Laser influence to biosystems

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Abstract

In this paper a continuous (cw) lasers in visible region were applied in order to study the influence of quantum generator to certain plants. The aim of such projects is to analyze biostimulation processes of living organisms which are linked to defined laser power density thresholds (exposition doses). The results of irradiation of corn and wheat seeds using He–Ne laser in the cw regime of 632.8 nm, 50 mW are presented and compared to results for other laser types. The dry and wet plant seeds were irradiated in defined time intervals and the germination period plant was monitored by days. Morphological data (stalk thickness, height and cob length) for chosen plants were monitored. From the recorded data, for the whole vegetative period, we performed appropriate statistical data processing. One part of experiments contains the measurements of coefficient of reflection in visible range. Correlation estimations were calculated and discussed for our results. Main conclusion was that there was a significant increment in plant's height and also a cob lenght elongation for corn.

Keywords: laser, biostimulation, biosystems, plants, doses, reflection coefficient, correlation.

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The influence of nuclear particles, electric fields, magnetic fields and electromagnetic (EM) radiation in the whole electromagnetic spectrum, including nuclear and atomic areas, belong to old problems, vastly studied, but not yet completely understood. Earlier investigations covered spontaneous radiation, while up-to--date investigations deal with stimulated radiation e.g. quantum generators (lasers, masers, uvasers, xrasers and gammarasers). There is not enough space for numbering positive or negative influences of electromagnetic radiations, generally speaking in contemporary civilization. In spite of administration and regulative, in dosimetric point of view and technical manuals, accidents are still present [1–8].

Bad influence to human health is in provoked thermal effects as well as electrochemical, mechanical and other effects. The radiation shows some kind of cumulative effects for the whole tissue, but it could be focused to lead to the processes in cells (cell surgery is known today, since first famous data were red blood cell drilled in nine places). Numerous are effects induced by electric and magnetic fields in everyday's life as well as in military and medical applications of RF (radio frequency) devices portion [9–11].

Local irradiation of high intensity RF leads to skin and muscle burns and to ocular damage of various degrees (lens blurring), damage of reproductive organs, changes in central nervous system, blood flow and immune defense mechanisms [12,13].

Well known accidents with radar beams caused by human negligence, deserve further analysis to prove the statement that long irradiation to RF beams of low intensity leads to health damage, cancer disease and lasting consequences to offspring. This could also be considered from the point of view of stimulated radiation in this range, due to maser (quantum generator or amplifier) and its implementation in a number of processes. Sometimes first letter m represents molecular and sometimes microwave, what is common for engineers and physicists.

Industrial growth and power engineering are always focused to power transmission (power lines, high voltage devices, etc.) and human health. Doses regulative are constantly changing in the areas of: influence of electric and magnetic fields, nuclear specified radiation, laser and other quantum generator thresholds and allowed radiation power densities (total cumulative doses). All numbered fields could influence human

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organs (heart, nervous system tissues, reproductive organs and other). Employees on power lines and transformers maintenance, electricians and other personnel in electrical based processes (induction furnace, electrified railways, etc.) are always in danger. Besides that, the influence of laser sources in communication purposes, various monitoring and measurements, laser therapy, mass media applications including open space laser shows present potential threat. Note that laser, maser, iraser, uvaser, xraser and gammaraser, work as quantum generators of stimulated radiation in various portions of the EM spectra, but the last two are not in commercial use.

Biological influences of various fields (electric and magnetic) are usually studied separately for DC and AC regimes, but also in transient regimes, which are very important due to high amplitude values. Some people consider that magnetic field has stronger effect to living creatures.

Usually, AC and DC regimes are vastly studied but transient regimes are very important in real applications of electric power. This is related to power network connections and usage of electrical devices. For dosimetry it is necessary to as precise as possible define this regime with possible overloading.

Electrical and magnetic fields in human body cause induced currents which, with defined density could affect organism, i.e. hearth functioning, nervous system tissues and others. In spite of tremendous work, there are still open issues in this area mainly targeting the fact, that these fields provoke other effects. Considering new methods of LLL therapy (low level laser) the novelties are biphasic dose responses [14–16].

The influence of modern electronic and electric systems is also particular problem of civilization and the questions of allowed doses, besides state regulative, are still opened, depending on various facts (television sets, video terminal, PC monitors, communication devices, GSM mobile devices, etc).

The levels of allowed EM radiation are often expressed by defined zones and allowed surface power densities. Note that both frequency and amplitude of electric field (intensity) have to be taken into account. Due to these facts all of these calculated numbers should be considered just as the quantitative values. Explicitly: the zone of very intensive radiation is with surface power density higher than 10 mW/cm^2 (15 min within 24 h), zone of moderate radiation 0.1–0.9 mW/cm^2 (3 h within 24 h) and zone of weak radiation with less than 0.1 mW/cm^2.

Allowed levels of electromagnetic radiation power density are regulated by national standards. In Sweden, the level of EM power density is 450 μW/cm^2 corresponding to the electric field E = 41 V/m. Serbian standard is more rigid, allowing levels of electromagnetic power densities of 200 μW/cm^2, *i.e.*, E = 27.5 V/m [9,10,17,18].

To compare mentioned values of electromagnetic power densities we remind that radiation of stars is aproximately14 pW/cm^2 and that for human body the value could be approximately 0.5 μW/cm^2.

The laser beam irradiation and allowed levels are new problems in spite of the fact that by frequency range they are parts of electromagnetic spectra. The main difference is that, depending on provoked processes, we could have all the effects as with spontaneous electromagnetic radiation, but many new ones as well.

In area of quantum generators dosimetry is not as detailed as in area of nuclear radiation where exposition and absorption are precisely defined and should be completed with different parameters, corrective factors and different units. Note that when high power lasers interact with the material many nuclear processes appear (X and gamma radiation, neutrons, etc.). Therefore, both dosimetries have to be united. Nuclear dosimetry worldwide still uses SI as well as other units, which is not allowed in our country. In presented paper we chose approach which is the most often implemented in references describing biostimulative (biomodulating) effects. We gave certain parameters for valuating possible maximal input energy levels. Our approach deals with optimal corrections for determining more precise doses and increasing of repeatability. It also deals with nominal laser power and time of exposition. It is interesting that in considering couplings between nuclear and laser radiation to objects from biosphere, laser beam can be used as means of measurement for evaluation of nuclear radiation influence by phosphorescent and other processes. Up to day it seems that only in nuclear dosimetry exist approaches with unit equivalent to biological tissues as rem (Röntgen equivalent man), but not in SI system. Interesting is that in laser influence to plant systems it could be used adequate unit per seed grain. [19–23].

Laser influence in biology and especially medicine usually is studied in four areas: for surgery, biostimulation (modulation, acupuncture), diagnosis and in production of drugs (generally) [16,24–27]. Minimal laser threshold for wound healing (influence to fibroblasts) and overall cell level influence for activation of targeted processes are the objects of numerous investigations [24].

Therefore, in this paper we start with experiments on plants and some objective physical processes which objectivize the point of view. In references, several tenths of cw mW, are used for biostimulation purposes and we wanted to see the influence of selected doses to plants, *i.e.*, plant seeds. The objective was to obtain data which could be correlated and could trace perspective for future manipulation of facts. The aim is to give experimental data for determination of minimal levels of irradiation doses for defined purposes and to categorize those levels of influence.

Significant number of references dedicated to interaction of laser beams with materials exist [28–31]. Although mainstream attitudes are formed for several tents of years, a lot of experimental and theoretical work is still needed to obtain reliable data with good repeatability. This is especially true when applied to different plant seeds, related to specific geographical area and to growth of plant during specific year, with defined meteorological situation. Besides this, vegetative periods in plant growth, considering certain plant species influence the level of meteorological influences and other factors (soil type). During previous periods papers often dealt with better wheat crop, corn crop and crop of other plants, than followed more precise research of genetic content, etc. [28–31].

In this paper authors tried to elaborate on performed experiments and to remind of complexity of data which follow experiments. This includes descripttions using coefficient of absorption and reflection, laser beam polarization state, etc. enriched with correlation coefficient.

EXPERIMENTAL

Here presented experiments including wheat (*Tricium aestivum*) and several types of corn (*Zea mays* var. *Amilacae*; var. *Identata*) were conducted in few phases. Wheat and corn seed were irradiated with various types and levels of coherent electromagnetic radiation. Here is presented one of the series of samples and corresponding results.

Certain irradiated seeds were wet, while as control were used dry and wet non-irradiated seed. This principle is applied to all experiments.

The first part of experiment had the following phases:

1. Wet and dry seed were irradiated with He–Ne lasers and different energies. The output power of He–Ne was 50 mW, and wavelength 632.8 nm.
2. Laser beam was expanded by beam expander BET 50 type Laser Collimator Eloma 6, so that obtained beam was of 4.5 cm diameter. The beam was then reflected from the flat mirror to Petry's cup towards samples.
3. The reflected laser beam was monitored by pyroelectric radiometer type Rk3440 with appropriate probe type RkP 345. The density of laser beam was 0.4 mW/cm^2.
4. Irradiation energy doses were 1, 2, 4 and 6 J/cm^2, with exposition times of 250, 500 and 1000 s.

Most of the seeds were planted outdoors, with small part (less than 5%) in laboratory environment. Control groups were planted with treated irradiated seeds in order to have more objective comparison of resulting data.

After germination, growth of the plants as well morphogenetic changes, were monitored and recorded (stalk thickness, height, cob length for the corn and in case of wheat plant height). Obtained changes were registered by days. Depending on plant family, vegetative periods were monitored (germination, blooming and fruit development) and at the end analyzed.

Results were processed and visual interpretation of time dependences of plant height, histograms and 3D relations were given. The correlation dependencies between cob length, height and stalk thickness were studied. All results are given by adequate graphical presentations.

The third part of the experiments was measurement of reflection coefficients of some selected seeds.

RESULTS

In Figure 1 the growth of the non-irradiated wet control group plants is presented in days. Characteristic corn plant sizes were measured for non-irradiated wet control seed and wet seed irradiated with He–Ne 6 J/cm^2 including plant thickness, heights and cob lengths Table 1. They were the subject of statistical analysis. Figure 2 represents histogram of plants sprouted from wet control seeds (height of plants).

In Figure 3 the results of the plant growth after wet seed irradiation using He–Ne laser 6 J/cm^2 are presented. Histogram of wet irradiated seed (He–Ne 6 J/cm^2) is presented in Figure 4.

3D presentation of mutual correspondence between characteristic plant sizes: height, plant thickness and cob length for corn irradiated with He–Ne 6 J/cm^2 is presented in Figure 5.

The results of coefficient of reflection for wheat are presented in Figure 6 [32]. Figure 7 shows data about growth of plants germinated from wet wheat control seed.

DISCUSSION

The wheat (*Tricium aestivum*) was selected as typical long day plant representative. Corn (*Zea mays*) is representative of daily neutral plants, although some varieties belong to long day plants. Depending on growth and the size of the fruits for corn samples (different varieties) it is possible to determine which variety belongs to which type of the plants (long day or daily neutral plants). Concerning wheat as long day plants representative, it is possible to draw certain conclusions about growth of the plants irradiated with different types of laser irradiation (as well as preconditions of those seeds – wet or dry) [33–35].

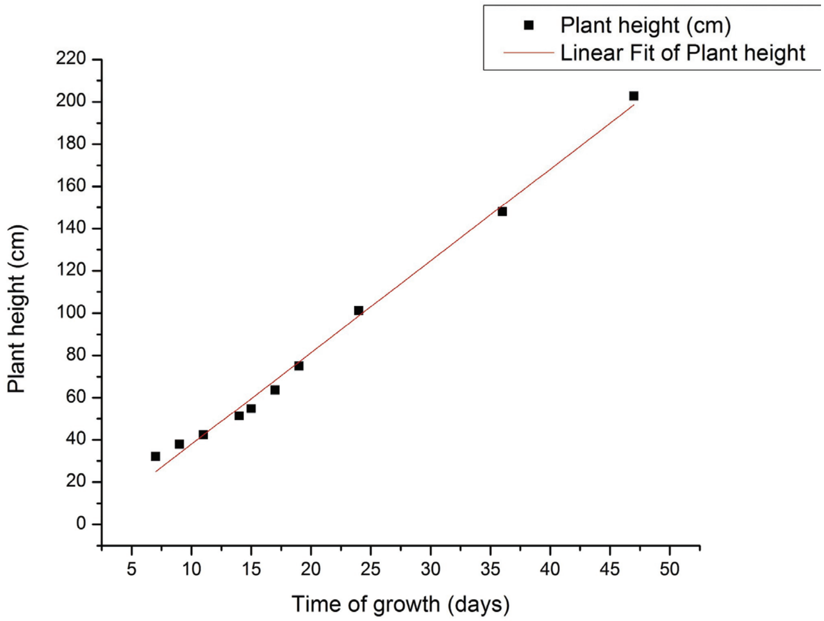

Figure 1. Corn control (non-irradiated) wet seed. Intercept value: –5.52; slope value: 4.34 cm/day.

Table 1. Corn comparison table of plant's characteristic dimensions of plants sprouted from non-irradiated wet control seeds and wet seeds irradiated with He–Ne 6 J/cm²

Parameter	Stalk thickness, mm	Plant height, cm	Cob length, mm
		Non-irradiated wet seed	
Mean	21.29	234.54	20.08
Std. dev.	2.8	36.02	7.14
Correlation coefficient	Stalk thickness – Plant height	Plant height – Cob length	Stalk thickness – Cob length
	0.764339	0.878498	0.791254
		Wet seed irradiated with He–Ne 6 J/cm^2	
Mean	21.42	256.56	22.23
Std. dev.	2.02	13.51	2.88
Correlation coefficient	Stalk thickness – Plant height	Plant height – Cob length	Stalk thickness – Cob length
	0.607617	0.395461	0.673945

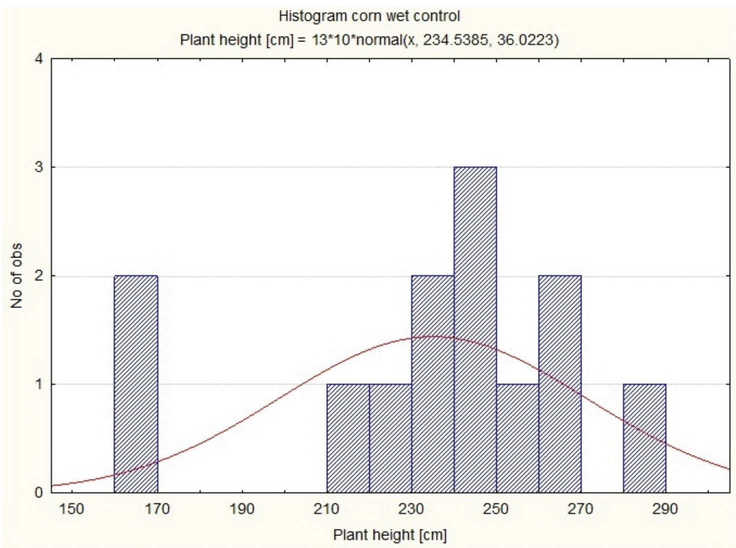

Figure 2. Histogram of wet control (non-irradiated) corn seeds.

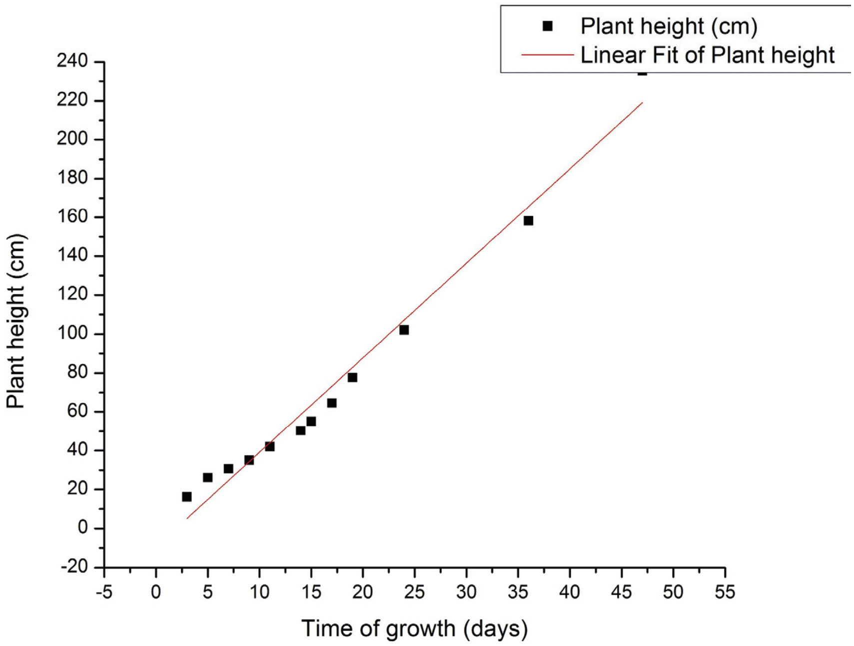

Figure 3. Corn irradiated wet seed (He–Ne laser, 6 J/cm^2). intercept value: –9.43; slope value: 4.86 cm/day.

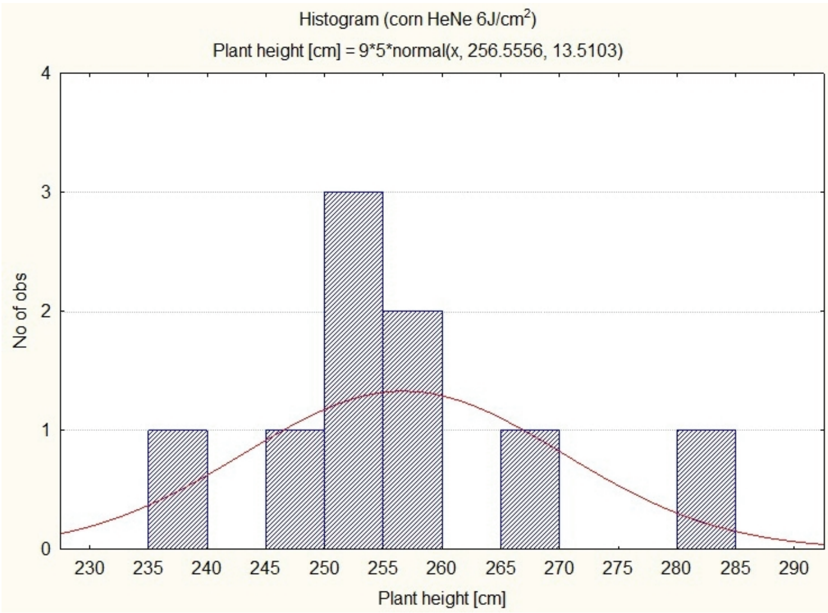

Figure 4. Histogram of wet irradiated corn seed (He–Ne laser, 6 J/cm^2).

In order to *in vivo* trace changes in living organisms, caused by various forms of electromagnetic radiation, mainly irradiated with laser beam, it is necessary to include a whole series of sciences, their methods and explanations. Domain in which results are commented is limited by used statistical methods, and plant characteristic monitoring, expected to be modified by irradiation (height, plant growth rate, fruit characteristics, organoleptic characteristics, genetic characteristics,...).

From the results obtained in this paper, it could be concluded that laser influence is certainly present. The real level of the induced changes could be the object of discussion because the principal question of the quantitative interpretation of results should be confirmed in various plant varieties. On the other hand the data from references in purely biological and biomedical areas do not present enough facts for objectivistic approach, from technical point of view. In spite of obeying the rules in those areas the concrete details were not given, i.e., the coefficient of reflection, absorption for some irradiation of living cells and systems including the situation with plants.

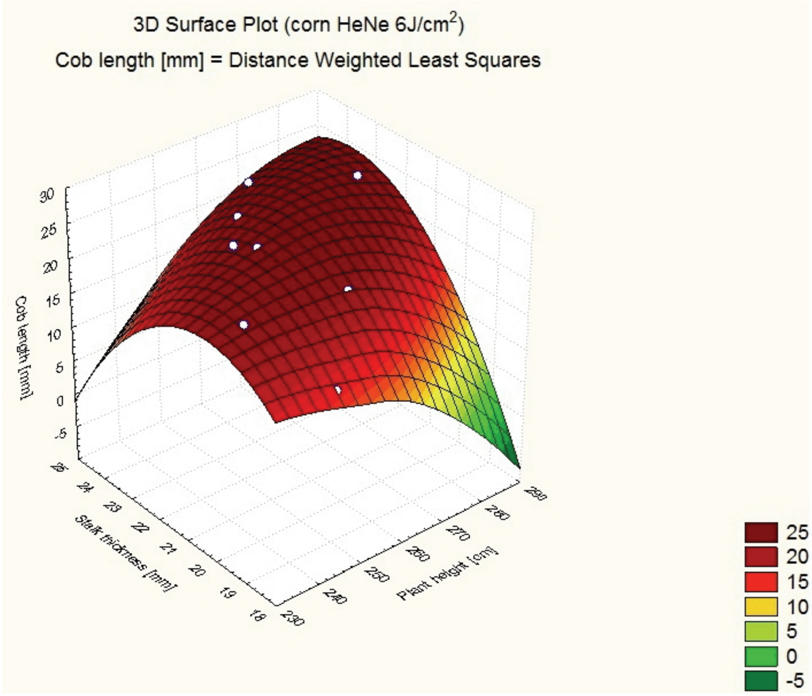

Figure 5. 3D dependence of plant height, stalk thickness and cob length for corn irradiated with He–Ne 6 J/cm^2.

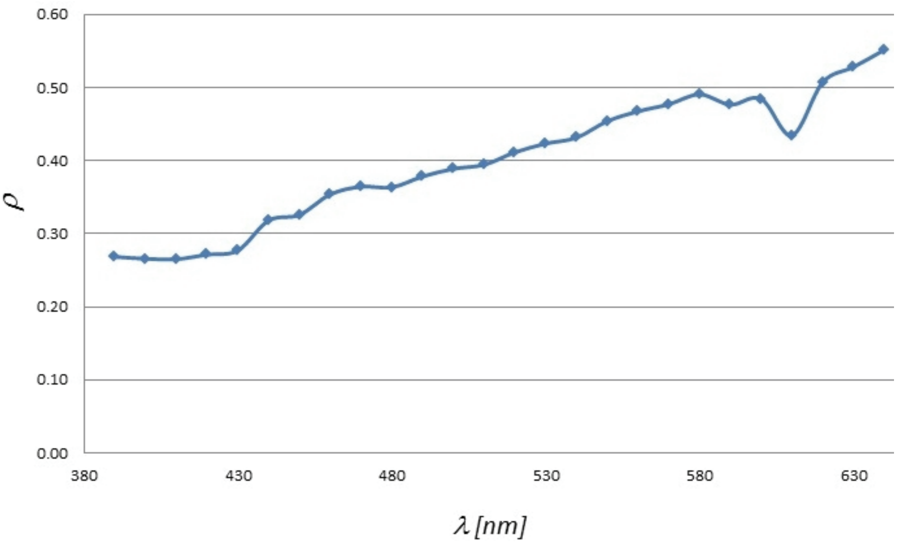

Figure 6. Reflection coefficient of wheat (regular sample).

The deepening of the discussion is related to question should the material whose coefficient of reflection is measured be in powder state (what is demand of the standard equipment) or left as grain.

Considering this paper, the aim of work was to present real parameters with correlation calculation. Some general conclusion could be: correlation dependencies show that from irradiated seed grow plants whose height is comparably greater than those germinated from control seeds. This is expected, concerning the fact that one of the first markers of genetic material change, due to irradiation, is elongation (contribution to height) of plant. Wet corn seed under influence of laser radiation are more susceptible to changes than dry ones. More generally speaking, we could talk about influence of electromagnetic irradiation in laser range of wavelength, power density, etc.

CONCLUSION

It is a general recommendation to avoid unnecessary exposure to magnetic and electric fields and this correspond to the general mainstream in which allowed exposition thresholds (doses) are constantly reducing.

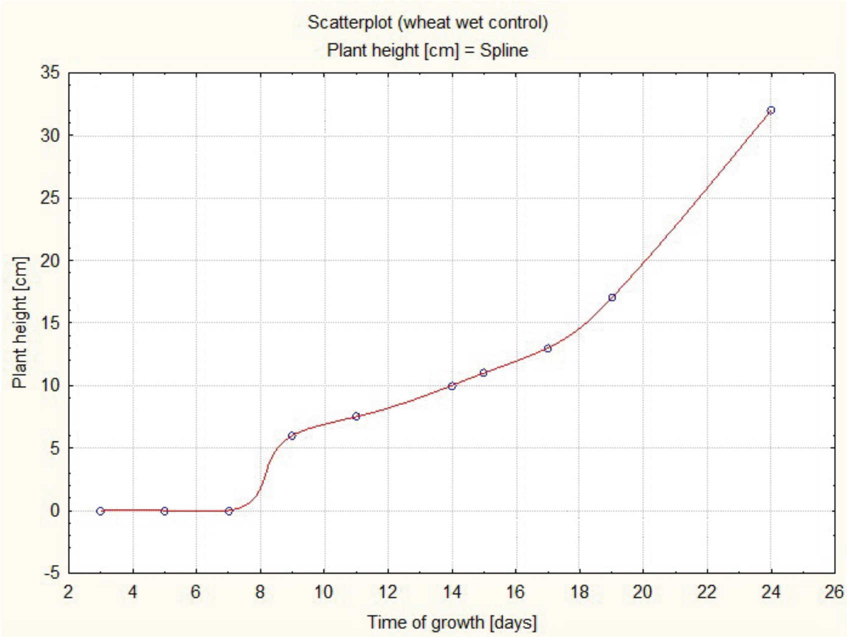

Figure 7. Wheat control (non-irradiated) wet seed.

It is possible to draw parallel between associated magnetic and electric fields of laser with other exposures to electric and magnetic fields (DC and AC). From correlation dependence of plant height and cob length it is possible to conclude that with plant elongation the crop is greater. Corresponding to our other experiments of similar type, here also the use of wet and dry control seed have shown to be a good practice. The results for He-Ne laser gave similar results as those in experiments with GaAs laser for same irradiation parameters, no matter the used wavelengths in visible and in NIR. These issues could be interpreted as quantitative values (real measurable magnitude values) for limited validity for selected exposition ranges – doses and to defined species.

Observing the plots obtained graphically by depicting the growth of plants sprouted from irradiated seeds one could conclude that plant irradiated with smaller irradiation energy (energy density) easily overcame crisis in growth between 30th and 40th day. Concerning plant growth, just by measuring height, it is obvious that irradiated plant needs more light for transition from second vegetative phase to third (germination, springing and flowering). Overall conclusion is that applied levels of energy influenced qualitative shift of plants.

For even further analysis of measured data for all types of plants and their varieties (wheat and corn) it is necessary, based on plant tissues parameters examined, to do appropriate model of seeds (i.e. wheat seed could be modeled using 7 layers with distinct parameters) before irradiation and then to compare real and theoretical cases.

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IZVOD

UTICAJ LASERA NA BIOSISTEME

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Uticaj laserskih snopova na žive organizme je već duže vremena predmet proučavanja koji zahteva multidisciplinarni pristup. Kako se podrazumevaju i vidljiva, UV i IC oblast, to se problem može posmatrati i kao proučavanje koje pripada široj problematici uticaja električnih, magnetnih polja i elektromagnetskih polja i talasa na žive organizme. U ovom radu su proučavani eksperimentalno procesi i rezultati delovanja He–Ne lasera na suva i vlažna semena pšenice (*Tricium aestivum*) i kukuruza– *Zea mays*, (var. *Amilacae*; var. *Identata*). Iako je uticaj elektromagnetnog (EM) zračenja na živa bića stari problem, koji je mnogo proučavan, on je još uvek sa mnogo nerešenih pitanja. Ako uz problematiku spontanog EM zračenja postoji veliki broj nerazjašnjenih procesa, utoliko pre to važi za oblast stimulisanih zračenja – kvantnih generatora, koji komercijalno (ili u eksperimentalnom stadijumu) rade od gama do RF (radio frekvencija) oblasti. Laseri su već odavno uključeni u praksu u okviru biomedicine, biologije, a u humanoj medicini odavno su uključeni u oblasti hirurgije, biostimulacije, dijagnostike, farmakologije, akupunkture, itd. Oblast primene lasera se vrlo brzo uključuje u mnogo svakodnevnih primena, a na delovanje zračenja na bioorganizme (biosisteme) se, i pored postojeće regulative, ne obraća dovoljno pažnje. Nove oblasti primene lasera vrlo kratkih impulsa i fenomeni nelinearne optike, otvaraju mnogo novih pitanja. Pragovi za terapeutska dejstva (biostimulaciju) se istražuju i pripadaju zadacima laserske dozimetrije i njenih preciznih stavova, koji se razlikuju od države do države. U radu su opisani izvršeni eksperimenti sa ozračavanjem semena pšenice i kukuruza (suvih i vlažnih). Korišćen je gasni He–Ne laser kontinualnog dejstva (cw, 632,8 nm, 50 mW). Posle klijanja praćen je rast biljaka po danima (debljina i visina stabljike, dužina klipa kukuruza, a za pšenicu visina biljke). Posle praćenja vegetativnih perioda i roda izvršena je analiza dobijenih rezultata sa statističkom obradom podataka. Korišćene su predstave u vidu histograma, 2D predstave i interpretacija u 3D na bazi metode najmanjih kvadrata i neke korelacione analize. U mnogo referenci se uključuje samo talasna dužina lasera i doza, bez osvrtanja na koeficijente refleksije i absorpcije biosistema. Zato je ovde dat koeficijent refleksije, tipičan za pšenicu, kao kvalitativna mera za uračunavanja pravilnije doze, prema tipu lasera. Koeficijenti refleksije biljaka mogu da posluže i za daljinske kontrole (LIDAR) stanja biljnih vrsta na terenu. Eksperimentalni rezultati sa He–Ne laserom su u dobroj saglasnosti (kvalitativnoj) sa rezultatima, dobijenih na bazi poluprovodničkih lasera sa uporedivim dozama. To važi i za uticaj vlažnosti semena, bez obzira da li se radi o He–Ne, gasnim ili poluprovodničkim laserima na opsegu od 800–900 nm.

Ključne reči: Laser • Biostimulacija • Biosistemi • Biljke • Doze • Koeficijent refleksije • Korelacija