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Research article

ALTERATIONS IN PHOTOSYNTHETIC ELECTRON TRANSPORT ACTIVITIES OF MAIZE THYLAKOID MEMBRANES UNDER THE INFLUENCE OF HIGH TEMPERATURE.

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ABSTRACT: High temperature treatment (30-50^{o}) induced alterations in photosynthetic electron transport activities of maize leaves in a preferential manner. From the electron transport measurements it is clear that high temperature causes inhibition in photosystem (PS) II catalyzed electron transport and enhances the PS I catalyzed electron transport activities. The possible reasons for the loss of PS II catalyzed activity could be alterations at water oxidation complex which is evidenced for whole chain catalyzed electron transport measurements. The reason for the enhancement of PS I catalyzed electron transport could be opening of new site for reduced ascorbate donation after Cyt b_6f.
Key words: Electron transport, High temperature, Maize thylakoids

INTRODUCTION

Exposure of the plants to the fluctuations in atmospheric temperature leads to the loss of functional ability of photosynthetic apparatus [1and 2]. The inhibition in whole leaf photosystem by high temperature has been found due to disruption of photosynthetic electron transport at chloroplast or thylakoid membranes level [3, 4 and 5]. Among other biological membranes, thylakoid membranes are more sensitive to heat stress [6]. The exposure of thylakoid membranes to the high temperature leads to the loss in electron transport activity and finally leads to the damage to photosystems [1 and 2]. Upto now studies related to the effect of high temperature on thylakoid membranes under *in vitro* condition are scanty. Therefore in this investigation an attempt has been made to characterize the effect of high temperature by isolating the thylakoid membranes from maize leaves and then exposing them to the high temperature ranging from 30- 50^{o} for 5min. The effect of high temperature was studied using O_2 electrode in terms of partial electron transport measurements to identify the target site in photosynthetic apparatus in maize thylakoids.

MATERIALS AND METHODS

Maize seedlings were raised on petri plates under continuous white light (16 Wm^{-2}) at 25^{o}C. Half strength Hoagland solution was supplied at 4-day intervals to the seedlings. Thylakoids have been isolated from 8-day old seedlings and they were exposed to various elevated temperatures (30-45^{o}C) for 5min in low light (1Wm^{-2}).The chloroplast isolation and the polarographic measurement of the partial photochemical activities were done as described earlier [7] with slight modifications. The leaves were homogenised in 25mM HEPES isolation buffer (pH 7.5) containing 400 mM sucrose, 10mM $MgCl_2$ and 5mM KCl. The assay mixture for the measurement of whole chain electron transport activity contained 0.5mM MV and 1mM Na-azide. The assay mixture for the measurement of PS II mediated oxygen evolution activity contained 0.5mM PBQ in three ml of the 25mM reaction buffer (pH 7.8) containing 100mM sucrose, 10mM $MgCl_2$ and 5mM KCl. For avoiding water donation to whole chain electron transport the donar diphenyl carbazide (DPC) has been replaced. DPC supported whole chain electron transport activity was measured by using 0.5mM DPC, 0.5mM MV and 1mM Na-azide.PS I activity mixture contained 0.1mM DCPIP, 5mM ascorbate , 1mM azide, 0.005mM DCMU and 0.5mM MV. In all the assays chloroplasts equivalent to 30 µg was used. The assays were conducted at 25^{o}C under saturating light intensity (400Wm^{-2}). Chlorophyll (Chl) was estimated according to Arnon [8].

RESULTS AND DISCUSSION

In this investigation an attempt has been made to study the effect of high temperature on whole chain catalyzed electron transport. Control thylakoids exhibited the activity equal to 136 µmoles of O_2 consumed.[Table 1]. The increase in the temperature caused gradual inhibition in whole chain electron transport and at 40°C of incubation 52% loss was noticed. This inhibition in whole chain electron transport could be due to alterations in oxygen evolving complex (OEC) or loss of Mn ions or extrinsic polypeptides of water oxidation complex (WOE) [9 and 10]. In addition to this there is a proposal that the inhibition could be due to alterations in non approved regions [11]. To resolve the above propositions PS II catalyzed electron transport has been measured using PBQ as Hill acceptor [12]. High temperature treatment (30-45°) induced inhibition in PS II catalyzed electron transport and 40° treatment induced 55% loss in electron transport activity of thylakoid membranes [Table 2]. The reason for the loss of PS II activity could be loss of Mn ions or extrinsic polypeptide related to water oxidation complex as proposed by Havaux [13]. To strengthen our argument whole chain electron transport activity has been measured by a avoiding water oxidation complex by replacing a donor DPC.

This chemical can donate the electrons after water oxidation complex [4]. High temperature treatment brought marginal loss in whole chain electron transport activity supported by DPC [Table 3]. These results are in agreement with the observations of Yamashita's group [14] who showed that the donor side of PS II is possible target for UV-B as well as high temperature stress. To establish the effect of high temperature on PS I catalyzed electron transport, the electron transport has been measured using reduced dichloro phenol indo phenol as donor an MV as a acceptor ($DCPIPH_2$ →MV) [Table 4]. In chloroplasts isolated from HT treated leaves, a significant enhancement in the PS I activity was observed. The heat induced of PS I activity was due to opening of a new site of donating to PS I near a site close to cyt b_6f as suggested by Thomas *et al* [15]. There was also a suggestion by Sundby *et al*[16] explaining that migration of LHC II from PS II to PS I is responsible for the enhancement in PS I activity. Thus HT causes the damage to WOC in PS II and enhancement in electron transport activity at PS I due to opening of new site in intersystem electron transport chain in maize thylakoid membranes.

Table 1: Effect of temperature (30-45°C) on whole chain electron transport (H_2O → MV) of the maize thylakoid membranes

Temperature (°C)	Whole chain electron transport activity (H_2O → MV) µ moles of O_2 consumed mg Chl^{-1} h^{-1}	Percentage of loss
25	155 ± 13	0
30	147 ± 8	5
35	129 ± 10	17
40	74 ± 6	52
45	60 ± 4	61

Table 2: Effect of temperature (30-45°C) on PS II catalyzed electron transport (H_2O → PBQ) of the maize thylakoid membranes

Temperature (°C)	PS II catalyzed electron transport (H_2O → PBQ) µ moles of O_2 evolved mg Chl^{-1} h^{-1}	Percentage of loss
25	181 ± 16	0
30	168 ± 11	7
35	134 ± 10	26
40	81 ± 7	55
45	61 ± 5	71

Table 3: Effect of temperature (30-45°C) on whole chain electron transport (DPC → MV) of the maize thylakoid membranes

Temperature (°C)	Whole chain electron transport activity (DPC → MV) µ moles of O_2 consumed mg Chl^{-1} h^{-1}	Percentage of loss
25	136 ± 11	0
30	131 ± 9	4
35	124 ± 11	9
40	116 ± 7	15
45	109 ± 5	20

Table 4: Effect of temperature (30-45^{o}C) on PS I catalyzed electron transport (DCPIP H_2 → MV) of the maize thylakoid membranes

Temperature (^{o}C)	PS I catalyzed electron transport (DCPIPH_2 → MV) μ moles of O_2 consumed mg Chl^{-1} h^{-1}	Percentage of enhancement
25	347 ± 31	0
30	385 ± 28	11
35	434 ± 38	25
40	493 ± 39	42
45	548 ± 47	58

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