tinuance for some time, the foregoing results are offered with the expectation of supplementing them later as opportunity permits.

In conclusion, I wish to express my appreciation of the kindness of Prof. G. T. Moore, of the Missouri Botanical Gardens, in placing at my disposal the resources of the gardens.

Conclusions.

The tissue of maple shoots, when mixed with solutions of malic acid or malates and exposed to sunlight, cause an increase in reducing power and a decrease of acidity in the solutions—which may be interpreted to mean a transformation of the malic acid into sugar.

A less pronounced change of the same kind is produced in darkness at 38°.

The active principle which produces the change is somewhat soluble in water, is destroyed by boiling and is therefore probably of enzyme nature.

The tissue of maple buds similarly treated brings about a decrease of reducing substance and an increase of acidity in solutions of malic acid or its salts.

[CONTRIBUTION FROM THE FOOD RESEARCH LABORATORY, BUREAU OF CHEMISTRY, U. S. DEPARTMENT OF AGRICULTURE.]

OSMOTIC ACTIVITY IN THE EGG OF THE COMMON FOWL.[1]

BY A. D. GREENLEE.

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Introduction.

Eggs, when fresh, contain a large percentage of moisture, and like all other highly aqueous substances they lose moisture on standing by evaporation to the external atmosphere. Chemical analyses of eggs by various investigators are fairly numerous, but little has been done to correlate the change in moisture content with the age or condition of the egg. König[2] reports an analysis of eggs by Bostock as early as 1855. Langworthy[3] determined the percentage in the whole egg with and without the shell, and on the white and yolk separate, and on boiled eggs. Lebbin[4] found the relative percentage of yolk, white, and shell. Cook[5] made a more extensive study of eggs, and, together with other changes, found that "eggs in storage for one year show a loss of weight equivalent to 10% of the total weight, which loss is largely water from the whites." He also found that "when fresh eggs are boiled a loss in weight

[1] Preliminary paper.

[2] *Chemie der Memnschlichen Nahrungs und Genussmittel*, 1, 98.

[3] U. S. Dept. Agr., *Farmers' Bulletin* 128.

[4] *Z. öffent. Chemie*, 6, 148 (1900).

[5] U. S. Dept. Agr., Bureau of Chemistry, *Bull.* 115.

occurs, while storage eggs gain on boiling. Apparently the whites lose more water than do the yolks and consequently gain more when boiled. The boiled yolks when fresh contain less than 50% of water; when cold-stored this percentage is increased, the figure reaching 64% in the last examination."

Pennington[1] in a rather extensive study of fresh eggs determined the moisture on several samples for the whites and yolks separately and established averages which have served as the basis of many calculations in this work.

Moisture Determinations.

The White Leghorn eggs used for this investigation were obtained from a poultry farm and were not more than 24 hours old when the experiment began. They were kept in the ordinary commercial 30-dozen egg crate at a temperature of 32° F.; those held at higher temperatures were in pasteboard cartons, each holding 1 dozen. The cartons were so arranged that they were not in juxtaposition, in order to give a uniform exposure to the atmosphere. An analysis of the fresh eggs was made and then of other samples from the lots held at 32° F. at intervals of from one to three months and from the higher temperature lots at intervals of from three to ten days. Two dozen eggs were usually taken for a sample in order to insure uniformity. The results of the moisture determinations are recorded in Table I.

Table I.—Percentage Change of the Moisture Content of White and Yolk on Holding at Different Temperatures for Varying Periods.

Expt. 196 (32° F.).			Expt. 197 (32° F.).			Expt. 356 (32° F.).			Expt. 357 (32° F.).		
Age in days.	White.	Yolk.	Age in days.	White.	Yolk.	Age in days.	White.	Yolk.	Age in days.	White.	Yolk.
41	87.42	49.15	43	87.54	48.80	0	88.25	47.35	14	88.75	47.17
76	87.15	49.77	83	86.42	49.25	14	87.59	47.87	21	88.06	47.54
166	86.65	50.25	197	86.30	50.54	35	87.55	48.05	35	87.94	48.08
201	86.19	49.73	268	85.96	50.81	49	87.10	49.35	49	88.16	48.37
266	85.35	50.60	...	...	...	..	...	...	..	...	...
Expt. 363 (52° F.).			Expt. 355 (65° F.).			Expt. 334 (70° F.).			Expt. 336 (80° F.).		
Age in days.	White.	Yolk.	Age in days.	White.	Yolk.	Age in days.	White.	Yolk.	Age in days.	White.	Yolk.
0	88.24	46.63	0	87.66	47.64	0	87.60	47.17	0	87.75	47.97
14	87.57	47.80	7	86.71	48.96	6	87.09	48.53	7	86.98	49.00
35	87.44	49.39	18	86.11	50.28	13	86.63	49.13	11	86.51	49.72
..	...	...	34	85.99	50.56	19	86.45	49.26	15	86.77	49.78
..	...	...	..	...	...	26	85.16	50.27	19	85.89	50.44
..	...	...	..	...	...	33	85.09	50.19	..	...	...
..	...	...	..	...	...	40	84.74	50.60	..	...	...

Interpretation of Results.

The decrease in moisture in the white appears to be easily accounted for by evaporation to the external atmosphere, but the loss in weight

[1] *J. Biol. Chem.*, **7**, 109 (1910).

as shown by means of the balance is not sufficient to account for the entire decrease in the percentage of moisture. This fact, together with the increased moisture in the yolk, suggests a transfer of water from white to yolk.

As far as the results given in Table I indicate, it can not be definitely proved that water passes from the white to the yolk. The same results would be obtained if the white took up solids from the yolk, and the yolk would apparently increase in water if, during the process of desiccation in the determination of solids in the oven at 100° C., some of the solids were volatilized. The two chief constituents in the yolk, aside from water, are fat and protein. A determination of the Reichert-Meissl number of the extracted fat indicates no increase of volatile fatty acids, from which it may reasonably be assumed that there is no volatilization of fatty substance. Furthermore, an analysis of the dried sample gives the same percentage of nitrogen as the percentage of nitrogen in the fresh sample calculated on the water-free basis, which would show that there is no loss of nitrogenous or protein matter. By a process of mathematical calculation it can easily be shown that solids have not passed from the yolk to the white. Although the yolk has decreased in the percentage of its solids by about the same number of points that the white has increased, nevertheless, since the percentage of solids in the yolk greatly exceeds that in the white, the amount which it would be necessary for the yolk to lose in order to account for the experimental data would be more than sufficient to raise the percentage of solids in the white to the experimental figures, regardless of the fact that there is almost twice as much white as yolk, and even if there were no loss of moisture to the external atmosphere.

This phenomenon of a transfer of water from the white to the yolk may easily be explained by the simple process of osmosis. The yolk, which contains a very high percentage of solids, is surrounded by a membranous tissue called the vitellin membrane, which in turn is surrounded by the egg white, a liquid much more dilute than the yolk. By osmosis the water passes through the membrane from the more dilute to the more concentrated solution until an equilibrium is obtained. In the egg this process continues until the vitellin membrane becomes so weak that it breaks, when the white and yolk begin to lose their identity. This action proceeds with such definiteness that by a process of calculation, knowing the original weight of the egg, the loss in moisture to the external atmosphere can be calculated with surprising closeness to the actual loss as shown by the balance. The following examples are given:

EXAMPLE I. No. 334 1–7 (40 days at room temperature).

(Fresh eggs = 59.35% white, 30.39% yolk, 10.26% shell.)

561 grams = original weight of 10 fresh eggs.

561 × 59.35% = 323.95 grams, original weight of white in 10 fresh eggs.
561 × 30.39% = 170.49 grams, original weight of yolk in 10 fresh eggs.
332.95 × 12.40% = 41.28 grams, solids in white of 10 eggs.
170.49 × 52.83% = 90.07 grams, solids in yolk of 10 eggs.
41.28 ÷ 15.26% = 270.52 grams, final weight of white.
90.07 ÷ 49.40% = 182.33 grams, final weight of yolk.
62.43 grams = total loss to white (calculated).
11.84 grams = total gain to yolk (182.33–170.49).
50.59 grams = loss to external atmosphere (calculated).
(Experimental loss to 10 eggs as taken from No. 334–7 shows an average of 49.53 gram .)

EXAMPLE II. No. 336 1–5 (19 days at room temperature).

556.6 grams = original weight of 10 fresh eggs.
556.6 × 59.35% = 330.34 grams, original weight of white in 10 eggs.
56.6 × 30.39% = 169.15 grams, original weight of yolk in 10 eggs.
330.34 × 12.25% = 40.46 grams, solids in white of 10 eggs.
169.15 × 53.02% = 88.01 grams, solids in yolk of 10 eggs.
40.46 ÷ 14.11% = 286.8 grams, final weight of white.
88.01 ÷ 49.56% = 177.6 grams, final weight of yolk.
43.54 grams = total loss to white (calculated).
8.40 grams = total gain to yolk (calculated).
35.14 = loss to external atmosphere (calculated).
35.83 = average loss to 10 eggs as shown by experiment No. 336–5.

TABLE II.—PERCENTAGE OF SHELL, WHITE, AND YOLK.

Expt. number and condition.	Weight of egg. Grams.	Shell. Per cent.	Yolk. Per cent.	White. Per cent.
Fresh.				
I	54.3153	10.13	31.35	58.52
II	58.5077	9.57	30.81	59.62
III	54.1700	10.83	30.00	59.17
IV	61.9017	10.53	29.40	60.07
Average	57.2237	10.26	30.39	59.35
Stale.				
I	51.3885	11.06	33.12	55.82

The change in water content increases its rate with the temperature and diminishes with the time. The rate or the average rate from the beginning of the experiment up to any given time is merely the loss in weight divided by the time. The original weight of the white or yolk is obtained, as shown in Examples I and II, and the weight of either at any stage in the experiment can be calculated on the basis of total solids. These rates are plotted in Figs. 1 and 2. It should be stated, however, that the curves are mathematically exact and consequently are slightly more uniform than could be obtained by experiment even under the most ideal conditions. Experimental results vary above and below the values indicated by the curves, but follow them very closely in general direction.

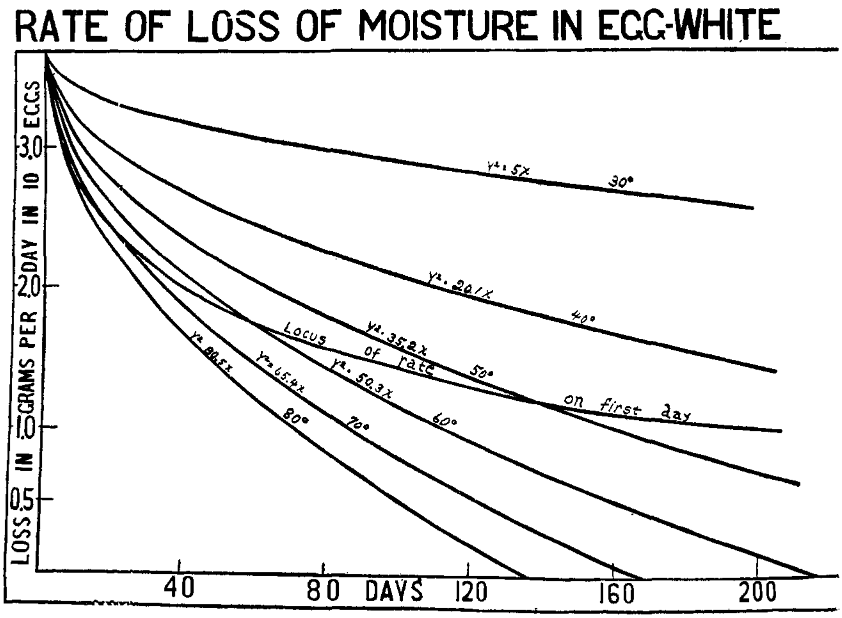
RATE OF LOSS OF MOISTURE IN EGG-WHITE
LOSS IN GRAMS PER DAY IN 10 EGGS
3.0
2.0
1.0
0.5
40
80
DAYS
120
160
200
30°
40°
50°
60°
70°
80°
Locus of rate on first day

Fig. 1.

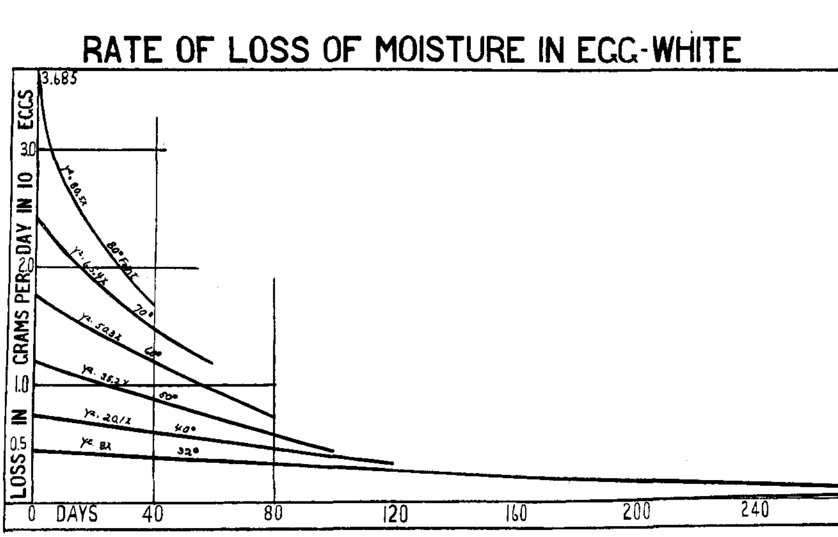
RATE OF LOSS OF MOISTURE IN EGG-WHITE
LOSS IN GRAMS PER DAY IN 10 EGGS
3.685
3.0
2.0
1.0
0.5
0
DAYS
40
80
120
160
200
240
70°
60°
50°
40°
32°

Fig. 2.

Development of the Rate Formula.

The formula is developed on the basis of the Fahrenheit scale. Days are plotted as abscissas, and grams per day or rate as ordinates. In the scale 1 unit of rate equals 20 units of the y ordinate; in the formulas y is negative and equals the distance downward from the 3.685 ordinate, or $R = 3.685 + y/20$ from which $y = -(73.7 - 20\,R)$; D = days = $2X$. The curve for the 80° experiment will take the shape of a parabola;[1] $y^2 = 80.5x$ with the axis on the y ordinate 3.685, and the vertex at $x = 0$. In order to be able to combine the curves for the different temperatures in one formula as a function of the temperature the curves must constitute a family. It is found that the 70° curve coincides almost exactly with $y^2 = 65.4x$ (if y is greater than 25.31) on the same axis, and the 32° curve in the same fashion coincides with $y^2 = 8x$ (if $y > 65$) also on the same axis. From this the following series is interpolated:

(1) At 80°, $y^2 = 80.5x$
(2) At 70°, $y^2 = 65.4x$
(3) At 60°, $y^2 = 50.3x$
(4) At 50°, $y^2 = 35.2x$
(5) At 40°, $y^2 = 20.1x$
(6) At 30°, $y^2 = 5.0x$
(7) At 27°, $y^2 = 0$

giving the general formula

$$y^2 = \left(\left(\frac{t-30}{10}\right) 15.1 + 5\right)x \quad \text{or} \quad y^2 = (1.51t - 40.3)x.$$

These are plotted in Fig. 1. The curve, however, for each temperature, does not start from $y = 0$, but from a variable origin, the locus of whose points is the parabola $v^2 = 39.1z$ (v = temperature and z = 4 plus 50 times the rate on the first day, giving the formula $t^2 = 39.1\,(50\,R_1 + 4)$). The parabola $v^2 = 39.1z$ is derived from a plotting of the rate for the first day at different temperatures, or R_1.

From

$$t^2 = 39.1\,(50\,R_1 + 4) \quad \text{or} \quad t^2 = 1955\,R_1 + 156.4$$

$$R_1 = \frac{t^2 - 156.4}{1955}.$$

In order to compare the curves for the different temperatures they are all started on the same ordinate of the vertex of the 80° curve, since this vertex happens to be at $D = 0$ (Fig. 2). Then in the general formula $y^2 = (1.51t - 40.3)\,x$, y has the same scale value at all times ($y = -(73.7 - 20\,R)$), but the value of x will be a function of the temperature.

[1] The parabolic curve was used in preference to the logarithmic, since diffusion rather than chemical action is responsible for the changes observed, and the parabola expresses in a more simple manner the facts observed.

$$x = \frac{y_1^2}{1.51t - 40.3} + \frac{D}{2} \qquad y_1 = y \text{ on the first day.}$$

$$y_1 = -(73.7 - 20\,R_1)$$

$$= -\left(73.7 - \frac{20t^2 - 3128}{1955}\right)$$

$$= -\left(75.3 - \frac{4t^2}{391}\right)$$

$$x = \frac{\left(75.3 - \frac{4t^2}{391}\right)^2}{1.51t - 40.3} + \frac{D}{2}$$

$$(73.7 - 20R)^2 = (1.51t - 40.3)\left\{\frac{\left(75.3 - \frac{4t^2}{391}\right)^2}{1.51t - 40.3} + \frac{D}{2}\right\}$$

$$73.7 - 20R = \sqrt{\left(75.3 - \frac{4t^2}{391}\right)^2 - \frac{(1.51t - 40.3)D}{2}}$$

$$R = 3.685 - \frac{1}{20}\sqrt{\left(75.3 - \frac{4t^2}{391}\right)^2 - \frac{(1.51t - 40.3)D}{2}}$$

No experiment has been conducted at a temperature lower than 32° F., but following the order of progression in the remainder of the series y becomes zero at about 27°, indicating no transfer of water. This is also approximately the freezing point of egg material, further confirming the nonprobability of a transfer of water.

The curves in Fig. 2 are the parts of the curves of Fig. 1 below the "locus of rates on the first day," all moved back to $x = 0$. These are in fact the experimental curves, whereas the prolongations in Fig. 1 merely indicate the mathematical relation.

Application of the Formula.

By means of the rate formula and one analysis of a sample of eggs it is possible to predict the condition of the eggs at any temperature for any given date within a reasonable holding period. The rate multiplied by the time gives the loss in weight, from which data it is a simple matter to find the percentage of moisture remaining.

The curves are all on the loss of water in the white. Similar results are obtained by plotting the rate of increased moisture in the yolk. More extensive work along this line is now being done.