

# Photoproduction of Hydrogen by Vishakapatnam Photosynthetic Bacteria and their Effect on Different Cultural Conditions

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## **Abstract:**

*Hydrogen, well advised by many scientists, as an ecofriendly and optimal fuel for the imminent future, were well out in plentiful applications. Rhodospseudomonas species have shown an immense grade of morphological and structural analogy, however, they are phylogenetically quite diverse. Therefore, the present study for Hydrogen production affirms that the influence consortium of purple non sulphur bacteria isolated from Vishakapatnam marine water. The different parameters like various Carbon, Nitrogen, Growth factors and  $P_H$  variation was identified for hydrogen production by the anoxygenic bacteria. The amount of hydrogen produced varies with various carbon, nitrogen, growth factors and  $P_H$  used in the medium. The bacterial culture was cultured in 50ml vessel within 30ml of culture to produce hydrogen in anoxygenic conditions and incubated in 2000lux light intensity for 196 hours. The best result for highest hydrogen productivity was 6ml/30ml of sodium benzoate, ammonium chloride, cyanocobalamine culture and at  $P_H$  7.0, 7.5 to purple non sulphur bacteria isolated from Vishakapatnam marine water.*

## **Keywords:**

Purple non sulphur bacteria; Hydrogen production; carbon; nitrogen sources; growth factors;  $P_H$

## **Introduction:**

In consonance with sustainable expansion and waste minimization issues, biological hydrogen production from renewable sources, also known as “green technology” has revived attention in

recent years. Morphological, cytological and physiological properties of the genus *Rhodospseudomonas* have been outlined [1-3]. Marine habitats serve fine niches for anoxygenic phototrophic bacteria, which are extensively assigned in diverse coastal marine habitats. Most common anoxygenic phototrophic bacteria have been isolated from estuarine salt pans, salt marshes, coastal lagoons with elevated salt concentrations, tidal waters; brackish waters and marine coastal sediments. They have even been found in the extreme marine habitats of Antarctica [4-11]. The peculiar substrates such as acetate, lactate, benzoate, malate, mannitol and starch are outlined by phototrophic bacteria as an electron donor for hydrogen production. Impact of various cultural conditions on hydrogen production is expressed [12-15]. Specially from hydrogen production multifold by-products are produced from photosynthetic bacteria makes a continuity for our earlier work on biotechnological applications [16-26]. The present aspects of the above data, reveals the impact of cultural conditions like various carbon, nitrogen, growth factors and pH variation on hydrogen production by anoxygenic bacterial consortium isolated from Vishakapatnam marine water was examined and explained.

## **Materials and Methods:**

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Purple non Sulphur an oxygenic Phototrophic bacterium was isolated from marine water samples by enrichment techniques by inoculating into the medium and incubated anaerobically under 2000 lux light intensity. A selected consortium isolate from Vizag marine water that had passed both screening tests was characterized using both morphological and physiological properties and identified according to Bergey's Manual of Systematic Bacteriology (1994). Growth conditions were measured at 660nm Optical density under UV-VIS Spectrophotometer. The harvested cells were obtained from the 30ml of bacterial culture centrifuged (10,000xg for 30mins) and washed with 0.3% saline and cells were suspended in basal medium. Based on experimental conditions provided at different concentrations of electron donors, nitrogen sources, ten days old cultures of phototrophic bacteria of 1 % (v/v) concentration into the basal medium were inoculated containing carbon sources Sodium benzoate, Glucose, Galactose, Mannose, Arabinose, Lactose, Mannitol, Malic acid, Citric acid, Sodium succinate with ammonium chloride as nitrogen source. Ammonium chloride, ammonium nitrate, glycine, sodium glutamate, histidine, tryptophan, tyrosine, threonine, alanine with sodium benzoate as carbon source, different growth factors like pantothenic acid, nicotinic acid, biotin, folic acid, riboflavin, cyanocobalamin and basal medium at different PH variations from 6.5, 7, 7.5, 8, 8.5, 9, 9.5 were also explored. The incubation period was 196 hours after inoculation of the consortium. The technique used for hydrogen measurement was water displacement method where as

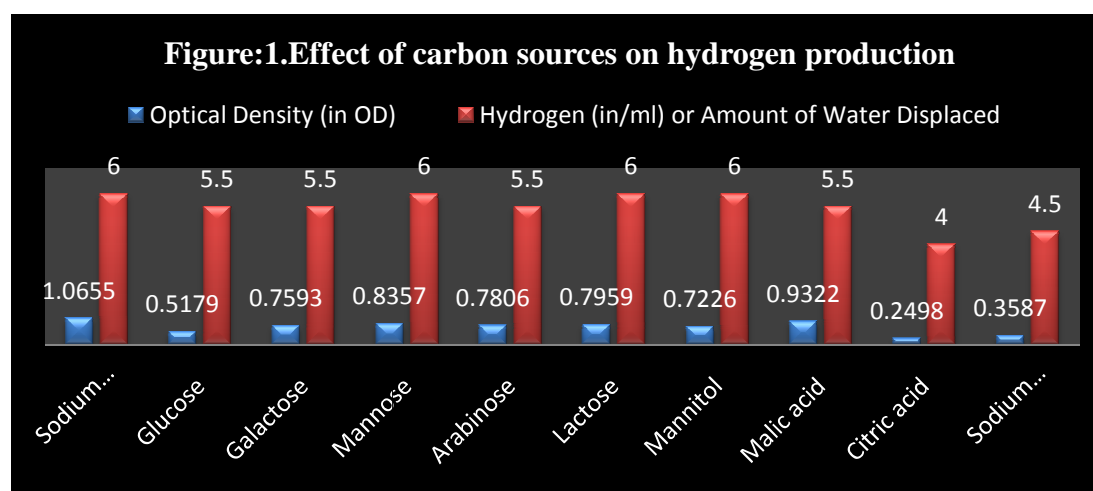
Gas Chromatography was used for gas analysis.

### **Results and Discussion:**

Ten day active cultures were used to assess their probability of producing hydrogen was shown in **Table 1 and Figure 1**. Photosynthetic bacterial consortium produced varying amounts of hydrogen with various carbon, nitrogen, growth factors and variation in  $P_H$  under anaerobic light. Sodium benzoate, mannose, lactose and mannitol showing maximum and followed by equal volumes in glucose, galactose, arabinose and malic acid were good carbon sources for production of hydrogen by photosynthetic bacterial consortium. Maximum production of hydrogen was 6ml /30ml culture was produced in presence of sodium benzoate. Effect of various nitrogen sources on hydrogen production was shown in **Table 2 and Figure 2**. In the presence of anaerobic conditions, ammonium chloride produces 6ml/30ml culture which shows a maximum amount of hydrogen followed by ammonium nitrate, sodium glutamate and tryptophan. Thus, maximum hydrogen production of both the carbon and nitrogen sources was observed in sodium benzoate, ammonium chloride. Glucose and galactose shows equal volumes of hydrogen and ammonium nitrate, sodium glutamate, tryptophan has shown the equal volumes of hydrogen production. Impact of  $P_H$  variation on hydrogen production was described in **Table 3 and Figure 3**. Maximum amount of hydrogen production was observed in 7.0 and 7.5  $P_H$ . Biotin, folic acid and riboflavin showed the equal amounts and maximum hydrogen production was observed in nicotinic acid and cyanocobalamin was shown in **Table 4 and Figure 4**.

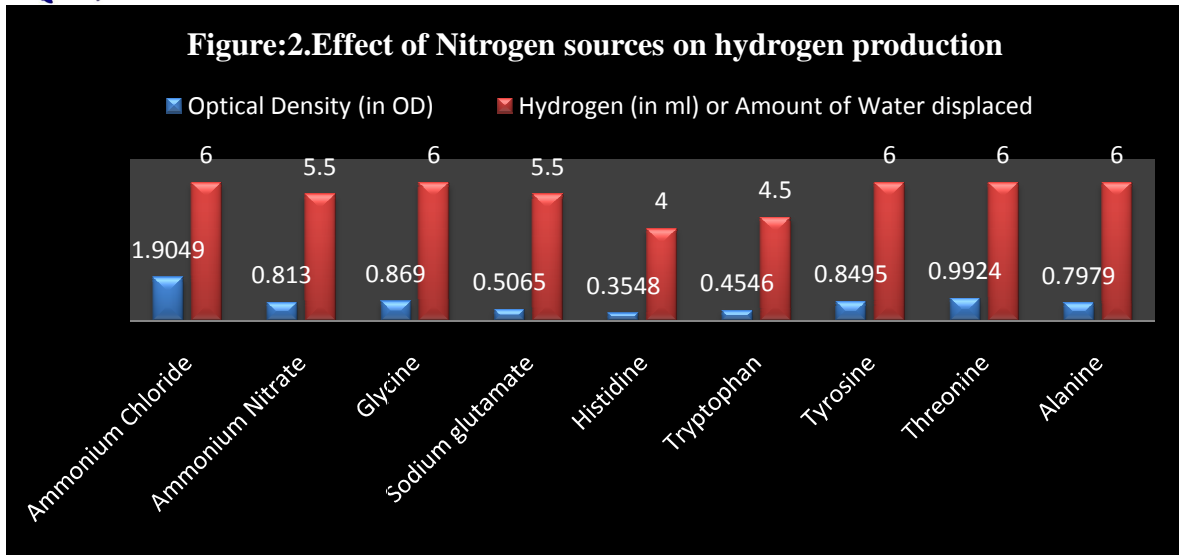
**Table 1. Effect of various Carbon Sources on Hydrogen Production (ml/30ml culture)**

Optical Density (in OD)	Carbon /Electron Donor	Hydrogen (in ml)
1.0655	Sodium Benzoate	6±0.0
0.5179	Glucose	5.5±0.1
0.7593	Galactose	5.5±0.2
0.8357	Mannose	6.0±0.1
0.7806	Arabinose	5.5±0.2
0.7959	Lactose	6±0.1
0.7226	Mannitol	6±0.0
0.9322	Malic acid	5.5±0.1
0.2498	Citric acid	4.0±0.6
0.3587	Sodium succinate	4.5±0.4



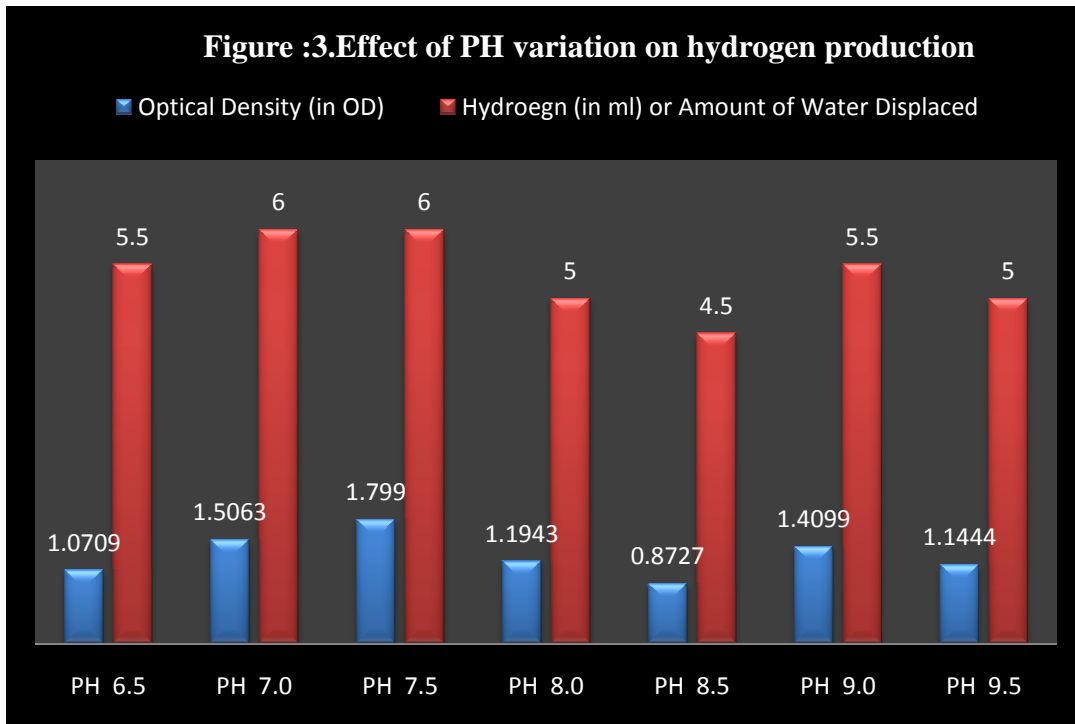
**Table 2: Effect of various Nitrogen sources on Hydrogen production (ml/30ml)**

Nitrogen Source	Optical Density (in OD)	Hydrogen (in ml)
Ammonium Chloride	1.9049	6±0.2
Ammonium Nitrate	0.8130	5.5±0.2
Glycine	0.8690	6.0±0.2
Sodium glutamate	0.5065	5.5±0.4
Histidine	0.3548	4±0.5
Tryptophan	0.4546	4.5±0.2
Tyrosine	0.8495	6±0.0
Threonine	0.9924	6±0.1
Alanine	0.7979	6±0.1



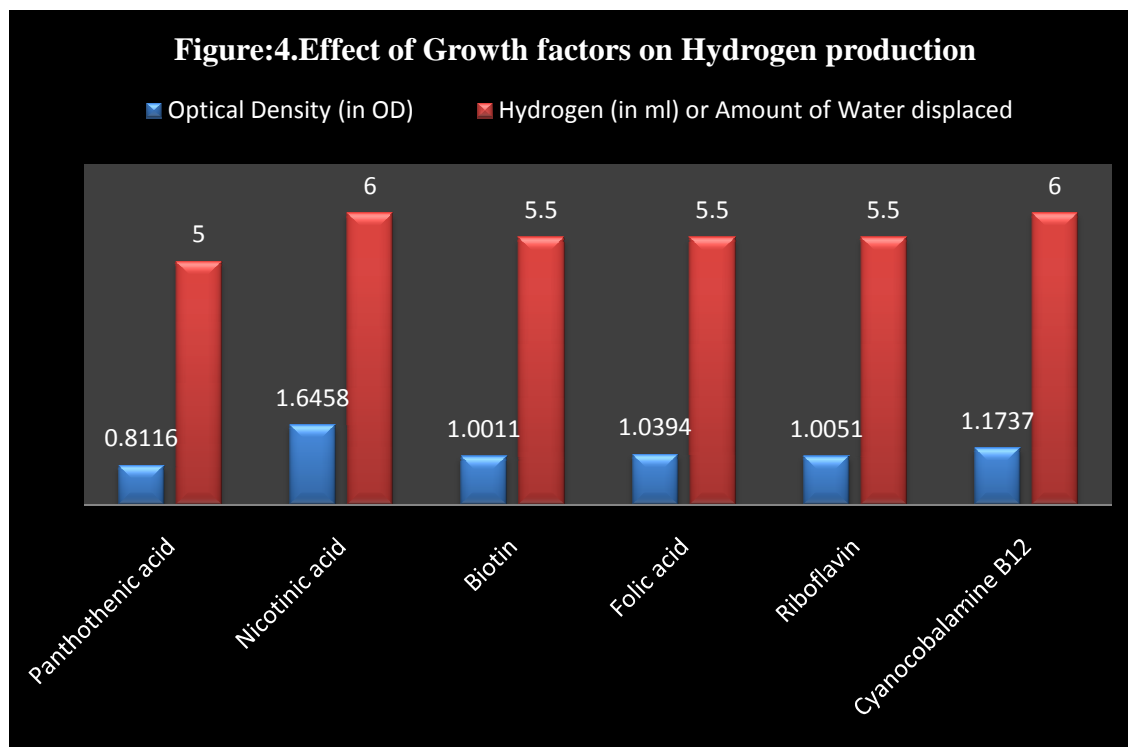
**Table 3: Effect of various P<sub>H</sub> on Hydrogen Production (ml/30ml)**

P <sub>H</sub>	Optical Density (in OD)	Hydrogen (in ml)
P <sub>H</sub> 6.5	1.0709	5.5±0.2
P <sub>H</sub> 7.0	1.5063	6.0±0.0
P <sub>H</sub> 7.5	1.7990	6.0±0.1
P <sub>H</sub> 8.0	1.1943	5.0±0.1
P <sub>H</sub> 8.5	0.8727	4.5±0.4
P <sub>H</sub> 9.0	1.4099	5.5±0.1
P <sub>H</sub> 9.5	1.1444	5.0±0.3



**Table 4: Effect of various Growth Factors on Hydrogen production (ml/30ml)**

Growth Factors	Optical Density (in OD)	Hydrogen (in ml)
Panthenic acid	0.8116	5.0±0.5
Nicotinic acid	1.6458	6.0±0.1
Biotin	1.0011	5.5±0.3
Folic acid	1.0394	5.5±0.3
Riboflavin	1.0051	5.5±0.2
Cyanocobalamine B <sub>12</sub>	1.1737	6.0±0.1



**Conclusion:**

The present study reveals that impact of various carbon, nitrogen, P<sub>H</sub> variation and growth factors shown that equals and maximum amounts of hydrogen production was shown in sodium benzoate, ammonium chloride, P<sub>H</sub> 7.0, 7.5 and nicotinic acid, cyanocobalamine and less amounts of hydrogen production in citric acid, histidine and panthothenic acid and at P<sub>H</sub> 8.5.

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