THE IMPACT OF UVB-B TREATMENT ON THE VARIATION OF NUCLEIC ACIDS CONTENT BY *VICIA SATIVA* L.

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Key words: *Vicia sativa* L., UV-B, nucleic acids, RNA, DNA.

Abstract: The ultraviolet radiations UV-B administrated on *Vicia Sativa* L. inhibate the nucleic acids biosynthesis. It can be concluded that it apears a plant defense reaction against UV-B. The nucleic acids quantity descrease directily proportional with irradiation time. Under the ultraviolet irradiation, the quantity descrease either for DNA or for RNA.

INTRODUCTION

The UV-B radiation (280-315nm) is part of the invisible solar spectrum. The stratospheric ozone lay from the upper Earth atmosphere absorbs short waved 290nm UV radiation (the potential damaging ones). The UV-C radiation is completely absorbed by ozone and atmospheric oxygen, the UV-B radiation is partially absorbed by ozone, and the UV-A radiation is just in a small amount absorbed (Tosserams, 1996; Frederic et. al., 1989). The stratospheric ozone depletion has like consequence the increase of UV-B radiation level which reach the Earth surface (Caldwell et. al., 1995).

MATERIALS AND METHODS

The experiment consists in UV irradiations, for different time periods, on *Vicia Sativa* L L, being in early ontogenetic stages, followed by biochemical tests (the content in nucleic acids).

25 mg vegetal tissue, were grinded in 2 ml percloric acid ($HClO_4$-0,2N) and keept at 4°C till next day. After samples centrifugation for 15 minutes at 3000 rpm, pellet was resuspended in 2ml mixture ethanol 70%:glacial percloric acid. After a new centrifugation in the same conditions, pellet was treated with 2 ml mixture ethanol 96%: etilic ether 3:1. After lipids elimination (10 minutes on water bath at 50°C), samples were centrifugated 15 minutes at 3000 rpm.5 ml percloric acid 1N was added. Next day, from supernatant was determinated RNA and from pellet DNA. For DNA dosing pellet was resuspended in 5 ml percloric acid 0,5N, samples being boiled in a reflux refrigerent for 30 minutes. After centrifugation extinsion were read by spectrophotometer, in UV light at 270nm and 290nm, reported to controle sample (percloric acid 1N for RNA and percloric acid 0,5N for DNA).

Calculation of DNA and RNA quantity was made after the formula:

$$RNA = \frac{5315 \times \Delta}{m} \times 0{,}1 \qquad DNA = \frac{5525 \times \Delta}{m} \times 0{,}25$$

Delta (Δ) represents the difference between extinctions read by 270nm and by 290nm, made with UV/VIS- Jasko, X-530. Values were expressed in mg/g fresh tissue.

RESULTS AND DISCUSSIONS

After the UV-A treatment (370nm), considered like controle for UV-B, aplied 12 hours on *Vicia Sativa* L L, it was observed the descrease of total nucleic acids quantity comparing with dark controle, due to descrease of DNA content (to 55,462 mg/g from 73,757 mg/g). The RNA quntity increase comparing with controle (to 18,311 mg/g from 11,734 mg/g).

For 24 hours irradiation time, the nucleic acids quantity dramaticaly descreased to 21,626 mg/g. The descrease was again due to DNA which reached a very low value (7,718 mg/g DNA comparing with 67,175 mg/g by controle).

The drastically inhibition of biosinthesys after 24 hours recovers at 48 hours and can be apreciated like a plant defense reaction against UV, for reparing the lezions at molecular levels.

For 12 hours irradiation with UV-B (305nm) could be observed a descrease of nucleic acids level (71,441 mg/g) comparing with controle (85,491 mg/g), again due to descrease of DNA

level. The RNA level increase to double value comparing with controle (20,621 mg/g instead 11,734 mg/g). We can conclude that UV-B aplied for 12 hours on seedlings descrease DNA synthesys, but stimulate RNA activity. For 24 hours treatment with UV-B, the nucleic acids quantity continue to descrease comparing to controle, due to DNA and RNA, same effect being observed also for UV-B controle.

For 48 hours irradiation time, the nucleic acids quantity increase, a very interesing fact which we can not explain yet.

For 12 hours irradiation time with UV-B (295nm), the nucleic acids quantity do not suffer quantitative changes, having close values to the controle(84,797 mg/g comparing with 85,491 mg/g). It can be seen (Fig.3) RNA descrease (9,756 mg/g comparing to 11,734 mg/g for dark controle) and a DNA increase (75,042 mg/g,comparing with 73,757 mg/g).

For 24 hours irradiation with UV-B 295nm, for *Vicia Sativa* L, it could be noticed a light descrease of nucleic acids descrease in fresh vegetal tissue. The descrease was also due to RNA quantity descrease (to 15,565 mg/g from 17,839 mg/g by controle), and also due to DNA (to 63,529 mg/g from 67, 175 mg/g).

For 48 hours irradiation time the nucleic acids quantity dramaticaly descrease (52,887 mg/g comparing to 84,700 mg/g). This time the descrease was also due to RNA(10,272 mg/g comparing to 16,259 mg/g),also DNA (42,615 mg/g comparing to 68,441 mg/g for controle).

Analising this data, we can conclude that UV-B with 295nm wavelengh,do not have a significant impact in first 24 hours, but for 48 hours irradiation time it can be noticed an inhibiting effect.

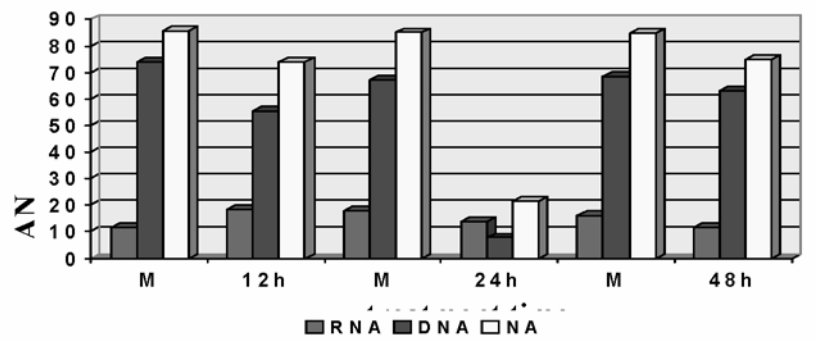

Figure 1. The variability of nucleic acids quantity under action of UV-B (λ=370 nm) at *Vicia sativa* L.species (mg/g fresh tissue).

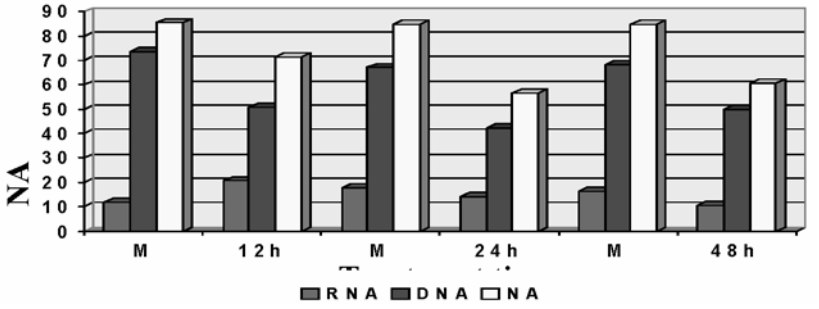

Figure 2 The variability of nucleic acids quantuty under action of UV-B (λ =305 nm) at *Vicia sativa* L.species (mg/g fresh tissue).

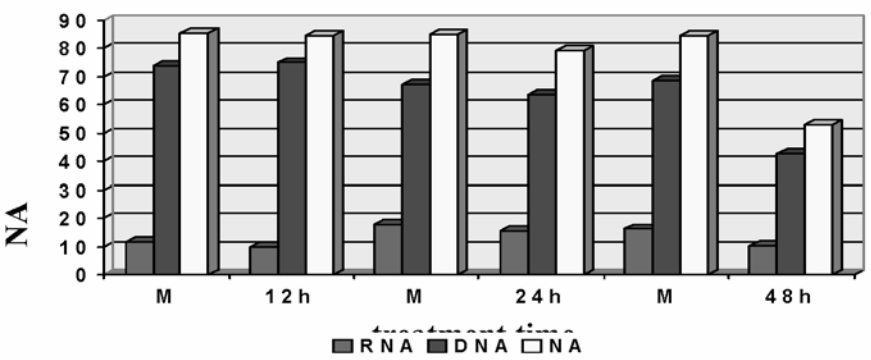

Figure 3. The variability of nucleic acids quantity under action of UV-B (λ =295 nm) at *Vicia sativa* L. species (mg/g fresh tissue).

CONCLUSIONS

The ultraviolet radiations UV-B with 305nm and UV-A with 370 nm wavelengh administrated for 24 hours on *Vicia Sativa* L. inhibate the nucleic acids biosynthesis, but inhibitio dissapears in the case of 48 hours irradiation.

It can be conclude that it apears a plant defense reaction against UV-B, having as result the repair of lezions at molecular level.

The nucleic acids quantity descrease directily proportional with irradiation time for UV-B 295nm.

Under the ultraviolet irradiation,the quantity descrease either for DNA or for RNA.

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