NOTE

**Bioconversion of Mango Processing Waste to Fish-feed by Microalgae Isolated from Fruit Processing Industrial Effluents**

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Fifteen different strains of blue green algae (including one green algae) collected from the wastewaters of fruit processing industrial areas were screened for production of algal biomass from mango processing waste. The utility of microalgae was evaluated based on feeding trials of the biomass to the fish *Tilapia mossambica*. Only the growth of fish in terms of body mass gain was measured. The mango waste after removing the stones was macerated, and then diluted with water to 15-20 per cent mango waste and inoculated with blue green algae. The cultures were continuously illuminated (4000 lux light intensity) for 15-20 d. Among various strains of algae tested, only two (*Scytonema sp* and *Microcystis sp*) were found to be toxic while several others gave promising results. Through such efforts mango waste could be converted into fish-feed and then through fish growth into important protein source for human consumption.

**Introduction**

Fruit processing industries generate considerable amount of waste during processing and its disposal is a big problem leading to pollution. The mango processing industry generates approximately 50 per cent of the fruit handled as waste comprising skin, stones and fibre that adds up to some 9.2 m t/y in India. Mahadevaswamy and Venkataraman reported that the mango waste contained (per cent, dry weight basis) moisture (6.4), crude protein (10.3), starch (16.2), hemicellulose (10.2), total reducing sugars (4.0), pectin (7.0), lignin (20.0) and ash (5.5). Thus, a rich source of sugars and proteins from the processing industry waste is an untapped source of energy and protein which when dumped as waste causes environmental hazard. It could otherwise be utilized for converting into value added products using biotechnological processes.

Microorganisms could convert wastes into biomass that might be a potential source of food, either directly or as a feed for fish and poultry. Fungi, algae and heterotrophic bacteria have been known from time immemorial; however, anoxygenic phototrophic bacteria with a proven potential are still largely untapped and underexploited. Earlier Noparatnarporn and Nagai reported that pineapple waste could be used as a potential feedstock for cultivation of anoxygenic phototrophic bacteria. Some earlier studies have reported the possibility of nitrogen fixing marine blue green algae as a potential source for feeding in aquaculture.

The present study was taken up for utilizing the industrial waste from a local mango processing unit by exploring the possibility of growing a few blue-green algae on the waste, and subsequently using the biomass as a source of feed for aquaculture. These studies serve a dual purpose — first to safely dispose off the mango processing waste, and secondly the waste is converted to biomass, which can be effec-
tively used as fish feed. Some reports in the literature indicated the acceptability of blue green algae as fish feed in aquaculture systems\(^a\). *Tilapia mossambica* was chosen as a model fish for feeding trials as they are known to thrive under conditions deleterious to many other fish and feed on organisms low in the food chain hierarchy, e.g., algae.

**Materials and Methods**

*Isolation and Identification of Blue Green Algae and Inoculum Preparation*:

Samples of wastewater from nearby areas receiving industrial effluents from fruit processing industries in Hyderabad were collected in sterilized glass bottles under routine microbiological precautions. Experiments were carried to grow the blue-green and green algae in Allen's medium and BG medium\(^b\). They were identified, purified and characterized according to the method of Rippka *et al.*\(^c\). All experiments were replicated three times and done under routine sterilized conditions.

*Preparation of Mango Waste Medium*:

Hard stones were removed from the mango waste before taking up for the growth of blue green algae. The remaining material comprised mostly mango-peel with some amount of pulp adhering to it. The mass was weighed and macerated in a domestic blender with known volumes of water into homogeneous thick slurry, autoclaved and kept in a refrigerator. Dilutions were made from the slurry with water as and when required depending upon the nature of the experiment. Normally, dilutions of the order of 20 per cent were employed.

*Growth on Mango Waste Medium*:

For preparing inoculum the strains were subcultured in synthetic BG11 medium in 1 L flask with a light intensity of 4000 lux at 26-28 °C. The flasks were provided with continuous aeration (50 mL/L/min) all through the experiment by means of an aerator. No additional micro- or macro-nutrients were added and the algae were harvested after 20 d. They were inoculated (6 g dry weight/L) into 20 L diluted (20 per cent) mango waste slurry. Sterilized carboys of 20 L capacity were used to grow the blue green algae. They were continuously illuminated by fluorescent bulbs (approximately 4000 lux) from a distance of 10-12 cm. The carboys were provided with continuous aeration (500 mL/min) all through the experiments, the temperature of the system varied with ambient temperature, which ranged from 26-30 °C. Sufficient quantity of biomass was harvested after 15-20 d which was used subsequently for feeding trials.

*Harvesting of Blue Green Algae*:

Most of the algae (filamentous forms) were collected on a four-fold thick cotton cloth. The remaining filaments which were drained in the filtrate were centrifuged at 10000 rpm common for collecting the unicellular forms. The algae were weighed (both for fresh and dry weights) and then fed fresh to the fish.

*Feeding Trials*:

*Tilapia mossambica* fish was chosen as a model fish for carrying out the feeding trials. The fingerlings were procured from a local government agency (Andhra Pradesh Fisheries Development Corporation, Hyderabad) where they were reared in fresh water ponds. They had an initial weight of 1-2 g. These were acclimatized indoors for 3 wk prior to use in experiments in a 60 L glass aquaria (61.5x30.5x31 cm) under constant illumination and were fed with a normal diet (control diet) comprising rice bran and oil-cake in the composition of 7:3 as specified by A P Fisheries Development Corporation. The fingerlings were transferred for feeding trials into aquarium containing fresh water and provided with illumination (4000 lux from top and side of the aquarium). The aquarium was aerated with an aquarium pump. Fish were starved for 24 h prior to initial and final weighing in all feeding trials.

Comparison was made between the fish fed on the control diet and those fed on algae. Water in the aquarium with algae feed was not exchanged (emptied and refilled) during feeding periods unlike the aquarium containing fish being fed on control diet (rice bran and oil cake). The water in this aquarium was changed every alternate day to minimize accumulation of ammonium ions generated from the droppings of the fish which might prove toxic to the fish and result in their mortality as experienced by Almeda *et al.*\(^d\).

Six fish were put into the 60 L glass aquarium (numbering six at a time) and a feed of 3 g/d/aquarium was maintained for 15 d initially. In a few cases, all of the algal diet was consumed
in less than 15 h. Hence the feed was increased to 5 g/d/aquarium till the termination of the experiments.

Results and Discussion

Most of the algae (filamentous forms) formed in the form of inter-linked mat. The COD of the medium was brought down to nearly 6000 mg/L from an initial level of 96000 mg/L. The colour of the medium, which was orange brown initially changed to light mustard yellow with no odour except faint ketonic smell of mango without much total dissolved solids in it.

The harvested biomass was fed to the fish in aquaria. The utility of the biomass as fish-feed was measured in terms of gain in fish body weight. The representative results and the screening trials involving fifteen blue-green algal strains are given in Table 1. Several strains of Anabaena, Nostoc and Oscillatoria supported the highest growth rates of the fish when the strains were selected for further testing in triplicate trials. Studies were conducted simultaneously by comparing the growth of fish fed on algal feed to that of those fed on the control diet. Majority of the blue-green algae supported growth of the fish to a great extent. Of the 15 strains tested, only four strains produced no increase in body weight while two proved to be fatal. The overall survival rate was 87 per cent.

The growth rate varied highly over strains of blue-green algae. Although the feeding trials were not of long duration, differences in growth responses of fish were readily discernible in control and those fed on algal diets.

It was noticed that two strains (Scytonema sp. and Microcystis sp.) were toxic and resulted in 100 per cent mortality due to subcutaneous haemorrhage in less than 96 h.

The results obtained so far demonstrated that the blue-green algae were generally acceptable as diet for Tilapia. The growth response of the fish fed on algae indicated that the algal feed had an edge over the control diet, which was apparently deficient in some nutrients, mainly vitamins, minerals and growth regulators. The results showed that 9 out of 15 strains of algae tested were effective in utilizing the mango processing waste. Chlorella, Anabaena, oscillatioria and Nostoc resulted in high average daily gain in the body weight of the fish that in turn is a good protein source for human consumption.

Conclusions

Some of the blue-green algae, which have the potential to convert fruit processing waste into biomass are extremely important in converting wastes into useful products which help reduce industrial pollution and protect the environment.

The harvested biomass is a good feed for Tilapia mosambica. The blue-green algae, which could utilize the waste from the processing industry could play an important role in the bioconversion of the processing waste into an alternative protein source for human consumption through aquaculture. The trials made in the present work, though for a shorter duration, are only a prelude to systematic studies to test further this hypothesis. The microbiological conversion of processing wastes into biomass in large scale could prove to be economical, safe, convenient and sustainable.

| Table 1—Tilapia mosambica fed on green and blue-green algae grown on mango waste |
|---|---|---|---|
| *S.* No. | Diet | Initial weight, g | Final weight, g | Weight gain, mg/d |
| 1 | Chlorella sp. | 1.981 | 32.280 | 142 |
| 2 | Chlorella sp. | 2.566 | 34.300 | 148 |
| 3 | Gloeotrichia sp. | 2.699 | 12.580 | 46 |
| 4 | Gloeotrichia sp. | 2.503 | 15.221 | 60 |
| 5 | Anabaena sp. | 2.892 | 40.041 | 174 |
| 6 | Anabaena sp. | 2.998 | 37.489 | 161 |
| 7 | Anabaena sp. | 2.621 | 38.021 | 166 |
| 8 | Scytonema sp. | 2.780 | Toxic | — |
| 9 | Scytonema sp. | 2.890 | 20.386 | 82 |
| 10 | Microcystis sp. | 3.801 | Toxic | — |
| 11 | Oscillatoria sp. | 3.621 | 40.021 | 170 |
| 12 | Oscillatoria sp. | 2.762 | 20.655 | 84 |
| 13 | Oscillatoria sp. | 2.861 | 38.760 | 169 |
| 14 | Oscillatoria sp. | 2.690 | 40.621 | 178 |
| 15 | Nostoc sp. | 2.742 | 40.220 | 175 |
| 16 | Rice bran + oil cake (control diet) | 2.850 | 16.109 | 62 |

Results are means of three replicates over 15-20 d feeding trial.
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