical amount of water in the body, as a whole, in the rehydrated test group D at time of autopsy was 30.18 grams. This is obtained from the average final body weight (36.74 grams) multiplied by the normal percentage of water in the whole body (82.14 per cent). If we subtract the total water actually found in the organs (24.46 grams) from the total (theoretical) water content of the body (30.18 grams), we get the theoretical amount of water in the lymph spaces (5.72 grams) of the rehydrated test group D. We can check this by subtracting the average fresh organ weight of test group D (31.13 grams, Table I) from the average final body weight for this group (36.74 grams). This gives 5.61 grams as the amount of water found in the lymph spaces.

In general, therefore, it appears that the lymph spaces of the desiccated test group C had given up approximately 80 per cent of their initial water content, while the lymph spaces of the rehydrated test group D had not fully refilled, but were still about 44 per cent below their estimated original water content. The explanation for this difference will be discussed later.

#### DISCUSSION

In general, it must be remembered that the number of frogs (10) in each of the four groups is relatively small. The results may therefore be influenced somewhat by individual variations, so that in some cases the conclusions are to be regarded as tentative rather than final.

#### GENERAL OBSERVATIONS

The average loss of weight (about 43 per cent) sustained by desiccation in our test groups C and D may be compared with the results of other investigators. Chossat (1843) noted a loss of 35 per cent in the body weight of frogs subjected to evaporation. Similar observations were made by Durig (1901). Hall (1922) subjected various species of animals to exsiccation, and found that frogs survived a desiccation period of 33 hours, with loss of 41 per cent in body weight, corresponding to 50 per cent of the entire water content. De Almeida (1926) reported that in frogs the loss in body weight before death from dehydration varies with the rapidity of evaporation. With rapid desiccation, death may occur with a loss of only 10 per cent, while with slower experiments the loss may be 30 to 40 per cent. Further data on dehydration in the frog are given by Iizuka (1926) and Hug (1927). Data for comparison as to storage and loss of water in other species and in other forms of inanition are cited by Jackson (1925, 1929). These include the effects of exsiccation on various invertebrates and vertebrates.

The curves of body weight during dehydration in our test groups C and D (Fig. 1) show a more rapid loss at the beginning, with a progressively slower rate of decrease later. The rate of rehydration followed by test group D appears slower at first, but is more nearly uniform from the third to the 26th hour (58th hour of the experiment), when the weight ceased to increase. This group, therefore, did not return to their original body weights, but maintained a nearly constant body weight averaging 36.74 grams, or 8.06 grams below their original weight. This difference is apparently due chiefly to the partially unrestored loss of water from the lymph spaces. It is probably caused by the change in room temperature, which was 8° to 11° up to the beginning of the experiment, but 25° C. during the test. Ott (1924) found that frogs on transfer to cooler environment may absorb water (chiefly in the lymph spaces), which is lost within a few days when they are removed to warmer quarters.

## INDIVIDUAL ORGANS AND PARTS

Fresh Weights.—The losses in fresh weights of the organs in the desiccated group C agree in general with the findings of Durig (1901). He reported that the musculature lost most heavily, but he did not measure the loss from the lymph spaces, which we find to be even greater. According to Durig, the brain and heart lost relatively less than any other organ; but our data indicate that the eyeballs, lungs and skeleton (organs not studied by Durig) lost relatively less fresh weight than the brain or heart. Durig also found that the kidneys, in spite of a marked decrease in water content, showed a relative increase in weight, which he ascribed to retention of insoluble urinary constitu-

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ents. The kidneys in our desiccated group C lost only 16.7 per cent of their fresh weight, so that they gained in relative weight, as did all the other organs losing relatively less than the body as a whole.

Also in other species under conditions of dehydration, certain organs such as the brain, eyeballs, skeleton and heart maintain their organ weights remarkably well (in some species even increasing their weight under these conditions); while other organs such as the skin, muscles, stomach-intestines, liver and spleen lose a large proportion of their fresh weight. (See review of the literature by Jackson, 1925, 1929.) A similar tendency was found by Ott (1924) in the frog during inanition.

Ott found that the excess water absorbed when frogs were transferred to a cooler environment did not appreciably affect the weights of the various organs, excepting the lungs and fat bodies. In the present experiments, all the organs excepting the skin, spleen, liver and fat bodies (as shown in Table I) in the rehydrated group D had returned to nearly normal weight, and all excepting the heart and skeleton (Table III) had returned to within two per cent of the original normal water content. Data for the water content of the fat bodies are lacking however.

Loss of Solids.—A comparison of the figures for fresh organ weights in Tables I and II shows certain discrepancies between the desiccated test group and the rehydrated test group D. All the organs of test group D returned to within 8 per cent of the normal weight, excepting the skin, liver and spleen. As previously mentioned, these discrepancies are attributed chiefly to loss of solids during the experiment. The liver during the brief period of inanition probably lost much of its glycogen content, which could not be restored by the mere addition of water. This would account for the nearly normal relative water content of the liver in spite of its marked subnormality of weight. The skin in test group D started shedding (ecdysis) three to four days after beginning rehydration, which probably accounts for its marked subnormality in weight, although the relative water content had returned to nearly normal. Similarly the spleen may have lost solids (through escape or destruction of cells), which would account for the subnormality in weight in spite of nearly normal water content. The other organs apparently lost but little or no solids, as was noted above.

Water Content.—The average percentage of water content in the organs of our control group A (Table III) is in general agreement with Ott's figures for his normal control frogs. The differences are within 3 per cent, excepting the spleen, which is higher in his series, and the liver, skeleton and stomach-intestines, which are somewhat lower.

In his fasting frogs with loss of 40 per cent in body weight, the percentage of water content is usually increased, in contrast with our desiccated group C showing marked loss in water content.

Table IV indicates that the greatest absolute sources of water during desiccation in the frog are from the lymph spaces, muscles, skin, blood, liver, "remainder," and stomach-intestines, decreasing in the order named. In relative loss, however, the corresponding order is the lymph spaces, blood, skin, stomach-intestines and muscles.

The escaped blood of the test group C contained 77.33 per cent of water, or 11.64 per cent below the water content of the escaped blood in control group A (Table III). Yet the escaped blood of the rehydrated test group D had regained practically the same relative amount of water as the control group A. The markedly thickened blood of the dehydrated frog is therefore rapidly restored to normal. Durig found that the number of red corpuscles per cu. mm. was more than doubled in the frog by desiccation.

After recovery from desiccation, nearly all the organs of the rehydrated group D return to within two per cent of their normal water content (Table III), although insufficient water has been absorbed to restore the original body weight. Ott (1924) similarly noted that the excess water absorbed by frogs at lower temperature did not affect the weight or water content of the organs, excepting the lungs and fat bodies. The only exceptions in our rehydrated series at higher temperature are the skeleton and heart, which gain an excess water content of 2.39 and 3.10 per cent, respectively. The lymph spaces of the test group D, however, are estimated (as previously mentioned) to have remained 44 per cent subnormal in water content. It is evident that the changes in water content of the frog's body with fluctuations of temperature affect the amounts in the interstitial spaces rather than the water content of the tissues and organs.

It is not the purpose of the present paper to compare the findings of other investigators in various species, but it may be noted that, in general, the main storage depots of water in the frog, aside from the subcutaneous lymph spaces, appear to correspond in general to those found in mammals. The amphibian is more tolerant of dehydration, however, and can survive relatively greater losses. Moreover, the water in the amphibian organism is stored in a more labile form, so that it can be more readily removed and restored. Data for comparison are cited by Jackson (1929).

#### SUMMARY

The principal results of the present investigation may be summarized briefly as follows: 1. Male frogs (*Rana pipiens*) weighing about 44 grams were able to recover after desiccation with average loss of about 43 per cent of their body weight or about 51 per cent of their total water content.

2. The eyeballs and lungs of the desiccated frogs remained nearly unchanged in average fresh organ weight, and the skeleton and brain lost less than 8 per cent. The kidneys, heart, spinal cord, testes, stomach-intestines and muscles formed a group losing moderately (16-30 per cent), but relatively less than the body as a whole. The skin, tongue, spleen, liver and (escaped) blood lost 43-81 per cent, or relatively more than the body as a whole.

3. The average percentage of normal water content in the entire body was 82.14 per cent (range 80.21-84.28) at temperature of 8 to  $11^{\circ}$  C.

4. In the normal organs the average percentage of water content ranged from 58.13 in the skeleton to 88.97 in the escaped blood.

5. The decreases in percentage of water content in the desiccated frogs averaged approximately as follows: skeleton 0.2, heart 1.8, eyeballs 2.5, brain 2.6, liver 4.0, spinal cord 4.0, spleen 4.3, lungs 4.6, "remainder" 5.9, kidneys 6.3, testes 7.5, muscles 7.6, tongue 9.3, stomach-intestines 9.8, skin 10.2, and escaped blood 11.6.

6. The average absolute amount (grams) of water given up by the various organs or parts of the desiccated frogs was approximately as follows: lymph spaces 8.5, muscles 5.0, skin 2.3, blood 1.1, liver 0.8, "remainder" 0.3, stomach-intestines 0.2, tongue 0.2, skeleton 0.1. The remaining organs each lost less than 30 mgm. of water.

7. In the rehydrated frogs, all the fresh organs with the exception of the skin, spleen and liver regained nearly normal weight (within 8 per cent). In all the rehydrated organs, except the skeleton and heart, the water content returned to within 2 per cent of the norm. However, the amount of water in the interstitial lymph spaces remained 44 per cent subnormal, probably due to the increased temperature of the room.

8. The following peculiarities were noted in the rehydrated organs: The fresh weight of the skin remained 18 per cent subnormal, apparently due to ecdysis. The fresh weight of the spleen was 22 per cent subnormal, probably through escape or destruction of cells. The liver weight remained 39 per cent subnormal, probably through loss of glycogen. The relative water content in each of these three organs in the rehydrated group was found to be within 0.9 per cent of the normal.

9. In general, the main storage depots of the frog, aside from the lymph spaces, appear to correspond in general to those found in mammals. The amphibian is more tolerant of dehydration, however, and can survive relatively greater losses. Moreover, the water in the amphibian organism is stored in more labile form, so that it can be more readily removed and restored.

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