

Plantlet regeneration from the mature embryo of *Bothriochloa ischaemum*

Na Yu , Kuan-hu Dong*

College of Animal Science and Technology , Shanxi Agricultural University , Taiyu , 030801 , China

E-mail : yuna1023@126.com , * Author for correspondence , E-mail : dongkuanhu@126.com

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Introduction *Bothriochloa ischaemum* is known to have a strong vitality , produce high yields , is tolerant to grazing and has a high feeding value . The regeneration from mature embryos was studied using the mature embryos of *Bothriochloa ischaemum* as explants . The effect of various hormone compositions on callus induction and growth state was studied .

Materials and Methods The explants were first immersed for 10min in distilled water , followed by immersion in 75% ethanol for 30s , then in 0.1% (w : v) HgCl₂ solution for 15 min with constant agitation , lastly extensive washing (four-five times) with sterile distilled water was undertaken . Murashige and Skoog's (MS) medium containing 3% sucrose and 0.5% agar was used as the basal medium . The MS medium supplemented with 2,4-D (0 , 1.0 , 2.0 , 3.0 , 4.0 , 6.0 mg/L) was tried in a single factorial treatment . Each treatment consisted of three replications with 15 explants . Callus was maintained at 25 ± 2°C , 60% relative humidity for 27d . Rapidly growing callus and embryogenesis were maintained in the callus subculture medium plus 1.0 mg/L 2,4-D for 5-6d . The embryogenic callus (EC) were subsequently sub-cultured on MS media supplemented with NAA (0.04 , 0.05 , 0.06 , 0.07 mg/L) in combination with 0.1 mg/L of 6-BA for inducing callus differentiation .

Results The frequency of callus induction reached 86.7% on the callus induction medium supplemented with 2.0 mg/L 2,4-D (Figure 1) . The rate of differentiation frequency from callus sub-cultured was 60.9% on the differentiation medium supplemented with 0.1 mg/L 6-BA and 0.05 mg/L NAA . Differentiation frequency from combinations of 0.1 mg/L 6-BA with 0.04 or 0.06 mg/L NAA was 43.6% and 34.6% , respectively (Table 1) .

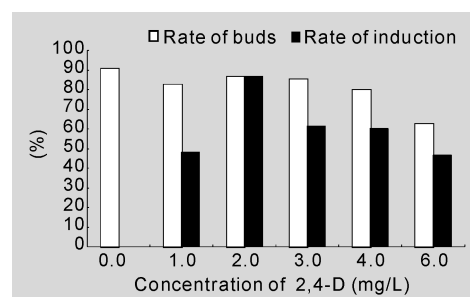


Figure 1 The growth of seeds in different concentrations of 2,4-D .

Table 1 Callus Differentiation in different combined of hormones .

Hormone composition(mg/L)		Number of inoculation	Number of EC	Induction frequency of EC (%)	Number of differentiation	Differentiation frequency (%)
6-BA	NAA					
0.1	0.04	86	71	82.6	31	43.6
0.1	0.05	87	46	52.9	28	60.9
0.1	0.06	88	81	92.0	28	34.6
0.1	0.07	86	73	84.8	21	28.7

Conclusions The callus induction Frequency reached 46.6%-86.7% on most of the media combinations during 20-27 days . It has been shown that the callusing in differentiation required a low auxin and comparatively high cytokinin level .

Reference

Parrott , W . A . , 1991 . Auxin-stimulated somatic embryogenesis from immature cotyledons of white clover . *Plant Cell Report* , 10 , 17-21 .