THE EFFECT OF X-RAY RADIATION ON THE TRANSPIRATION OF KITCHEN ONION (ALLIUM CEPA L.) IN THE COURSE OF ONTOGENESIS

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The problems of the water regime of plants are rarely dealt with in the radiobiological literature and the data available are often contradicting (among others Biebl 1956, Mathur and Lewis 1961, Herčík 1963, Nazirov 1963). The situation is even worse in the literature devoted especially to questions of radiation influence on transpiration. Biebl and Hölzl (1958) did not find any marked differences between the transpiration of soya leaves irradiated by X-rays and the control leaves. Also Höhn and al. (1958) studying guttation of oat seedlings irradiated by a dosis of 12,000 r of X-rays, did not find any differences as compared with the non-irradiated controls. Bowen and Cawse (1960), however, reported a marked temporary effect of high doses of gamma-rays (12,500-100,000 r) on the course of transpiration in tomato leaves. The same authors compared the intensity of transpiration in irradiated plants in the light and in the dark. In the latter case, evaluation was impossible owing to very small values. Dependence on radiation dosis was earlier observed by Härtel and Vukovits (1952). These authors established an increased cuticular transpiration in young plants of Phaseolus, Vicia faba, maize, oat and others with increasing doses of UV-radiation. A decrease in transpiration in Vicia faba plants irradiated by a dosis of 10,000 r of gamma-rays, as compared with the control, was found by Karmanov and Savin (1963). It is explained by an accelerated ageing of leaves and by general decrease in the course of all the vital reactions in irradiated plants.

The stated reports, which are often contradicting, show a lot of problems on the water regime of irradiated plants, especially when the literare data for comparison of experiments carried out in the course of the whole ontogenesis are lacking. Not a report on the changes of water regime of *Allium cepa* L. after irradiation has been found as yet. That is why we began to study these problems. The first results are presented in this communication.

Material and Methods

Healthy onions of Allium cepa L. seedlings, Vsetaty variety, delivered by Sempra, national enterprise Prague, were irradiated by X-rays on April 9, 1963 (Supersanax, 180 kV, 15 mA, without filter, flow 180.3 r/min) in doses of 100 r. 500 r and 1,000 r. Non-irradiated seedlings were used as the control. Planting was carried out on April 13 into the clayey -sandy soil (depth 5 cm) on the land of the Nine-Year School at Svobodné Dvorv near Hradec Králové. In the autumn of 1962 the soil was dunged and then ploughing was carried out. Powders according to the norm were used in spring. Agrotechnical measures in the course of vegetation were also in agreement with the current demands (Podešva et al., 2nd volume, 1959). Beds were 4 m long and 0.9 m wide (152 onions were planted on the area of 3.64 m²). The distance between the rows was 25 cm and there was 10 cm space between individual seedlings. The control onions were grown on four beds, irradiated (for each dosis separately) on five beds. Thus the initial number was 608 non-irradiated onions and 760 onions in each radiation dosis.

Intensity of transpiration (IT) was studied by means of Ivanov's gravimetric method, modified and often used by Penka and al. (Penka 1953 a, b, c, d, 1958 a, b, c, 1963, 1964, Penka, Hloušková and \overline{R} ezáč 1955, Penka, Kocábová, Srb and Šebová 1962, Cetl and Penka 1958). On the day of measurement (eight times altogether in the course of vegetation: 7.5., 21.5., 28.5., 4.6., 11.6., 26.6., 4.7., 18.7. 1963) IT was established at 9, 11, 13, 15 and 17 o'clock; in the last two measurements at 8, 10, 12, 14, 16 and 18 o'clock, so that the daily rhythm of IT could be found, too. All the measurements were carried out in two repetitions. The average whole day values of IT made for each measurement were then calculated mathematically on the basis of individual data.

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Results

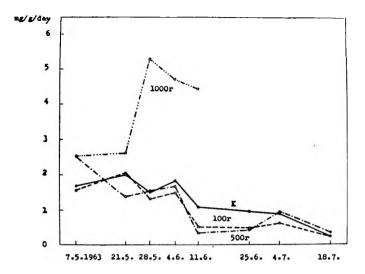
Some meteorological data and their course during the 1963 vegetation period are given in Table I. The average vegetation temperature was 16.6° C, being by 1.6° C higher as compared with fifty years' average. In June and July, when the growth of onions is markedly dependent on warm weather, the average month's temperature was by about 2° C higher. Similarly was the case of water precipitations. In the first ontogenetic phases (shooting and rapid growth) there was a sufficient amount of easily accessible moisture (in May the average precipitations were 91.9 mm and in June 116.4 mm), as compared with the fifty years' average (58 mm and 65 mm). It formed a sufficient reserve for further phases of development, when the surplus of water was undesirable. The average of July reached 33.7 mm only — a value almost 45 mm lower than for the fifty years' average. In August the precipitations were on the level of 1901—1950 years, in September by about one half higher (our experiment, however, ended as early as on 6th September). Summarizing, the main meteorological conditions of the year 1963 were favourable for the growth and development of the kitchen onion.

Changes in the daily averages of IT in the course of 1963 vegetation are given in graph 1. An almost identical course of IT is apparent in the initial phases of ontogenesis for the control and for the plants irradiated by a dosis of 100 r: from the phase of shooting (May 7th) to the beginning of the phase of rapid growth (May 21st) a small increase, a slight fluctuation in the course of this phase, changed by a slight decrease lasting till the last measurement can be observed (July 18th). From May 28th, however, the data of IT for the control and the dosis of 100 r are differentiated. Till June 25th, IT in plants irradiated by 100 r approached towards the data for the plants irradiated by a dosis of 500 r. In plants irradiated by a dosis of 500 r IT increased above the control (July 4th) and fluctuated here till the last measurement (July 18th). The course of IT in plants irradiated by a dosis of 1,000 r was a specific one: during the measurements it was many times higher (especially from May 28th) as compared with IT of the control plants and those irradiated by lower doses.

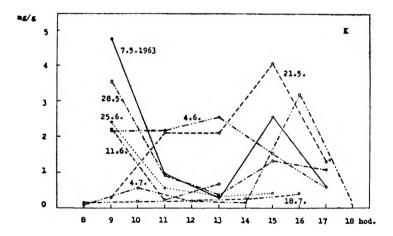
The dynamics of the daily course of IT of the non-irradiated control plants of kitchen onion in individual measurements during 1963 vegetation are given in Graph 2. As a rule, IT was the highest in the morning hours, in the course of the forenoon it decreased till the midday minimum, increasing slightly in the afternoon and decreasing again to the evening. These one-top curves with the midday minimum were recorded mainly in the initial phases of growth and development (on May 7th and 28th). In the course of phase of rapid growth, a one-top curve with the midday maximum was, however, also observed (on June 4th) or even with the afternoon maximum (on May 21). In the measurements of July 7th and 18th carried out in the course of phase of onion growth, the established values of IT were substantially lower than in the phase of rapid growth. Also fluctuations during the day were much milder.

Graph 3 shows IT values in plants irradiated by a dosis of 100 r. Practically all possible curves of daily transpiration dynamics can be traced in it: one-top curves with the midday minimum (May 7th), one-top curves with the forenoon (July 4th) or afternoon (May 28th, July 18th) maximum, the two-top curves with the forenoon and afternoon maximum (June 4th).

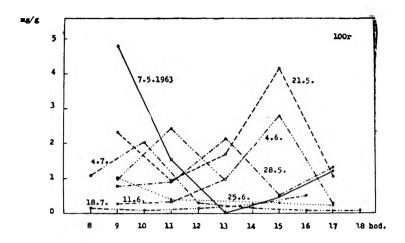
Similar, but also different courses of IT were observed in plants irradiated by a dosis of 500 r (Graph 4) and 1,000 r (Graph 5). In plants irradiated by a dosis of 500 r the course of transpiration differed mainly in the forenoon hours. IT increased at the beginning (practically in all the measurements during ontogenesis), reached its maximum at about 11 o'clock and decreased to minimum at about 13 o'clock. The afternoon course was irregular. In the last case (see Graph 5 - 1,000 r) the values were anarchic ones and were much higher than the data of IT in plants irradiated by the previous doses.



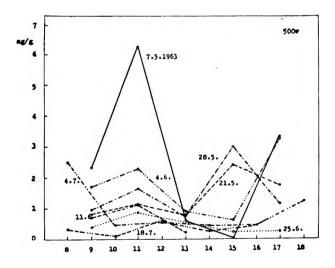
Graph. 1. Daily averages of transpiration intensity of above-ground parts of Allium cepa and their changes in the course of 1963 vegetation due to X-irradiation. Abscissa: days of measurements, ordinate: loss of water in mg per gramm of weight of fresh mass per day.



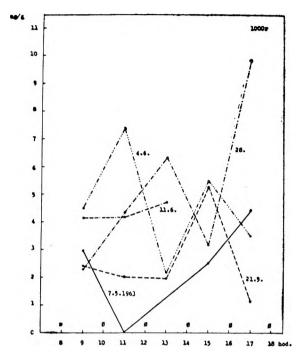
 Graph. 2. Daily IT changes in non-irradiated above-ground part of Allium cepa in individual measurements in the course of 1963 vegetation. Abscissa: hours of measurements during the day, ordinate: loss of water in mg per 1 gramm of weight of fresh mass in 1 min.



Graph. 3. Dailly 1T changes in above-ground part of Allium cepa irradiated by a dosis of 100 r of X-rays, in individual measurements in the course of 1963 vegetation. Abscissa: hours of measurements during they day, ordinate: loss of water in mg per gramm of weight of fresh mass in 1 min.



 Graph. 4. Daily IT changes in above-ground part of Allium cepa irradiated by a dosis of 500 r, in individual measurements in the course of 1963 vegetation. Abscissa: hours of measurements during the day, ordinate: loss of water in mg per gramm of fresh mass in 1 min.



Graph 5. Daily IT changes in above-ground part of Allium cepa irradiated by a dosis of 1000 r, in individual measurements in the course of 1963 vegetation. Abscisa: hours of measurements during the day, ordinate: loss of water in mg per gramm of weight of fresh mass in 1 min.

Discussion

Summarizing, the doses of ionizing X-ray radiation used (100 r, 500 r, 1,000 r) influenced the course of transpiration both in the course of vegetation and in the daily dynamics of IT. The degree of individual radiation doses asserted itself. Curves representing IT of the part of the kitchen onion above the earth had on the whole decreasing character from the beginning to the end of vegetation period (except for the data of plants irradiated by a dosis of 1,000 r). This is in agreement with the data of Felklová (1959) for Allium cepa, Benšová (1955, 1960) for Lactuca sativa, Penka, Kocábová, Srb and Šebová (1962) for Levisticum officinale who also observed a general decrease in transpiration in the course of vegetation.

The evaluation of the transpiration curves during ontogenesis of Allium cepa as regards the doses used gave a marked deviation for a dosis of 1,000 r (see Graph 1) in substantially higher values of IT. Plants irradiated by a dosis of 1,000 r grew and developed badly so that they died in the course of vegetation (S r b 1964a). A strongly inhi-

Table 1. Days of measurements, phases of growth and development and some meteorological characteristic in the course 1963 vegetation of Allium cepa L. — Vsetaty variety.

Month (1963) day of measurement		April	May			June				July			August	Spe- tember
		13.4.	7.5.	21.5.	28.5.	4.6.	11.6.	18.6.	25.6.	4.7.	11.7.	18.7.		6.9.
phase of growth and development		planting	shoo- ting	1		owth of above- round part				groving of onion				.harwest
average tempera- ture (°C)	daily	7,0	13,3	12,5		18,4	17,8	-	20,0	20,0	20,1	23,0	_	15,3
	monthly	9,5	13,3			17,4				19,8			18,1	15,3
	vegetation period 1963	15.6												
	1901—1950	7,4	12,8			15,0				17,4			16,8	13,5
relative moisture of the air $\binom{0}{10}$	daily	79	68	63	88	43	86	-	75	64	-	70	72	83
	monthly	65	77			68				66			70	78
precipita- tions (mm)	daily	0	0	0	0,1	0	26,1		7,0	0	-	9,0	2,3	3,2
	monthly	15,8		91,9			116,4			33,7			72,8	94,6
	1901—1950	43,0	58,0			65,0				78,0			70,0	48,0
average sunshine (hours)	daily	_	5,1	12,5	5,6	14,3	3.7		8.7	2,1	-	9,4	6,7	5,9

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bitory or even lethal dosis of 1,000 r affected the physico-chemical properties of the cells, especially the viscosity of the protoplasm and permeability. The cells were unable to keep water (this process is didirectly dependent on the protoplasmic visosity — S i m on o v a 1954), the content of free water increased in them. The plant transpirated the water in a greater amount. On the other hand, however, this decrease in the water content in the protoplasm meant a decrease in the physiological activity. The loss of turgescence induced a slowering up and then total stopping of growth — the onion died. This explanation is in agreement with K ramer (1959) and with the fact that IT depends greatly on the content of water in the plant (P e n k a 1958 c, F e l k l o v å 1959 and others). Many authors report Cell permeability to be also a very important part of metabolism, ensuring mutual connection of the organism and the media (H o a g l a n d and Broyer 1942, K ramer 1945, K o n a r e v 1948, S r b 1964 b).

For the other doses, 100 r and 500 r such great differences from the control curve were not apparent. Only some of the plants irradiated by a dosis of 500 r transpirated more than the control plants. At this phenomenon was not met with in plants irradiated by a dosis of 100 r, rather on the contrary, it is suggestive of a possibility of existence of a direct relation of IT to the degree of radiation dosis applied.

It remains, however, incomprehensible why high doses of radiation, exceeding many times lethal doses, are used by some authors (see authors quoted in the introduction). We suppose than the values obtained in plants irradiated by such high doses ($12\ 000\-100\ 000\ r$) can not be used for comparisson at all! All our experiments — and not only those of ours (Shull and Mitchel 1933, Sax 1955, Herčík 1959) show that the plant, too, is a very sensitive organism reacting even on the so called small doses ($100\ r$ and lower). Vasiljev (1962) is right saying that the present ideas on radiosensitivity of plants require critical revision. A revision, however, not always in his conception because he claims, e. g., that it is only growth which is radiosensitive while all the other physiological processes are resistent even to the doses of about 100,000 r.

Various processes are known in biology, taking their course in certain rhythms. One of them is also a twelve-hours' rhythm of transpiration dynamics: plants transpirate during the day, IT is, however, negligible at night (among others Maximov 1952, Bowen and Cawse 1960). Morover, the rhythm mentioned consists of partical rhythms which are subordinated and take their course in far from ideal way. As early as more then 50 years ago, the fluctuation of IT in individual hours during the day was described, and this problem has been solved till the present time (Livingston 1906, Briggs and Shantz 1916, Loftfield 1921, Vasiljev 1927, Berger-Landefeldt 1949, Maximov 1952, Felklová 1958, Benšová 1955, 1960, Benko 1963 a, b, Penka 1964 etc.). Two types of daily LT curves have been described so far: one-top curves with the midday maximum and curves with the midday minimum (Schratz 1931, Penka 1958 b, Penka, Hloušková and Řezač 1955, Penka, Kocábová, Srb and Šebová 1962). Two-top curves have been also recorded (Maximčuk 1923, Maximov 1952) or those with another course of IT — the morning forenoon, afternoon or evening maximum or minimum (Žemčužnikov 1924, Felklová 1959).

A fluctuation of IT in the course of the day was observed also in our experiments for all measurements during vegetation. In the control plants (Graph 2), we mostly recorded curves with a tendency to the midday minimum, though shifted to the forenoon or afternoon hours. As far as a different course of daily IT dynamics was established, i.e. one-top curve with the afternoon maximum (May 21) and the midday maximum (June 4), their course was presumably influenced by meteorological factors (mainly by the amount of sunshine — IT is substentially influenced by the light — Maximov 1952), as it follows from Table 1.

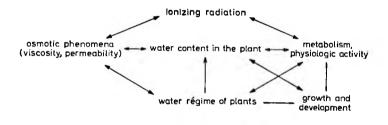
It changes in the course of a day are much variable (Felklová1959) complicated and cannot be expressed by a simple scheme (Penka, Kocábová, Srb and Šebová 1962). They depend on a lot of outer and inner factors (e. g. light, temperature, moisture, wind, soil water, plant species, intensity of metabolism, phase of growth and development, changes in permeability etc.).

During the day IT is strongly influenced especially by light, free water content in the plant and by the dynamics of the stomata (Livingston and Brown 1912, Iljin 1913, Maximov 1952, Loftfield 1921, Mina and Butovskij 1923, Vasiljev 1927, Stålfelt 1956, Petinov and Prusakova 1957, Penka 1958 a, c, 1963, 1964, Felklová 1959, Kramer 1959, Benko 1963, a, b). Penka, Hloušková and Rezáč (1955) even suppose that IT changes in plants in the course of a day and during individual development may be explained mainly by quantitative and qualitative changes in the water content of plants.

Morover, the complex course of IT is complicated by ionizing x-radiation both during the day and the entogenesis. Already in nonirradiated plants in the course of ontogenesis there are changes in metabolism, water regime of plants, resistance to cold and drought, changes in sugars and nitrogen substances, enzymatic activity, in physico-chemical properties of the protoplasm, need for mineral nutrition elements, state of chlorophyl in the leaves, saying nothing of a certain need for temperature, length of day and intensity of illumination (Gunar and Krastina 1952). The considerably varying radiosensitivity of organism in the course of ontogenesis has also a certain effect (Breslavec, Berezina, Ščibrja and Romančikova 1956, Biebl 1959 a, b, 1961, Barnetzky 1960, Šmálik, Drozd, Kubíková and Hončariv 1960, Šormová 1961). Transpiratory curves practically inexplicable then result — see Graphs 3, 4, 5 — in which all possible types of daily curves of IT are met with

Evaluating the gravimetric Ivanov's method of cut off above ground parts of plants used for the establishment of IT, we state that it proved to be good in our experiments. Also its evaluation by Cihova (1960) shows that the described different courses of IT in the irradiated plants are not caused by an unsuitability or inaccuracy of this method but by the radiation action.

On the basis of the present radiobiological knowledge we know that there exists a close, direct dependence between the intensity of metabolism and radiosensitivity, or the degree of radiation damage (Sisakjan 1955, Bilquez 1956, Kuzin 1962 a, b, Nuždin and Dozorceva 1962). It is also proved that this mutual close dependence and conditional character assert themselves between the metabolism and water activity of plants (Hoagland and Broyer 1942, Kramer 1945, Cetl 1957, Penka 1964). By describing the existence of a certain dependence also between the water regime of *Allium cepa* and the ionizing X-radiation we think we have closed the partial system of correlations "ionizing radiation — metabolism — water régime" of plants according to the scheme:



SUMMARY

Very little attention was paid to the study of water régime of plants irradiated by ionizing radiation. No literary data have been found for *Allium cepa*. The authors have established that the ionizing X-radiation (100 r, 500 r, 1,000 r) affects the course of transpiration of the part of kitchen onion above ground in the course of whole ontogenesis. They have also recorded changes in the daily dynamics of transpiration. The curves of transpiration intensity were on the whole od descending character from the beginning to the end of vegetation period in the control plants and in the plants grown from the onions irradiated by "lower" doses. The transpiration intensity was many times higher in the plants irradiated by a dosis of 1,000 r. In the control plants the curves characterizing the daily course of transpiration showed a tendency to one-top curves with the midday minimum. The ionizing X-radiation affected the daily transpiration dynamics to such a degree that we were unable to explain the curves obtained.

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SADRŽAJ

DJELOVANJE X-ZRAČENJA NA TRANSPIRACIJU CRVENOG LUKA (Allium cepa L.) U TOKU ONTOGENEZE

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Studiju vodenog režima biljaka ozračenih inonizirajućim zračenjem bilo je dosad posvećeno vrlo malo pažnje. Literaturni podaci za crveni luk nisu bili nađeni.

Autori su utvrdili da ionizirajuće zračenje (100 r, 500 r, 1000 r) utječe na transpiraciju nadzemnog dijela crvenog luka u čitavom toku ontogeneze. Oni su također zabilježili promjene u dnevnoj dinamici transpiracije. Krivulje intenziteta transpiracije, od početka do kraja vegetacijskog perioda, uglavnom su pokazivale pad, kako u kontrolnih biljaka tako i u biljaka izraslih iz lukovica koje su bile ozračene »nižim« dozama. U biljaka ozračenih dozom od 1000 r intenzitet transpiracije bio je, međutim, mnogo puta viši. Krivulje koje karakteriziraju dnevni tok transpiracije pokazivale su u kontrolnih biljaka tendenciju krivulja s jednim šiljkom s opodnevnim minimumom. Ionizirajuće X-zračenje utjecalo je, međutim, na dinamiku dnevne transpiracije u tolikoj mjeri da se dobivene krivulje nisu mogle protumačiti.