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THE EFFECT OF COVERING WITH A TRANSPARENT PLASTIC SHEET, ON THE TISSUE STRUCTURE OF THE LEAVES OF BEAN PLANTS

SZERÉN M., PATAKY and I. HORVÁTH

Department of Botany, Botanical Garden, Attila József University, Szeged

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Abstract

Field experiments demonstrated that covering plants with colourless and orange plastic sheets resulted in considerable increases in the dry weight of shoots, and the crop, and changes in the tissue structure of the leaves. There is a close connection between the ratio of spongy parenchyma and the dry weight of shoot. As a consequence of being covered, the cells of the palisade parenchyma became nearly isodiametrical, the number of stomata and the thickness of epidermis are reduced. Underneath the orange sheet, the amphistomatic leaves became almost hypostomatic.

Introduction

Among the plant tissues, the structure of the assimilating parenchyma of the leaf and that of the epidermis are the most influenced by light. The effect of light is considerable because there is a close relationship between the tissue structure of the leaf and the organic-matter production.

It was established by HESSELMANN, as early as 1904, that the leaves of southern exposure of a shrub *Corylus avellana* L., developed under natural conditions, were thick, with a double layer of palisade parenchyma. Inside the shrub, where the intensity of illumination was ten times lower, leaves were much thinner, and the palisade parenchyma consists of a single layer of cells.

It was demonstrated by us, too, in our earlier investigations (PATAKY, 1969), that within the foliage of *Salix alba* L., there are significant changes in the tissue structure of the leaves as a result of changing light conditions.

There are several other reports demonstrating that the tissue structure of the leaf is generally related to the intensity of illumination, from ferns up to the flowering plants:

GRAHL & WILD (1973) observed two different phenotypes of *Sinapis alba* L., as a result of light intensity. Under intense light the plants developed a thick mesophyll with a stratified palisade parenchyma and a thinner spongy parenchyma.

A still more considerable change in the mesophyll of the leaf was observed by STARZECKI (1958). The mesophylls of the leaves of *Asplenium trichomanes* L. and *A. ruta-muraria* L., when grown in full sunshine, were well-developed and consisted of both palisade and spongy parenchymas. The leaves of plants, living in a cave, 20 m from the entrance, were thin and their mesophyll consisted only of spongy parenchyma.

TAKÁCS (1973), using *Lactuca sativa* L., *Spinacia oleracea* and *Rumex acetosa* L., established that, as a function of light intensity, leaves of different thickness developed and the proportions of palisade and spongy parenchymas changed. The structure of the epidermis is also influenced by light intensity. Shenikov's establishment (1953), according to which, reduction of the intensity of illumination results in enlargement of the epidermal cells, reduction in the number of stomata, shortening of the vascular system of the leaf, and the cell wall becoming more winding: is generally accepted.

In this paper, the effect exerted on the leaf-tissue structure by covering the plant with different nets of mesh, is investigated. Primarily, the structures of mesophyll and epidermis have been followed.

Materials and Methods

The investigations were performed using beans (*Phaseolus vulgaris L.* — a kind of dodder). Sowing took place on July 2, 1973, in the Botanical Garden of the Department of Botany, Attila József University, Szeged, in three repetitions, 2×3 m plots, in a random block arrangement. Row and stem distances were 30 cm.

Three plots were covered with colourless and three with orange plastic sheets. The plots remained covered from sowing until elaborating the material.

According to our earlier investigations of micro-climate (HORVÁTH, 1965), the air and soil temperatures, as well as the relative air humidity are not changed appreciably by a similar covering. The intensity of illumination was reduced by the colourless sheet by about 15 per cent, and by the orange one by 25 per cent. The distribution of spectral energy was not changed by the colourless covering, but the orange covering resulted in reduction of the energy falling in the shorter wave range and increased that falling in the longer one. While the percentage of energy falling in the orange and red wave ranges in the control plots and those covered with the colourless sheet was 39 per cent, that under the orange sheet was 49 per cent (Table 1).

Although not measured, it is probable that the carbon dioxide concentration of the air was increased by the covering, because the air movement was reduced.

Table 1

treatment	violet	blue	green	yellow	orange	red	per cent
control	6	20	27	8	14	25	
colourless plastic sheet	6	19	27	9	13	26	
orange plastic sheet	4	15	24	8	17	32	

The morphological and phenological investigations were carried out on August 8 and 29, and September 7 and 17, 1973, using ten plants from each plot. The leaf surface was calculated by taking into consideration the fresh weight and surface of five leaves from each plot, as well as the fresh weight of all the leaves of the plants collected. Leaves were collected for the histological examinations from the middle region of the stem, on September 7, 1973. The examinations were made from the central part of the middle leaflet of the triple leaf. The material collected was fixed in 60 per cent alcohol and macerated directly or embedded in celloidin for sectoining.

Under the light-microscope, the thickness of the spongy and palisade parenchymas of the leafl, and the length and breadth of the cells of the palisade parenchyma, were measured. From the cell count of the stoma and epidermis, a stomatic index was calculated. In every case, the arithmetical mean of fifty measurements was taken.

The connection between the tissue properties investigated and the production of organic matter was analysed, using a correlation calculation.

A survey and evaluation of results

The main aim of our work was to analyse the connection between the tissue structure of leaves and the organic-matter production. Our results concerning the dry weight of the shoot of a plant, on the basis of a survey on September 7, 1973, are given in Table 2.

Table 2

treatment	dry weight (g)			leaf-surface (sq. cm)
	shoot	crop	total	
control	10.8	3.6	14.4	952.2
colourless plastic sheet	12.5	7.5	20.0	1723.8
orange plastic sheet	14.7	10.0	24.7	2517.2

The data of September 7, 1973 were chosen because it was established during the morphological-phenological elaboration of the material (KÉRI, 1975) that the effects were increasing with time. In the last elaboration, however, the quantity of crop only was given exactly, because many of the leaves had already fallen.

It can be ascertained that the dry weight was increased by covering considerably; this increase was about 75 per cent under the influence of the orange net.

The difference in dry weight first appears in the crop — approximately 200 per cent — and in practical relation the importance of this way of covering is also increased.

Before reporting on the influence exerted on the tissue structure of the leaf, we notice that the leaf surface was considerably increased by covering. As compared with the control, the increase under the colourless net of foil mesh was 90 and under the orange one 170 per cent. The effect can only partly be explained by the lower intensity of illumination. An important factor is — on the basis of our earlier investigations (HORVÁTH, 1965) — also the distribution of spectral energy.

The correlation between leaf surface area and dry weight is positively linear. This is, therefore, not influenced by either light intensity or the distribution of spectral energy (Fig. 1).

Table 3

treatment	thickness of the mesophyll (μ)			percentage		cells of the palisade parenchyma (μ)		
	spongy	palisade	total	spongy	palisade	length (l)	breadth! (b)	l/b
			parenchyma					
1	89.0	103.7	192.7	47	53	24	4.3	
2	103.0	81.4	184.4	56	44	81.7	23.3	3.5
3	90.0	58.3	148.3	63	37	58.4	26.5	2.2

1 = control, 2 = colourless plastic sheet, 3 = orange plastic sheet

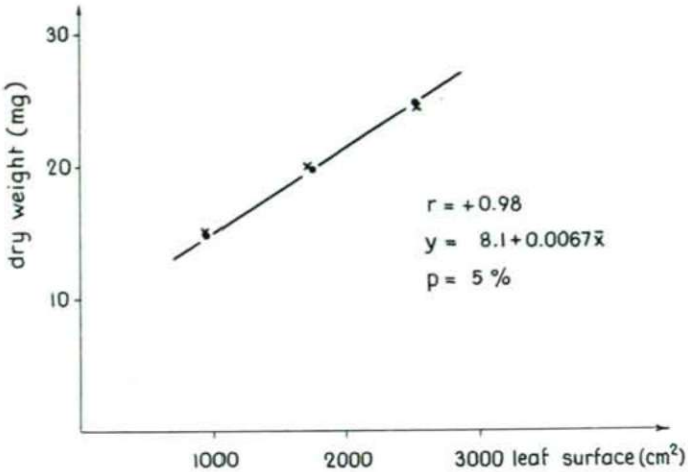


Fig. 1. The results of our histological investigations are summarized in Table 3.

It should be noted that, as a result of covering, the thickness of the leaf-blade is diminished. The diminution caused by the colourless sheet is slight, not more than 4 per cent, but that caused by the orange one is 17 per cent.

This effect — as well as other tissue changes to be discussed below — cannot be explained only by the reduction in light intensity. It is rather to be attributed to a change in the distribution of spectral energy.

The leaves of plants grown in the phytotron in an earlier experiment (HORVÁTH, 1975) in orange light, were thicker than those of plants grown in white, blue or red light. This may be interpreted on an ecological basis, because the light rays belonging to the orange wave range are absorbed to a comparatively lesser extent, ensuring sufficient light energy even in the lower cell-layers of a thick leaf, while the blue and red rays are absorbed to a higher degree, mostly by the upper layers of the leaf mesophyll.

The mesophyll of the leaf of bean consists of the unistratal, well differenctiated palisade parenchyma of heterogeneous structure, and the multi-layer spongy parenchyma.

The cells of palisade parenchyma are columnar, their length-breadth ratio (l/b), is 5 to 1. The spongy and palisade parenchymas comprise approximately identical proportions of the mesophyll.

Under the influence of being covered with plastic sheets, the shape of the cells of palisade parenchyma has considerably changed. They have become less "columnar". This was most evident under the orange sheet where the length/breadth ratio (l/b) was reduced to 2.2. Light is, therefore, one of the main factors involved in the formation of the palisade parenchyma. This has also been shown out by STARZECKI (1962), according to whose statement the main form of the assimilating tissue is the palisade parenchyma. The palisade parenchyma is expressed in leaves receiving intensive light. From among the two kinds of assimilating parenchymas, the spongy parenchyma is, therefore, more general and important.

This is also indicated by our investigations, because the ratio of spongy and palisade parenchymas is considerably shifted — as a result of being covered with the plastic sheets — towards the spongy parenchymas. In the uncovered plots, the proportion of spongy parenchyma was 47 per cent, while under the orange nets it was 63 per cent.

The ratio of spongy parenchyma and the dry-matter production calculated for a single plant are also closely connected (Fig. 2) : $r=0.99$, and the reliability of the connection is : $P=0.01$ per cent.

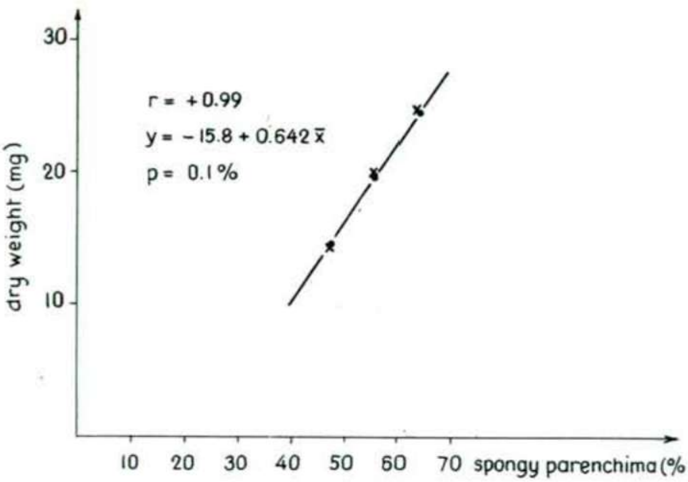


Fig. 2

The change taking place in the structure of the mesophyll is still more considerable if the absolute values of the thicknesses of spongy and palisade parenchymas are taken into consideration. While the thickness of the spongy parenchima has, despite the decrease in the thickness of leaf-blades, changed to only a small extent (Table 3), the thickness of the palisade parenchyma was reduced almost by half.

As a result of the change in form of the cells of the palisade paranchyma, the structure of the mesophyll became more homogeneous, and this may primarily be attributed to the decrease in light intensity. This is also suggested by the comparatively close positive correlation between the change in length-breadth ration and the intensity of illumination (Fig. 3).

The leaves of beans are amphistomatic. The stomata are, apart from a few exceptions, of paracytic type (VAN COTTHEIM 1971), occurring sporadically in the islands of leaf veins. Their number on the upper surface is considerably smaller than on the lower surface. On both upper and lower epidermes, there are to be found unicallular coating hairs and polycellular glandulair hairs. The radial wall of epidermal cells meanders, and the number of cells is considerably lower on the upper surface than on the lower one.

The effect of being covered with the plastic sheets manifests itself first of all on the upper-surface epidermis, and is considerable particularly on the number of stomata. The stoma count is greatly reduced, particularly by the orange covering.

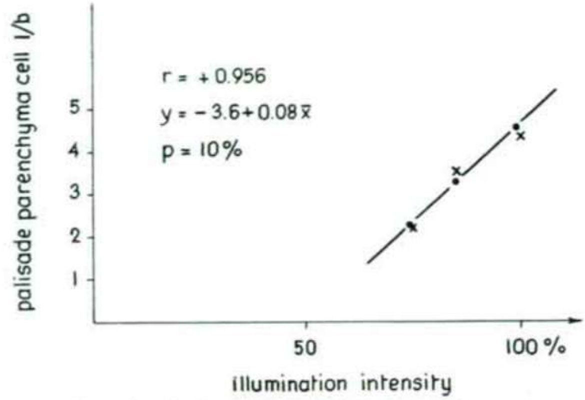


Fig. 3. The changes taking place in the structure of the epidermis are summarized in Table 4.

Table 4

treatment	stoma count sq. mm upper	lower	epidermal cell count sq. mm upper	lower	stomatal index upper	lower
			surface			
control	55.3	375.0	506.9	1066.4	9.8	26.0
colourless plastic sheet	34.5	273.9	433.5	920.2	7.3	22.9
orange plastic sheet	3.9	248.9	329.7	906.9	0.2	21.6

The leaf becomes nearly hypostomatic. The epidermis cell count at the upper surface is also reduced, *i. e.,* the basis cells of the epidermis became larger. The effect on the lower-surface epidemis is smaller. The reduction in the number of epidermal basic cells and in that of stomata can be ascribbed to the decrease in light intensity. A considerable change in the stomatal index, however, is rather to be considered as an effect of the distribution of spectral energy.

* * *

The influence of the covering of colourless or orange plastic sheets, upon the tissue structure of the leaf and the dry weight of the shoot, investigated in field experiments. The intensity of illumination was reduced by 15 and 25 per cent, respectively, as a result of covering. The distribution of spectral energy was also changed by the orange plastic sheets; the percentage of the energy falling to the orange and red wave ranges, rising from 39 to 49 per cent.

It is to be established as a result of our investigations that:

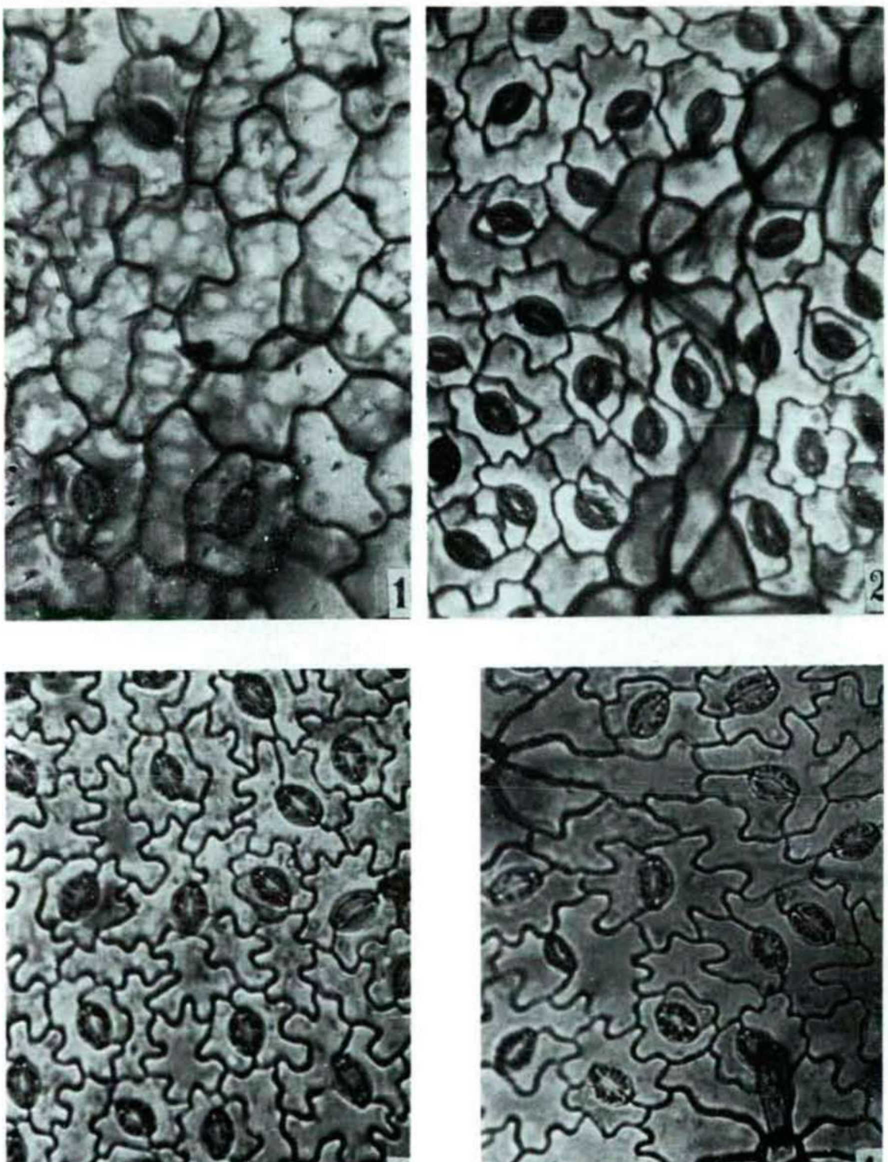
1. The dry weight of shoot and crop, as well as the leaf surface, were considerably increased by covering.
2. The thickness of the leaf-blade was reduced and the proportion of spongy

Plate 1. Upper and lower epidermis of *Phaseolus vulgaris L.* ►
(Magnified ×280)
1. Control, upper epidermis
2. Control, lower epidermis
3. Covered with a colourless plastic sheet, lower epidermis
4. Covered with an orange plastic sheet, lower epidermis.

parenchyma increased. The connection between the proportion of spongy parenchyma and the dry weight is close.

3. The shape of the cells of the palisade parenchyma changed, as they became nearly isodiametrical.
4. The number of stomata on the upper surface epidermis, and the epidermal cell count, were reduced. As a result of the orange covering, the amphistomatic leaves became almost hypostomatic.

Plate 1



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Address of the authors:

DR. SZERÉN PATAKY
Prof. DR. I. HORVÁTH
Department of Botany, A. J.
University, H-6722 Szeged, P. O. Box 428,
Hungary