

The Ability of Plants Leaves Tissue to Change Polarization State of Polarized Laser Radiation

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Abstract

The main purpose of this work was to confirm the previously proposed model of the laser radiation interaction with the epidermal layer of monocotyledons cells. The other purpose was to show that plant age affects polarization parameters and polarization direction affects plant development.

The methods used in this work include the development of LED light sources, the polarization parameters assessment (polarization index) and statistical analysis. The maize plants (*Zea mays* L.), variety “Early gourmet 121”, optical setup and software TXP Series Instrumentation were used in the work. Plants were grown under white light (WW) at an intensity of $200 \mu\text{mol m}^{-2} \text{s}^{-1}$.

The study established that the harmonic ellipticity polarization change by $\pm 12.0^\circ$ occurs when the linearly polarized light passed through the maize leaves depending on the axis rotation angle of the epidermal cells. In addition, it was shown that the maize leaves of different ages could have different values of the refractive index anisotropy, and, consequently, different polarization ellipticity index.

It has been established that the maize leaves with ordered structure of epidermal cells are able to change the polarized radiation ellipticity. That confirms the previously proposed model of the polarized light interaction with plant leaves. We established that linearly polarized light passing through the leaves of maize plants turns into elliptically polarized light, which is able to interact more effectively with the photosensitive protein structures of the leaf inner cells.

1 Introduction

The light significantly affects plants, being an energy source through photosynthesis process. The electromagnetic energy converts into the energy of chemical bonds of the organic substances (Nakonechnaya et al. 2021a). However, in addition to this, light also plays a regulatory role. The light signals registration in the visible light spectrum by photoreceptors allows plants to launch certain development programs, control the processes of morphogenesis and photosynthesis, and to adapt to changing environment. Consequently, controlling the light spectral characteristics in which plants grow, it is possible to manage plants ontogenesis processes (Kulchin et al. 2020).

In recent years, the most popular artificial light sources, including those for agronomic purposes are LEDs due to their large number of advantages (Olle and Viršile 2013). The other popular quantum radiation sources are lasers. There are wide range of works that describes the laser radiation stimulating effect to various cultures seeds (Hasan et.al. 2020; Podleśna et al. 2015; Reyes et al. 2015) and crops (Budagovsky et al. 2015; Filina et al. 2020; Grishkanich et al. 2016; Hernandez et al. 2010; Sevostyanova et al. 2021a). These works show the practical applicability of such radiation. Many works describes the impact results, but relatively small number of works devoted to explain the mechanisms behind these effects. Such works shows that coherent laser radiation exposure to seeds, as well as the plants themselves, with a duration of one and tens of seconds is enough to cause responses of the

photoreceptor system that triggers the processes of morphogenesis (Hasan et.al. 2020, Sevostyanova et al. 2021b).

Recently, many authors are studying the light spectra effect on plant growth, as well as on the various useful substances content accumulation (Chen et al. 2020; Grishchenko et al. 2022; Micheeva et al. 2021; Nakonechnaya et al. 2021a). Subsequently, the data obtained that closed a number of gaps existed in this field of knowledge. In addition, there are works describing the impact of light spectra and intensity, as well as radiation level changing dynamics, on different plants development (Kulchin et al. 2018; Nakonechnaya et al. 2019; Nakonechnaya et al. 2021b). At the same time, the light polarization and its effect on plants insufficiently presented in papers.

The radiation which intensity vector oscillation directions are somehow ordered called polarized radiation. Polarized radiation, imperceptible to the naked eye, is nevertheless widespread in nature. The radiation polarization occurs as a result of the natural light reflection from dielectrics (for example, reflection from the water surface or leaves), as well as the solar radiation scattering on molecules and the atmosphere inhomogeneities. When sunlight is scattered in the atmosphere, the scattered light is mainly polarized at a right angle relative to the direction to the sun (Zvereva 1988). Thus, when the sun is low on the horizon, the radiation coming from the zenith point of the celestial sphere is highly polarized. Consequently, in the morning and evening hours, linearly polarized radiation reaches the earth's surface. This is the basis for the assumption that plants, which have developed under such conditions throughout the multi-million-year history of their evolution, were able to adapt to the polarized radiation use for their own purposes, as many animals did (Chiou et al. 2008; Horvath et al. 2014; Wehner 1976).

There are only a few works describing plant organisms' response to polarized light. For example polarotropism observed by Kataoka et al. (2000) in *Vaucheria algae*, but this reaction occurs only after a few days since irradiation, and the mechanism itself is not clear. In addition, Lamparter et al. (2004) established that the terminal cells of moss grow in the direction perpendicular to the electric field strength vector of the light. The positive effect of polarized radiation on the plant seeds germination was observed by Perveen et al. (2010), Podleśny et al. (2012), and Podleśna et al. (2015).

It is well known that the use of laser light sources makes it possible to control such radiation characteristics as wavelength, intensity, exposure time, dose and period of exposure, as well as polarization. Therefore, the use of lasers opens up the possibility of controlling the radiation polarization state and thereby studying the polarized light interaction with plants.

Kulchin et al. (2021) established that the *Allium cepa* L. epidermis layer is capable of converting linearly polarized laser light into elliptically polarized light. This explained by the refractive index anisotropy presence in the *A. cepa* cells epidermal layer. That explanation allows to propose the model for the polarized laser radiation interaction with plant leaves. Later, similar phenomena discovered by Kulchin et al. (2023) in the *Phalaris arundinacea* leaves study. It was established that the ability to change the polarization state and degree is specific for monocotyledons characteristic, as a rule, which epidermal cells have an approximate rectangular shape and form an ordered structure. Such cells order in the

structure of plant tissue, in combination with their mechanical stress, are capable of imparting the properties of optical activity to the cells layers. Yang et al. (2020) established that in the field of plant cell polarity, many unresolved questions remain regarding how protein polarization is initiated, maintained, and regulated in plants. In particular, by the maize cells example, it is demonstrated that in maize, before the SMC asymmetric division, the SCAR/WAVE regulatory complex first polarizes to the membrane adjacent to the guard mother cell (GMC) contact sites, where PAN2 polarizes subsequently. After PAN2 polarization, PAN1 and Rho of Plants (ROP) proteins are polarized, followed by the formation of an actin patch and the directional migration of the pre-mitotic SMC nucleus.

As it was established by Kulchin et al. (2023), the layer of epidermal cells can lead to the transformation of the polarization state of laser radiation acting on cells from linear to chiral, which can lead to a change in the nature of its interaction with the molecular system of the cell. Presumably, the result of such linearly polarized radiation transformation provides the easier interaction possibility of polarized light with polarized proteins.

Kulchin et al. (2021) proposed optical model of the polarized radiation interaction with the leaf epidermis layer. According to the proposed model and data by Rytov (1956) and Tuchin (2016) the cell cytosol refractive index can vary from 1.34 to 1.42, and the cell membrane refractive index can vary within 1.40–1.50. Therefore, the anisotropy of the cell layer refractive index can take values in the range from 0.002 to 0.07, which are significant values for linearly polarized light passing through a plant leaf. Therefore, when linearly polarized light interacting with the epidermis layer, the light is converted into elliptically polarized light, which, proposedly, should be more efficiently absorbed by the light-sensitive molecules chromophores lying deeper relative to the parenchyma cells. Thus, understanding of the polarized light interaction with plant leaves could help in choosing the best conditions for plant cultivation.

Our work purpose was to study the ability of the leaf tissue of monocotyledon maize plants to change the state of polarization of laser radiation, as well as the factors affecting this ability.

2 Material and methods

2.1 Optical setup

To study the optical properties anisotropy of the maize leaves, the optical setup shown below was used (Fig. 1). In the setup, radiation from a He-Ne laser (1) with the 632.8 nm wavelength passes through a $\lambda/4$ plate (2) and is expanded by the lens (3) to the beam with 5 mm diameter. Next, the light beam passes through the attenuator (4) to control the irradiance level of the sample (5). After that, the light beam is collected on the polarimeter (PAX5710 Thorlabs, USA) (6) to analyze the radiation polarization state.

The samples studied were rectangular fragments of maize leaves with 15 × 15 mm in size. The size was selected taking into account that the defocused laser beam did not go out the sample during its rotation. For measurements, the sample was fixed between two cover slips. The plant leaf samples were placed

between two glass plates 0.15 mm thick. For correct calibration and interpretation of the measurement results, measurements of the radiation polarization state were carried out with and without a plant leaf sample between the glass plates. The results of measurements of light transmission through glass were taken as control. The results of polarization measurements were processed using the TXP Series Instrumentation software (Starter, Server Control, TPX Polarimeter), USA.

The main parameter in the analysis of the polarization state was the ellipticity index k (Tatarinov et al. 2012). For linearly polarized light it is equal to 0° , and for circularly polarized light it is equal to 45° .

2.2 Plant material

To study the change in the light polarization state the seeds of maize (*Zea mays* L.), variety “Early gourmet 121” were used. This is an early ripe, high-yielding variety of sweet maize, demanding on the quality of light. The choice of monocotyledon maize plants was determined by the shape of the epidermal cells of their leaves (Fig. 2), which, as a rule, is characteristic of the epidermal cells structure for monocotyledons.

Plant samples for research were prepared as follows. Maize seeds were soaked in distilled water for 24 hours. Then the germinated seeds were sown in pots (W 9 cm × H 10 cm, Sady Primor'ya, Ussuriysk, Russia) that were filled with the commercial universal growing substrate (N1:P1:K1, mg/L: 160–240:145–215:180–290, organic matter, mg/L: 35, pH 5.5–7, Terra Master, Novosibirsk, Russia). The plants (11 samples) were cultivated in phytoboxes for 60 days under warm white LEDs with the light intensity $300 \mu\text{mol m}^{-2} \text{s}^{-1}$, where the relative humidity was maintained at $70 \pm 5\%$ and a temperature of $24 \pm 2^\circ\text{C}$. Irrigation was provided one time in three days.

3 Results

The microscopic photograph of the maize epidermis layer cells is shown in Fig. 2. As it possible to see, the cells in the epidermis layer are arranged in an orderly manner and have the shape of rectangles with wavy walls. In compliance with the model proposed by Kulchin et al. (2021) and in accordance with the measured cell parameters, the mean value of the refractive index anisotropy of the maize leaves epidermis layer should be $\delta_n = 0.016$, which indicates a rather high order of the epidermal layer optical activity.

The results of measurement of the laser radiation anisotropy passed through the maize leaves, which were 7 days old, are shown at Fig. 3. The plots show the mean values of five samples radiation ellipticity measurements. The observed ellipticity parameters indicate a high anisotropy of the maize leaves epidermis layer. It can be seen from the aforementioned dependences that the linear polarization ellipticity of the light passed through the of epidermis cells layer of the 7-day-old maize leaves varies within $\pm 12.0^\circ$. The circular polarization ellipticity varies within $\pm 2.5^\circ$. The linear and circular polarization ellipticity of the light passed through the glass plate varies within $\pm 0.5^\circ$.

3.1 The age impact

To study the maize leaves age impact on the ability to change the degree of light polarization, 5 maize plants samples aged 7, 19, 28, 49, and 60 days were used. Photographs of the epidermal layer cells are shown at Fig. 4. As can be seen from Fig. 4, the shape of the epidermis cells changes slightly with age, but in younger plants, the epidermis cells are more elongated.

The maize epidermal cells mean sizes of different ages, as well as the calculated mean values of the refractive index anisotropy, are presented below (Table 1). The table also shows the ellipticity change values for linear and circular polarization in plants of each age.

Table 1 Mean sizes of maize cells of different ages and polarization indices

Age, days	Mean values				
	Length, μm	Width, μm	δn	Ellipticity change (polarization), degrees	
				Linear	Circular
7	113,13	26,46	0,016	24,0	5,0
19	91,67	34,22	0,010	5,0	2,0
28	72,07	35,95	0,007	5,0	3,0
49	88,75	29,49	0,012	20,0	3,0
60	86,95	25,76	0,015	15,0	2,0

According to Table 1, the leaf epidermis layer of young maize plants shows the larger refractive index anisotropy, the value of which decreases slightly with age. This, apparently, is due to the greater mechanical anisotropy of cells that occurs in developing plants.

Figure 5 shows the dependences of the ellipticity parameter change in the plants of different ages using linearly and circularly polarized light.

As it follows from the dependences comparison, the light ellipticity changes according to the harmonic law in maize leaves samples of different ages. In this case, the maximum values of these fluctuations reach the value of ± 12 degrees. Consequently, at the given relative position of the sample and the radiation intensity vector, light with the highest polarization ellipticity penetrates to the inner leaves cells. Such light is able to most effectively interact with photosensitive protein structures. It can be noted that the largest values of the radiation ellipticity change, as a rule, are observed in samples with the highest values of the refractive index anisotropy.

According to Fig. 5, samples of each age showed greater ellipticity index change with linearly polarized light passed through rather than with circularly polarized light. Linearly polarized light passed through

maize leaf, is able to transform into elliptically polarized light with ellipticity index values within $\pm 12.0^\circ$, while the changes in ellipticity for circularly polarized light were within $\pm 2.5^\circ$. In addition, the highest ellipticity index values were observed in samples aged 7, 49, and 60 days.

The results obtained allow us to conclude that the epidermal layer age of the maize leaves affects the ellipticity of the circularly polarized light, but to a small extent. The layer does not affect the amplitude of changes in ellipticity depending on the sample rotation angle, but changes the ellipticity index value. Figure 5 shows that the ellipticity of plants aged 19 and 28 days varies from the values of 43° within $\pm 2.0^\circ$. The ellipticity indices of plants aged 7, 49 and 60 days vary from 33° to within $\pm 2.5^\circ$. This difference is presumably due to the higher cell size values ratio (width/length) in comparison with the plants aged 7, 49 and 60 days. Therefore, more elongated epidermal cells demonstrates the higher ellipticity index of circularly polarized light. This fact presumably indicates that at the initial stage of development, the maize plant is most susceptible to polarized light, which can positively affect its development.

4 Discussion

It is known that the pigments responsible for the absorption of light energy in plants, such as chlorophylls, carotenoids, flavonoids, are provide photosynthesis (Kulchin et al. 2020). Photoreceptors, consisting of the apoprotein and its associated chromophore, which directly serves as an antenna that receives electromagnetic radiation lying in any of the visible range regions, are responsible for receiving light signals that ensure the regulation of plant development (Golovatskaya 2016). A completely set of such photosensors, spectrally complementing each other, ultimately allows plants to receive signals from the entire visible radiation band. The light energy absorbed by chromophores at different wavelengths further converted into conformational changes using various mechanisms, which as a result leads to corresponding changes in plant development (Glantz et al. 2016; Kneuttinger 2022).

It has been established that in nature biomolecules, for example, proteins and sugars, are characterized by homochirality (Potapov 1988; Davankov 2018). This means that there is only one of two possible symmetrical mirror versions of their development. Plant chromophore proteins can have a chiral structure, therefore, as a result, they are able to show circular dichroism. Thus, depending on the elliptically polarized light, they can absorb light differently. In addition, the presence of chirality in plant chromophore proteins should lead to a different response of the light-regulating apparatus of plant cells to radiation with elliptical polarization.

Previously, Kulchin et al. (2021, 2023) established that the epidermis of plant leaves can change the state of linearly polarized radiation into elliptically polarized. Therefore, we can assume that plants are able to respond to polarized radiation, which is probably observed in the morning and evening hours of daylight under natural conditions.

To date, there are several works devoted to verifying the latter fact. Thus, Lkhamkhuu et al. (2020) found that red light with right circular polarization promotes the germination of lettuce and *Arabidopsis thaliana* seeds, as well as greater values of weight in white light. At the same time, hypocotyl elongation

under red light with such polarization was slower in comparison to left-twisted light. Shibayev and Pergolizzi (2011) established that the *Pisum sativum* and *Lens culinaris* growth was most stimulated by light with circular polarization twisted to the left. Anderson (2018) found that the *Brassica napus* growth was more favored by light with left circular polarization compared to the irradiation variant with right twisted circularly polarized light. In the case when *B. napus* being irradiated with linearly polarized light, the result of the stimulating effect was much inferior to the variant with left circular polarization. This suggests that light with a greater polarization ellipticity may have a greater physiological effect on plants.

As it was established by Kulchin et al. (2021, 2023), when linearly polarized light passes through the epidermis of the *A. cepa* L. skin and the leaves epidermis cells of the monocotyledonous *P. arundinacea* plant, the radiation polarization ellipticity changes. At the same time, Kulchin et al. (2023) found that when linearly polarized light passed through the leaves epidermis cells of the dicotyledonous plants, no change in the polarization ellipticity is observed. Probably, the absence of a polarization change could be explained by the fact that monocotyledons have differences in the epidermal cells structure and shape compared to dicotyledons.

Besides epidermis monolayers that cover leaf from above and below, leaves have two more cell layers of photosynthetic tissue inside: the palisade and spongy mesophyll (Borsuk et al. 2022). The first contains the main photosynthetic cells of a cylindrical shape, optimally positioned to absorb light, and the second consists of loosely packed irregularly shaped cells with a significantly smaller number of chloroplasts (Lotova 2007). When radiation propagates inside the leaf tissue, scattered light arises, the polarization state of which depends on the properties of the leaf surface and does not depend on its internal structure (Vanderbilt et al. 2014).

Optical anisotropy in the leaves epidermis layers develops because of cells directional deformation, which explained by their mechanical stretching during growth (Bringmann and Bergmann 2017). The plant leaves epidermis cells size can vary from 10 to 300 μm , the anisotropy of cells size deformation can reach 10–30%, and the cells membrane thickness can vary from five to 30% of the longitudinal cells size. The shell of plant leaf cells has a refractive index in the range of 1.40–1.50 (Landry et al. 2011; Lehmuskero et al. 2018; Shackelford 2000; Tamada et al. 2014). The main cell component is the protoplast, which is an aqueous solution of carbohydrates, proteins, amino acids, organic acids and their salts, mineral ions, alkaloids, glycosides, pigments, tannins, etc. Because of this, the refractive index of the internal contents of the cell can vary from 1.34 to 1.42 (Hassani and Kreysing 2019; Liu et al. 2016).

Study of the maize leaves of different ages showed that when linearly polarized light passes through the cells epidermal layer, it is transformed into elliptically polarized light. This is explained by the refractive index anisotropy in the cellular structure of the leaves epidermis, which occurrence, apparently, is due to the epidermis layer structure. The results obtained confirm the previously obtained results by Kulchin et al. (2023) for *P. arundinacea*, which epidermal layer is also capable of significantly changing the polarized radiation ellipticity index.

We studied how the light polarization ellipticity changes in two different monocotyledons plants when polarized light passes through the leaves. The results obtained shows that plant leaves with an ordered structure of the cells epidermal layer are able to change the light polarization state. Given the fact that light-absorbing plant proteins have chiral chromophores that predominantly absorb light with a certain direction of twisting the polarization angle, and the epidermis layer ability to change the ellipticity, it could make the light interaction process with deeper plant cells more efficient. For example, Anderson (2018) established that *B. napus* plants developed better under circularly polarized light twisted to the left. However, the study results were somewhat worse than those of control samples grown under unpolarized light. This could be explained by the fact that *B. napus* is the dicotyledonous plant that does not respond to polarized light.

If linearly polarized light falls on maize plants, this can affect the rate of plant development, especially at an early age. The light passes through the upper active layers of the leaf epidermis, making it elliptically polarized, which allows better interaction with the cells under the epidermal layer. These effects often occur at mornings and evenings, when linearly polarized radiation reaches the Earth's surface (Zvereva 1988).

Our study established that the plant age is able to affect the ellipticity index of linearly and circularly polarized light. Consequently, the use of polarized light for growing crops is most justified during certain time periods of plant development.

We studied the polarization ellipticity dependence of the radiation passed through the epidermal layer of maize leaves irradiated with low-power laser radiation with linear and elliptical polarization. The results obtained showed that maize leaves have optical activity and are able to change the light ellipticity. This is explained by the ordered structure of the cells epidermal layer, which has the refractive index anisotropy. The results obtained confirm the previously proposed model by Kulchin et al. (2021), which describes the interaction of polarized laser radiation with plant leaves.

The ellipticity and azimuth changes dependencies from the maize age also were studied. It was found that both young and old plants have leaves capable of changing the light polarization state to varying degrees.

In addition, the relationship between the maize age and the polarization ellipticity change was studied. It was found that the leaves of different ages are capable of changing the polarization state of linearly and circularly polarized light to varying degrees. The leaves epidermal layer at the initial stage of development had the highest refractive index anisotropy and, consequently, the highest ellipticity indices.

The processes of transformation of linearly polarized light into elliptically polarized light by the maize leaves epidermis layer have been studied. The data obtained suggests that such a transformation contributes to a more efficient interaction with radiation and can favorably affect plants development.

Declarations

Author contributions

Conceptualization: Yuriy N. Kulchin, Evgeny P. Subbotin; Methodology: Yuriy N. Kulchin, Evgeny P. Subbotin, Sergey O. Kozhanov, Vadim V. Demidchik, Yuriy V. Trofimov; Formal analysis and investigation: Evgeny P. Subbotin, Sergey O. Kozhanov, Alexander S. Kholin, Natalia I. Subbotina; Writing – original draft preparation: Yuriy N. Kulchin; Writing – review and editing: Sergey O. Kozhanov, Evgeny P. Subbotin, Natalia I. Subbotina, Alexander S. Kholin, Vadim V. Demidchik, Yuriy V. Trofimov, Kirill V. Kovalevsky, Andrey S. Gomolsky; Funding acquisition: Yuriy N. Kulchin, Evgeny P. Subbotin, Kirill V. Kovalevsky, Andrey S. Gomolsky; Resources: Evgeny P. Subbotin, Kirill V. Kovalevsky, Andrey S. Gomolsky; Supervision: Yuriy N. Kulchin, Evgeny P. Subbotin. All authors read and approved the final manuscript.

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Conflict of interests

The authors declare that they have no conflict of interest.

Financial or non-financial interests

The authors have no relevant financial or non-financial interests to disclose.

Competing interests

The authors have no competing interests to declare that are relevant to the content of this article.

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Figures

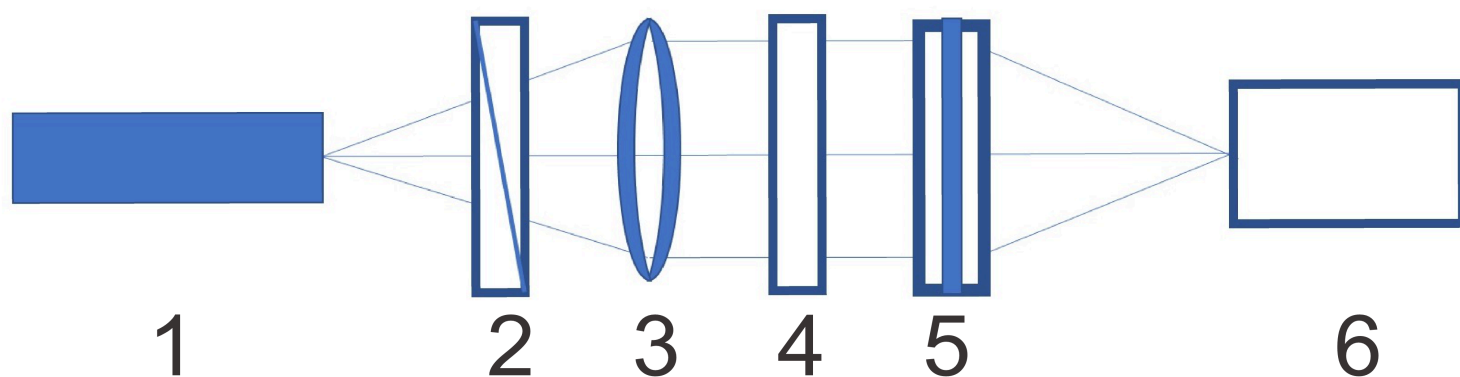


Figure 1

Optical scheme of the experimental setup: 1 – 633 nm laser; 2 – plate $\lambda/4$; 3 – collimating lens; 4 – attenuator; 5 – test sample on the rotator; 6 – polarimeter (PAX5710 Thorlabs, USA)

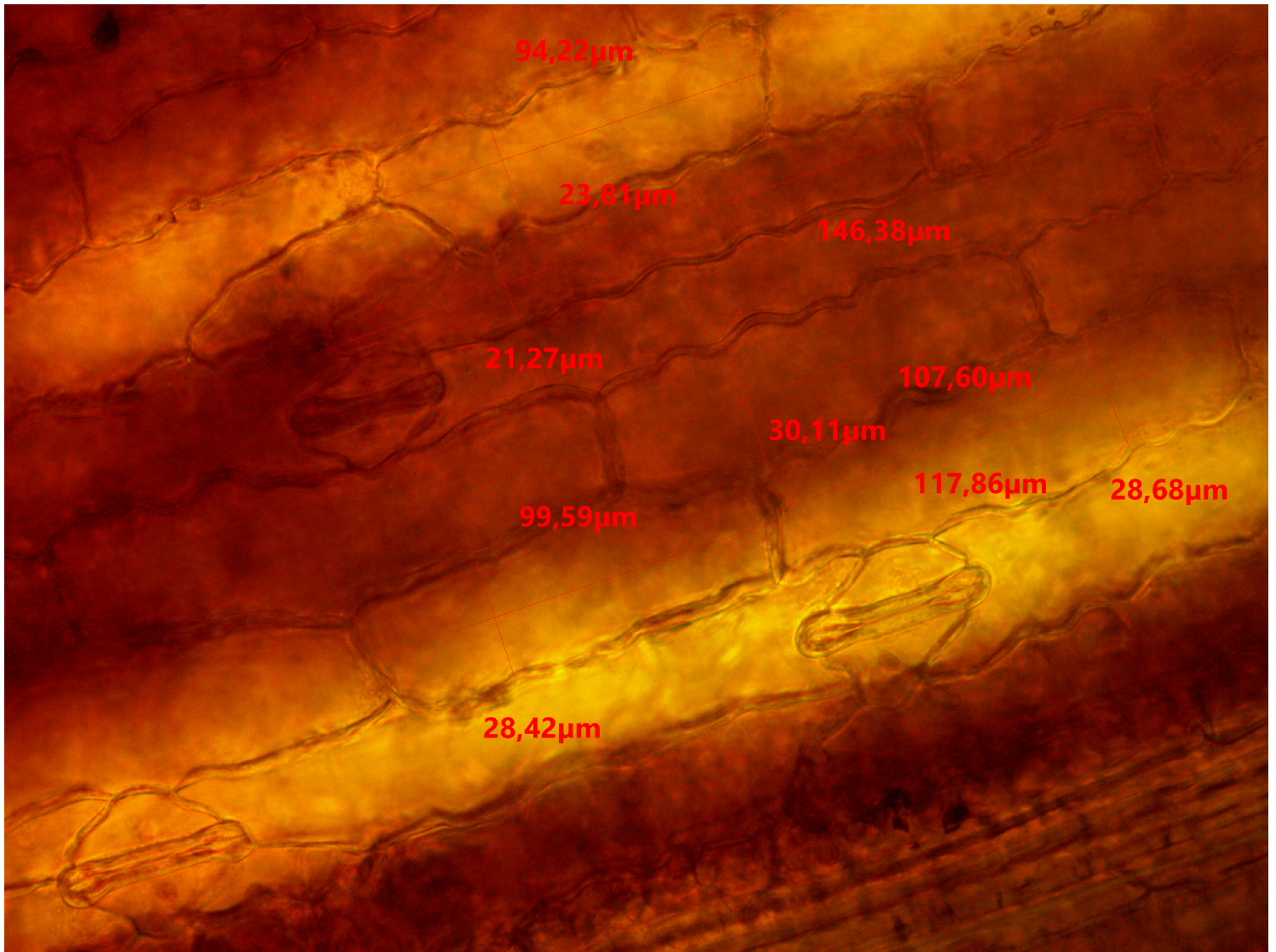


Figure 2

Microscopic photograph of the cell structure of the epidermis of maize leaves (numbers show the values of cell sizes in the longitudinal and transverse directions)

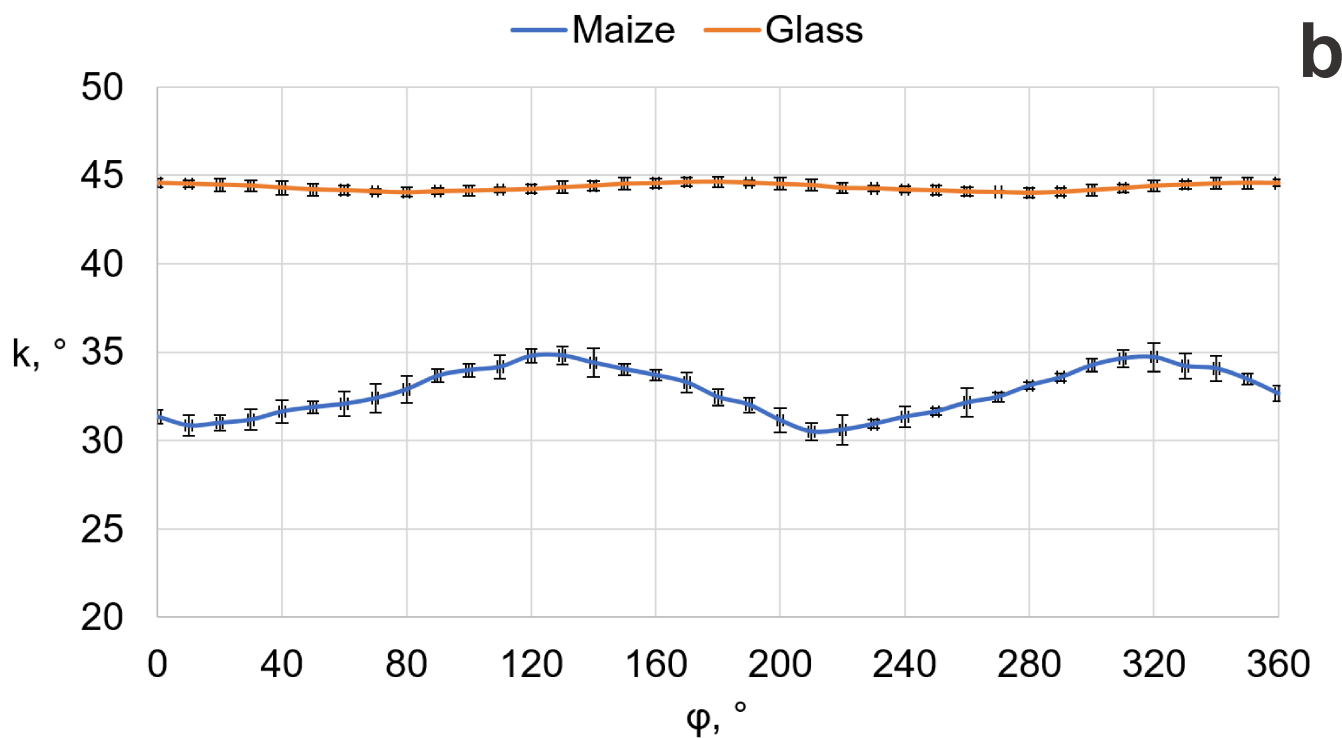
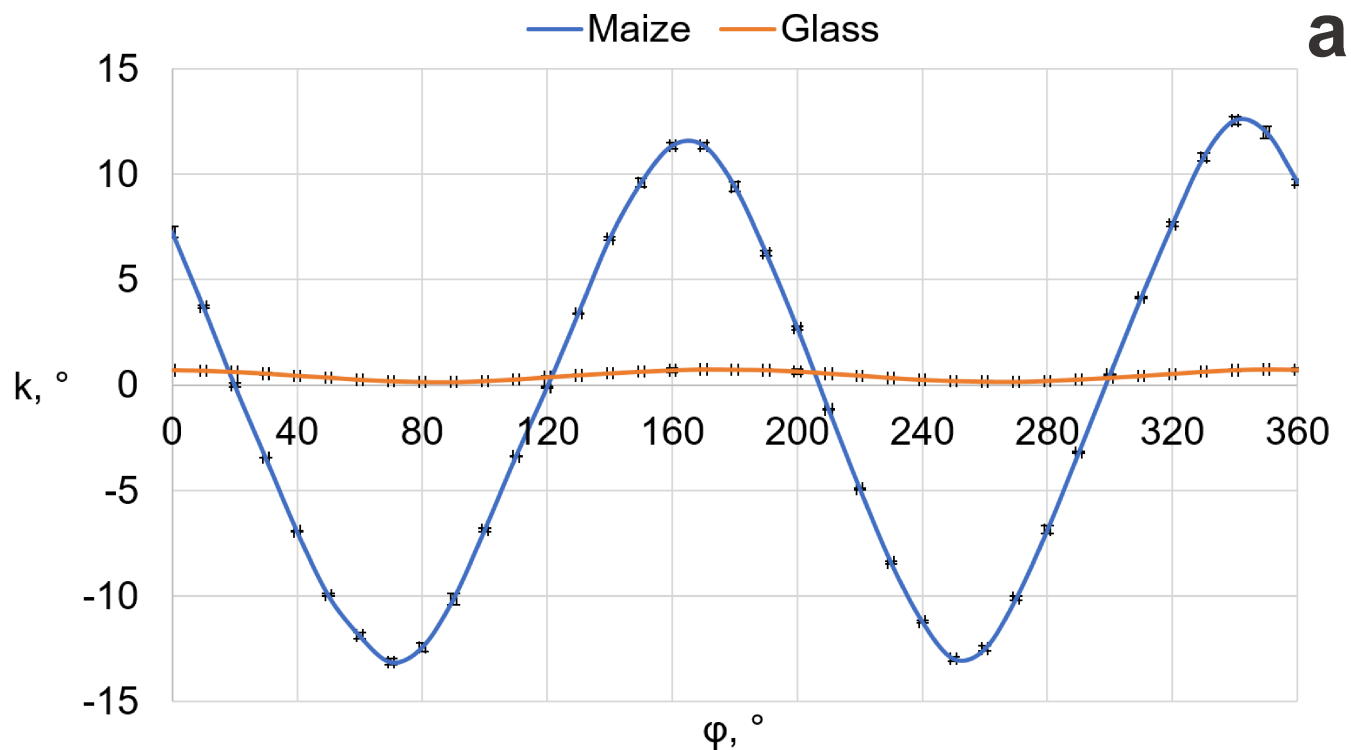


Figure 3

Results of measurements of linear (a) and circular (b) polarization ellipticity of light passed through maize leaves samples and clear glass plates

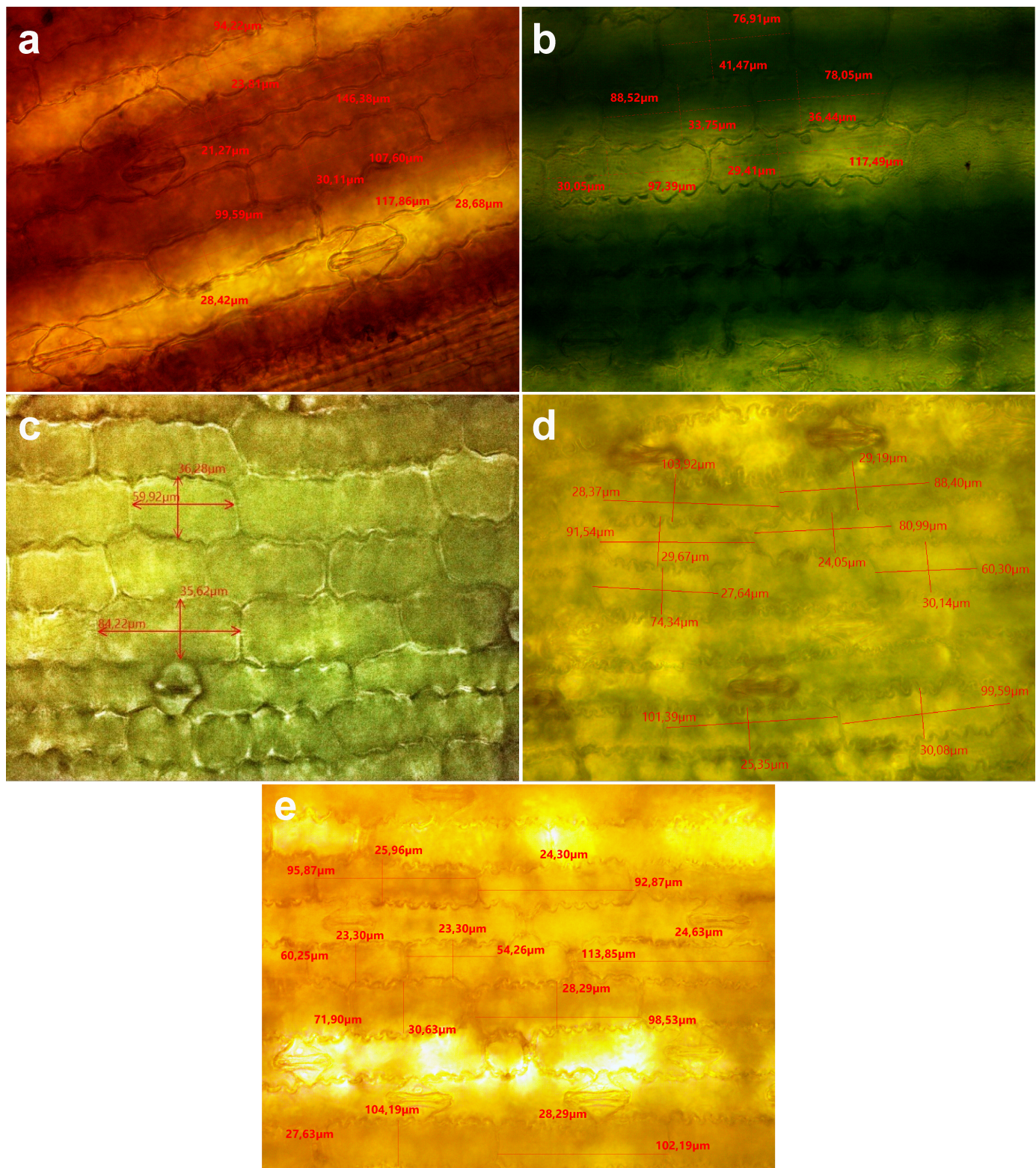


Figure 4

Epidermal maize cells of different age in order from left to right and top to bottom – 7 (a), 19 (b), 28 (c), 49 (d), and 60 (e) days old

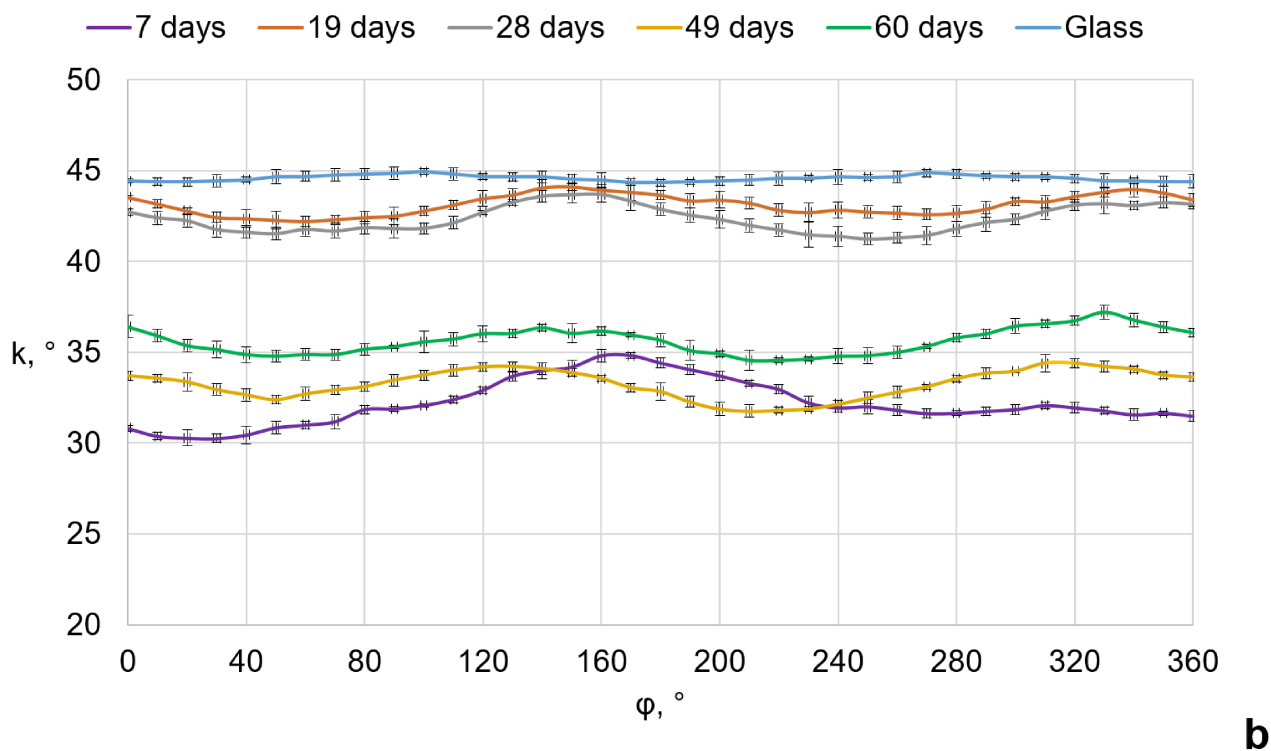
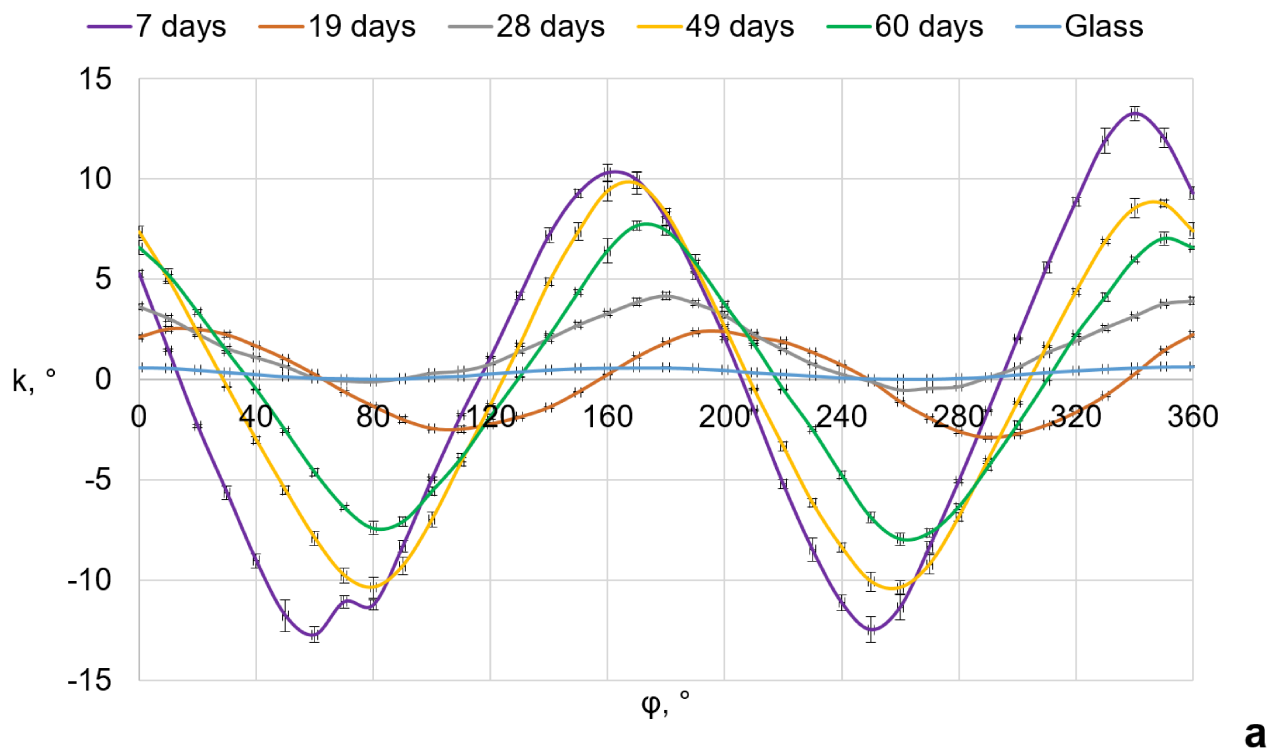


Figure 5

Dependences of the ellipticity parameter change from the samples rotation angle for maize of different ages under irradiation with linearly (a) and circularly (b) polarized light