

Populus nigra L.

In vitro

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Populus nigra L.

IBA / 0.3 BA / 1 MS
(8 6 4 2) / 1000 500 250 125
/ 125

/ 250 125

%18

/ 500

%50.66

. 2 / 125

/ 250 125

/ 500

. 2

/ 250

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**Doubling the Chromosome Number of Black Poplar Trees (*Populus nigra L.*)
by Colchicine *In vitro***

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ABSTRACT

The possibility of inducing chromosomal polyploidy was studied for Black poplar (*Populus nigra L.*) which micropropagated from stem cutting by exposure to different colchicine concentrations 125, 250, 500 and 1000 mg / l and different exposure periods (0.0, 2, 4, 6, 8 days) in

MS medium supplemented with 1mg/l BA and 0.3 mg/l IBA. After 4 weeks of exposure results showed the significant superiority of 125 mg / l colchicine treatment for stem cutting survival percentage transplant survival percentage after acclimatization and diploid plants percentage characteristics, also the results of polyploidy showed that the highest percentage of tetraploid plant (18%) was recorded for both treatments 125 and 250 mg/l colchicine, when 50.66% of plants treated with 500 mg/l colchicine has a mixed polyploidy and it was significantly different from the rest treatments. but the interaction effect of colchicine concentrations and exposure periods showed that the highest survival percentage was recorded for the treatments of non exposure and exposure to 125 mg/l colchicines for 2 days. After 16 weeks of acclimatization and field growing the results showed the significant superiority for both treatments of exposure with 125 and 250 mg/l colchicine for stem length, stem diameter, leaves number, vegetative system dry weight, stomata length, stomata width and total chlorophyll contents characteristics, this in comparison with 500 mg/l colchicine treatments, and this is in combined with finally the interaction effects of colchicine for the treatment of 250 mg/l colchicine for 2 days.

Keywords: *Populus nigra*, tissue culture, polyploidy, colchicines.

salixaceae *Populus nigra* L.
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(1992) (2003

Tang *et al.* ; Liu *et al.*, 2009) (Tetraploid)
(*al.*, 2010)
(Dhooghe *et al.*, 2011)
MS *Paulownia tomentosa* (Tang *et al.*, 2010)
(70 48 24) (/ 1000 500 100)
48 / 500 (40)
) . 72 48 / 1000
(
MS *Populus pseudo-simonii* (Cai and Kang, 2011)
(3 2 1) (/ 30 20 10) (Murashige and Skoog, 1962)
/ 20
. 1 / 10 14.6 3

MS

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/ /

.2012 / 8 /17 2011 / 6 /15

/ 1 (Murashige and Skoog,1962) MS

2) (/ 1000 500 250 125)

IBA / 0.3 BA

.(8 6 4

IBA / 0.3 BA / 1 MS

()

/ 0.5 MS (2)

.IBA

(Guofeng *et al.*, 2007)

(1 : 1)

(20)

(/)

(CRD) .(Knudsen *et al.*, 1977)

5 (2000)

(SAS, 1996) %

(1)

/ 125 (% 78.79)

/ 1000 (% 18)

2 (% 51.48) (% 90)

.(% 34.22) 8 6 4

(% 90)

/ 125 2

6 4 / 125

Populus

: 1

IBA / 0.3 BA / 1

MS

nigra

	/				()
	1000	500	250	125	
A 90	A 90.00	A 90.00	A 90.00	A 90.00	
B 51.48	F	C-E 52.09	BC 63.86	A 90.00	2
BC 44.32	F	DE 39.08	CD 58.57	AB 79.63	4
BC 41.38	F	E 35.18	C-E 52.09	AB 78.23	6
C 34.22	F	E 35.18	C-E 45.59	CD 56.09	8
	D 18	C 50.37	B 62.02	A 78.79	

.% 5

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(2)

/ 125

% 56.56

/ 500

/ 250

.(% 39.15)

2

(% 70.35)

% 42.74 % 41.09 % 44.00

6 4

8

(% 37.15)

(% 70.35)

250

% 26.56 % 26.56 % 34.80

8 6

/ 500

4

/

(3)

125

/ 250

(/

3.81)

.(/ 1.85)

/ 500

/

(/ 3.77)

4

8

(/ 2.36)

(/ 5.44)

2

/ 250

6

/ 125

4

/ 250

8

/ 500

(/

1.22)

.....

: 2

IBA / 0.3 BA / 1 MS *Populus nigra*

	/			()
	500	250	125	
A 70.35	A 70.35	A 70.35	A 70.35	
B 44.00	A B 36.15	A B 46.31	A B 49.54	2
B 41.09	A B 36.15	B 34.80	A B 52.33	4
B 42.74	B 26.56	A B 41.15	A B 60.53	6
C 37.15	B 26.56	A B 36.15	A B 50.08	8
	B 39.15	AB 45.75	A 56.56	

.%5

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Populus nigra

:3

IBA / 0.3 BA / 1 MS

	/			()
	500	250	125	
C 2.58	C D 2.49	C D 2.44	C D 2.83	
C 2.69	C D 2.44	A 5.44	C 3.21	2
A 3.77	C D 1.77	A 5.33	B 4.22	4
B 3.10	E 1.33	C 2.99	A B 4.99	6
D 2.36	E 1.22	C D 2.88	C 2.99	8
	B 1.85	A 3.81	A 3.64	

.%5

*

(diploidy)

-

(4)

/ 125 %78

/ 500 % 47.34

%100

%40

6

% 100

8 6

/ 500

%

(tetraploidy)

(4)

-

250 125

%18

.% 3

/ 500

/

2

% 21.66

%

2

/

250

6

/

125

% 40

. 8 6 4 / 500

(mixploidy)

(4)

-

/ 500

% 50.66

/ 125

% 4

%48.33

%

8

8 6

/ 500

% 100

%

. 6 4 2 / 125

Populus nigra

: 4

/ 1

MS

IBA / 0.3 BA

	/			()
	500	250	125	
A 100	A 100	A 100	A 100	
B 70	D 70	H 50	B 90	2
C 68.9	E 66.7	G 60	C 80	4
D 40	I	G 60	G 60	6
E 41.66	I	F 65	G 60	8
	C 47.34	B 67	A 78	

.% 5

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Populus : 4
MS *nigra*

IBA / 0.3 BA / 1

	/			()
	500	250	125	
D	F	F	F	
A 21.66	D 15	A 40	E 10	2
B 16.66	F	B 30	C 20	4
B 16.66	F	E 10	A 40	6
C 10.00	F	E 10	C 20	8
	B 3	A 18	A 18	

.% 5

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Populus : 4
BA / 1 MS *nigra*

IBA / 0.3

	/			()
	500	250	125	
E	G	G	G	
D 10	E 20	F 10	G	2
C 14.44	B 33.33	F 10	G	4
B 43.33	A 100	C 30	G	6
A 48.33	A 100	D 25	E 20	8
	A 50.66	B 15	C 4	

.% 5

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(5)

/ 125 % 90
 / 500 % 81

4 2 % 90

8 % 75

% 90

/ 500 8 / 250

%63.44 %71.56 %71.56 8 6

()

(6)

/ 250 / 125 29.066

. 20.066 / 500

29.666 4

25.777 25.222 8 6 2

(6) . 21.222

2 / 250 38.667

4 / 250 6 / 125

. 8 / 500 16.333

:5

Populus nigra

	/			()
	500	250	125	
A 90	A 90	A 90	A 90	
A 90	A 90	A 90	A 90	2
A 90	A 90	A 90	A 90	4
B 83.85	B 71.56	A 90	A 90	6
C 75	C 63.44	B 71.56	A 90	8
	C 81	B 86.31	A 90	

.% 5

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Populus nigra

:6

	/			()
	()			
	500	250	125	
B 25.222	C D 25.667	C D 25.667	C D 25.667	
A 28.333	E 20.667	A 38.667	C D 25.667	2
A 29.666	E 21.000	A 36.333	B 31.667	4
B 25.777	F 16.667	C-E 24.333	A 36.333	6
C 21.222	F 16.333	E F 21.333	C 26.000	8
	B 20.06	A 28.99	A 29.06	

.% 5

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()

2.572

(7)

/ 125

/ 250

. 2.271 / 500

2.588 4

2.305 2.474 2.438

8 6

2

2.856

2 / 250

4 / 250 6 / 125

. 8 / 500 2.113

15.196

(8)

/ 250

/ 125

/

. / 9.864 / 500

4 / 15.21

10.99 13.55 12.77

8 6

2

/

/ 20.66

2 / 250

4 / 250 6 / 125

. 8 / 500 / 7.66

Populus nigra

:7

	/			()
	()			
	500	250	125	
B 2.438	C 2.430	C 2.446	C 2.440	
A 2.544	E 2.313	A 2.856	C 2.463	2
A 2.588	E 2.313	A 2.823	B 2.630	4
B 2.474	F 2.186	C D 2.406	A 2.830	6
C 2.305	F 2.113	D E 2.330	C 2.473	8
	B 2.271	A 2.572	A 2.567	

Populus nigra

:8

	/			()
	500	250	125	
B 12.77	CD 13.00	C-E 12.66	C-E 12.66	
AB 14.55	E-H 10.00	A 20.66	CD 13.00	2
A 15.21	E-G 10.33	AB 18.66	B 16.66	4
B 13.55	HI 8.33	C-E 12.66	A 19.66	6
C 10.99	I 7.66	D-F 11.33	C 14.00	8
	B 9.864	A 15.194	A 15.196	

% 5

*

(²)

(9)

/ 125

² 21.60

/ 500

² 10.19

8

2

² 18.54

² 13.15

² 26.81

6 / 125

4 2

/ 250

4

/ 125

² 7.03 8

/ 500

:9

Populus nigra

	/			()
	(²)			
	500	250	125	
A 16.81	DE 16.44	CD 17.46	D 16.55	
A 18.54	GH 9.42	A 26.63	BC 19.59	2
A 16.38	GH 9.38	A 25.72	A 24.56	4
A 16.38	H 8.70	EF 13.65	A 26.81	6
B 13.15	H 7.03	FG 11.93	B 20.51	8
	C 10.19	B 19.07	A 21.60	

.%5

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(/)

(10)

125

/ 500

1.423

.(1)

1.251 1.293

/ 250

8

1.498

1.286 1.238 1.255

6 4 2

/ 125

2

/ 250

4

/ 250

6

/ 125

1.539

:10

Populus nigra

	/			()
	500	250	125	
A 1.498	A-C 1.476	A-C 1.480	A 1.539	
B 1.255	C-E 1.362	G 1.009	B-D 1.394	2
B 1.238	B-D 1.409	G 1.094	F 1.212	4
B 1.286	A-D 1.448	DE 1.341	G 1.070	6
AB 1.336	A-D 1.423	DE 1.335	EF 1.251	8
	A 1.423	B 1.251	B 1.293	

.5%

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*Populus nigra*

:1

6 / 125 (1)
 2 / 250
 /) 1.009 1.070
 .1.539 (

(11)
 / 125 2.684
 1.407 / 500 / 250
 2.800
 . 1.586 8 6 4 2
 2.950
 / 250 2 / 250
 1.113 1.440 8 6 4 2 / 500 8
 . 0.900 0.990 1.056
 2 /

(12)
 250 125 / 500 2 / 1749.8
 . 2 / 1394.3 1349.7 /
 1367.03 4 2 2 / 1771.3
 . 8 6 2 / 1317.66

2 / 814.7 2 / 250
 / 250 2 / 916.4 1175.8 6 4 / 125
 . 2 / 1046.1 4

:11

Populus nigra

	/			()
	()			
	500	250	125	
A 2.800	A 2.980	A 2.660	A 2.760	
A 2.145	C 1.113	A 2.950	A B 2.373	2
A 2.290	C 1.056	A 2.883	A 2.933	4
A 2.184	C 0.990	A 2.626	A 2.936	6
B 1.586	C 0.900	B 1.440	A B 2.420	8
	B 1.407	A 2.511	A 2.684	

% 5

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Populus nigra

:12

	/			()
	2 /			
	500	250	125	
A 1771.3	A1805.3	A1786.9	A1721.7	
B 1367.03	A1758.9	D814.7	AB1527.5	2
B 1317.66	A1731.1	CD1046.1	B-D1175.8	4
AB 1422.7	A1712.8	A1638.9	D916.4	6
A 1610.83	A1740.3	A1685.0	A-C1407.2	8
	A 1749.8	B 1394.32	B 1349.7	

% 5

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()

(13)

/ 250 125

0.060

0.061

0.041

/ 500

4 2

4

0.045 0.043

8

0.083

2 / 250

0.076

0.077

4

/ 250 6

/ 125

()

(14)

/ 250

0.030

0.015 / 500

/ 125

0.031 4

0.018 8

6 2

4

0.050 / 250

2

/ 125

6 / 250

/ 500

4

0.014

Populus

:13

nigra

	/			()
	()			
	500	250	125	
B 0.043	DE0.044	E 0.041	DE 0.044	
A 0.060	E 0.041	A 0.083	C 0.058	2
A0.061	E 0.039	AB 0.076	B 0.070	4
A 0.058	E 0.043	CD 0.056	AB 0.077	6
B 0.045	E 0.040	DE 0.044	C-E 0.053	8
	B 0.041	A 0.060	A 0.060	

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Populus nigra

:14

	/			()
	()			
	500	250	125	
B 0.018	DE 0.016	DE 0.016	CD 0.023	
A 0.030	DE 0.016	A 0.050	C 0.025	2
A 0.031	DE 0.014	A 0.044	B 0.035	4
A 0.027	DE 0.016	C-E 0.020	A 0.046	6
B 0.018	C-E 0.016	C-E 0.020	C-E 0.020	8
	B 0.015	A 0.030	A 0.029	

.%5

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(15)

/ 250 / 0.727

. / 0.421 / 500 / 125

4 / 0.681

0.545 0.588 8 6 2

0.931

6 / 125 /

4 2 / 250 4 / 125

. / 0.356 2 / 500

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:15

Populus nigra

	<i>I</i>			()
	(/)			
	500	250	125	
B 0.588	B 0.632	B 0.565	B 0.568	
A 0.611	C 0.356	A 0.916	B 0.561	2
A 0.681	C 0.366	A 0.860	A 0.817	4
A 0.657	C 0.379	B 0.662	A 0.931	6
B 0.545	C 0.373	B 0.632	B 0.631	8
	B 0.421	A 0.727	A 0.701	

.% 5

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.(Mohammadi, 2012 2005)

Jatropha curacas

Oliveira *et al.*, (2013)

MS

/ 250 125

/ 500

.(2005)

.(Dhooghe, 2011)

.(2005)

/ 250 125
(4)

(Dhooghe *et al.*, 2011)

/ 250 125

- Ewald *et al.*,
18 / 1000 MS (2009)
- Cai and Kang, (2011)
Populus pseudo-simonii
MS
(2005)
(2000)
(1992)
() (2003)
- Cai, X.; Kang, X.Y. (2011). In vitro tetraploid induction from leaf explants of *Populus pseudo – simonii* Kitag. *Plant Cell Rep.* **30**,1771-1778.
- Dhooghe, E.; Van Laere, K.; Eckhaut, T.; Leus, L.; Van Huylenbroeck, J. (2011). Mitotic chromosome doubling of plant tissues in vitro. *Plant Cell Tiss. Organ Cult.* **104**, 359-373.
- Ewald, D.; Ulrich, K.; Naujoks, G.; Schroder, M.B. (2009). Induction of tetraploid poplar and black locust plants using colchicine: chloroplast number as an early marker for selecting polyploids in vitro. *Plant Cell Tiss. Organ Cult.* **99**, 353-357.
- Liu, X.Z.; Lin, H.; Mo, X.Y.; Long, T.; Zhang, H.Y. (2009). Genetic variation in colchicine – treated regenerated plants of *Eucalyptus globules* Labill. *J. Genetics*, **88**(3), 345-348.
- Mohammadi, P.P.; Moieni, A.; Ebrahimi, A.; Javidfar, F. (2012). Doubled haploid plants following colchicine treatment of microspore – derived embryos of oilseed rape (*Brassica napus* L.) *Plant Cell Tiss. Organ Cult.* **108**, 251-256.
- Murashige, T.; Skoog, F. (1962). A revised medium for rapid growth and bio assays with tobacco tissue cultures. *Physiol. Plant.* **15**, 473-497.
- Olivera, S.C.; Nunes, A.C.P.; Carvalho, C.R.; Clarindo, W.R. (2013). In vitro Polyploidization from shoot tips of *Jatropha curcas* *Plant Growth Regul.* **69**, 79 – 86.
- SAS. (1996). Statistical Analysis System, Washington, USA.
- Tang, Z.O.; Chen, D.L.; Song, Z.-J.; He, Y.C.; Cai, D.T. (2010). In vitro induction and identification of tetraploid plants of *Paulownia tomentosa*. *Plant Cell Tiss. Organ Cult.* **102**, 213-220.