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Changes of micronutrients, dry weight and plant development in canola (*Brassica napus* L.) cultivars under salt stress

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This study was carried out to determine the effects of salt stress on the growth, dry weights and micronutrient contents of canola (*Brassica napus* L.) cultivars grown in greenhouse conditions. 12 canola cultivars (Marinca, Kosa, Spok, Semu DNK207 NA, Tower, Liraspa, Star, Tobin, Helios, Semu 209/81, Regent and Lirawell) were exposed to salinity treatments (150 mM NaCl and control). Shoot, leaf and root dry weights of all the cultivars at 45-day-old plants were determined. Micronutrient contents (Fe, Mn, Cu and Zn) of the leaves, stems and roots were also analyzed. Salinity stress negatively affected the canola cultivars and the extent of effects varied depending on the salt tolerance of the cultivars. Generally, salinity reduced the plant growth and dry weights. Fe, Mn, Cu and Zn concentrations were high in the roots when compared with those in the leaves and shoots in the salt applied samples. It was observed that, micronutrient contents showed some variation in the different plant parts of the canola cultivars as a result of salt applications to the growing media. Iron (Fe), manganese (Mn) and copper (Cu) content increased in all the plant parts with salt applications except for some cultivars. On the other hand, when mean data of the cultivars were considered, it could be said that zinc (Zn) content of the leaves was not significantly affected by the salt stress.

Key words: Canola, *Brassica napus*, dry weight, micronutrient accumulation, salt stress.

INTRODUCTION

The family Brassicaceae includes a number of species that have considerable nutritional and economic values and that have been under cultivation since 1500 B.C. These crops are extensively grown as cash crops, fodder and industrial/medicinal crops (Ashraf and McNeilly, 2004). The most common *Brassica* oilseed crops grown in the world for industrial purpose are rape-seeds, *Brassica campestris* and *Brassica napus*.

Soil salinity is one of the major problems of agriculture throughout the world. Due to this, large areas of arable lands are substantially or partially unproductive. There is evidence that irrigation systems and type of irrigation water have contributed to a large extent in converting arable lands to saline lands (Ashraf and McNeilly, 2004). Saline environments affect plant growth in different ways,

including a decrease in water uptake, an accumulation of ions to toxic levels and a reduction of nutrient availability (Ashraf, 1994). Salinity stress is often associated with nutritional imbalance. The interaction between salt stress and other environmental factors influence the plant's response to the stress (Ashraf and McNeilly, 2004).

Tolerance of oilseed *Brassicaceae* to salt stress is a complex trait, which is greatly modified by cultural, climatic and biological factors (Kumar, 1995; Minhas et al., 1990). The amphitetraploids *Brassica* species, including *B. napus*, *Brassica carinata* and *Brassica juncea* are more tolerant to salinity and alkalinity than their respective diploid progenitors such as *Brassica campestris*, *Brassica nigra* and *Brassica oleracea* (Kumar, 1995). Stage-specific tolerance components could then be incorporated individually or simultaneously into modern *Brassica* cultivars to develop genotypes that are tolerant at all stages of plant development. *Brassicaceae* exhibit susceptibility to salinity at seedling emergence and at early stages of growth (Puppala et al., 1999); therefore,

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these stages of growth can be useful for identifying Salinity tolerance of the genetic materials (Rameeh et al., 2004)

Salinity stress is a major environmental constraint to crop productivity in the arid and semiarid regions of the world. High concentrations of salts cause ion imbalance and hyper osmotic stress in plants. As a consequence of these primary effects, secondary stresses such as oxidative damage often occur. High salt stress disrupts homeostasis in water potential and ion distribution. This disruption of homeostasis occurs at both the cellular and the whole plant levels. Drastic changes in ion and water homeostasis lead to molecular damage, growth arrest and even death (Zhu, 2001).

Although, Brassica species produce maximum yield under normal soil and environmental conditions, their growth, seed yield and oil production are markedly reduce due to environmental stresses such as drought, water logging, salinity, low or high temperature, nutrient deficiency or excess, etc. In particular, for these crops, there is a great magnitude of interspecific variation for salinity tolerance (Ashraf and McNeilly, 1990). While assessing the comparative salt tolerance of some Brassica species at the early growth stages, *B. napus*, followed by *B. carinata* and *B. juncea*, were found to be more salt tolerant than *B. campestris* (Ashraf and McNeilly, 1990).

The most common adverse effect of salinity on Brassica crops is the reduction in plant height, size and yield, as well as deterioration of the quality of the product (Kumar, 1995). *B. napus* is the most salt tolerant, whereas *B. nigra* and *B. campestris* are the most salt-sensitive crops (Kumar, 1995). Different plant cultivars with particular genetic structures are supposed to have various reactions to environmental factors and salt stress. Other salt stress capturing capability of minerals by plants from the soil may also be different. Thus, in this study, we aimed to investigate the effects of salt stress on the growth, dry weights and micronutrient contents of different canola cultivars.

MATERIALS AND METHODS

The experiments were conducted in pots filled with soil in the greenhouse at the Horticulture Department of Agriculture Faculty of Yuzuncu Yil University Van (Turkey) during April to June 2007. The experiments were carried out using a completely randomized plots design containing non salinity and sodium chloride (NaCl) (0 and 150 Mmol) groups with three replications. 12 canola (*B. napus* L.) cultivars (Marinca, Kosa, Spok, Semu DNK207 NA, Tower, Liraspa, Star, Tobin, Helios, Semu 209/81, Regent and Lirawell) were used as the plant materials.

Ten seeds of each cultivar were sown directly in plastic pots containing 4 kg of loam soil, which was collected from an agricultural field and passed through a 2 mm mesh screen. The texture of the soil was based on sand-clay-silt, 1.96% total organic matter, 0.035% total salt, pH 7.30, 0.9% total nitrogen, 28 mg kg⁻¹ dry soil available phosphorus, and 180 mg kg⁻¹ dry soil exchangeable potassium. All the pots were fertilized with urea as a nitrogen

fertilizer equivalent to 150 kg ha⁻¹ and triple-superphosphate (80 kg P₂O₅ ha⁻¹) were incorporated into the soil before seeding. The daily air temperature ranged from 30°C (maximum at day) to 10°C (minimum at night), with the daily average temperature being about 25°C. Relative humidity fluctuated between 30 and 85%; the average value was about 60%.

For salinity treatments, non-salt-treated plants were kept as the control groups and salt-stressed plants were subjected to 150 mM NaCl 30 days after sowing and were maintained until final harvest. The pots were randomly arranged in a greenhouse. After sowing, soils were irrigated immediately and irrigation was carried out regularly at a day interval during the experiment (45 days). Plants were irrigated until saturated, with the excess solution allowed to drain into collection pans.

Thinning was carried out 15 days after planting leaving four plants from each pot, and 30 days later, salt-treatment started. After 45 days, the plants were lifted and samples were washed in deionized water to remove salts from the tissue surfaces; plant and root height (cm) were measured. After this, leaves, roots and shoots were separated. Their dry weights were determined after drying for 48 h at 75 to 80°C in a forced air oven.

For micronutrients determination, dry samples of roots, shoots and leaves were extracted in concentrated HNO₃ and HClO₄. Fe, Mn, Cu and Zn contents were determined by atomic absorption spectrometry (AAS) (Kacar, 1984). Data were analyzed by an analysis of variance using SAS (1985) software to test the significance of the main effects. Means were compared using LSD multiple range tests. Terms were considered as significant at the level of $p < 0.05$.

RESULTS AND DISCUSSION

The effects of salinity treatment on the plant height, root length, dry root weight, dry shoot weight and dry leaf weight of 12 canola cultivars are shown in Table 1. The results of the study showed that, the differences of plant height, root length, root dry weight, shoot dry weight and leaf dry weight in both salt treatment and cultivars were significant ($p < 0.01$). The salt stress caused significant reductions in all the growth variables including dry weights for all the canola cultivars.

Plant heights recorded 45 days later from planting were significantly affected by salt treatment (Table 1). Lirawell cultivar members were taller than the other cultivars in the control group. Salinity decreased the plant height of all the canola cultivars. Plant height of Semu DNK207 NA was more affected (25% reduction compared with controls) than those of the other cultivars. A comparison of the responses of the different cultivars indicated that root length was reduced significantly by salt stress except those of Liraspa and Semu 209/81. Tower cultivar was more affected (17% reduction compared with controls) than the other cultivars in terms of root length. Root dry weight was reduced by salinity in all the cultivars except for Liraspa. Shoot dry weight was reduced by salinity in all the cultivars; there was a less decrease in Semu DNK207 NA and Liraspa cultivars. Leaf dry weight decreased significantly by salt treatment in all the cultivars, but the reduction was less in Kosa cultivar. It seemed that, salinity affected (21% reduction when compared with control group) more significantly the leaf dry

Table 1. The effects of NaCl treatment on leaf, shoot and root dry weights, plant height and root length in canola cultivars.

Varieties	Plant height		Root length		Root dry weight		Shoot dry weight		Leaf dry weight	
	0	150	0	150	0	150	0	150	0	150
Marinca	22.2 ^d	19.7 ^{de}	17.3 ^{cd}	15.7 ^{cd}	0.41 ^c	0.34 ^c	0.55 ^c	0.46 ^{cd}	0.86 ^{cd}	0.73 ^{c-d}
Kosa	23.7 ^c	19.0 ^e	15.7 ^{de}	14.7 ^{de}	0.38 ^{cd}	0.33 ^c	0.48 ^e	0.42 ^{de}	0.79 ^{de}	0.75 ^{b-d}
Spok	20.0 ^e	16.2 ^f	15.0 ^e	14.5 ^e	0.31 ^{ef}	0.24 ^f	0.51 ^d	0.45 ^{cd}	0.73 ^{e-g}	0.61 ^e
Semu (1)	25.2 ^{bc}	19.0 ^e	15.8 ^{de}	15.3 ^{cd}	0.28 ^f	0.24 ^f	0.54 ^c	0.50 ^{bc}	0.86 ^{cd}	0.79 ^b
Tower	27.0 ^b	24.0 ^b	20.0 ^a	16.7 ^c	0.46 ^b	0.41 ^b	0.59 ^{bc}	0.53 ^b	0.89 ^c	0.73 ^{cd}
Liraspa	29.3 ^a	23.5 ^c	18.7 ^b	20.0 ^a	0.33 ^{d-f}	0.31 ^{cd}	0.53 ^{cd}	0.49 ^{bc}	0.72 ^{fg}	0.65 ^e
Star	23.0 ^d	20.0 ^{de}	18.5 ^b	17.8 ^b	0.41 ^c	0.29 ^{de}	0.50 ^d	0.44 ^d	0.97 ^b	0.77 ^{bc}
Tobin	28.3 ^{ab}	25.8 ^a	17.7 ^c	14.8 ^{de}	0.33 ^{d-f}	0.27 ^{ef}	0.50 ^d	0.44 ^d	0.67 ^g	0.59 ^e
Helios	19.0 ^f	17.8 ^{ef}	18.3 ^{bc}	15.7 ^{cd}	0.29 ^f	0.25 ^{ef}	0.49 ^{de}	0.40 ^e	0.75 ^{ef}	0.71 ^d
Semu (2)	21.7 ^{de}	20.3 ^{de}	17.5 ^d	17.3 ^{bc}	0.30 ^{ef}	0.25 ^{ef}	0.57 ^{bc}	0.48 ^c	0.89 ^c	0.79 ^b
Regent	23.8 ^c	21.5 ^d	15.8 ^{de}	14.8 ^{de}	0.35 ^{de}	0.24 ^f	0.61 ^b	0.49 ^{bc}	0.75 ^{ef}	0.64 ^e
Lirawell	29.7 ^a	25.0 ^{ab}	17.8 ^c	15.0 ^d	0.53 ^a	0.48 ^a	0.73 ^a	0.65 ^a	1.06 ^a	0.99 ^a
Means	24.4 ^a	20.9 ^b	17.1 ^a	15.9 ^b	0.36 ^a	0.30 ^b	0.55 ^a	0.48 ^b	0.83 ^a	0.73 ^b
LSD	1.52		1.58		0.029		0.036		0.042	

*Mean values indicated by the same letter were not significant different ($p < 0.05$); * Semu (1); Semu DNK207 NA; Semu (2); Semu 209/81.

weight of the Star cultivar than those of the other cultivars.

Salt treatment effects were different in the early growth stages of the plants. Salinity has both osmotic and specific ion effects on plant growth (Dionisio-Sese and Tobita, 2000). In the study, salt stress caused a significant decrease in the plant height, dry weights of root, shoot and leaf of cultivars (Table 1). Reduction in plant growth as a result of salt stress has also been reported in several other plant species (Ashraf and McNeilly, 1990; Mishra et al., 1991; Ashraf and O'leary, 1997; Turkmen et al., 2008). The uptakes of some mineral nutrients dissolved in water are also restricted in plants under salt stress. Thus, growth and development of plants are inhibited due to occurring defect in metabolism. Some investigators thought that because of ion accumulation by changing membrane permeability, metabolism was negatively influenced (Cramer et al., 1985; Grieve and Fujiyama, 1987). Most crop plants suffer after exposure to saline conditions and showed decline in growth. The deleterious effect of salinity was suggested as a result of water stress, ion toxicities, ion imbalance or combination of all these factors (Kurth et al., 1986).

The findings related to micronutrient contents of roots, shoots and leaves of different canola cultivars are shown in Table 2. As shown in Table 2, differences in the amounts of Fe, Mn, Cu and Zn were significant among all the cultivars. Significant differences were determined among the varieties in terms of Fe, Mn, Cu and Zn contents (Table 2). Fe, Cu and Zn concentrations were higher in the roots when compared with shoots and leaves in the salinized samples. However, Mn concentrations were high in the shoot samples.

When compared with the control plants, salt treatment caused significant increases in the Fe content of the

leaves (except for Omaha, Semu DNK-207-NA and Lirawell), shoots (except for Marinca, Tobin, Helios, Regent and Lirawell) and roots (except for Kosa, Spok, Tobin, Semu209/81 and Liraspa) of all the varieties. Similar result was reported for tomato cultivars. Fe concentration in some cultivars decreased and in the others was increased under salt stress (Martinez et al., 1987). However, Sanchez-Raya and Delgado (1996) suggested that, Fe transport decreased from seed to seedling under salt stress in sunflower. On the other hand, Lazof and Bernstein (1999) showed that salinity had no effect on the Fe content of the leaf in lettuce.

In the root part of the plants, Mn content increased for Omaha, Spok, Semu DNK-207NA, Tower, Semu209/81 and Lirawell in the salt stress treatment, however, its concentration decreased in the other cultivars. Mn content decreased with salt stress in the shoot of plants in Marinca, Kosa, Spok and Helios cultivars. However, it increased in the other cultivars shoots. Mn content of the leaves increased in the Tower, Tobin, Helios, Regent and Lirawell cultivars, but decreased in the other cultivars. It was reported that, salinity significantly increased the uptake and concentration of Mn in the shoots and leaves of alfalfa plants (Wang and Han, 2007). On the other hand, it was found that, salinity had no effect on Mn content of the root and aerial part of strawberry (Turhan and Eris, 2005)

Copper contents of the plant parts were different under salt stress, its concentrations decreased in the leaves of Semu DNK-207NA, Tower and Tobin and its content increased in the roots of the other cultivars. Salt application decreased the Cu concentration in the shoots of Star, Semu 209/81 and Regent; the other cultivars increased as a result of the treatment. Copper concentration decreased in roots of only Marinca cultivar; the other

Table 2. In roots, shoots and leaves of canola varieties micronutrient accumulations (ppm) under salt treatment and non salt treatment.

M.N	P. O	T	Marinca	Kosa	Spok	Semu DNK207 NA	Tower	Liraspa	Star	Tobin	Helios	Semu 209/81	Regent	Lirawell	TM	P.O.M	
Fe	Leaf	0	36.9 ^e	76.1 ^a	42.8 ^e	45.6 ^d	37.3 ^e	62.3 ^b	37.0 ^e	54.8 ^c	53.4 ^c	44.4 ^d	38.4 ^e	63.8 ^b	49.4	54.94	
		150	74.7 ^b	85.4 ^a	47.5 ^e	35.2 ^f	48.2 ^e	77.1 ^b	47.7 ^e	63.6 ^c	74.5 ^b	52.6 ^d	63.5 ^c	55.8 ^d	60.5		
	Shoot	0	55.5 ^{cd}	66.4 ^b	49.9 ^d	43.1 ^e	21.8 ^g	26.6 ^g	53.6 ^{cd}	56.6 ^c	37.6 ^{ef}	32.5 ^f	96.2 ^a	33.6 ^f	47.8	49.03	
		150	44.7 ^e	86.5 ^a	77.2 ^b	60.1 ^{cd}	34.2 ^f	37.3 ^f	57.9 ^d	43.2 ^e	35.7 ^f	34.3 ^f	64.00 ^c	28.0 ^g	50.3		
	Root	0	45.1 ^e	79.8 ^c	99.5 ^b	64.8 ^d	44.6 ^e	62.6 ^d	62.5 ^d	110.7 ^a	78.6 ^c	98.3 ^b	64.9 ^d	107.1 ^a	76.6	74.13	
		150	73.9 ^d	36.1 ^g	42.3 ^f	82.4 ^c	68.6 ^e	82.3 ^c	74.7 ^d	91.8 ^b	100.2 ^a	38.8 ^{fg}	102.5 ^a	67.1 ^e	71.7		
	Mn	Leaf	0	101.5 ^c	104.2 ^c	103.3 ^c	106.0 ^c	94.3 ^c	39.9 ^e	76.7 ^d	75.4 ^d	42.2 ^e	140.1 ^a	51.8 ^e	121.9 ^b	88.0	81.10
			150	43.9 ^f	91.6 ^d	37.4 ^{fg}	42.4 ^{fg}	102.4 ^c	32.6 ^g	61.1 ^e	114.8 ^b	84.3 ^d	42.3 ^{fg}	102.1 ^c	136.0 ^a	74.2	
Shoot		0	106.8 ^c	160.8 ^a	86.1 ^d	135.1 ^b	90.2 ^d	24.4 ^f	34.9 ^f	73.6 ^e	88.6 ^d	61.9 ^e	35.9 ^f	123.2 ^b	85.1	87.15	
		150	33.2 ^f	135.1 ^b	55.2 ^e	201.4 ^a	117.5 ^c	32.4 ^f	37.1 ^f	143.4 ^b	50.5 ^e	91.6 ^d	38.8 ^f	133.8 ^b	89.2		
Root		0	134.3 ^{ab}	47.8 ^d	30.4 ^e	123.4 ^b	42.7 ^{de}	140.0 ^a	125.8 ^b	111.6 ^c	142.7 ^a	45.2 ^d	35.7 ^{de}	38.5 ^{de}	84.9	81.02	
		150	104.9 ^b	44.5 ^d	37.3 ^f	150.5 ^a	84.7 ^c	110.7 ^b	106.6 ^b	68.2 ^d	44.7 ^{ef}	77.8 ^{cd}	50.5 ^e	46.3 ^{ef}	77.2		
Cu		Leaf	0	4.64 ⁱ	6.2 ^d	5.71 ^{eg}	5.77 ^{ef}	5.48 ^{f-h}	5.22 ^h	5.3 ^{gh}	8.06 ^a	7.23 ^b	6.67 ^c	6.03 ^{de}	4.07 ^j	5.9	6.14
			150	6.48 ^d	6.9 ^{bc}	6.69 ^{cd}	4.95 ^f	5.31 ^{ef}	7.34 ^b	5.5 ^e	5.64 ^e	8.77 ^a	7.36 ^b	6.47 ^d	5.53 ^e	6.4	
	Shoot	0	4.34 ^f	6.4 ^{ab}	6.80 ^a	5.99 ^{bc}	5.21 ^{de}	4.34 ^f	6.5 ^{ab}	6.10 ^{ac}	4.68 ^{ef}	5.60 ^d	5.54 ^{cd}	3.18 ^g	5.4	5.59	
		150	4.74 ^e	7.8 ^b	8.24 ^a	6.46 ^c	5.35 ^d	5.05 ^{de}	6.2 ^c	6.34 ^c	6.33 ^c	5.20 ^d	4.13 ^f	3.71 ^g	5.8		
	Root	0	4.78 ^e	8.4 ^d	8.95 ^{cd}	9.95 ^b	10.1 ^b	8.85 ^{cd}	8.6 ^{cd}	9.36 ^{bc}	10.09 ^b	8.28 ^d	11.1 ^a	9.90 ^b	9.0	8.14	
		150	6.57 ^d	4.5 ^f	5.90 ^e	8.19 ^{bc}	8.80 ^a	8.85 ^a	7.7 ^c	7.68 ^c	8.01 ^c	4.23 ^f	8.7 ^{ab}	7.79 ^c	7.3		
	Zn	Leaf	0	40.2 ^e	32.7 ^f	69.5 ^{bc}	35.0 ^{ef}	62.9 ^c	90.5 ^a	64.4 ^c	74.4 ^b	73.5 ^b	57.6 ^d	53.4 ^{de}	70.3 ^{bc}	60.4	60.5
			150	23.6 ^g	37.4 ^f	57.7 ^d	45.7 ^e	51.3 ^{de}	66.7 ^{cd}	50.4 ^{de}	85.5 ^b	85.9 ^b	74.9 ^c	57.3 ^d	91.1 ^a	60.6	
Shoot		0	62.6 ^{bc}	65.1 ^b	61.1 ^{bc}	42.5 ^{de}	81.7 ^a	43.2 ^{de}	61.1 ^{bc}	47.5 ^d	40.3 ^e	28.1 ^f	83.9 ^a	51.6 ^c	55.7	55.79	
		150	76.6 ^{ab}	79.3 ^a	79.6 ^a	51.4 ^{cd}	51.7 ^{cd}	47.7 ^d	54.9 ^c	31.7 ^f	42.3 ^e	32.7 ^f	56.4 ^c	66.2 ^b	55.9		
Root		0	29.5 ^f	41.7 ^e	90.9 ^{bc}	101.4 ^a	78.0 ^d	93.4 ^b	98.4 ^{ab}	102.9 ^a	88.5 ^c	83.9 ^{cd}	72.9 ^{de}	100.6 ^a	81.8	73.42	
		150	37.4 ^e	61.4 ^c	72.5 ^{bc}	70.0 ^c	72.4 ^{bc}	75.8 ^b	73.5 ^{bc}	94.6 ^a	42.1 ^{de}	46.9 ^d	76.9 ^b	58.9 ^c	64.9		

Mean values indicated by the same letter are not significant different ($p < 0.05$). Abbreviations : M.N, micro nutrient; P.O, plant organs; T, treatment; T.M, treatment mean; P.M.O, plant organs mean.

cultivar increased. Similarly, Wang and Han (2007) showed that, salinity reduced the uptake and concentration of Cu in alfalfa plants but significantly increased Zn content in the roots, shoots and leaves. On the other hand, Alpaslan et

al. (1998) and Martinez et al. (1987) suggested that, salinity caused increased Cu content in rice, wheat and tomato plants.

Zinc contents of the plant organs were different under salt stress, while its content decreased in

the leaves of Marinca, Spok, Tower, Liraspa and Star but in the other cultivars it increased. Zinc concentration decreased in the shoots of Spok, Star, Tobin and Regent, but increased in those of other cultivars. As its content increased in roots of

Marinca, Kosa, and Regent, it decreased in the other cultivars. Previously, varying results were obtained in other plants for the differences of Zn content in salt stress conditions. In most cases, salinity increased the content of Zn in the plant tissue as in pepper (Cornillon and Palloix, 1997), wheat and rice (Alpaslan et al., 1998), zucchini (Villora et al., 2000), strawberry (Turhan and Eris, 2005), alfalfa (Wang and Han, 2007) and sunflower (Achakzai et al., 2010).

Conclusions

Significant differences were determined among the canola cultivars for plant growth and micronutrient contents of different plant parts under the salt stress. It could be concluded that salt stress in the canola cultivars was affected negatively in all the yield components studied. High Na⁺ content generally disrupted the nutrient balance, thereby, causing specific ion toxicity despite disturbing the osmotic regulation (Greenway and Munns, 1980). The effect of salinity on the micronutrient composition of plant tissues was different in the cultivars. Salt stress caused ion imbalance in the canola cultivars. The results indicated that, micronutrient contents and distribution in the plant tissues lost their balance. The cultivar Lirawell showed the best development under salt stress compared with the other cultivars investigated. Fe, Mn and Cu content increased in all the plant parts with salt applications except for some cultivars. There were no significant differences for the Zn content of the leaves by salt stress.

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