

## THE GROWTH OF CUCUMIS MELO SEEDLINGS AT DIFFERENT TEMPERATURES

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### I

The work of Julius Sachs (10) published in 1860, and in particular his discussion of the results, served to introduce two concepts into plant physiology, that of minimal, optimal, and maximal temperatures, and that of the grand period of seedling growth. Each of these phenomena has received the attention of numerous workers since that time but there have been relatively few attempts to combine the two fields of interest and to follow the course of growth of seedlings cultured in darkness at several constant temperatures throughout their entire growth period. The work of Sierp (11), Hamada (4), and Silberschmidt (12) on the growth of oat coleoptiles belongs in this class, as does that of Edwards, Pearl, and Gould (1) on *Celosia cristata* seedlings. Silberschmidt also tested pea and rice seedlings and contributed an interesting analysis of the problem.

The present paper deals with the growth of *Cucumis melo* seedlings in darkness at seven constant temperatures between 15° and 40°C. inclusive. While we have accumulated in this laboratory during the past 10 years a considerable volume of unpublished observations on the effect of temperature on the growth of canteloup seedlings, so far as is known the only published work on the temperature relations of growth in this species are those of de Vries (14) who found greater growth at the end of a 48 hour period at 37.2° than at any of the other three temperatures he tested. Gregory (3) has made interesting temperature tests of another sort on *Cucumis sativus* seedlings grown in light which indicate a much lower optimal temperature range than was found in de Vries' experiments or in those presented here.

The cultural methods used in the experiments reported here are essentially those used in other experimentation on canteloup seedlings reported from this laboratory (2, 6, 8) and they may be stated briefly as follows: Seeds from a single melon were freed from their testas, weighed, and only those whose weights fell between 0.0200 and 0.0240 gm. were used. They were immersed 1 minute in 1:1000 HgCl<sub>2</sub> solution, rinsed once, and soaked 3 hours in sterile distilled water in individual vials. They were planted in previously sterilized glass tubes, 44 cm. long and 2 cm. in diameter, containing 25 cc. of 1 per cent agar (the 30° tubes contained 40 cc.) made up in Knop's mineral salt nutrient solution, instead of distilled water as in the other experiments. Another difference in the procedure used here consisted of forcing 80–90 cc. of air into each tube daily, but comparative tests (8) showed that this did not improve growth. The tubes were kept in darkness in constant temperature chambers and the lengths of the straight portion of the hypocotyl, exclusive of the curved portion at the top, were measured under non-actinic ruby light regularly at 24 hour intervals from the time of planting of the seeds.

The purpose of the present paper is to record the description of certain experimental observations made in the course of development of the program of investigation of seedling growth and duration of life, which has been in progress in this laboratory since 1925, and to analyze quantitatively the effect of temperature upon the growth of *Cucumis melo* seedlings under a particular set of experimental conditions. A more extensive series of experiments of this kind will be reported later, in which *Cucumis* seedlings were grown with and without mineral salts, at five constant temperatures, and in which the respective durations of life were observed.

## II

Table I shows the mean lengths of *Cucumis melo* hypocotyls for various intervals after planting at six constant temperatures. Germination and a little root growth occurred at 40° but the hypocotyls did not become differentiated. There is a little irregularity in the 20° data near the end of the growth cycle because observations on two series of experiments were combined; in one series no readings were made on the 16th day and in the other none were made on the 17th. Although the numbers of seedlings used in these tests are small, nevertheless there was good agreement between the values within each series and those obtained by repetition. Except at 15° two or more series were run at each temperature and the data were combined.

The data show that during the first 2 days of observed elongation of the hypocotyl both the 37.5° and 35° seedlings were taller than those at 30°, but their rapid initial growth was not sustained long, and the

TABLE I  
*Mean Height of Cucumis melo Seedlings at Six Constant Temperatures*

Interval after planting	15°	20°	25°	30°	35°	37.5°
<i>days</i>	<i>mm.</i>	<i>mm.</i>	<i>mm.</i>	<i>mm.</i>	<i>mm.</i>	<i>mm.</i>
2	—	—	2.6	3.1	8.9	5.5
3	—	—	14.1	24.0	41.4	25.0
4	—	2.3	51.1	77.0	89.1	57.2
5	—	7.8	101.8	121.5	127.4	91.5
6	—	13.7	142.3	166.4	159.2	118.5
7	—	31.4	176.6	205.3	178.2	136.0
8	—	59.0	203.4	225.5	187.5	146.9
9	—	92.3	212.1	236.9	191.5	151.9
10	—	120.6	219.0	240.4	192.8	153.9
11	—	143.8	221.2	242.8	194.2	154.2
12	—	159.5	223.6	243.8	194.8	—
13	1.0	167.9	224.1	—	—	—
14	1.0	172.2	—	—	—	—
15	—	173.1	—	—	—	—
16	2.6	178.7	—	—	—	—
17	3.6	177.5	—	—	—	—
18	5.3	177.8	—	—	—	—
19	7.1	—	—	—	—	—
21	13.0	—	—	—	—	—
23	21.9	—	—	—	—	—
25	35.1	—	—	—	—	—
27	49.0	—	—	—	—	—
29	61.4	—	—	—	—	—
31	69.1	—	—	—	—	—
33	72.3	—	—	—	—	—
35	73.0	—	—	—	—	—
37	73.4	—	—	—	—	—
No. of seedlings...	7	13	17	18	17	11

30° cultures finally grew taller than any others. Although the 25° cultures grew more slowly than at 30° nevertheless they finally grew nearly as tall. At 20° about 4 days were required for differentiation and the first appreciable elongation of the hypocotyl. Growth was

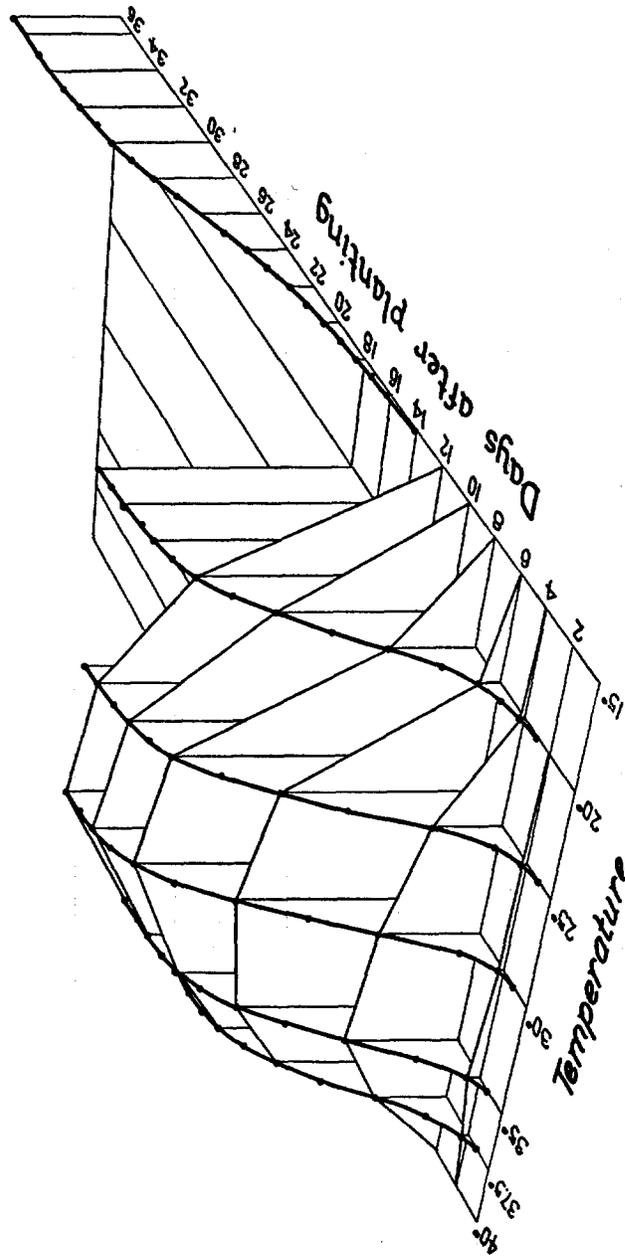


FIG. 1. Three dimensional diagram showing the observed heights of *Cucumis melo* hypocotyls (represented by heights of plotted points) as observed at six constant temperatures for total growth periods of different durations.

still slower than at 25° and the final height was less but they grew more rapidly and to a greater height than at 15°.

If, for purposes of comparison, one considers these sigmoid growth curves as made up of three portions, a lower part which is convex downwards, a nearly straight portion on either side of the point of inflection, and an upper part which is concave downward some interesting differences in form among the curves appear. The lower portion is most prominent at the lower temperatures, and as one examines the curves for successively higher temperatures it becomes less conspicuous. An examination of the slope of the central portions shows that the 30° seedlings grew most rapidly of all, and the farther removed from 30° either above or below that a tested temperature lay, the lower was the maximal growth rate observed. As for the upper parts of the curves, little difference in form is to be seen. A study of the increments of growth serves to confirm these relations but this does not require further discussion here.

The three dimensional diagram in Fig. 1 has been constructed as though the curves of Table I had been traced on some material like cardboard, then cut out and made to stand perpendicularly and in proper order. Intersecting these planes at right angles is another set of graphs which shows the heights attained by the hypocotyls after growth periods of certain durations at the several constant temperatures. This gives rise to a system of three coordinates; the temperature scale is laid off along one axis, the number of days after the planting of the seeds along another, and the heights of the plotted points above the plane formed by the first two coordinates represent the mean heights of hypocotyl grown at a given temperature for the period of time indicated. The curves passing through these points have been smoothed graphically. An attempt has been made to allow for the effect of perspective in drawing this diagram; a separate scale of height, proportionate to the distance from the vanishing point on the left, has been used for each temperature, and all the lines of the time and temperature coordinates have been made to converge on two vanishing points, one at the left, and one behind the diagram.

Three dimensional diagrams of this sort have been used by Rahn (9) to show the temperature relations of enzyme action and similar phenomena, and have long been a commonplace of statistical litera-

ture, particularly in the writings of Karl Pearson and his associates, and of A. J. Lotka.

An inspection of the transverse planes connecting culture periods of the same duration provides a means for comparing graphically the relative heights of the seedlings. The high initial growth rates prevailing at 35° stand out clearly, and the greater relative growth that occurs at 30° as time goes on is shown by the slopes of the lines that connect the two growth curves. Shifts of the optimal temperature with time were first reported by Lehenbauer (5) for maize seedlings, and they occur in the data of Talma (13) for *Lepidium sativum* roots, in the work of Silberschmidt (12) for oat coleoptiles, and in particular in his experiments on pea epicotyls which had their highest initial growth rates at 25° but which attained their greatest length at 12.5°C. The data of Edwards, Pearl, and Gould (1) for *Celosia* show similar relations as do the present data for *Cucumis*.

### III

While Fig. 1 permits a rough visual appreciation of the effect of temperature on growth in these experiments, further analysis is essential to an understanding of the matter. It is obvious that there are many variables involved in the growth of any organism. In the present case the records taken permit an analysis of three of the basic variables in all growth phenomena; namely, *yield* (here measured by length of hypocotyl), *time*, duration of whole growth period, defined as the time from planting to cessation of elongation of the hypocotyl and, derivatively from these two, the total *time rate* of growth (*i.e.* the amount of yield per unit of time). Study of the data of Table I brings out the following relationships.

1. As the temperature deviates from the observed optimum<sup>1</sup> (30°),

<sup>1</sup> Since the literature indicates a considerable confusion of thought and diversity of usage regarding the concept of the optimum temperature for growth, it may be well to state that in this paper we use the term to indicate that temperature at which the greatest total amount of growth activity occurs. Under the conditions of our experiments with *Cucumis melo* it is a matter of indifference as to whether (*a*) mean time rate of growth (yield per unit of time spent in growing) or (*b*) mean absolute yield (length of hypocotyl) at the end of growth, be taken as the index or measure of "total amount of growth activity." This may not necessarily be the case with other species or under other experimental conditions.

in either the plus or minus direction the *yield* is reduced. The minus deviations in yield, measured in units relative to the yield at the optimum, are fairly closely proportional to the relative deviation in temperature (neglecting signs and having regard to errors of sampling) at temperatures near the optimum, but are in excess, proportionally to the temperature deviations, at the outer temperatures remote from the optimum.

2. As the temperature deviates in the minus direction from the optimum the *duration* of the whole growth period tends to be prolonged, again about proportionally (relatively) to the temperature deviations at temperatures near the optimum, but in excess at the temperatures more remote. At temperatures above the optimum (so far as the data go) the duration of the growth period appears to be approximately constant and the same as that at the optimum.

3. The above relationships at once suggest that the total *time rate* of growth (yield per unit of time over the whole growth period) will be found to follow a parabolic relation to temperature, of the general form

$$R = a + bT + cT^2, \quad (i)$$

where  $R$  = rate as above defined, and  $T$  = temperature in  $C^\circ$  above zero.

This suggested relationship turns out to be the fact, when the rates computed from the data of Table I are fitted by least squares with such a parabola. The equation so computed is

$$R = 4.864T - 0.082T^2 - 53.274, \quad (ii)$$

where  $R$  = average growth in mm. of hypocotyl *per diem* over whole period of growth, and  $T$  = temperature in  $C^\circ$ . Putting  $\frac{dR}{dT} = 0$  from equation (ii) it appears that the temperature at which the average time rate of growth of *Cucumis melo* is at its maximum under the conditions of these experiments, is  $29.74^\circ C$ .

The observations and fitted curve are shown graphically in Fig. 2.

The fit is an obviously reasonable one, considering the magnitude of the data.

In the discussion up to this point we have taken as a single numerical measure of total growth activity of the seedling—a logically necessary

statistic if growth activity is to be related mathematically to temperature—the mean total time rate of growth (yield per unit of time) as defined above. This procedure involves two postulates; namely, (1) that the process called growth is a continuous one, at, however, widely varying rates, from the time the dry seed is planted and begins

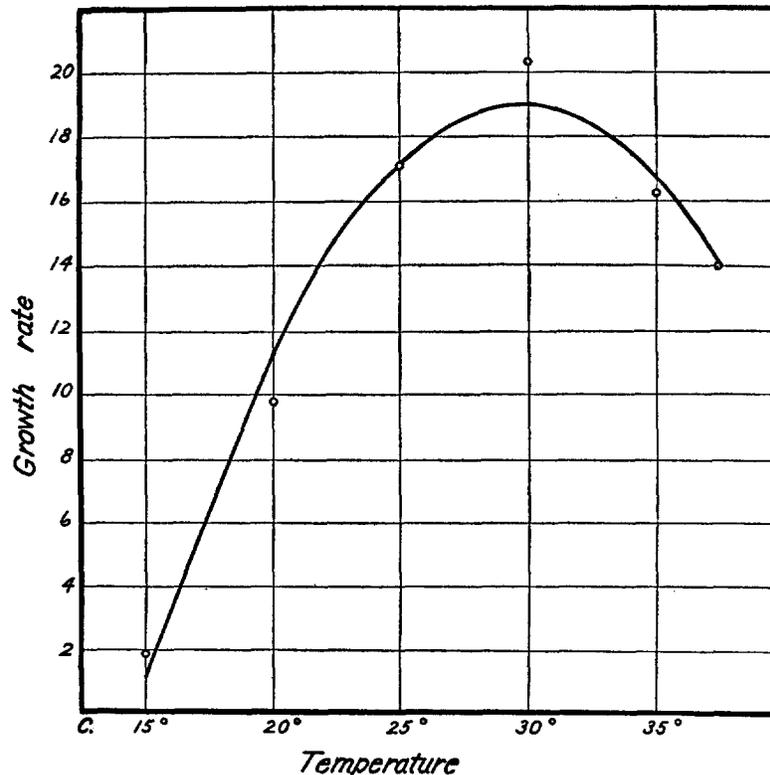


FIG. 2. Change of time rate of growth of *Cucumis melo* with temperature. Circles give the observations, and the smooth curve is the graph of equation (ii).

the imbibition of water, until the process ends, with the cessation of enlargement of the plant. In other words this postulate assumes that there is no biological discontinuity between the processes of germination on the one hand and *visible* growth of the plant and its organs on the other hand. (2) That the mean total time rate of growth (as defined earlier and computed in the present instance by dividing the final

height of the hypocotyl by the whole number of days of growth from planting to the cessation of growth) is a reasonable and efficient *single* numerical index of total growth activity. While these two postulates seem reasonable, it is evident that they are by no means the only ones possible in the premises. Other measures of growth rate might be taken; germination might conceivably be regarded as something

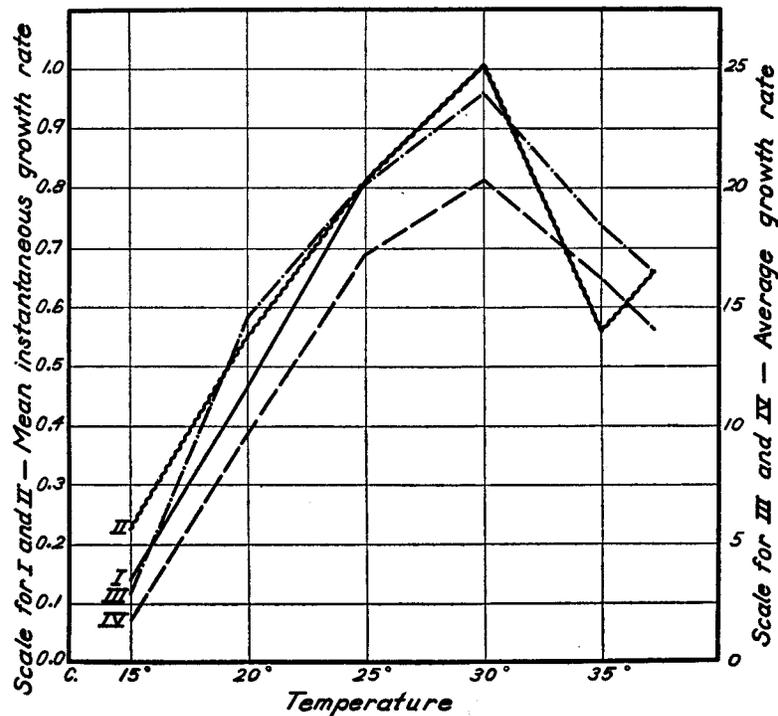


FIG. 3. Showing the similarity of results under different postulates as to growth rates and times.

biologically different from visible growth, disparate, and discontinuous with it. Would the results be different so far as concerns the main point at issue—the relation of growth of the seedling to temperature—if postulates different from those used above were adopted?

To test this question three different procedures were tried, in addition to the one already described above. These were:

I. To compute for each temperature the mean daily relative growth rate for that day by finding the fraction which the absolute mean increment for the day was of the growth already attained at the beginning of the interval. Then their daily mean relative growth rates were averaged for the whole period from planting to the cessation of growth. This procedure corresponds in principle to determining the mean instantaneous growth rate over the whole period, but is done on the basis of the gross finite differences of the observations, rather than upon differentials.

II. The same procedure as in I, except that only the period of observed, measured elongation of the hypocotyl is used, instead of the whole period from planting to the cessation of growth as in I.

III. The same procedure as was used in computing the data of Fig. 2, except that the rates were calculated on the basis of only the period of observed, measured elongation of the hypocotyl.

The results of all four methods of dealing with the data (the original method and I, II, and III) are shown graphically in Fig. 3, the scales of plotting being so adjusted as to bring all the lines near together, since the interest in the case pertains to relative rather than absolute values.

It is evident from Fig. 3 that such differences in theories of the growth process, and views of an appropriate measure of the growth rate as are embodied in it make no essential or important difference in the net result regarding the relation to temperature of growth in *Cucumis melo* seedlings under the conditions of these experiments.

#### IV

The data of Table I and Fig. 1 indicate that temperature affects growth differently in different parts of the whole growth period or cycle. The nature of these changes is clearly shown in Table II, in which two sets of percentages are presented. The first of these gives the approximate percentage of the final mean total yield at each temperature which the plants in that temperature series have achieved at the end of the first quarter, the first half, and the first three-quarters of their own total growth periods. If the growth proceeded uniformly with time, and was the same relatively in all temperature series, the figures in the left half of Table II would be all 25 on the first line, 50

on the second line, and 75 on the third. The second set of figures, in the right half of Table II, gives the approximate percentages which the achieved yield in each temperature series was of the achieved yield in the 30° series (the optimum temperature in respect of total yield) in one-quarter, one-half, and three-quarters of the total growth period of each series. The figures show, for example, how the yield of the 20° series compared with that of the 30°, when each had accomplished one-fourth of its total growth period. The percentages are approximate in Table II because, in interpolating times and yields, we have assumed that growth proceeded at a constant rate between any two recorded observations. This is not strictly true, especially in some parts of the cycle, but the error implicit in this assumption is negligible

TABLE II  
*Approximate Relative Yields (Mean Hypocotyl Lengths) at Stated Relative Times, and in Proportion to 30° Yields at the Same Relative Times*

Percentages of total growth period	Percentages of own total yield						Percentages of own yield to 30° yield at same relative times					
	15°	20°	25°	30°	35°	37.5°	15°	20°	25°	30°	35°	37.5°
25	0	2.8	10.4	9.8	21.3	13.1	0	21.0	97.3	100	172.5	83.9
50	8.4	51.9	71.2	68.3	81.7	68.1	3.7	55.5	95.8	100	95.7	63.1
75	73.1	95.6	97.0	97.2	98.3	96.1	22.6	71.8	91.7	100	80.8	62.5
100	100	100	100	100	100	100	30.1	72.9	91.9	100	79.9	63.2

for present purposes. In all of the computations the total length of the growth period is taken as from planting to the cessation of growth. That is to say, germination is counted, in respect of time, as a part of growth.

From Table II the following points may be noted.

1. At no temperature did the seedlings in these experiments attain 25 per cent of their final total yields in the first quarter of their growth period. They came nearest to it in the highest temperatures, above the optimal.

2. When a half of the total growing period has been completed the seedlings in all the series except the 15° have achieved more than 50 per cent of their final total yields. The same thing is true, *mutatis mutandis*, for the first three quarters of the total growth period, except

that there the 15° series with 73.1 per cent of its total yield achieved is not significantly below the even 75 per cent.

3. In general it is plain that in these experiments at all temperatures above 20° the greatest growth activity was concentrated in the second quarter of the total growth period. At 20° the same thing was true, taking the figures literally at their face value, but the difference between the growth activities in the second and the third quarters at this temperature was so small as to be practically insignificant. At 15° the greatest growing activity was in the third quarter of the cycle. Also at this temperature the activity was much greater in the fourth quarter than at any other of the observed temperatures. In short, as the temperature increased in these experiments the time of greatest growing activity tended to be pushed farther and farther back towards the beginning of the cycle.

4. The same rule is manifested in another way in the second half of Table II. At the end of the first quarter of their respective growth cycles the 35° seedlings, for example, had an average yield nearly twice as great as the 30° seedlings, though in the end their yield was only 79.9 per cent of the 30° lot.

5. In these experiments temperatures far below the optimum (15° and 20°) affected growth activity more adversely in the first quarter of the cycle than in any other period of the cycle. Temperatures above the optimum (35° and 37.5°) affected growth activity more adversely in the third and fourth quarters of the cycle than in any other part of the cycle.

6. At all temperatures tested and in all parts of the growth cycle (save only the first quarter at 35°) growth as measured by yield was less than in the corresponding part of the cycle at 30° in these experiments.

7. It is possible that the short duration of the relatively high growth rates observed at 37.5° and 35° may be due to a rapid oxidation of the available food materials by the high rates of respiration which one may presume these temperatures induce, and which renders the food supply inadequate for further growth fairly early in the growth cycle. The present data, however, furnish no material by which this possibility may be crucially tested, and it is therefore idle to discuss it further.

## V

The results of this study suggest that anything approaching a thorough understanding of the phenomenon of growth is not likely to be reached until we have comprehensive and exact observations available regarding the cellular activities going on during the process. In so comparatively simple a biological structure as a seedling (simple, that is to say as compared with a human fetus) growth is far from a homogeneous phenomenon. Not all parts of the growing organism are growing at the same time, or at the same rate. While much progress has been made in recent years in getting an understanding of the gross quantitative relations of growth, we still know next to nothing of the real underlying biology of the process, the cellular activities involved, etc.

*Summarizing* the results it appears that seedlings of *Cucumis melo*, grown under carefully controlled experimental conditions at seven constant temperatures, show the optimal temperature to be approximately 30°C. (29.74° from the graduated data). The mean time rate of growth (millimeter increase in length of hypocotyl *per diem* over the whole growth period) is a parabolic function of temperature. As the temperature increases, within the limits of these experiments, the period of maximum growth activity tends to fall earlier in the whole growth cycle.

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