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ON THE NUTRITION OF EXCISED RICE EMBRYO[1]

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Abstract

The optimum temperature and light intensity for excised rice embryo culture were 32°C and 25,000 Lux, respectively.

When anyone of the micronutrient elements was absent from the white's medium, chlorosis developed on the leaves accompanied by poor development, reduced seedling growth and dry weight. In the absence of boron and manganese, the chlorotic leaves showed brown spot symptom.

Vitamins were not necessary for excised rice embryo growth. However, vitamins were required when embryos had not produced chloroplasts.

The optimun concentrations of the plant hormones used alone were 0.5 mg/l for IAA, 0.01 mg/l for kinetin and 0.3 mg/l for gibberellic acid. Different effects on embryo growth were observed by interaction between the hormones. This interaction also varied with different concentrations of hormones.

Carbohydrate was the most important factor for excised rice embryo culture. Two percent glucose or 4% sucrose in the medium was the best concentration for culturing the rice embryo. Embryo grew poorly in soluble starch medium. But when gibberellin was added into soluble starch medium, the growth of seedlings was slightly increased. Xylose was uneffective for embryo growth.

Most amino acids used singly did not have a marked effect on excised rice embryos. However, glycine enhanced whereas aspartate or glutamate inhibited embryo development at the concentrations of 0.19-1.17 mg N/l. The dry weight of seedlings was increased markedly by the addition of coconut milk in the white's medium.

Ammonium as the nitrogen source was more effective for seedling growth than nitrate. Ammonium, howerver, was unfavorable for root growth.

Introduction

Hanning (1904) was the first to demonstrate that embryos (of *Raphanus* and *Cochearia*) could be successfully cultured in a medium containing certain mineral salts and sucrose.

LaRue (1936) reported that indoleacetic acid (0.5 mg/l) enhanced the growth

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of small excised embryos *in vitro*. Solacolu and Constantinesco (1936) observed that indoleacetic acid induced tumors on cultured embryos of *Phaseolus* and *Ricinus*.

Narayanaswami (1957) investigated the responses of *Pennisetum* embryo in liquid media containing 2, 4-D (0.5 mg/l) and IAA (1 mg/l), and found that no initiation of the embryo occurred when either substance was used alone. Raghavan & Torrey (1963) reported that *Capsella* proembryos could be cultured on media containing IAA, kinetin and adenine in combination, but not separately. Raghavan also observed that 0.0001 mg/l of IAA was the optimum concentration for *Capsella* embryo growth. According to Schooler (1960), gibberellic acid, in concentration of 1 to 3 mg/l, enhanced the growth of roots and shoots of excised barley embryos. Abnormal plants developed from *Eronumus* embryos cultured on media containing 1 mg/l gibberellin (Bulard and Monin, 1960), and germination wheat embryos exhibited a differential growth response to gibberellin when cultured in light and darkness (Bulard, 1954). Lee (1962) found that root initiation results from a high-auxin/low-kinin situation, whereas shoots formed when the balance is reversed.

Bonner and Axtman (1937) have reported that amino acid promoted the growth of cultured pea embryos. Later, Rijven (1952) observed that glutamine at a concentration of 400 mg/l stimulated the growth of embryos of *Capsella*. Norstog and Smity (1963) reported that glutamine and alanine were active in the promotion of growth of small barley embryos. Spoerl (1948) reported that arginine and aspartic acid supported good growth of orchid embryos. Amemiya *et al.* (1956) tried several amino acids in rice embryo cultures, and found them, for the most part, to be inhibiting to normal embryo development. Nitrate-nitrogen proved to be superior to ammonium nitrogen in these cultures.

Van overbeek *et al.* (1944), Lofland (1950) and Rijvan (1952) found that sucrose was better than other sugars in the embryo culture. Ball (1959) had demonstrated that sucrose enhanced shoot growth and that glucose produced the greatest increment for roots. Mauney (1961) with barley embryos reported that sucrose produced the greatest growth.

The purposes of these experiments were to study the effects of environmental factors, such as temperature and light, on development and growth of excised rice embryo; and to investigate the relationship between the excised embryo growth and the medium's composition, such as micronutrient elements, plant hormones, vitamins, amino acids or carbohydrates.

Materials and Methods

Seeds of *Oryza sativa* (variety Tainan No. 5) were surface-sterilized with 0.1% mercuric chloride, washed with sterile distilled water three times, and

then transferred to moistened sterile Petri dishes. After two days at 25°C, embryos, about 0.3 mm in length, were excised and were used for all experiments.

The White's medium was used as a basal medium for culturing the excised embryos. This basal medium was modified as followings: (1) one of the micro-elements (Zn, Mn, B, Mo or Cu) or of vitamins (thiamine, pyridoxine or nicotinic acid) was omitted from the basal medium; (2) the basal medium was supplemented with various hormones (IAA, gibberellic acid and kinetin); (3) Different carbohydrates (glucose, soluble starch or xylose) was substituted for sucrose in the basal medium; and (4) the basal medium was supplemented with different kinds of amino acids (glycine, aspartate, glutamate or arginine) or coconut milk. In the amino acid experiments, NH_4^+—nitrogen substituted for NO_3^-—nitrogen in the basal medium was also used as a basal medium. The compositions for macronutrients of these two basal media are shown in Table 1.

Table 1. *Compositions for macronutrients (mg/l) of two basal media used in amino acid nutritional experiments*

	Basal medium containing NH_4^+	Basal medium containing NO_3^-	Molarity
$CaCl_2$	133.2		1.2×10^{-3}
$Na_2SO_4 \cdot 10H_2O$	450.0	450.0	1.4×10^{-3}
KCl	65.0	65.0	9.0×10^{-4}
$NaH_2PO_4 \cdot 2H_2O$	20.5	20.5	1.3×10^{-4}
NH_4Cl	172.2		3.2×10^{-3}
$KHCO_3$	80.0		8.0×10^{-4}
$Ca(NO_3)_2 \cdot 4H_2O$		208.0	1.2×10^{-3}
KNO_3		80.0	8.0×10^{-4}

The pH of all media were adjusted to 5.5. Then, 25 ml of the medium were pipetted into each culture tube (100 ml) and autoclaved for 20 minutes at 121°C and 15 pounds. In the case of gibberellic acid, it was sterilized by filtration through a millipore filter and added into tubes containing media.

Two weeks after cultivation, the seedlings were taken from the melted agar carefully, the number of roots, shoot height, root length and dry weight were measured.

This investigation was conducted in the phytotron of National Taiwan University, Taipei, from August, 1969 to July, 1971. Each experiment was repeated three times, each time with triplicate treatment. The data presented was the average of nine treatments.

Result

Effect of environmental conditions on the growth of excised embryos

To determine the favorable condition for the cultivation of excised embryos, the embryos were cultured on White's medium and maintained at different temperature and light intensity for 10 hrs. photoperiod in artificially lighted (fluorescent light and mercury vapour lamp) rooms.

Room 1: Light intensity, 5,000 Lux, and temperature, 18±1°C.

Room 2: Light intensity, 13,000 Lux, and temperature, 25±1°C.

Room 3: Light intensity, 25,000 Lux, and temperature, 32±1°C.

Among three artificially lighted rooms, room 3 gave the best condition for rice embryo culture growth (Table 2). Thus, room 3 condition was used in all subsequent experiments.

Table 2. *Growth of excised rice embryos cultured at the White's medium in three environmental conditions*

	Room 1	Room 2	Room 3
Final root length/Initial root length	29.0	49.0	51.3
Final dry weight/Initial dry weight	3.0	6.9	10.3
Final shoot height/Initial shoot height	55.3	131.0	147.8

The role of vitamins and micronutrient elements in the nutrition of the excised embryo of rice

When any of the micronutrient elements (Mn, Zn, B, Mo or Cu) was absent from the White's medium, the embryos did not grow well. Their seedling height, root length and dry weight were less than those cultured in whole White's (CK) medium (Table 3). The poorest growth was obtained when boron (B) was omitted.

Table 3. *The growth of rice embryos cultured in micronutrient omitted White's media*

	CK	-Zn	-Mn	-B	-Mo	-Cu
No. of root	10	6	8	4	9	10
Shoot height (cm)	12.67	7.60	5.03	2.20	5.30	8.0
Root length (cm)	7.93	4.80	3.27	0.75	1.05	3.20
Dry weight (mg)	17.10	6.80	4.10	3.05	5.55	8.50

Generally, micronutrient deficiency caused chlorosis on leaves. Manganese deficient seedlings, in particular, showed both chlorosis and spots on the leaves.

Before the production of chloroplasts, vitamins were required for the development of embryos. However, when the developed shoots became green, vitamins in the medium did not show noticeable effects on seedling growth. Embryos cultured at vitamin deficient media did not have serious symptom, but their seedling height, root length and dry weight were less (Table 4).

Table 4. *Growth of rice embryos cultured at vitamin deficient White's media*

	CK	-Thiamine	-Pyridoxine	-Nicotinic acid
No. of root	7	7	8	9
Shoot height (cm)	14.10	12.10	14.10	11.83
Root length (cm)	7.50	4.75	6.50	4.20
Dry weight (mg)	10.30	6.10	10.00	8.20

Effects of hormones (auxin, gibberellin and kinetin) on growth of excised rice embryo

Indoleacetic acid (IAA) at the concentration of 0.5 mg/l gave the best growth for rice embryo seedling. But 0.1 mg/l IAA was better than 0.5 mg/l for root growth (Table 5).

Table 5. *Effect of IAA concentration in White's medium on the growth of excised rice embryo*

	IAA concentration (mg/1)				
	0	0.05	0.1	0.5	1.0
No. of root	3	3	7	5	4
Shoot height (cm)	15.4	16.15	17.94	18.65	16.62
Root length (cm)	10.0	10.40	12.80	11.70	10.64
Dry weight (mg)	4.9	5.81	6.90	7.00	4.60

The optimum concentration of gibberellin (GA_3) for seedling height, root length and dry weight of embryo was 0.3 mg/l, but that for root formation was 0.5 mg/l (Table 6).

Table 6. *The growth of excised rice embryo at various concentrations of gibberellin*

	GA_3 concentration (mg/1)				
	0	0.1	0.3	0.5	1.0
No. of root	2	3	3	4	3
Shoot height (cm)	15.25	16.35	20.75	20.00	15.16
Root length (cm)	7.50	8.16	10.00	9.65	8.50
Dry weight (mg)	4.90	5.40	8.80	8.66	3.45

Kinetin at very low concentration such as 0.01 mg/l enhanced the rice embryo growth. When the concentration higher than 0.50 mg/l kinetin was used, the growth was notably reduced. (Table 7).

Table 7. *The growth of excised rice embryo at various concentrations of kinetin*

	Kinetin concentration (mg/1)				
	0	0.01	0.03	0.05	0.50
No. of root	2	8	8	4	4
Shoot height (cm)	13.15	20.55	19.56	16.80	10.55
Root length (cm)	5.00	9.00	8.65	8.55	3.15
Dry weight (mg)	4.50	8.72	8.00	6.70	4.00

As mentioned above 0.50 mg/l kinetin alone notably reduced the growth of rice embryo, but when it was combined with IAA, there were no unfavorable effects observed as the concentration of kinetin increased to 0.5 mg/l. The data also showed that root development with a high-auxin/low-kinetin situation (Table 8).

Table 8. *The growth of excised rice embryo in media containing both IAA (I) and kinetin (K) at various concentration (mg/l)*

	0 0	0.05(K) 0.5 (I)	0.5 (K) 0.5 (I)	0.05(K) 1.0 (I)	0.5 (K) 1.0 (I)
No. of root	3	7	5	8	3
Shoot height (cm)	9.44	17.45	20.00	16.00	20.00
Root length (cm)	5.70	19.50	16.00	21.00	13.55
Dry weight (mg)	4.32	6.70	8.62	5.92	8.23

Interaction between gibberellin and IAA was also noted. On combination of IAA (0.5 mg/l) and gibberellin (0.05 mg/l), a superior growth rate was manifested. When the concentration of IAA was increased to 1.0 mg/l, the gibberellin had to be increased to 0.3 mg/l simultaneously for promoting the height and dry weight of the seedlings (Table 9).

Table 9. *The growth of excised rice embryo in media containing both IAA (I) and gibberellin (G) at various concentration (mg/l)*

	0 0	0.05(G) 0.5 (I)	0.3 (G) 0.5 (I)	1.0 (G) 0.5 (I)	0.3 (G) 1.0 (I)
No. of root	3	7	4	3	6
Shoot height (cm)	8.2	22.55	17.53	14.35	20.45
Root length (cm)	4.3	16.54	8.54	7.00	13.53
Dry weight (mg)	4.9	8.45	6.60	6.42	7.90

Effects of carbohydrates on the culture of excised rice embryo

The optimum sucrose concentration for rice embryo culture was 4.0%. Seedling cultured at 2.0% and 8.0% were smaller than those cultured at 4.0%. But root length and dry weight of seedlings cultured at 8.0% were longer and larger than cultured at 4.0%. Sucrose concentration at 4.0% was optimum for root formation. Seedling at 4.0% and 8.0% sucrose media showed dark green color and strong. The seedling grew best at 2.0% glucose and seedling root developed well at 4.0% glucose. The dry weight of the seedlings was progressively increased with an increase in the concentration of glucose in the basal medium. The seedling color also became progressively more green and seedling showed stronger. Glucose concentration at 4.0% was optimum for root formation. Embryos grew poorly in soluble starch medium. Seedlings height and root length were short. The dry weights were less. Embryo cultured at media containing xylose showed very poor growth. Embryos cultured at 4.0% and 8.0% xylose media had little growth and died after cultured for one week (Table 10).

Table 10. *Effect of various concentrations of carbohydrates on rice embryo culture*

	Concentration (g/100 ml)	No. of root	Shoot height (cm)	Root length (cm)	Dry weight (mg)
Sucrose	0.5	6	12.14	4.36	3.46
	2.0	7	15.14	10.21	7.95
	4.0	9	19.16	11.65	14.81
	8.0	8	14.47	16.95	16.31
Glucose	0.5	5	14.20	7.57	4.74
	2.0	8	20.57	23.50	17.67
	4.0	9	18.73	25.00	28.47
	8 0	6	15.33	13.93	33.40
Soluble starch	0.5	3	7.08	3.10	1.78
	2.0	4	6.55	3.01	1.80
	4.0	5	6.26	2.20	2.16
	8.0	3	4.66	1.24	2.16
Xylose	0.5	2	3.25	0.87	1.22
	2.0	2	2.40	0.95	1.55
	4.0	1	1.33	0.3	1.13
	8.0	1	0.80	0.3	0.7

Some enhancing effect on embryo growth was noticed when 0.3 mg/l GA_3 was added to the media containing soluble starch. The media without this carbohydrate but with the same amount of GA_3 gave no such effect.

Table 11. *Effeect of combination of gibberellin (0.3 mg/l) with various concentrations of soluble starch in the White's medium on the growth of rice embryos*

	Without gibberellin and starch	Concentration (g/100 ml) of soluble starch				
		0	0.5	2.0	4.0	8.0
No. of root	1	1	3	3	4	5
Shoot height (cm)	0.8	0.8	6.95	6.16	7.30	7.07
Root length (cm)	0.3	0.3	3.05	4.45	3.02	2.07
Dry weight (mg)	0.7	0.7	2.15	2.20	2.60	2.73

When various amount of GA_3 were added separately to medium containing 2.0% soluble starch, however, it only slightly increased the embryo growth (Table 12).

Table 12. *Effect of combination of soluble starch (2%) with various concentrations of GA_3 in White's medium on the growth of rice embryos*

	Concentration of GA_3 (mg/1)				
	0	0.3	3.0	6.0	12.0
Shoot height (cm)	6.23	7.10	7.25	9.64	13.35
Root length (cm)	2.50	3.30	2.78	3.08	3.70
Dry weight (mg)	4.30	4.20	3.30	3.82	4.58

Effects of amino acid and coconut milk on the growth of excised rice embryos on the White's medium containing nitrate-nitrogen source or ammonium-nitrogen source

Embryos cultured in media containing glycine were better than those in the other amino acid media, 0.19 mg N/l as glycine in the medium contained NH_4^+ nitrogen source or 1.71 mg N/l as glycine in the medium contained NO_3^- nitrogen source supported best growth of seedlings.

The media containing higher concentration of aspartate or glutamate were unfavorable for seedling growth and root development. But the dry weight of the seedlings were not much affected when aspartate or glutamate concentration was increased. Arginine did not have promoting or inhibiting effect on rice embryo culture. (Table 13).

Coconut milk markedly increased dry weight of the seedlings. Seedlings cultured at media containing coconut milk were strong. But leaves of seedlings showed slight yellow (Table 14).

Table 13. *Effect of various concentrations (mg N/l) of glycine on rice embryo growth in White's medium containing ammonium-nitrogen or nitrate-nitrogen*

Amino acid	Nitrogen form in medium	Amino acid concentration (mg N/1)	No. of root	Shoot height (cm)	Root length (cm)	Dry weight (mg)
Without amino acid	NH_4^+		5	12.23	5.01	3.83
	NO_3^-		6	10.02	6.01	2.80
Glycine	NH_4^+	0.19	6	17.20	7.40	7.93
		0.57	6	16.40	5.85	7.01
		1.71	6	15.08	6.08	5.04
	NO_3^-	0.19	5	13.06	7.44	4.28
		0.57	5	14.24	8.40	4.30
		1.71	4	14.91	10.93	4.80
Aspartate	NH_4^+	0.19	6	11.84	3.92	5.52
		0.57	6	9.65	3.10	4.58
		1.71	8	11.40	1.75	5.01
	NO_3^-	0.19	5	11.00	5.90	3.94
		0.57	5	7.40	3.20	3.30
		1.71	6	7.70	2.08	3.80
Glutamate	NH_4^+	0.19	6	12.04	3.20	3.68
		0.57	6	9.01	2.90	3.46
		1.71	5	8.00	1.84	3.26
	NO_3^-	0.19	6	14.50	7.23	4.67
		0.57	7	10.98	5.98	4.92
		1.71	4	6.71	2.17	2.91
Arginine	NH_4^+	0.19	6	16.13	7.12	6.48
		0.57	4	12.64	5.42	4.84
		1.71	7	15.60	7.23	6.56
	NO_3^-	0.19	4	12.45	8.08	3.97
		0.57	5	11.43	7.90	4.00
		1.71	4	10.43	8.91	4.30

Table 14. *Effect of various concentrations (g/100ml) of coconut milk on rice embryo growth in White's medium containing ammonium-nitrogen or nitrate-nitrogen*

	NH_4^+ series				NO_4^- series			
	0	7.5	15.0	30.0	0	7.5	15.0	30.0
No. of root	5	7	6.5	6	6	6	5	7
Shoot height (cm)	12.23	11.81	12.10	11.50	10.02	11.53	10.10	14.88
Root length (cm)	5.01	6.51	7.91	6.87	6.01	7.50	7.77	9.56
Dry weight (mg)	3.83	9.00	13.05	17.08	2.80	7.67	8.00	19.14

Among several amino acids used in this experiment, glycine had the best enhancing effect on seedling growth, whereas aspartate and glutamate had inhibitory effect on seedling growth. Aspartate and glutamate induced root formation, but they restrained root development. Besides coconut milk, glycine had the best effect on increasing dry weight of seedling.

The seedlings cultured on NH_4^+ media were superior to that cultured on NO_3^- media. It has been demonstrated that mature rice embryos were favored more by ammonium-nitrogen than by nitrate-nitrogen. But it is particularly noted that NH_4^+ medium had unfavorable effect on root development. Root length of seedling grown at NH_4^+ series media was shorter than that at NO_3^- series media.

Discussion

According to the present investigation, temperature at 32°C and light intensity at 25,000 Lux was the best environment for rice embryo culture. This environment is similar to that for *Vanilla* embryo reported by Kundson (1950) and to that for small cotton embryo reported by Manuney (1960).

The results showed that most micronutrient deficiency caused chlorosis, and manganese deficiency caused brown necrosis on the chlorotic leaves developed from the embryo culture. Micronutrients often offered as compositional elements and activators of enzymes. Therefore microelements are also absolutely necessary for embryo growth. Eltinge (1941) reported that the chloroplast of tomato leaves are the first part of plant affected by manganese deficiency.

Promoting effect of vitamins on Dutura embryo culture has been demonstrated by Van Overbeek *et al.* (1944). However, we demonstrated that vitamins were required for rice embryo development only before formation of chloroplasts. Therefore, synthesis of thiamine, pyridoxine and nicotinic acid probably took place in the part of plant having chlorophyll. Pyridoxine (Vitamin B_6) deficiency was characterized by less dry weight and poor embryo growth. It is known that vitamin B_6 functions as a coenzyme in amino acid metabolism.

When IAA and gibberellin were applied simultaneously, their effect on rice embryo were more pronounced. It was also observed that high IAA when used with low kinetin had an enhancing root development as Lee (1962) reported. The results demonstrated that the hormones within rice embryo had a very complex interaction. Interaction between auxin and gibberellin in pea stem elongation was reported by Ocherse *et al.* (1967).

Carbohydrates were the most important factor for excised rice embryo culture. Sucrose was the most commonly used carbohydrate for plant embryo

culture. However, rice embryos grew better on glucose medium than on sucrose medium. Although 8.0% glucose was not the optimum concentration for seedling growth and root development, it produced the greatest increment in dry weight. This result of sucrose accords with that reported by Amemiya *et al.* (1956) According to this work, glucose could be directly utilized for carbon metabolism in rice embryo culture, although Zimmermann (1957) reported that sucrose but not hexose was the most prominent sugar translocated.

The utilization of soluble starch by rice embryo growth was enhanced by gibberellic acid. It was possible that the activity of α-amylase formed which degrades starch was stimulated by gibberellic acid in rice embryo, although α-amylase was generally produced in aleuron layer as that reported by Varner (1964) in barley endosperm.

Amino acid was not absolutely necessary in rice embryo culture when ammonium ion or nitrate ion was used as nitrogen source. Although many workers reported that many single amino acids were quite inactive as sources of nitrogen, results of this study showed that glycine had some promoting effect, whereas aspartate and glutamate at the concentrations between 0.19–1.71 mg N/l tested had some inhibitory effect on rice embryo growth. Increasing the concentration of aspartate or glutamate above 0.002 M, the growth inhibition was markedly reduced but reappeared at much higher concentrations.

Coconut milk markedly increased the dry weight of seedling. Coconut milk was known to contain many amino acids (Spoerl, 1948). The promoting effect caused by coconut milk was therefore probably due to the balanced mixture of amino acids. Gorter (1955) reported that the coconut milk contains "embryofactor" necessary for embryo growth of *Cyclamen.* The "embryofactor" of coconut milk has not been chemically identified. However, Lia (1970) reported that amino acid mixture could increase the cell yield by 50% as compared with that in absence of both coconut milk and amino acid mixture in cell suspension culture. Therefore, amino acid and coconut milk were useful in rice embryo culture medium.

Although many workers reported that ammonium ion is only effective at pH value close to neutral in cell and callus culture (Hannay *et al.*, 1959; Nihgingale, 1937), the result of this work showed that White's medium, except supplimented with coconut milk, with ammonium as nitrogen source at pH 5.5 was more effective than that with nitrate for rice embryo culture. The root length of seedling cultured on ammonium medium, however, was shorter than that cultured on nitrate medium. There are two possible explanations for that: (1) ammonium ion has some inhabitory effect on root development; (2) seedling growing in ammonium ion medium is not necessary to have long root for absorbing nutrient.

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水稻切離胚的營養研究

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1. 水稻切離胚的培養試驗，以光度 25,000 Lux 及溫度 32°C 時，對幼胚的生長發育最爲適當。

2. 培養基中缺乏任何一種微量元素時，幼葉黃化，生長率降低，根發育不良，乾量減輕。

3. 維他命對水稻幼胚生長的影響僅限於葉綠素未形成之前，因培養基中維他命的供給與否都沒顯著的效果。此可能幼苗因自行光合作用後，自身可合成維他命。

4. 各生長激素最能促進幼胚發育的濃度；IAA 爲 0.5 ppm，Kinetin 爲 0.01 ppm，而 GA_3 爲 0.3 ppm。如生長激素混合使用時，它們的濃度必須有適當的調節配合，激素間濃度會相互影響。

5. 培養基中碳水化合物的濃度過低，對切離胚的乾重量顯著減少。就種類來說，以葡萄醣 2.0% 效果最好，次爲蔗醣 4.0%。最不易被利用的是木醣，次爲水溶性澱粉。但如加入 GA_3 於含有水溶性澱粉之培養基中，則胚的生長略有增加。

6. 氨基酸在水稻胚培養試驗中，效果不一致，glycine 有促進生長的作用，而 glutamate 及 aspartate 在 0.19–1.71 mg N/l 的濃度，則有抑制生長的反應。

7. 培養基中氮源以氨態比硝酸態爲優，但氨態對根的發育亦不理想。

8. 培養基中加入椰子汁者比沒加入者能增加切離水稻幼苗的乾量約四倍。