FURTHER OBSERVATIONS ON THE NERVOUS SYSTEM OF THE AMERICAN LEOPARD FROG (*RANA PIPIENS*) COMPARED WITH THAT OF THE EUROPEAN FROGS (*RANA ESCULENTA* AND *RANA TEMPORARIA*)

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WITH TWO FIGURES

In a paper under the general title given above (Donaldson '08) I discussed some observations made in 1904 on *R. esculenta* at Zurich and *R. temporaria* at Liverpool.

On comparison with the American frog, *R. pipiens*, it was found that although the European species were very similar to the latter in form and proportions, nevertheless the weight of the central nervous system was significantly smaller in the European species, and in the case of *R. esculenta*, the number of medullated fibers in the spinal nerves was much less than in *R. pipiens*.

These observations made it possible to correct the records of Fubini ('81) on the weight of the brain and spinal cord, which had alone been available for the European forms, and to call attention to the possible bearing of the anatomical differences on physiological results obtained from the two European species on the one hand, and the American species on the other.

In view of the fact that on the basis of rather few observations I had ventured to designate Fubini's records as untrustworthy, and also to suggest possible physiological differences in the responses of the central nervous system, it seemed desirable to repeat the observations on the European forms.

This I did during the past summer. For a second time I am indebted to Professor Gaule for the hospitality of the Physiological Institute at Zurich, where I had examined *R. esculenta* in 1904, and to Professor Sherrington for similar privileges at the

Physiological Laboratory of University College at Liverpool, where I had examined *R. temporaria* in the same year.

To both these gentlemen I desire to express my obligations for their courtesy and aid.

The results of these latest observations support completely the conclusions based on the records of 1904.

In the present communication therefore it is not necessary to repeat the entire argument of the earlier paper, but merely to present the evidence for the similarity of the earlier and later records.

For this purpose it will be desirable to print in full only the original measurements for both years, while the important ratios can be given in condensed tables accompanied by a few charts.

The following are the tables of the principal measurements as made on the three species in 1904 and 1909

TABLE 1
Data on R. pipiens, Chicago 1904. 12 specimens

BODY WEIGHT IN GRMS.	SEX	TOTAL LENGTH IN MM.	BODY LENGTH IN MM.	WEIGHT IN GRAMS OF			PERCENTAGE OF WATER	
				C. N. S.	Brain	Sp. C.	Brain	Sp. C.
11.6	M.	130		.0918	.0666	.0252	84.4	79.4
16.0	M.	150		.1148	.0796	.0352	85.2	80.7
17.0	F.	159		.1054	.0714	.0340	84.0	80.6
20.8	M.	170		.1232	.0844	.0388	85.2	81.6
22.5	M.	162		.1165	.0807	.0358	84.5	80.4
26.4	M.	180		.1372	.0946	.0426	84.4	78.4
27.6	F.	179		.1416	.1014	.0402	84.8	80.1
30.6	M.	180		.1454	.0998	.0456	84.6	79.8
34.2	M.	190		.1518	.1056	.0462	85.6	81.6
41.8	M.	197		.1652	.1146	.0506	86.9	82.2
43.9	M.	200		.1708	.1210	.0498	85.8	80.7
47.0	M.	198		.1664	.1140	.0524	84.4	80.5

TABLE 2

Data on R. esculenta, Zurich 1904. 11 specimens

BODY WEIGHT IN GRMS.	SEX	TOTAL LENGTH IN MM.	BODY LENGTH IN MM.	WEIGHT IN GRAMS OF			PERCENTAGE OF WATER.	
				C. N. S.	Brain	Sp. C.	Brain	Sp. C.
12.40	F.	131		.0818	.0577	.0241	84.2	78.4
16.75	F.	144		.0926	.0634	.0292	83.4	79.1
18.43	F.	144		.0928	.0650	.0278	83.2	78.2
20.00	F.	161		.1103	.0756	.0347	82.5	79.2
22.00	.	164		.1107	.0769	.0338	84.0	79.0
24.10	M.	167		.1217	.0841	.0376	83.4	78.4
33.85	M.	175		.1327	.0895	.0432	83.2	78.2
36.30	M.	177		.1478	.1004	.0474	83.4	78.6
37.56	F.	188		.1490	.0993	.0497	82.9	78.8
37.96	F.	194		.1427	.0953	.0474	82.8	77.8
45.03	F.	196		.1578	.1078	.0500	83.9	78.4

TABLE 3

Data on R. esculenta, Zurich 1909. 11 specimens

BODY WEIGHT IN GRMS.	SEX	TOTAL LENGTH IN MM.	BODY LENGTH IN MM.	WEIGHT IN GRAMS OF			PERCENTAGE OF WATER	
				C. N. S.	Brain	Sp. C.	Brain	Sp. C.
18.9	M.	143	57.3	.1047	.0707	.0340	83.6	78.6
24.7	F.	167	65.0	.1065	.0719	.0346	83.6	79.3
26.5	M.	167	63.4	.1120	.0737	.0383	83.6	77.8
30.9	M.	177	69.2	.1198	.0830	.0368	83.9	78.5
32.3	F.	183	68.0	.1301	.0873	.0428	83.6	78.1
33.0	F.	184	70.5	.1275	.0845	.0430	83.4	77.0
35.5	F.	188	72.5	.1435	.0985	.0450	83.8	78.2
47.4	F.	204	80.0	.1593	.1063	.0530	83.1	79.4
48.4	F.	193	79.3	.1545	.1027	.0518	83.8	77.7
52.4	F.	205	82.0	.1589	.1105	.0484	83.6	79.1
58.0	F.	216	87.0	.1858	.1278	.0580	83.6	77.6

TABLE 4

Data on R. temporaria, Liverpool 1904. 12 specimens

BODY WEIGHT IN GRMS.	SEX	TOTAL LENGTH IN MM.	BODY LENGTH IN MM.	WEIGHT IN GRAMS OF			PERCENTAGE OF WATER	
				C. N. S.	Brain	Sp. C.	Brain	Sp. C.
14.05	F.	144		.0881	.0596	.0285	82.3	78.2
16.10	F.	151		.0991	.0690	.0301	82.7	79.0
17.65	M.	154		.0916	.0618	.0298	83.0	78.5
21.75	M.	171		.1045	.0671	.0374	82.8	78.2
23.45	M.	162		.0947	.0628	.0319	82.1	77.0
24.17	F.	173		.1333	.0864	.0469	81.9	76.5
27.05	M.	173		.1298	.0874	.0424	82.4	77.1
28.15	M.	168		.1018	.0687	.0331	82.5	76.7
28.95	M.	174		.1324	.0813	.0511	81.3	76.8
28.95	M.	178		.1485	.0928	.0557	81.3	76.8
32.15	M.	173		.1321	.0890	.0431	80.9	78.6
32.81	F.	178		.1161	.0766	.0396	82.7	78.0

TABLE 5

Data on R. temporaria, Liverpool 1909. 16 specimens

BODY WEIGHT IN GRMS.	SEX	TOTAL LENGTH IN MM.	BODY LENGTH IN MM.	WEIGHT IN GRAMS OF			PERCENTAGE OF WATER	
				C. N. S.	Brain	Sp. C.	Brain	Sp. C.
14.5	F.	148	53.3	.0772	.0522	.0250	82.2	76.8
17.2	M.	154	56.0	.0903	.0598	.0305	83.6	79.6
19.1	F.	155	58.5	.0907	.0621	.0286	82.4	76.3
21.0	M.	162	60.3	.1014	.0663	.0351	83.8	79.5
24.2	M.	176	64.5	.1099	.0736	.0363	83.9	78.5
25.4	M.	164	60.0	.0994	.0672	.0322	84.5	78.2
26.0	M.	162	60.8	.1066	.0702	.0364	84.0	79.4
26.1	F.	174	65.8	.1191	.0787	.0404	84.0	79.4
26.9	F.	163	63.2	.1092	.0737	.0355	84.1	76.3
27.9	F.	175	66.7	.1149	.0786	.0363	83.8	79.5
29.2	F.	174	66.5	.1114	.0744	.0370	83.8	79.5
29.4	M.	170	66.0	.1356	.0887	.0469	83.8	78.9
29.8	M.	168	62.5	.1167	.0751	.0416	83.8	78.9
32.1	F.	184	73.2	.1314	.0864	.0450	84.3	79.2
33.3	M.	167	64.0	.1373	.0900	.0473	84.3	79.2
39.1	F.	196	76.8	.1452	.0964	.0488	82.3	77.8

The foregoing tables (1-5), representing five series, contain the fundamental data.

The plan was to have twelve specimens in each series. In the case of *R. esculenta* 1904 and also 1909, there are, however, only eleven in each. The absent records were excluded because the percentage of water, which was not calculated until my return home, showed the excluded specimens to be in abnormal condition.

In the case of *R. temporaria* 1909 sixteen records were made. In general, the grouping of these data is by threes. There are however three exceptions: In *R. esculenta* 1904, with a total of 11 specimens, there is one group of two (Records 7 and 8) and in *R. esculenta* 1909, there is one group of two (Records 10 and 11).

In *R. temporaria* 1909 there is one group of four. In each case this departure from the rule is indicated in the condensed tables (6, 10, 12, 13,) by a bracketed number following the average for body weight.

It will be noted that in the 1904 series, the column under the heading "Body length" is vacant. This measurement was not made in that year, but was made in the specimens collected in 1909.

It represents the length of the frog from the tip of the nose to the tip of the urostyle, the skin over the urostyle having been split in order to expose its cartilaginous tip; the measurement being taken with vernier calipers.

In the previous paper (Donaldson '08) some measurements on preserved material were introduced without correction for the effects of the reagents used. These cases were explicitly noted. It is of interest to state therefore that, in this paper, the data apply to the fresh material only. Indeed all the measurements were made on the material when fresh except in the case of the leg bones of the two 1909 series. In these cases the legs were brought to this country from Europe in 60 per cent alcohol and then the bones were measured.

A long series of control observations on the legs of *R. pipiens* treated in the same way and for the same time have shown that

this treatment reduces the length of the femur by 0.70 per cent making it 99.30 per cent of the fresh length, the tibia by 0.73 per cent making it 99.27 per cent of the fresh length; the foot (tarsus-pes) by 1.54 per cent making it 98.46 per cent of the fresh length.

These corrections were applied before the data were used in tables 7 and 8.

TABLE 6
Body weight per millimeter of total length. Averages from groups of three

	BODY WEIGHT IN GRAMS	BODY WEIGHT PER MILLIMETER, IN GRAMS
R. pipiens.....	{ 14.9 23.2 30.8 43.2	.102 .135 .168 .218
R. esculenta 1904.....	{ 15.9 22.0 35.0 [2]* 40.2	.114 .134 .199 .208
R. esculenta 1909.....	{ 23.4 32.1 43.8 55.2 [2]	.146 .177 .225 .262
R. temporaria 1904.....	{ 15.9 23.1 28.0 31.3	.107 .137 .162 .177
R. temporaria 1909.....	{ 18.0 [4] 25.2 26.9 29.5 34.8	.116 .151 .159 .173 .191

(A) AVERAGE AMOUNT OF BODY WEIGHT FOR EACH MILLIMETER
OF TOTAL LENGTH

The general form of the specimens examined is obtained by dividing the body weight by the total length (table 6). The data in this table are given in Chart 1 and show that in the years

1904 and 1909, the European frogs were similar in their general form.

The records for *R. pipiens* are not entered on this chart. They would run a trifle below those for the European species, showing that *R. pipiens* was more slender in its general build. This character of *R. pipiens* taken alone would imply a slightly smaller nervous system, but as we know the contrary is the case.

TABLE 7

Percentage of the total length represented by the combined lengths of the leg bones

	SPECIMENS	PERCENT
<i>R. pipiens</i>	9	66.6
<i>R. esculenta</i>	11	65.1
<i>R. temporaria</i>	16	66.2

(B) PERCENTAGE OF TOTAL LENGTH REPRESENTED BY THE
COMBINED LENGTHS OF THE LEG BONES

The absolute values of the percentages in this table are on the average less by 3.5 than those given in the previous paper (see Donaldson '08, table 2). This is the result of a change in the technique of measurement. Previously the total length of the frogs was taken when the animals were suspended, and under this condition a certain amount of flexion persisted in the legs.

In the present case the frog was measured when stretched out on the table and lying on its ventral surface. By this treatment the amount of flexion was reduced, and the total length thereby slightly increased. This naturally reduced the percentage value of the sum of the lengths of the leg bones, the measurements of which were made in the same way in both cases. The above mentioned change in technique is the only one which has been made.

The point of importance is that the percentages are nearly the same for the three species which are here compared.

(C) THE PROPORTIONAL LENGTHS OF THE SEVERAL LEG BONES

These are shown in table 8 in which the 1904 records have been repeated and a complete series of 1909 records added. It will be seen that there is no essential difference between the obser-

uations made at the contrasted dates, and that in both instances the proportional lengths are nearly the same for the three species compared.

TABLE 8

The proportional lengths of the several leg bones

	NO. OF SPECIMENS	FEMUR	TIBIA	FOOT (TARSUS AND PES)
		<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
R. pipiens 1904.....	12	25.5	29.3	45.2
R. pipiens 1909.....	19	26.0	29.3	44.7
R. esculenta 1904*.....	5	26.3	28.2	45.5
R. esculenta 1909.....	12	26.8	28.2	45.0
R. temporaria 1904*.....	6	26.1	28.7	45.2
R. temporaria 1909.....	16	25.7	28.3	46.0

* Leg bones from frogs of the so-called "Zurich series of 1898." These frogs had been carefully fixed in 4% formaldehyde and then preserved in 80% alcohol. The effect of this on the lengths of the several leg bones was not at the time determined. (See Donaldson '08, p. 127).

(D) PERCENTAGE VALUE OF THE LENGTH OF THE ENTIRE CENTRAL NERVOUS SYSTEM—THE TOTAL LENGTH OF THE FROG BEING TAKEN AS THE STANDARD.

In the case of this character we have grouped the 1904 data (see Donaldson '08, table 5) into three entries and added the measurements on the new material for the 1909 groups.

The table shows that the length of the entire central nervous system is slightly greater in the European species. As this excess in length is associated with a deficiency in absolute weight, it follows, as was previously noted (Donaldson '08, p. 128) that the nervous system in *R. pipiens* must exceed that of the European species in its transverse diameters.

TABLE 9

Percentage value of the length of the entire central nervous system—the total length of the frog being taken as the standard

NO. OF SPECIMENS	AVERAGE TOTAL LENGTH IN MM.	PERCENTAGE VALUE OF THE LENGTH OF THE ENTIRE CENTRAL NERVOUS SYSTEM		
		<i>Rana pipiens</i>	<i>Rana esculenta</i>	<i>Rana temporaria</i>
9 (1904).....	152	17.5		
4.....	155			17.6
3.....	159		18.4	
3.....	167			17.6
6.....	171			17.3
9 (1904).....	176	16.7		
3.....	181		16.9	
3.....	182			17.2
3.....	195		16.6	
4 (1904).....	196	16.3		
2.....	210		16.2	

(E) THE WEIGHT OF THE CENTRAL NERVOUS SYSTEM

Turning now to the main character under consideration, the weight of the central nervous system, the condensed records are presented in table 10.

When these data are put in the form of a chart, (chart 2) several interesting relations between the observations of 1904 and those of 1909 at once appear. In the first place the later records follow the same line as the earlier; second, the record for each species in 1909 is somewhat less than in 1904, and as a consequence still further below the records of 1904 for *R. pipiens*. This result serves to establish the main conclusion, namely that *R. pipiens* has a heavier nervous system than either of the European forms. The fact that the values for the weight of the central nervous system in the European species as determined in 1909 are less than those determined in 1904, calls for a word of comment.

Some unpublished studies which are being made on *R. pipiens* at the Wistar Institute relative to the change in the weight of the central nervous system with season, indicate that in this species the greatest weight is attained about the end of July.

CHART 2. Showing the weight of the entire central nervous system.

TABLE 10

Weight of the central nervous system in grams. Averages from groups of three

	BODY WEIGHT	WEIGHT OF CENTRAL NERVOUS SYSTEM
R. pipiens.....	{ 14.9	.1040
	{ 23.2	.1256
	{ 30.8	.1463
	{ 43.2	.1674
R. esculenta 1904.....	{ 15.9	.0890
	{ 22.0	.1142
	{ 35.0 [2]	.1402
	{ 40.2	.1498
R. esculenta 1909.....	{ 23.4	.1077
	{ 32.1	.1258
	{ 43.8	.1524
	{ 55.2 [2]	.1724
R. temporaria 1904.....	{ 15.9	.0929
	{ 23.1	.1108
	{ 28.0	.1213
	{ 31.3	.1323
R. temporaria 1909.....	{ 18.0 [4]	.0899
	{ 25.2	.1053
	{ 26.9	.1144
	{ 29.5	.1212
	{ 34.8	.1380

If this observation applies, as it probably does, to the European species, then the differences in weight as shown in chart 2 are susceptible of the following explanation:

The esculenta of 1904 were examined August 1—5, when it may be assumed that the nervous system of *R. esculenta* had attained approximately its maximal seasonal weight. In 1909 the examination was from July 5-7, or some four weeks earlier. Under these circumstances, a somewhat smaller weight was to be expected, and the records show this.

The temporaria of 1904 were examined July 11 and 12, before the central nervous system had reached the maximum for the season.

In 1909 the examination was from August 17 to 21, or some

three weeks after the assumed maximum, and at a time when the seasonal weight has begun to diminish. Here the difference is less than in the case of the esculenta, but is susceptible of a similar explanation.

The relation of these two series of observations can be conveniently shown in still another way.

I have been able to point out (Donaldson '02) that a fairly accurate determination of the weight of the central nervous system in frogs can be made by the formula

$$C. N. S. = (Log. Bd. W. \sqrt[4]{L}) C$$

where *C. N. S.* is the weight of the central nervous system, *Bd. W.* the body weight in grams, *L* the total length in mm. and *C.* a constant to be determined for each species. Since publishing this formula I have found that the most convenient way of expressing seasonal variations on the weight of the central nervous system is by the variations in *C.*

Applying this method to the series before us, and remembering that the increase in the relative weight of the central nervous system is measured by the increase in *C.*, and vice versa, we obtain the following:

TABLE 11

To show the values of "C" for each of the several series

	AVERAGE BODY WEIGHT	VALUE OF C.
R. pipiens 1904 } Average of 12 records }	28.0	26.2
R. esculenta 1904 } Average of 8 records }	32.4	24.6
First "weight group" omitted }		
R. esculenta 1909 } Average of 9 records }	33.1	23.0
Last "weight group" omitted }		
		Difference 1.6
R. temporaria 1904 } Average of 12 records }	24.6	22.8
R. temporaria 1909 } Average of 16 records }	26.8	21.9
		Difference 0.9

As is to be seen by inspection of the foregoing table 11 the value of C for the 1904 records is greater in both the European species than for the corresponding 1909 records, and as noted above, the greatest difference (1.6) is in the case of *R. esculenta*.

In connection with this table a word of explanation is required. It has been found that there is a slight increase in the value of C as the absolute size of the frog increases. This is a relation previously overlooked, but which will be discussed elsewhere. The bearing of it on the present case is that in making a comparison of the values of C in any pair of records, it is necessary in order to get trustworthy results, to compare the determinations for frogs of approximately the same range in size. In the present instance this makes it necessary in the case of *R. esculenta* to omit the value of C for the first weight group of the 1904 series, because there is no corresponding weight group on the 1909 series, and similarly to omit the determinations for the last weight group of the 1909 series.

A glance at chart 2 will serve to supplement the explanation.

In the case of the records for *R. temporaria*, the values for C in all the weight groups of both years have been used in making up the averages. It is because of this influence of the absolute size that the average body weights for each series are entered in table 11.

All through the present paper the data on *R. pipiens* used in 1904 have been repeated without revision. In the former communication (Donaldson '08. pp. 132-133) it was noted that the weight of the central nervous system in the series of this species was low in comparison with other data which we had. This statement still holds good, but it was thought wiser to leave the standard as represented by 1904 records on *R. pipiens* unchanged at this time.

As evidence that the weights here used were low for this species, I give below two other series of determinations of C on Chicago frogs as follows:

NUMBER OF SPECIMENS	DATE ABOUT AUG. 1	AVERAGE BODY WEIGHT GMS.	AVERAGE VALUE FOR C .
48.....	1902	22.3	28.6
4.....	1909	27.7	29.5

It will be seen on comparison with the value of C . for the series of *R. pipiens* here used ($C = 26.2$) that these are much higher. This implies an increase in the weight of the central nervous system proportional to the differences in the values of C after correction for the differences in body weights in the several series.

Why the particular series of *pipiens* used by me as a standard falls below that for the two other series is a point the discussion of which must be reserved for a future paper.

In this connection it is desirable to refer to one modifying condition affecting the value of C which has not heretofore been mentioned, and the data on which are still unpublished. I find that the value of C is not the same for specimens of *R. pipiens* from different parts of our own country. For example those coming from northern Minnesota give a value sensibly greater than that found for the so-called "Chicago frogs" and the specimens taken about Philadelphia give a value less than that found for the "Chicago frogs," as a rule, but almost identical with that of the series used as a standard in this paper.

R. pipiens extends much farther south in this country of course, being found both in Florida and Texas. What the relation of C may be in specimens from stations farther south than Pennsylvania, has still to be determined, but the possibility of variation in this character with latitude is a matter of much interest.

(F) THE RATIO OF THE WEIGHT OF THE BRAIN TO THAT OF THE SPINAL CORD

Omitting the tabulation of the absolute values for the brain and cord, as these can be readily found in the full tables, I give below in table 12 a condensed statement of the ratios.

It will be seen that in both 1904 and 1909, that relative weight of brain (the value given under "ratio") is higher in *R. esculenta* than in *R. temporaria*, although the difference is not so great in the later as in the earlier records. Further, this ratio in *R. pipiens* is always greater than in either of the European forms.

Finally it is to be noted that the ratios which I find for the

European species are much higher than those determined by Fubini (see Donaldson '08, table 20) and so confirm my earlier conclusions concerning the untrustworthy character of his records.

TABLE 12

Ratios of the weight of the brain to the weight of the spinal cord. Averages from groups of three

	BODY WEIGHT	RATIO
R. pipiens.....	{ 14.9	2.33
	{ 23.2	2.22
	{ 30.8	2.32
	{ 43.2	2.28
R. esculenta 1904.....	{ 15.9	2.29
	{ 22.0	2.22
	{ 35.0 [2]	2.09
	{ 40.2	2.05
R. esculenta 1909.....	{ 23.4	2.03
	{ 32.1	2.09
	{ 43.8	2.05
	{ 55.2 [2]	2.24
R. temporaria 1904.....	{ 15.9	2.15
	{ 23.1	1.86
	{ 28.0	1.87
	{ 31.3	1.87
R. temporaria 1909.....	{ 18.0 [4]	2.02
	{ 25.2	2.02
	{ 26.9	2.06
	{ 29.5	1.90
	{ 34.8	1.93

(G) THE PERCENTAGE OF WATER IN THE BRAIN AND SPINAL CORD

Table 13 shows the condensed results on the percentage of water. In my former communication I called attention to the differences in this character in the several species (Donaldson '08, p. 139.)

While the percentages of water in both the brain and spinal cord as determined for both European species in 1909 are less than that found in *R. pipiens*, they are nearly alike, and also similar to the 1904 determination for *R. esculenta*, so that it is

not desirable to give any weight to the differences as observed in 1904.

The value of this table as it stands is to show that we were dealing in all cases with healthy frogs, as the frog readily shows by changes in the amount of water in the nervous system, the effect of infections or disturbing conditions.

TABLE 13

Showing the percentage of water in the brain and in the spinal cord. Averages from groups of three

	BODY WEIGHT	PERCENTAGE OF WATER IN Brain Spinal Cord	
R. pipiens.	{ 14.9	84.5	80.2
	{ 23.2	84.7	80.1
	{ 30.8	85.0	80.5
	{ 43.2	85.7	81.2
R. esculenta 1904.	{ 15.9	83.6	78.6
	{ 22.0	83.3	78.9
	{ 35.0 [2]	83.3	78.4
	{ 40.2	83.2	78.3
R. esculenta 1909.	{ 23.4	83.6	78.6
	{ 32.1	83.6	77.9
	{ 43.8	83.6	78.4
	{ 55.2 [2]	83.6	78.3
R. temporaria 1904.	{ 15.9	82.7	78.6
	{ 23.1	82.3	77.2
	{ 28.0	82.1	76.9
	{ 31.3	81.6	77.8
R. temporaria 1909.	{ 18.0 [4]	83.0	78.1
	{ 25.2	84.1	78.7
	{ 26.9	84.0	78.4
	{ 29.5	83.8	79.1
	{ 34.8	83.6	78.7

The foregoing tables and the comments on them are intended to demonstrate that a second series of observations on *R. esculenta* and *R. temporaria* made in 1909 at an interval of five years yield results substantially similar to those first obtained in 1904

and that therefore so far as the conclusions of the earlier paper depend on the observations which have been repeated, they may be considered as well founded.

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