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Abstract The effects of the cultivation medium plant growth regulators and inoculum size on the cell growth and 20-hydroxyecdysone production in suspension cultures of Vitex glabrata R. Br. were investigated. The cell growth and 20-hydroxyecdysone formation reach the highest when cells are cultured in the Gamborg's B5 medium supplemented with 2.0mg/L BA, 6-benzylaminopurine and 1.0mg/L 2,4-D. The maximum 20-hydroxyecdysone productivity of about 1.1mg/L/day was observed in the culture with 20% PCV packed cell volume of inoculum size. These data also show that the increment of the inoculum size to 20% PCV could increase the productivity in 7-folds.

Key words 20-hydroxyecdysone suspension cultures Vitex glabrata
1.1 V. glabrata

MS Murashige and Skoog and 1.0mg/L 2,4-D 30g/L, pH 5.8

121℃ 20min, 120r/min, 2000lx 25℃

1.2

1.3 HPLC

0.3g soxhet 180mL 95% V/V

6h 3mL 2 mL Sep- pak C18 10mL 20%

V/V 80% 1.0ml/min 254nm

Y = 9E - 0.6X - 7925.2 R² = 0.998

2.0mg/L BAP 1.0mg/L 2,4-D 12.1g/L B5 3 1/2 MS

B5 1/2 MS B5 0.038% DW 24% B5 1/2 MS

2.2 V. glabrata

0.3g 180mL 95% V/V

6h 3mL 2 mL Sep- pak C18 10mL 20%

V/V 80% 1.0ml/min 254nm

Y = 9E - 0.6X - 7925.2 R² = 0.998

2.0mg/L BAP 1.0mg/L 2,4-D 30g/L, pH 5.8

121℃ 20min, 120r/min, 2000lx 25℃

2.3 PCV

10% PCV 21d 20% PCV

0.040% DW 20% PCV 1.10mg/l E 21d 10% PCV 7
悬浮细胞在不同培养基配方中培养时细胞干重和羟基蜕皮激素产生的时间曲线。另外，接种量对细胞生长以及次级代谢物形成的影响可能与参与代谢途径的酶活性的提高有关。同样的，通过提高接种量来提高次级代谢产物的含量的方法也在以下方面有应用：

生产紫杉酚，

生产葫芦素，

产生人参皂甙以及

产生甘草。但是，接种量对次级代谢物生物合成的影响的具体机制还不明确。

结论！悬浮细胞培养对细胞生长和羟基蜕皮激素形成的最佳条件为

培养基，添加

和

两种不同的植物生长素

和

的IA培养基中培养时的干重和羟基蜕皮激素产生的时间曲线。

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V. glabrata

Gamborg’s B5

1.0mg/L 2,4-D 2.0 mg/L BAP 20 % PCV

Table 1 Effect of inoculum size on the biomass and 20-hydroxyecdysone productivity in suspension culture of V. glabrata in B5 medium supplemented with 2.0mg/L BAP and 1.0mg/L 2,4-D

<table>
<thead>
<tr>
<th>Inoculum size</th>
<th>% PCV</th>
<th>% DW</th>
<th>Maximum cell growth (g/L)</th>
<th>Time of maximum production (d)</th>
<th>Maximum productivity (mg/L·d)</th>
</tr>
</thead>
<tbody>
<tr>
<td>10</td>
<td>0.033</td>
<td>11.2</td>
<td></td>
<td>21</td>
<td>0.176</td>
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<tr>
<td>20</td>
<td>0.040</td>
<td>11.0</td>
<td></td>
<td>4</td>
<td>1.1</td>
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