

# Assessment of metribuzin effects on potatoes using a method of very rapid fluorescence induction

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## ABSTRACT

Effects of increasing doses of the herbicidal preparation Sencor 70 WP (metribuzin a.i.) on selected potato varieties were studied in a field and pot experiments using very rapid fluorescence induction (vrFI). Tuber yield, and in pot experiments a tuber number and characteristics of the aboveground biomass were assessed. The curve of rapid induction goes through the O-J-I-P phases. The parameters  $F_o$ ,  $F_v/F_p$  and  $rF_j$  were measured. Based on changes in the vrFI parameters in comparison with controls, varieties were ranked according to sensitivity to metribuzin in field experiments. Keřkovské rohlíčky and Ukama were the most sensitive varieties and Impala the least sensitive variety. In pot experiments in the growth chamber, significant decrease in tuber weight vs controls was found in sensitive varieties Keřkovské rohlíčky (in 1998,  $0.75 \text{ kg} \cdot \text{ha}^{-1}$  Sencor 70 WP decreased tuber weight by 28% and  $1.5 \text{ kg} \cdot \text{ha}^{-1}$  by 89%) and Ukama (in 1997, at  $1.5 \text{ kg} \cdot \text{ha}^{-1}$  by 35%). Under stable conditions in the growth chamber, there were lower differences in the  $rF_j$  parameter in these varieties in comparison with controls than in resistant ones. Based on this finding it can be assumed that the sensitivity of varieties can be determined according to  $rF_j$  changes under stable ambient conditions.

**Keywords:** potatoes; metribuzin; chlorophyll fluorescence; tuber yield; aboveground biomass

Metribuzin is an active ingredient of the triazinone group, which is widely applied in weed management in potatoes. It is contained (70%) in the preparation Sencor 70 WP that is registered in the Czech Republic for preemergence and postemergence application. The primary mode of action is to inhibit photosynthetic electron transmission (Corbet 1994).

Some varieties of potatoes are injured by metribuzin, therefore if Sencor 70 WP is applied, the recommended dose and application method are necessary to consider or to omit the treatment (Daniel 1979, Vokál et al. 1985, and following more recent special publications). Determination of varietal sensitivity in potatoes is therefore very important. The sensitivity of potato varieties can be expressed by the change in photosynthesis intensity. Here, the measurement of chlorophyll fluorescence is suitable. If the photosynthetic process is inhibited, chlorophyll fluorescence rises. Van Orshot and Van Leeuwen (1988), Kuks et al. (1992) and others have described effects of herbicides on fluorescence parameters.

The objective of this paper is to evaluate relationships between chlorophyll fluorescence in selected potato varieties after postemergence application of increasing doses of Sencor 70 WP and characteristics of tubers and aboveground biomass.

## MATERIAL AND METHODS

Field experiments were conducted on the School Experimental Farm of Mendel University of Agriculture and Forestry (MUF) Brno at Žabčice and pot experiments in the growth chamber at MUF in 1997 and 1998.

In 1997, varieties Impala and Koruna designated as resistant to metribuzin and variety Ukama designated as sensitive were examined in both field and pot experiments. In 1998, Ukama and Keřkovské rohlíčky, in which sensitivity was confirmed by vrFI parameters (Dvořák and Remešová 1999), and Krystala reported as sensitive were studied in the pot experiment. All the varieties mentioned, i.e. Impala, Koruna, Ukama, Krystala and Keřkovské rohlíčky, were examined in the field experiment in 1998. Sencor 70 WP was used in postemergence application at doses of  $0.75$ ,  $1.5$  and  $2.25 \text{ kg} \cdot \text{ha}^{-1}$  in field experiments and at doses of  $0.75$  and  $1.5 \text{ kg} \cdot \text{ha}^{-1}$  in pot experiments. The plants of individual varieties were treated at growth stages from 25 to 31 (BBCH scale, stalk development, beginning of elongation growth).

Parameters of very rapid fluorescence induction (vrFI) were measured at time intervals after herbicide application using the analyser PEA (Plant Efficiency Analyser, Hansatech Ltd. King's Lynn, England).

The vrFI method is rather novel since the first information published Strasser and Govindjee (1991, 1992). This method has contributed to the previous techniques for chlorophyll fluorescence measurement because it allows detecting a new wave J. The used exciting light has to be of a high intensity at wavelength of max. 650 nm. The curve of rapid induction is enriched by one more wave, that means it goes through phases O-J-I-P (O = the initial point of measurement, J-I = waves in the curve course, P = peak of the curve). This differentiation is described by Matoušková et al. (1996) and Lazár (1999). Accurate measurements of the curve part with the J wave can correct some findings (for example, Chodová et al. 1995) that resistant species (biotypes) exhibit higher  $F_o$  values. It

is probably a higher J wave erroneously presented as  $F_o$ . A level of the assessed value of  $F_o$  changes the  $F_o/F_p$  ratio that characterizes photosynthesis yield (Srivastava et al. 1995).

The following parameters were measured in the presented paper:  $F_o$ , i.e. initial fluorescence,  $F_v/F_p$ , i.e. variable fluorescence ( $F_v = F_p - F_o$ ) divided by maximum fluorescence ( $F_p$ ),  $rF_j$ , i.e. relative height of the J wave [ $rF_j = (F_j - F_o)/F_v$ ,  $F_j$  = peak of the J wave].

The excitation intensity was  $3000 \mu\text{mol.m}^{-2}.\text{s}^{-1}$ , time of fluorescence reading 2 s, time of leaf adaptation to dark in Hansatech clamps was 30 min.

The increase in  $rF_j$ , i.e. at the first stage of herbicide effects on the leaf, is very sensitive. Therefore, the vrFI method can be considered more sensitive for examination of herbicidal effects than some techniques applied until now (Matoušková et al. 1999). Moreover, the advantage of this method is more accurate measurement of the parameters  $F_o$  and  $F_p$ , and thus more precise conclusions on changes induced by ambient factors, among others, herbicides. In comparison with a classical method of rapid induction (Nauš and Melis 1992), this method enables to extend a time scale of measurements.

### Establishment and conditions of the experiments

The experiment location Žabčice is 176 m above sea level. Soil substrate is formed by non-calcareous alluvial deposits. The topsoil humus horizon is to the depth of 35 cm; the content of size fractions to 0.01 mm is 66.3%. Variants were in four replicates in complete randomized blocks. Potatoes were planted in rows 75 cm apart, the area of one replicate was 10 m<sup>2</sup>. Planting dates were 9 April 1997 and 15 April 1998, herbicides were applied on 29 May 1997 and 28 May 1998, the harvest was carried out on 23 August 1997 and 25 August 1998. During the growing season of 1998, effects of Sencor 70 WP on aboveground organs were visually evaluated (according to the linear scale 0–100% joining all expressions of phytotoxicity in one datum) in the field experiment.

Pot experiments were carried out in Mitscherlich pots (a tuber of each variety per pot) in four replicates. Controlled conditions in the growth chamber were as follows: daylight in the 1<sup>st</sup> month of the experiment lasted from 5.00 to 19.30 of CET, in the 2<sup>nd</sup> month from 4.30 to 20.30. Air temperature during the daylight ranged from 15 to 20°C in the 1<sup>st</sup> to 2<sup>nd</sup> week and from 20 to 25°C in subsequent weeks, and 15°C in dark. Relative air humidity was 85/90% day/night. Potatoes were planted on 20 March 1997 and 21 April 1998, herbicides were applied on 22 April 1997 and 23 May 1998, tubers were harvested on 2 June 1997 and 19 June 1998. Individual plants were analyzed for tuber weight and number, numbers of stalks and leaves, weight of fresh leaves and stalks (1997, 1998) and dry weight (1998).

The vrFI parameters were measured on selected plants in the experimental field. Ambient conditions of measurements were influenced by the weather. In 1997, for in-

stance, daily sums of global radiation on the 1<sup>st</sup> day of measurement were 15.90 and on the 3<sup>rd</sup> day 20.56 MJ.m<sup>-2</sup>, in 1998, the sums ranged between 17.20 (4<sup>th</sup> day) and 28.80 MJ.m<sup>-2</sup> (6<sup>th</sup> day). In the growth chamber, the vrFI parameters were measured in all plants. Ambient conditions were in accordance with the constant chosen temperature and light regime. Daily sums of global radiation were around 20 MJ.m<sup>-2</sup>. The point of measurement (a top leaflet of the uppermost leaf) was designated on each examined plant when first measured. These points remained constant during the experiment.

### Evaluation of experiments

Since earlier studies showed that normal distribution was not valid for the vrFI parameters, our results are presented using medians. Ten measurements per variant were performed in field experiments and eight measurements in the growth chamber. Fluorescence intensity is expressed by deviations in parameters (in %) found in treated variants vs the untreated variant (Nauš and Melis 1992).

To assess significance of differences in examined fluorescence parameters; a two-sampling median test was used. Calculations were carried out using Unistat 4.53 software. Treated variants were compared with the untreated control. Significance of differences among values demonstrating characteristics of potato plants was assessed by analysis of variance with calculation of a minimum significant value of differences and confidence intervals. Relationships between characteristics of potato plants cultivated in the growth chamber and a level of vrFI parameters were assessed by correlation analysis. Correlation coefficients were calculated for all varieties on all dates of measurements. Statistical significance of the examined relationships was verified.

## RESULTS AND DISCUSSION

### Field experiments

The vrFI parameters were measured at 5 and 9 intervals in 1997 and 1998, respectively. In 1997, maximum differences in parameters  $F_o$  and  $F_v/F_p$  were found 72 h after application, in  $rF_j$  6 and 24 h. In 1998, maximum differences in  $F_o$  and  $F_v/F_p$  were assessed 48 and 72 h after application, maximum values of  $rF_j$  were found at different intervals, they ranged in dependence on varieties and application doses from 6 up to 168 h after application.

In some cases, lower differences in vrFI parameters were determined after application of 2.25 kg.ha<sup>-1</sup> than 1.5 kg. In 1997, that was found in resistant varieties Koruna (4 cases) and particularly in Impala (11 cases). In 1998, it was more frequent in the sensitive variety Keřkovské rohlíčky and resistant variety Koruna (six cases per each). This case appeared in Impala only once.

Table 1. Changes in medians of vrFI parameters as compared with the control (%), field trial at Žabčice, 1997

Variety	Parameter	Sencor 70 WP (kg.ha <sup>-1</sup> )	Hours after application					Mean	Final ranking
			6	24	48	72	312		
Ukama	Fo (+)	0.75	47	51	50	62	-6	40.8	2
		1.50	62	66	67	70	-8	51.4	
		2.25	83	80	80	85	-4	64.8	
	Fv/Fp (-)	0.75	23	24	26	28	+1	20.0	
		1.50	27	25	26	28	+1	21.0	
		2.25	38	39	39	42	0	31.6	
	rFj (+)	0.75	65	61	64	65	-4	50.2	
		1.50	78	72	69	72	2	58.6	
		2.25	96	86	86	86	0	70.8	
Impala	Fo (+)	0.75	-2	45	54	60	36	38.6	3
		1.50	0	80	84	86	72	64.4	
		2.25	-2	55	65	72	54	48.8	
	Fv/Fp (-)	0.75	0	19	22	23	18	16.4	
		1.50	0	35	36	38	31	28.0	
		2.25	1	30	35	39	27	26.4	
	rFj (+)	0.75	2	69	64	63	64	52.4	
		1.50	10	88	87	85	87	71.4	
		2.25	1	84	85	84	84	67.6	
Koruna	Fo (+)	0.75	66	86	92	89	-9	64.8	1
		1.50	82	98	112	102	-7	77.4	
		2.25	88	99	109	110	-3	80.6	
	Fv/Fp (-)	0.75	28	34	40	42	+1	28.6	
		1.50	32	37	41	45	1	31.2	
		2.25	32	37	40	41	0	30.0	
	rFj (+)	0.75	92	95	91	95	-5	73.6	
		1.50	100	106	98	101	1	81.2	
		2.25	109	108	99	100	1	83.4	

untreated = 100%; (+) increased parameter; (-) decreased parameter

Based on the data in Tables 1 and 2, the varieties were ranked according to their sensitivity to metribuzin (Tables 1 and 2). Final ranking that was assessed on the basis of ranking at particular intervals for all parameters and application doses was as follows: in 1997, Koruna (most sensitive), Ukama, Impala (least sensitive), in 1998, Keřkovské rohlíčky (most sensitive), Ukama, Koruna, Krystala, Impala (least sensitive). The final ranking of the variety (except for Koruna and Keřkovské rohlíčky at 0.75 kg.ha<sup>-1</sup>) was identical with their ranking at individual application doses.

The percentage of plant injury assessed visually is given in Table 3. The most damaged variety was Keřkovské rohlíčky. On 3 June 1998 (i.e. 5 d after application), the percentage of injury in dependence of the application dose ranged from 18 to 49.5%, and on 19 June 1998 (i.e. 16 d after application) from 1 to 10%. Injury of the other varieties was less important. The injury was characteristic of leaf yellowing, drying (from edges), and retarded growth.

The stand of potatoes was strongly damaged by floods in 1997. This year, incomplete data on tuber yields were obtained which are for orientation only. In the variety Ukama, for instance, the yield at 0.75 kg.ha<sup>-1</sup> decreased

by 17%, at 1.5 kg.ha<sup>-1</sup> by 33% whilst in Krystala and Impala (which was the most resistant variety against metribuzin based on evaluation of vrFI parameters) the yield was not reduced at 1.5 kg.ha<sup>-1</sup>. In 1998 (Table 4), low and insignificant yield loss was assessed in Impala at the dose of 2.25 kg.ha<sup>-1</sup> (7%). Higher insignificant yield decrease was assessed in the variety Ukama. The highest yield loss was in Keřkovské rohlíčky. A statistically significant yield decrease was assessed between variants treated with Sencor 70 WP and the untreated control (at 0.75 kg.ha<sup>-1</sup> by 25%, 1.5 kg.ha<sup>-1</sup> by 35%, 2.25 kg.ha<sup>-1</sup> by 28%). Differences among the yields obtained in treated variants were not significant.

### Pot experiments

The vrFI parameters were measured at 6 and 11 intervals in 1997 and 1998, respectively. Deviations in parameters (in %) in comparison with the control were usually higher after the application of 1.5 than 0.75 kg.ha<sup>-1</sup> Sencor 70 WP. In 1997, these were assessed in 48 cases of the total number 54 (3 varieties with 3 parameters each at

Table 2. Changes in medians of vrFI parameters as compared with the control (%), field trial at Žabčice, 1998

Variety	Parameter	Sencor 70 WP (kg.ha <sup>-1</sup> )	Hours after application									Mean	Final ranking
			6	24	48	72	120	168	216	264	312		
Ukama	Fo (+)	0.75	135	124	267	255	49	67	16	8	0	102.33	2
		1.50	118	96	269	253	91	103	34	24	-1	109.67	
		2.25	123	103	252	248	151	166	56	45	0	127.11	
	Fv/Fp (-)	0.75	37	32	69	67	9	14	5	3	0	26.22	
		1.50	33	32	65	68	22	30	11	8	+1	29.78	
		2.25	36	29	75	73	38	43	20	14	0	36.44	
	rFj (+)	0.75	139	123	96	68	101	101	41	41	8	79.78	
		1.50	153	130	67	62	159	154	82	88	10	100.56	
		2.25	149	125	85	66	277	166	94	92	13	118.56	
Impala	Fo (+)	0.75	135	132	219	223	11	26	4	4	0	83.77	5
		1.50	101	90	206	213	58	60	7	5	-2	82.00	
		2.25	132	137	221	225	95	87	19	18	-1	103.67	
	Fv/Fp (-)	0.75	37	40	54	55	3	7	4	2	+1	22.33	
		1.50	30	30	56	57	16	16	2	2	+1	23.11	
		2.25	41	42	56	59	24	23	7	6	0	28.67	
	rFj (+)	0.75	123	114	66	60	67	71	28	23	4	61.78	
		1.50	125	116	76	62	133	122	35	38	2	78.78	
		2.25	128	117	67	63	156	144	51	48	4	86.44	
Koruna	Fo (+)	0.75	173	167	254	258	21	36	17	9	-1	103.78	3
		1.50	141	114	249	256	87	117	34	24	-2	113.33	
		2.25	148	130	273	248	134	148	62	55	2	133.33	
	Fv/Fp (-)	0.75	42	42	62	60	6	8	5	4	0	25.44	
		1.50	41	32	68	68	19	26	9	7	0	30.00	
		2.25	43	36	65	59	30	35	18	17	0	33.66	
	rFj (+)	0.75	139	120	87	59	88	90	34	33	4	72.67	
		1.50	150	128	91	51	161	164	63	67	3	97.56	
		2.25	145	125	75	59	176	183	102	108	8	109.00	
Krystala	Fo (+)	0.75	80	97	225	229	28	38	3	0	-2	77.56	4
		1.50	87	94	262	281	70	89	27	18	-1	103.00	
		2.25	143	151	252	262	102	112	29	23	-1	119.22	
	Fv/Fp (-)	0.75	22	24	49	51	6	9	1	0	+1	17.88	
		1.50	23	23	72	64	15	20	9	5	+1	25.56	
		2.25	36	33	75	64	24	26	10	8	+1	30.56	
	rFj (+)	0.75	115	104	80	68	74	74	20	21	1	61.89	
		1.50	122	112	72	79	120	113	48	49	6	80.11	
		2.25	126	119	79	74	141	129	68	60	5	89.00	
Keřkovské rohličky	Fo (+)	0.75	101	86	267	256	77	79	26	20	-1	101.22	1
		1.50	119	97	262	244	121	102	29	16	-1	109.89	
		2.25	176	150	247	233	121	100	40	38	1	122.89	
	Fv/Fp (-)	0.75	29	23	77	73	20	19	8	7	0	28.44	
		1.50	41	31	81	77	49	44	10	8	0	37.89	
		2.25	42	44	80	77	48	45	22	17	0	41.67	
	rFj (+)	0.75	155	132	88	104	136	145	64	61	6	99.00	
		1.50	153	135	65	102	164	160	83	85	8	106.11	
		2.25	151	134	82	100	172	163	95	93	7	110.77	

6 intervals, Table 5) and in 1998, in 76 cases of 99 (3 varieties with 3 parameters each at 11 intervals).

In 1997, percentage deviations were highest in rFj parameters (for example, means in the variety Koruna at 0.75 kg.ha<sup>-1</sup> were Fo 71.17%, Fv/Fp -24.43% and rFj 84.67%,

Table 5). The lowest deviations (-) were found for the Fv/Fp parameter.

Similar data were obtained in the variety Krystala in 1998. In Keřkovské rohličky the highest difference was found in the Fo parameter and high values were also as-

Table 3. Injury of plants (%), mean of four assessments

Sencor 70 WP (kg.ha <sup>-1</sup> )	Date of evaluation	Variety				
		Ukama	Impala	Koruna	Krystala	Keřkovské rohlíčky
0.75	3. 6. 1998	1.0	1.8	1.3	1.5	18.0
	10. 6. 1998	0.5	2.0	0	1.5	7.3
	19. 6. 1998	0	0	0	0	1.0
1.50	3. 6. 1998	2.0	1.3	4.3	3.3	29.5
	10. 6. 1998	2.8	2.0	0.5	1.0	18.8
	19. 6. 1998	0	0	0	0	5.0
2.25	3. 6. 1998	12.8	4.5	2.0	1.8	49.5
	10. 6. 1998	4.5	6.0	2.5	1.5	23.8
	19. 6. 1998	0	0	0	0	10.0

sessed in the Fv/Fp parameter (Table 6). Differences in rFj between treated variants and the untreated control were markedly low. The average values were 21.82% (0.75 kg.ha<sup>-1</sup>) and 24.09% (1.5 kg.ha<sup>-1</sup>), whereas in Krystala these were 56.45 and 53.09%.

After application of Sencor 70 WP, medians of vrFI parameters (in relative units) mostly significantly differed from the control. A time slope of medians of vrFI parameters (original measured values) for the resistant variety Krystala and sensitive variety Keřkovské rohlíčky is illustrated in Figure 1. Slight differences (after application of 0.75 kg.ha<sup>-1</sup>) in Fv/Fp were assessed in Krystala (6 up to 72 and 264, 312 h after application), rFj in Keřkovské rohlíčky (24, 48, 408 and 504 h). Differences vs the controls were small in the mentioned cases.

Maximum values of Fo and Fv/Fp parameters were assessed at different intervals after application. In 1998, for instance, these were in the Fv/Fp parameter after 48 h in the variety Ukama and after 264 h in Keřkovské rohlíčky (1.5 kg.ha<sup>-1</sup>). Maximum values of rFj were obtained in both years in the studied varieties and at both application doses (except for Koruna at 0.75 kg.ha<sup>-1</sup> in 1997) at measurements till 48 h. The herbicide affected the examined parameters even 504 h (21 d, Table 6) after application, particularly rFj in Ukama and rFj and Fo in Keřkovské rohlíčky. The examples presented in Figure 1 illustrate differences during fluorescence in varieties

Krystala and Keřkovské rohlíčky. Marked differences are in values of the Fv/Fp parameter. While in the variety Krystala slightly lower values were assessed after application of Sencor 70 WP vs the untreated control (around 0.68 when measured 120 and 168 h after application, about 0.84 in the control), considerable reduction was determined in the variety Keřkovské rohlíčky (0.20 up to 0.10 in 120 up to 312 h after application). Furthermore, Figure 1 shows that value of Fo and Fv/Fp in Krystala changed very little 6 to 72 h after application of Sencor 70 WP, but the rFj parameter notably increased. The sensitive variety Keřkovské rohlíčky had markedly increased Fo, decreased Fv/Fp but less response of rFj during the first measurements.

In 1997, there were no significant differences in above-ground biomass (a number and weight of fresh stalks and leaves). More apparent effects were found in Impala when the dose of 1.5 kg.ha<sup>-1</sup> Sencor 70 WP decreased a number of stalks (by 27%) and leaves (by 22%). Differences in a tuber number and weight are illustrated in Figure 3. The sensitive variety Ukama after application of 1.5 kg.ha<sup>-1</sup> significantly decreased tuber weight by 58 and 38% as compared with the control and a dose of 0.75 kg.ha<sup>-1</sup>, respectively. The decrease in a tuber number at 1.5 kg.ha<sup>-1</sup> (by 38% vs the control) is close to significance limit. The insignificant decrease in tuber weight by 35% was assessed in Impala at 1.5 kg.ha<sup>-1</sup>.

Table 4. Tuber yield in field experiments, Žabčice (10 m<sup>2</sup>)

Variety	Sencor 70 WP (kg.ha <sup>-1</sup> )							
	0.00		0.75		1.50		2.25	
	kg	%	kg	%	kg	%	kg	%
Ukama	20.19	100	19.20	95	18.95	93	17.37	86
Impala	20.95	100	20.90	100	19.25	92	19.56	93
Koruna	21.18	100	20.10	95	20.45	96	18.43	87
Krystala	17.70	100	14.10	80	15.88	90	15.70	89
Keřkovské rohlíčky	10.23	100	7.68*	75	6.67**	65	7.27*	72

Keřkovské rohlíčky: d at  $P_{0.05} = 2.07$  kg\*,  $P_{0.01} = 3.14$  kg\*\*

Table 5. Changes in medians of vrFI parameters as compared with the control (%), growth chamber, MUAF in Brno, 1997

Variety	Parameter	Sencor 70 WP (kg.ha <sup>-1</sup> )	Hours after application						Mean
			6	24	48	72	144	240	
Ukama	Fo (+)	0.75	14	36	23	48	64	73	43.00
		1.50	15	39	28	49	60	137	54.67
	Fv/Fp (-)	0.75	4	17	15	20	23	18	16.17
		1.50	5	19	16	16	22	37	19.17
	rFj (+)	0.75	53	44	84	67	61	57	61.00
		1.50	73	47	89	68	62	70	68.16
Impala	Fo (+)	0.75	15	41	30	72	54	67	46.50
		1.50	42	62	38	91	139	131	83.83
	Fv/Fp (-)	0.75	4	12	12	18	10	12	11.33
		1.50	16	23	17	31	45	87	36.50
	rFj (+)	0.75	40	43	94	89	67	52	64.17
		1.50	124	60	102	98	76	56	86.00
Koruna	Fo (+)	0.75	10	53	40	92	109	123	71.17
		1.50	21	69	47	114	125	119	82.50
	Fv/Fp (-)	0.75	4	22	16	22	31	51	24.33
		1.50	10	23	22	36	56	76	37.17
	rFj (+)	0.75	69	88	104	104	88	55	84.67
		1.50	100	94	114	111	87	39	90.83

In 1998 (Figure 4), most characters of the variety Ukama showed lower values after application of Sencor 70 WP in comparison with the untreated control. Significant differences or differences at the limit of significance were assessed in dry weight of stalks (19% at 0.75 kg and 33% at 1.5 kg.ha<sup>-1</sup>) and leaves (30% at 1.5 kg.ha<sup>-1</sup>). The decrease in tuber weight at 1.5 kg.ha<sup>-1</sup> vs the control was 40% and was not statistically significant.

In the variety Krystala treated with 1.5 kg.ha<sup>-1</sup>, a significant decrease vs the control was assessed in a stalk number (43%), stalk dry weight (44%) and a leaf number (36%). Tuber weight was insignificantly lower by 13%.

After application of Sencor 70 WP in the variety Keřkovské rohlíčky, all characters decreased in comparison with the control. Differences in characteristics of above-ground biomass were not statistically significant. Differ-

Table 6. Changes in medians of vrFI parameters as compared with the control (%), growth chamber, MUAF in Brno, 1998

Variety	Parameter	Sencor 70 WP (kg.ha <sup>-1</sup> )	Hours after application											Mean
			6	24	48	72	120	168	216	264	312	408	504	
Ukama	Fo (+)	0.75	53	44	49	41	97	104	42	7	-3	5	4	40.27
		1.50	65	58	86	90	123	113	71	37	8	19	19	62.63
	Fv/Fp (-)	0.75	19	16	18	13	17	17	6	0	0	0	0	9.63
		1.50	21	20	24	23	21	16	10	4	1	1	2	13.00
	rFj (+)	0.75	54	79	84	72	46	56	48	2	0	5	9	41.36
		1.50	65	95	95	72	48	55	54	7	30	21	32	52.18
Krystala	Fo (+)	0.75	25	19	27	40	118	127	-17	12	9	14	12	35.09
		1.50	30	20	32	63	118	139	-18	21	11	-4	1	37.54
	Fv/Fp (-)	0.75	9	8	9	10	18	17	2	0	1	2	1	7.00
		1.50	8	8	9	17	18	19	7	2	1	+3	+1	8.45
	rFj (+)	0.75	103	111	60	75	51	82	39	38	26	19	17	56.45
		1.50	87	93	61	76	53	89	44	43	37	-2	3	53.09
Keřkovské rohlíčky	Fo (+)	0.75	48	39	43	62	72	71	-40	32	27	3	3	32.73
		1.50	56	52	75	68	111	122	-23	72	195	17	27	70.18
	Fv/Fp (-)	0.75	27	24	22	27	39	38	8	7	3	0	1	17.82
		1.50	27	26	37	48	76	75	89	91	76	6	11	51.09
	rFj (+)	0.75	41	40	27	12	28	31	15	9	34	-2	5	21.82
		1.50	42	42	29	9	35	34	8	-13	38	19	22	24.09

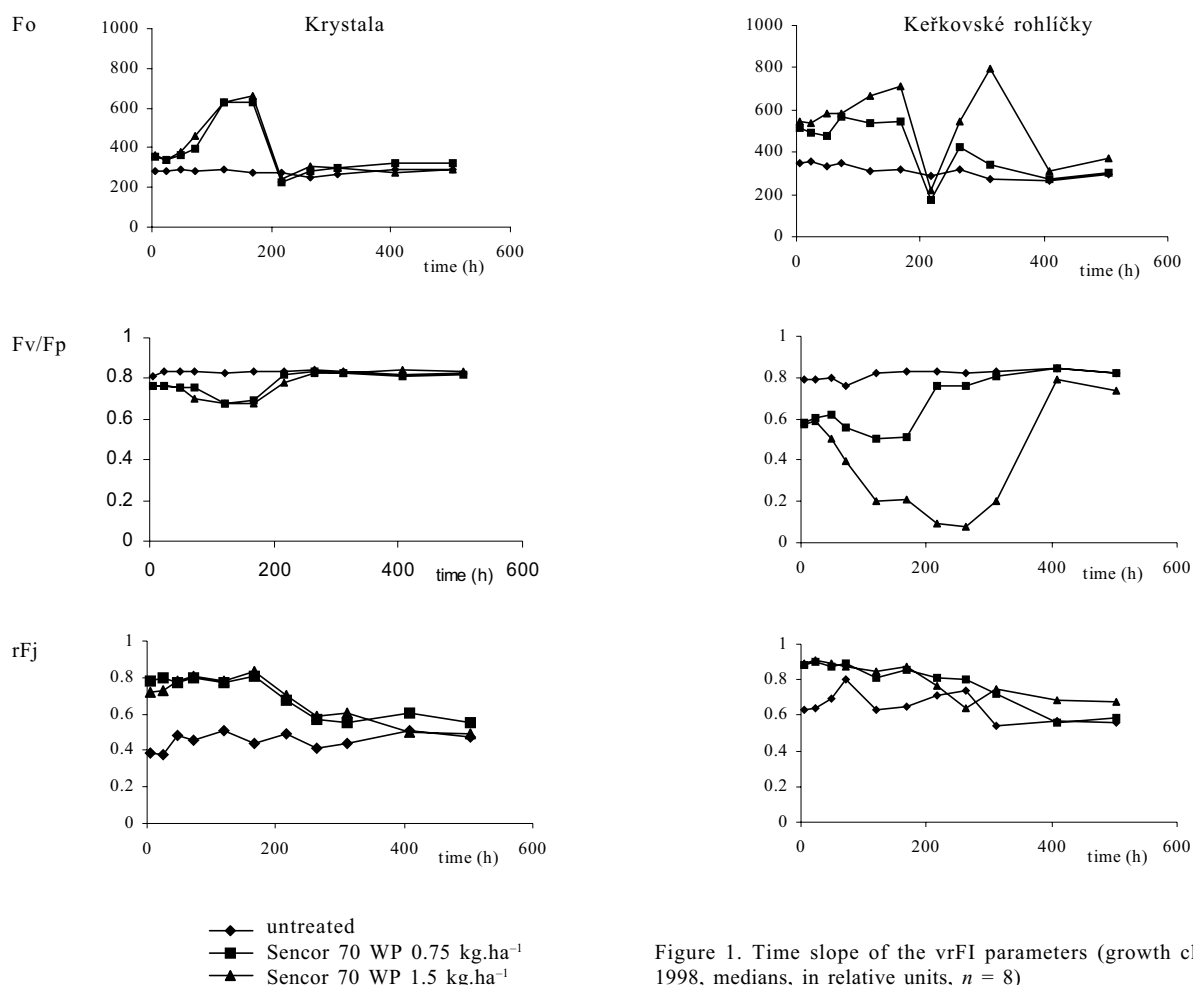


Figure 1. Time slope of the vrFI parameters (growth chamber 1998, medians, in relative units,  $n = 8$ )

ences in a tuber number were significant at 1.5 kg.ha<sup>-1</sup> by 71% (at 0.75 kg by 42% at a significance limit) and tuber weight was significantly lower at 1.5 kg.ha<sup>-1</sup> (by 89%).

Mostly negative correlations were found between Fo, rFj parameters and plant characteristics (the higher parameter, the lower value of plant characteristics). There were positive correlations between the Fv/Fp parameter and plant characteristics (the lower level of the parameter, the lower value of plant characteristics).

In 1997, the correlations were mostly weak (correlation coefficients  $r$  up to  $\pm 0.33$ ) to medium strong ( $r = \pm 0.33$  up to  $\pm 0.66$ , a scale according to Bravais). Statistically significant correlations ( $P = 0.05$ ) between the vrFI parameters and tuber number and weight were calculated for the sensitive variety Ukama. For instance, 48 h after application correlation coefficients between a tuber number and Fo were ( $r = -0.60$ ), Fv/Fp ( $r = 0.57$ ) and rFj ( $r = -0.60$ ), 240 h after application between tuber weight and Fo ( $r = -0.61$ ), Fv/Fp ( $r = 0.62$ ) and rFj 144 h after application ( $r = -0.57$ ).

In 1998, from 792 coefficients 60% showed weak correlation dependence (most frequently in the rFj parameter). There were 705 cases when the increase in Fo and rFj or decrease in Fv/Fp correlated with reduced values of plant characteristics. In 105 correlation coefficients, the examined relationship was at significance level of 95 and 99%.

There were such 4, 50 and 49 correlation coefficients in the rFj, Fo and Fv/Fp parameters, respectively. In the variety Keřkovské rohličky the correlations between Fo, Fv/Fp and numbers, weight of tubers mostly significant and highly significant (at measurements 6 and 504 h after application, correlation coefficients for a tuber number and Fo were  $-0.74$  and  $-0.58$ , Fv/Fp 0.76 and 0.68, correlation coefficients for tuber weight and Fo  $-0.81$  and  $-0.53$ , Fv/Fp 0.71 and 0.53). The course of the relationship between the vrFI parameters and tuber yield of the variety Keřkovské rohličky is illustrated in Figure 2.

This study presents results of chlorophyll fluorescence measurements together with tuber yield and characteristics of the aboveground biomass of potatoes. The vrFI parameters were measured on identical plants in the same stand that were harvested at the stage of technological maturity. Data on harvest were evaluated. There is a lack of such investigations until now. For instance, similar problems were studied in cereal crops (Flašarová et al. 1999) when a day prior to herbicide treatment, the plants were transplanted from the field experiment into pots filled with wet sand and placed in the not-heated greenhouse where fluorescence was measured. Yield characteristics were evaluated in plants growing in the field experiment. Hrabalová et al. (2000) cultivated the

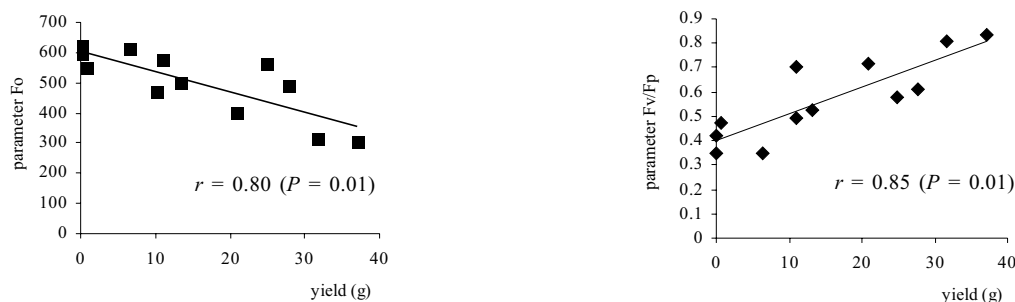


Figure 2. Relationship between tuber yield and values of vrFI parameters in the variety Keřkovské rohličky (growth chamber 1998, 72 h after application)

examined plants of winter wheat in the greenhouse for 25 days when their dry weight was assessed.

A variety of effects in the growth chamber and in the field could influence the values assessed in potatoes grown up to their harvest. There were differences between conditions in the growth chamber and in the field that apparently affected also values of the vrFI parameters.

Changes in all examined vrFI parameters in field and pot experiments after application of Sencor 70 WP mostly result in higher differences along with the increasing application dose in both sensitive and resistant varieties vs untreated controls. Based on average data in Tables 5 and 6 (growth chamber), these higher differences were not always in relation to varietal sensitivity to the active ingredient. The highest increases in the growth chamber in 1997 were assessed in the resistant variety Impala where the Fv/Fp parameter at 0.75 kg.ha<sup>-1</sup> vs the control differed by -11.33% and at 1.5 kg.ha<sup>-1</sup> by -36.50%, in 1998, the values in the sensitive variety Keřkovské rohličky were -17.82 and -51.09%, respectively. These increases were not as distinct in the field experiments. In Keřkovské rohličky in 1998 the difference increased from -28.44 to -37.89% (Table 2), etc. It can be concluded that a level of the application dose showed higher effects on changes in the vrFI parameters under stable conditions of the growth chamber than under variable conditions in the field.

The attention should be paid to the fact that was observed in both experimental years in the growth chamber. If Sencor 70 WP was applied, tuber weight significantly decreased vs untreated controls, but there was no significant decrease in characteristics of the above-ground biomass (Ukama 1997, Keřkovské rohličky 1998, Figures 3 and 4). By contrast, when significant decreases were in characteristics of the aboveground biomass, there was no significant reduction of tuber weight (Ukama, Krystala 1998, Figure 4). Under specific conditions, phytotoxic effects of metribuzin could express more on the above-ground or belowground biomass of potatoes.

The varieties were ranked according to their sensitivity to metribuzin in the field based on percentage changes in the vrFI parameters (1997, 1998). The presented ranking is in accordance with our earlier results (Dvořák and Remešová 1999). The fact that the variety Krystala can be considered resistant to metribuzin in relation to described changes in the vrFI parameters is contradictory to the published opinion that it is a sensitive variety.

Greater changes (in %) in the vrFI parameters indicate high inhibition of photosynthesis, i.e. higher phytotoxic effects of the herbicide. This relationship is apparent in the field in reduced tuber yields after application of Sencor 70 WP (Table 4). In 1997, the flood due to extreme rainfall damaged the experiment; therefore, the data are incomplete. Yield results of field experiments show that

Figure 3. Plant characteristics in the growth chamber, 1997 ( $n = 4$ ,  $P = 0.05$ )

Stalk number, pc

Stalk fresh weight, g

0 – untreated  
0.75 – Sencor 70 WP 0.75 kg.ha<sup>-1</sup>  
1.5 – Sencor 70 WP 1.5 kg.ha<sup>-1</sup>

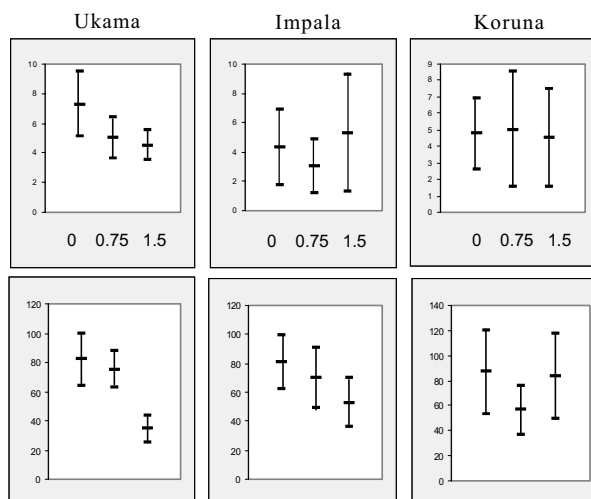
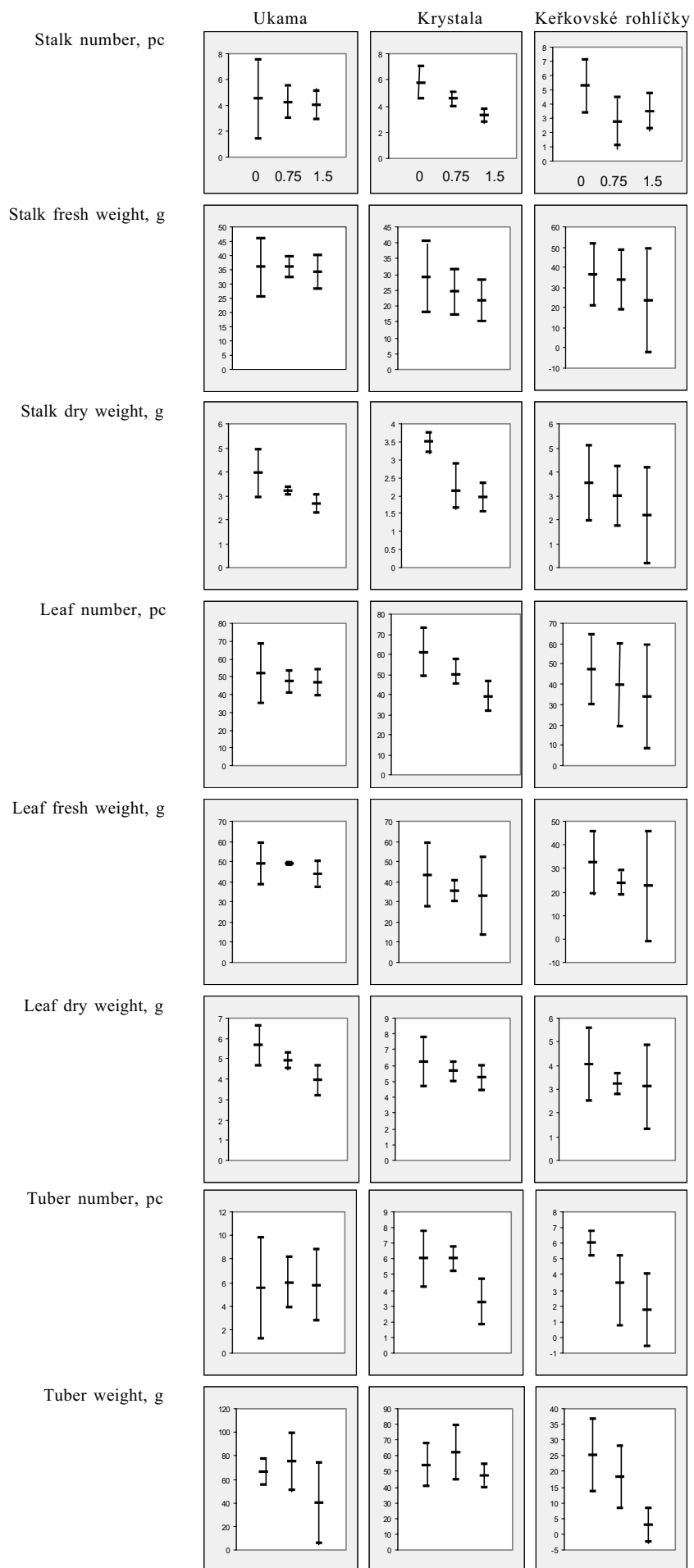




Figure 4. Plant characteristics in the growth chamber, 1998 ( $n = 4$ ,  $P = 0.05$ )



0 – untreated

0.75 – Sencor 70 WP 0.75 kg.ha<sup>-1</sup>

1.5 – Sencor 70 WP 1.5 kg.ha<sup>-1</sup>

phytotoxic effects of metribuzin were strongest on the variety Keřkovské rohlíčky. Here, the highest differences (in %) in the vrFI parameters (Table 2, for example, rFj on average according to application doses 99.00, 106.11 and 110.77%) and at the same time, the highest decrease in tuber yield (by 25, 35 and 28%) were assessed. The least differences in the vrFI parameters were in Impala (Table 2, average values of differences in rFj were at 0.75 kg.ha<sup>-1</sup> Sencor 70 WP 61.78%, at 1.5 kg 78.78% and at 2.25 kg 86.44%). Tuber yield decreased only slightly (0% at 0.75 kg, 8% at 1.5 kg and 7% at 2.25 kg.ha<sup>-1</sup>, Table 4). These values correspond with percentage of plant injury (Table 3).

The above-mentioned results suggest that if changes in the vrFI parameters do not exceed a certain limit, the decrease in tuber yield cannot be expected. In percentage change in the rFj parameter vs the control under variable field conditions, a safe situation can be considered if an average value assessed in the period from 6 to 312 h after application does not exceed 70%. These problems have been discussed earlier (Remešová and Dvořák 2000).

The comparison of changes in the vrFI parameters and effects on tuber yield in the growth chamber does not always show proportional relation. In 1997, the decrease in tuber weight was significant in the variety Ukama where changes in the vrFI parameters were lower than in varieties Impala and Koruna that did not reduce tuber weight significantly. It was apparent in rFj (on average, in Ukama 61.00 and 68.16%, in Impala 64.17 and 86.00%, in Koruna 84.67 and 90.83% at 0.75 and 1.5 kg.ha<sup>-1</sup>, respectively, Table 5).

The most injured variety in 1998 was Keřkovské rohlíčky. After the application of Sencor 70 WP, it markedly reduced tuber weight and in comparison with the control by 28% (at 0.75 kg.ha<sup>-1</sup>) and 89% (at 1.5 kg.ha<sup>-1</sup>). Most measurements on this variety showed higher differences in Fo parameters and namely in Fv/Fp. The mean of changes in the Fv/Fp parameter calculated from individual measurements was at 0.75 kg.ha<sup>-1</sup> -17.82% (-9.63% in Ukama, -7.0% in Krystala) and at 1.5 kg.ha<sup>-1</sup> -51.09% (-13% in Ukama, -8.45% in Krystala, Table 6). The rFj parameter, however, showed lower values than in the other varieties and it was observed at all intervals of measurements. The values were on average at 0.75 kg.ha<sup>-1</sup> 21.82% and at 1.5 kg.ha<sup>-1</sup> 24.09%. In the variety Ukama these were 41.36% and 52.18%, in Krystala 56.45% and 53.09% (Table 6).

The results obtained in the growth chamber suggest that differences in the rFj parameter vs the untreated control were the lowest in such varieties (Ukama and Keřkovské rohlíčky) that produced markedly decreased tuber weight, it means the highest phytotoxicity of metribuzin.

The above finding is interesting and indicates that if sensitivity of potato varieties to metribuzin is higher, lower changes in the rFj parameter can be expected in experiments conducted under stable conditions. This is also in accordance with partial results of laboratory experi-

ments in 1997. Stalks of 8 varieties were put in metribuzin solution ( $5 \cdot 10^{-3}$  and  $1 \cdot 10^{-2}$  mol.l<sup>-1</sup>) for 1 s. If the parameters vrFI were measured 18 and 24 h after contamination, the differences in rFj were lowest in the sensitive variety Ukama. It corresponds with results obtained by Strasser and Govindjee (1991) that the J wave is very sensitive to a number of factors, including herbicides. Srivastava et al. (1995) determined that the J wave considerably increased in 7 resistant mutants (D1 protein change) of unicellular alga *Chlamydomonas reinhardtii* vs a non-resistant biotype if photosynthesis inhibitor (DCMU) was added. It cannot be foreclosed that higher plants can behave similarly under specific conditions. Therefore, it can be assumed that the J wave will be higher in resistant varieties and lower in sensitive varieties.

The results obtained in the growth chamber in 1997 and 1998 correspond with the above-mentioned assumption. Sensitive varieties (Ukama and especially Keřkovské rohlíčky) exhibited lower values of changes in the rFj parameter in comparison with resistant varieties (Impala and Koruna). The data from rather stable conditions of the growth chamber differ from results gained under variable conditions in the field where rFj values in sensitive varieties increased. In spite of the mentioned difficulties, the investigated method of the vrFI parameters can be considered perspective for evaluation of varietal sensitivity to herbicides whose mode of action is inhibition of photosynthesis.

Significant correlations were found between values of plant characteristics and Fo and Fv/Fp parameters. These correlations were frequent at measurements up to 264 h, and in Ukama and Keřkovské rohlíčky (1998) even 408 and 504 h after application of Sencor 70 WP. Correlations in the rFj parameter were lower and insignificant except for some cases (Ukama in 1997).

The most important criterion for assessment of herbicide phytotoxicity is yield performance in exact field experiments. Due to an increasing number of varieties and newly registered herbicides, however, requirements in this respect are often difficult to meet. It can also happen that under specific conditions (extreme weather and others) results from field experiments are not reliable. Therefore, the possibility of testing the phytotoxicity using a non-invasive method for measuring the vrFI parameters is necessary.

This study was conducted as a part of the project CEZ: J08/98:432100001 supported by the Ministry of Education, Youth and Physical Training of the Czech Republic.

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Received on July 12, 2001

## ABSTRAKT

### Stanovení účinku metribuzinu na brambory metodou velmi rychlé fluorescenční indukce chlorofylu

V polních pokusech a ve fytotronu byl pomocí velmi rychlé fluorescenční indukce (vrFI) chlorofylu sledován vliv odstupňovaných dávek herbicidu Sencor 70 WP (ú. l. metribuzin, 70 %) na vybrané odrůdy brambor v letech 1997 a 1998. Křivka rychlého indukčního jevu probíhá ve fázích O-J-I-P (Strasser a Govindjee 1991, Lazár 1999). Pomocí fluorimetru PEA (Plant Efficiency Analyzer) byly měřeny parametry Fo, Fv/Fp a rFj. V pokusech byly použity odrůdy Impala, Koruna, Ukama, Krystala a Keřkovské rohlíčky. V polních pokusech byl Sencor 70 WP aplikován v dávkách 0,75; 1,50 a 2,25 kg.ha<sup>-1</sup>, ve fytotronu v dávkách 0,75 a 1,50 kg.ha<sup>-1</sup>. V polních pokusech bylo podle změn parametrů vrFI oproti kontrolám stanovení pořadí odrůd podle citlivosti na metribuzin. Nejcitlivější byly Keřkovské rohlíčky a Ukama, nejméně citlivá byla Impala (tab. 1 a 2). Uvedeným hodnotám odpovídají výnosy hlíz (tab. 4, průkazné difference byly u odrůdy Keřkovské rohlíčky). Podle hmotnosti hlíz na pokusech ve fytotronu byly metribuzinem nejvíce poškozeny citlivé odrůdy Keřkovské rohlíčky (1998) a Ukama (1997, 1998), kde bylo většinou zjištěno průkazné snížení (obr. 3 a 4). Byly nalezeny korelační závislosti parametrů vrFI a výnosů hlíz. V případech, kde byly oproti neošetřeným kontrolám zjištěny po aplikaci Sencoru 70 WP statisticky průkazné poklesy hmotností hlíz, nebyly nalezeny statisticky významné poklesy údajů charakterizujících nadzemní biomasu a naopak. Fytotoxické účinky se za určitých okolností mohou více projevit na nadzemní nebo podzemní biomase brambor. V konstantních podmínkách fytotronu byly na odrůdách s nejmenší hmotností hlíz menší difference parametru rFj ve srovnání s kontrolní variantou (tab. 5 a 6) než u odolných odrůd. Podle tohoto zjištění lze usuzovat na možnost stanovení citlivosti odrůd podle změny rFj při stálých vnějších podmínkách.

**Klíčová slova:** brambory; metribuzin; fluorescence chlorofylu; výnos hlíz; nadzemní biomasa

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