VOL 7 ISSUE 4 December 2015

AQUAFEED

ADVANCES IN PROCESSING & FORMULATION

An Aquafeed.com publication

Functional Feed Additives

▪ India's Waterbase ▪ Using Haemocytes in the Battle Against WSSV and AHPND

Published by: Aquafeed.com LLC. Kailua, Hawai'i 96734, USA. ▪ www.aquafeed.com ▪ info@aquafeed.com

AQUAFEED
Advances in Processing & Formulation

Volume 7, Issue 4

Contents

** Cover story*

AQUAFEED
Advances in Processing & Formulation
Volume 7, Issue 4

Index to advertisers

aquafeed.com
~1998~

From the Editor

As we head rapidly towards the end of 2015, we take a look at some of the multitude of functional feed additives that are available to aquafeed producers today, from phytonutrients to organic acids, clays and algae. We also focus on mycotoxin contamination, which has been found at high levels in aquafeeds, often with more than one type, posing serious potential health threats to aquatic species. Stories

Our next issue will tackle the important issue of water activity and feeding marine fish. We will also have a sneak peek at the FIAAP/VICTAM/GRAPAS Asia exhibition, taking place in Bangkok at the end of March. Advertising and editorial copy deadline is March 1, 2016.

All of us here would like to thank you for being part of our Aquafeed.com community and wish you a very happy, healthy and prosperous 2016.

Suzi Dominy, Editor & Publisher

SUBSCRIBE

Subscription is free to industry. Subscribe from Aquafeed.com or by clicking the links below:

- Aquafeed: Advances in Processing & Formulation
- The Aquafeed Newsletter (weekly)
- Hatcheryfeed Annual Guide to Hatchery Feeds & magazine

Published by Aquafeed.com, LLC.
Kailua, Hawaii 96734, USA.

Contact us

Editor/Publisher:
Suzi Dominy
editor@aquafeed.com

Associate Editor:
Roger Abbott
editor@aquafeed.com

Advertising:
sales@aquafeed.com

Technical feed consulting:
Warren Dominy, Ph.D.
consulting@aquafeed.com

Protecting shrimp from early mortality syndrome (EMS)

By Ylva Sundstrom, Kashinatham Alisala, Alejandro E Nyul, Sara I Hash and Suresh Menon

Menon Renewable Products, Inc., San Diego, CA, USA.

A study was conducted to evaluate the survivability of shrimp fed MrFeed® when exposed to the pathogen responsible for causing Early Mortality Syndrome (EMS), *Vibrio parahaemolyticus*. Tests were performed by the University of Arizona to compare MrFeed® produced by Menon Renewable Products, Inc. alongside a commercial brand feed. The shrimp were fed for 21 days, and then exposed to an infectious dose of EMS-causing *V. parahaemolyticus*. The shrimp control group fed the commercial feed had confirmed shrimp mortalities from the first day after disease exposure, and showed an average of 10% survivability. All shrimp fed MrFeed® in this study demonstrated high survivability numbers, averaging 90% at day seven after pathogen exposure.

BACKGROUND

The shrimp farming industry has been expanding rapidly because of high consumer demand. To maximize profits, farmers have pushed for higher density farms to produce more shrimp per hectare. Crowded conditions increase the yield per acre, and potential profits to the farmer, but also increase the risk of disease. Shrimp throughout South East Asia and other important shrimp farming centers around the world have been seriously affected by early mortality syndrome (EMS). EMS, also known as Acute Hepatopancreatic Necrosis Disease (AHPND), is caused by a pathogenic strain of the bacterium Vibrio parahaemolyticus which colonizes the shrimp gastrointestinal tract and causes destruction and dysfunction of the hepatopancreas, a digestive organ. Infection leads to elevated levels of shrimp mortality and causes devastating economic losses to farmers.

Proper monitoring and management of shrimp ponds can reduce the severity of the disease, but once an area has been struck by the infection it is difficult to eliminate the disease. A central part of shrimp farming is to provide the shrimp with a complete and nutritious diet to manage and reduce disease.

MENON RENEWABLE FEED, MRFEED®

Menon Renewable Products, Inc. has developed a feed product which is produced using a unique and patented fermentation process. The fermentation process converts plant-based carbon sources such as grains and other cellulosic and hemicellulosic materials into more easily digestible forms, and provides essential micronutrients in the form of prebiotics. MrFeed® also provides valuable probiotic properties, which help establish a healthy microbiome in the shrimp gut as well as in the pond. The fermented product is formulated into a complete feed product by adding supplemental vitamins and nutrients, according to the specific feed requirements of shrimp.

Three feeds were tested in this trial: A control diet consisting of a commercial brand feed used in aquaculture (Rangen Inc. Shrimp Production Feed 40 3/32 Buhl, Idaho) and two formulations of the MrFeed® diet, referred to as MrFeed OB1 and MrFeed OB2. OB1 and OB2 diets

differed in the feed formulation, namely the total amount of fats and soluble carbons.

MATERIALS AND METHODS:

All experiments were carried out at Aquaculture Pathology Laboratory, University of Arizona and performed by its personnel.

Tank set-up and feeding:

Specific-pathogen free (SPF) *P. vannamei* were obtained from Shrimp Improvement System, and reared at University of Arizona. A total of three 1,665L capacity tanks were filled to 1,382L volume with artificial seawater (Chrystal Sea Marine Mix, Marine Enterprises International). All tanks were equipped with air spargers to maintain oxygen levels in tanks and pre-acclimated biological filters to maintain water quality. Temperature of tanks was maintained at 26 °C. Tanks were monitored weekly to verify that temperature, salinity, and nitrite levels remained constant. At the start of the trial, shrimp weighing an average of 1.3 g were weighed and transferred to the experimental tanks. Each tank was fed one of three experimental feeds (Rangen, MrFeed OB1, or MrFeed OB2) twice daily, with 2.5% of their average body weight at each feeding, adding up to the daily total feeding at 5% of the shrimp body weight.

After 21 days, survivability was calculated before transferring the shrimp in smaller groups into glass aquariums in preparation for the disease exposure test.

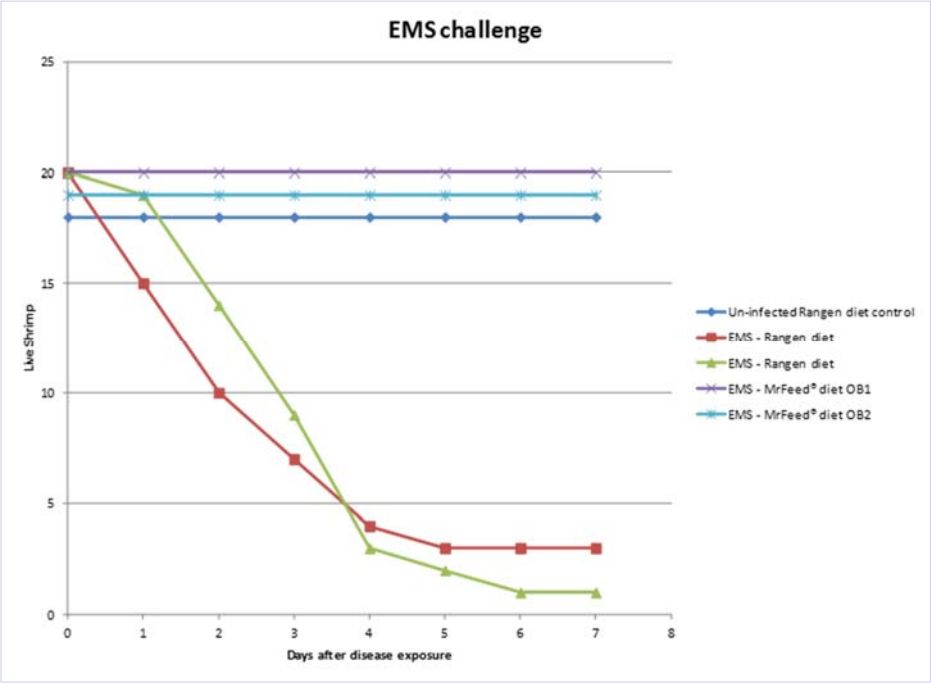
Disease challenge:

The disease challenge was carried out in glass aquariums filled to 70L volume. Each aquarium was outfitted with a pre-acclimated biological filter, and aerated. Tanks were covered to reduce the risk of cross contamination or loss of shrimp from jumping. The EMS disease challenge was started after dividing the shrimp into experimental glass aquariums, each containing 19-20 shrimp. Each aquarium was fed the same experimental feed as during the initial 21 day period (Rangen, MrFeed® OB1, or MrFeed® OB2).

To get a positive control of survivability during the disease study, two control aquariums were used to determine if the dose of *V. parahaemolyticus* was sufficient to cause EMS disease. Mortalities were expected in both positive control tanks within the first 24h. If mortalities were not observed within this timeframe, all tanks in the experiment would get a second infectious dose of V. parahaemolyticus. MrFeed® OB1 and OB2 fed tanks had one replicate each. Negative control tanks from each feed condition were intentionally never infected.

A consistently virulent strain of *V. parahaemolyticus* (strain 13-028A/3) was incubated for 18h in tryptic soy broth plus

2% sodium chloride. The absorbance at OD600 was measured at 1.847 and the colony forming units were 2.15*109 cfu per mL. The infective broth was directly introduced to the challenge tanks. After the addition of the infectious bacteria, daily observations were made, and the twice daily feedings continued in the same pattern as before the disease exposure for a total of 7 days. Daily observations were made to note dead or moribund shrimp. Moribund animals were noted, and dead animals were removed from the tank, counted, and noted as confirmed mortalities.



RESULTS

During the initial feeding period, diet group OB1 had 99% survivability, OB2 had 98% survivability and the control Rangen fed group had 99% survivability. The high survivability for all the different feeds indicates that all the feeds tested in the study were consumed, and that the shrimp stayed overall healthy throughout the 21 days. Based on this initial survivability under standard growth conditions, the difference in survivability between the feed formulations OB1 and OB2 are insignificant.

After disease exposure, the first shrimp mortalities were observed in both of the exposed Rangen-fed tanks on the first day of observation after EMS infection. The total recorded mortalities over time are summarized in Figure 1. Mortalities were recorded daily until day 7, until no more daily mortalities were observed. The disease challenge was terminated on day 7 after exposure.

There was a combined survival of 10% in the Rangen fed positive control group. In the aquariums that housed shrimp that were acclimatized to MrFeed®, there were no confirmed observed mortalities. On day one, one potentially moribund animal was observed and noted for the OB2 fed tank, but on day two, there were no visible remains of dead shrimp. This could be due to cannibalization of the shrimp. At the end of the feed trial, three animals were unaccounted for. These animals could have been cannibalized or escaped during handling of shrimp and initial stocking of tank. Neither of the MrFeed® fed aquariums had any noted mortalities throughout the 7 day study. Survivability for the MrFeed fed tanks at the conclusion of the study was 95% for MrFeed® OB1 and 84% for MrFeed® OB2. For the control tanks which were not exposed to the pathogen, the Rangen control had overall survival of 89% while MrFeed® OB1 feed had 90% survival, and MrFeed® OB2 had 100% survival.

CONCLUSION

MrFeed®, a unique feed product produced through fermentation of cellulosic and hemicellulosic materials, provides shrimp with increased resistance to EMS pathogen exposure. EMS has afflicted hundreds of thousands of tons of shrimp over the last 5 years and has decimated shrimp production in regions of South East Asia. In combination with good farming techniques, use of MrFeed® can lead to more sustainable shrimp farming.

More information

Traci Johnson, Menon Renewable Products

E:tjohnson@menon.us