

ISSN 1000-0933
CN 11-2031/Q

生态学报

Acta Ecologica Sinica



第34卷 第10期 Vol.34 No.10 **2014**

中国生态学学会
中国科学院生态环境研究中心
科学出版社

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, 2014, 34(10): 289-298.

Du X K, Li J M, Zhong Z C, Dong M. Proteomic analysis of *Arachis hypogaea* leaf in response to ultraviolet-B radiation. *Acta Ecologica Sinica*, 2014, 34(10): 289-298.

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UV-B

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studies of the stress responses in plant species, including *Arabidopsis*, soybean, rice, wheat, maize, potato, tomato, and many other species. A wide range of abiotic stresses have been examined such as drought, nutrient deficiency, temperature, oxidative stress, herbicide wounding, anoxia, salt, and heavy metals. These investigations have provided a wealth of important information on the physiological processes involved in plant stress responses. To explore the molecular mechanisms of the decreased photosynthetic rate and the resistance of peanut (*Arachis hypogaea*) when exposed to natural UV-B radiation, 2-D PAGE and MS were used to identify the differentially expressed proteins in peanut seedling leaves in response to supplemental UV-B radiation (4 $\mu\text{W cm}^{-2}$) for 24 h. A total of 39 protein spots were differentially expressed by at least 2-fold compared with the controls (22 proteins were downregulated and 17 were upregulated) after treatment with supplemental UV-B radiation. Of these protein spots, 27 were successfully identified by MALDI-TOF MS after the data was searched. These 27 proteins could be classified into eight categories according to their functions: class I, photosynthesis (plastocyanin, ribulose-5-phosphate carboxylase small subunit, oxygen-evolving enhancer protein 1, Ps P domain-containing protein 6, and fructose-1,6-bisphosphate aldolase); class II, carbohydrate metabolism (malate dehydrogenase); class III, nitrogen metabolism (TPS1); class IV, amino acid biosynthesis (cysteine synthase, Vb protein synthesis (ribosomal protein S5), proteinase inhibitor 1, and proteinase inhibitor 2); class V, defense response (chitinase, peroxidase, Cu-Zinc superoxide dismutase, calcium-binding protein, and proteinase inhibitor 2); class VI, signal transduction (protein kinase C δ); class VII, protein synthesis (ribosomal protein S5); class VIII, protein synthesis (ribosomal protein S5). First, supplemental UV-B radiation induced a

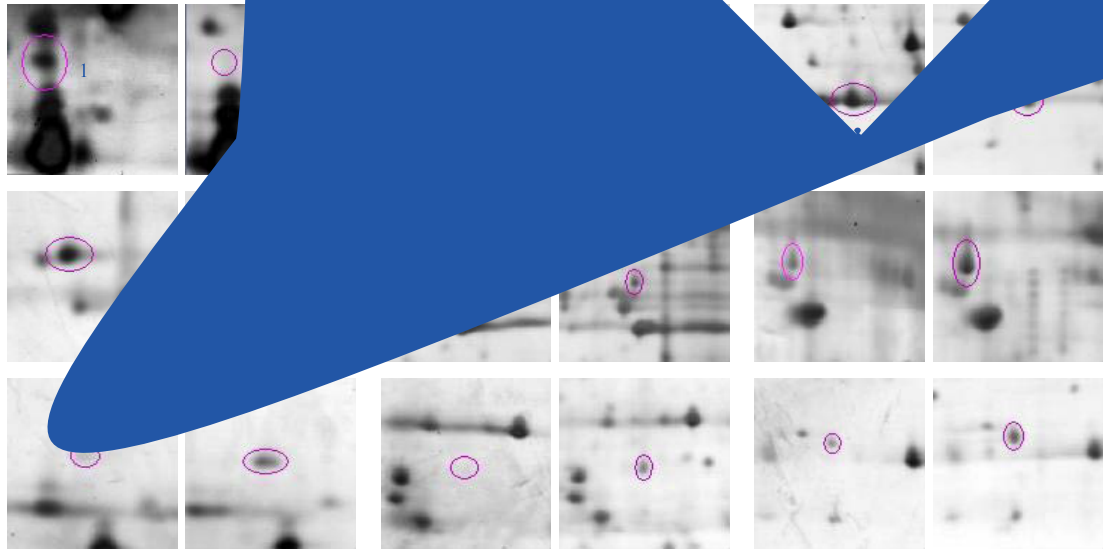
bioRxiv preprint doi: <https://doi.org/10.1101/2017.07.26.180111>; this version posted July 26, 2017. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY-NC-ND 4.0 International license.

[9], , UV-B , -20 °C 1h; 4°C, 1 000 g 5 1 min, , ; 20 mg 1 ml A (9 mol/L Gb, 4% CH PS, 1% IP uff r, e UV-B 24 h , 1% DTT), 30 °C 1h, ; 5 1 000 r /min 5 1 min, , UV-B , , : 1) UV-B , Bradford , 2) UV-B -80 °C .

1.3.2

pH (Iso l ctric e e G focusing, I F): G H althcar e eBP E (24 cm, pH= 3—10), 200 μg ((A. hypogaea L.) , 5 4 0 μL) 。 20 °C , 5 : 0 V 12h, 00 V 1h, 1000 V 1 1.2 h, 1000—10000 V 1 h, 10000 V 10 h 。 , : pH 7. 7、 A 26.21G SDS-P : , E g/kg、 1.08 g/kg、 551. g/kg、 5 1 .20 g / , 10 ml I [6 mol/L 5、 0 mmol / kg、 36.1 mg/kg、 112.26 mg/kg、 L Tris· HCl (pH= 8.8)、 30% 、 2% SDS、 1% DTT 551 .62 mg/kg。 20125 1 50.0 %] 10 ml II [6 mol/L 、 5 2 °C 24 5、 0 mmol/L Tris· HCl (pH= 8.8)、 30% 、 2% h, , 2 , 40 cm, 20 cm。 SDS 2. % 50.0 %] UV-B , UV-B A G 5 1 min。 SDS-P (12%) 5 (2 mmol/L Tris、 192 mmol/L (, 0.1% SDS 50. %) , SDS-P A G , SDS-PE 280—320 nm, 308 nm) 。 G SDS-P A G , SDS-PE UV-B 5 4 μW / . , cm² (UV-B 20%, UV , 5 1 °C : 100 V nhanc đ UV đ, 8 h, 3d; 5 4 min 200V, . (CK) , . **1.3.3** , , 2 , 8 Sh vch nkoe [10] , . , -80 °C , : 5 min; (30% 、 10%) 30 min; 10% , 5 min; 1.3 , 5 min; (0.02% 5 , 1 g) 1 min; , 1 min; (0.1% , , ; 1 40) 20 min; , 30 s; 10% , -20 °C 1h; 4 °C, 0.2 % , 0.04%) 3 min; 5 1 000 r /min 5 1 min, , ; 5 (%) ; 3 ,

CK CK



2

Fig.2 Enlarged images of partial differentially expressed protein spots

1

Table 1 Identification of differential proteins in peanut leaves by MALDI-TOF-TOF MS

Function	Spot No.	Accession number	Protein name	Molecular weight (Th o. / xpe) (kD)	Isoelectric point (Th o. / xpe)	Score	Species	Regulation
Photosynthesis	1	gi15279	51, -	56.16.40	4.82/3.24	177	<i>Glycine max</i>	↓
	3	gi11079736		19.98/16.43	8.80/5.9	374	<i>Glycine soja</i>	↓
	7	gi11079736		19.98/16.73	8.80/4.14	374	<i>G. soja</i>	↓
	18	gi1131386	1	53.2/57.0	5.5/5.8/4	616	<i>Spinacia oleracea</i>	↓
Carbohydrate	22	gi15671888	PsP	528.1/29.91	5.73/6.33	409	<i>G. max</i>	↓
	37	gi13172481		38.66/48.44	6.7/8.1	676	<i>G. max</i>	↓
	32	gi137293		37.39/30.8	7.01/8.29	699	<i>Pisum sativum</i>	↓
	33	gi2827080		36.00/44.28	8.80/7.97	634	<i>Medicago sativa</i>	↓

m

A Function	Spot No.	Accession number	Protein name	Molecular weight (Th o. / xpe) (kD)	Isoelectric point (Th o. / xpe)	Score	Species	Regulation
	5 1	gi17104066	Cu-Zn	51.20/88.0	5.27/.42	284	<i>A. hypogaea</i>	↓
	24	gi37470923	3-O	29.54/1.39	5.5/.95/6.4	328	<i>Medicago truncatula</i>	↑
	28	gi3713969		23.08/27.33	8.96/7.87	263	<i>M. truncatula</i>	↓
	5 3	gi3713969		23.08/27.93	8.96/8.30	263	<i>M. truncatula</i>	↓

11 12) , (23) , PS II

G , SH b Ps P ,

, H₂ Q₂ ; Cu / ;

Zn-S O (5 1) , Q₂⁻ ; 5l, -

, Q₂⁻ -1, 6-

o ,

G (LPs) 、 UV-B o

, UV-B o

o ,

; ,

, [31] o UV-B

(*Capsicum chinense* Jacq.) LPs UV-B o

(*Nicotiana tabacum* Xanthi nc) ,

G [32], LPs

(*Sclerotinia sclerotiorum*) [33], A

LP ,

(*Manduca sexta*) [34] o

LP (28 5 3) ,

UV-B , 、

o

(P R)

, , β-1, 3-

P R 5[3] o

(Chitinas) e N- -D-

[36], 、

, ,

, UV-B

, (4 31)

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o

, , HSP18.2 (

16) HSP70 (14) ,

HSP

、 ,

UV-B , Du [37]

UV HSP70 ,

Murakami [38] HSP17.1

UV-B o

, UV-B

: UV-B

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Population Community and Ecosystem

2014

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ACTA ECOLOGICA SINICA

Edited

Editor-in-chief
Supervised
Sponsored

科 学 出 版 社

Published

科 学 出 版 社

Printed

Distributed

Domestic
Foreign

ISSN 1000-0933

