An Investigation of the Survival Level of *Oreochromis mossambicus* fry variably kept in a Closed System: Laboratory Experiment

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An Assignment presented in partial fulfillment of the requirements for the degree of Masters of Philosophy in Livestock Industry Management (Aquaculture) at the University of Stellenbosch.

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I, the undersigned, herby declare that the work contained in this assignment is my own original work and that I have not previously in its entirety or in part submitted it at any university for a degree.

Signature:…………………………....   Date:………………………..

Abstract

This paper contains literature on tilapia culture, feeding and nutritional factors of prime consideration to survival and growth of *Oreochromis mossambicus*. Results are presented for a three-phase laboratory experiment on survival of *O. mossambicus* fry in an attempt to evaluate the use of live *Spirulina platensis*. The experiment was conducted on fry in a closed system in an effort to maximize the use of live Spirulina and also optimize growth and production.

Fry were tested for tolerance levels of salinities, 0-35 g/lt, and showed favourable survival rates up to 15 g /lt salinity without being fed. Manipulation of input in freshwater turned high fry mortalities with increasing rates without difference for physical form of Spirulina. Growth was not significantly affected by types of input. However fry grew well at 0-40% rates with considerable survival performance. It is noted good quality of water that allow improved survival and growth of fry in a closed system may be assured with rates of input up to 5 or 10% of bodyweight. These input rates can guide use of live Spirulina in saline water tilapia culture if Spirulina proves good productivity at the consistency of fry tolerance to the salinity levels established in this paper.

Opsomming

Hierdie skripsie bied 'n oorsig oor die beskikbare literatuur rakende die kweek van tilapia met verwysing na voeding en die belang daarvan by die oorlewing en groei van Oreochromis mossambicus. *Dit bied ook resultate van 'n drie-fase laboratorium eksperiement oor die oorlewing van jong* O. mossambicus *vissies wat in afsonderlike houers aangehou is in 'n poging om die voedings profiel van* Spirulina *platensis beter te benut. Die prestasie van die klein vissies in 'n geslote sisteem was nuttig om die gebruik van lewendige* Spirulina *asook groei en produksie te optimaliseer. Die verdraagsaamheid van ongevoerde vissies jeens vlakke van southeid tussen 0-35 g/lt was goed en oorlewing is waargeneem tot en met southeid vlakke van 15 g /lt. 'n Toename in mortaliteit van vissies is waargeneem met 'n toename in voedings persentasie tydens die manipulering van die fisiese vorm (poeier, korrels of vlokkies) en persentasie* Spirulina *gevoer. Die fisiese vorm van* Spirulina *en die tipe rantsoen (*Spirulina *en tilapia rantsoen gekombineerd) het geen invloed op die mortaliteit gehad nie. Soortgelyk is groei ook nie betekenisvol beïnvloed deur die fisiese vorm van* Spirulina *nie. Vissies het egter goed gegroei by 'n voedingsvlak van 0-40% met 'n betekenisvolle hoër oorlewings persentasie. Opmerklik was die feit dat goeie water kwaliteit bevorderlik is vir oorlewing en groei van vissies in 'n geslote sisteem kan verseker word deur voedingspersentasies tot en met 5 of 10% van liggaamsmassa. Indien* Spirulina *weerstandigheid toon by southeids vlakke soos bepaal in die kweek van tilapia in brak- of seewater, kan bogenoemde voedingspersentasies van droë* Spirulina *as 'n riglyn dien vir die gebruik van lewendige* Spirulina *tydens die kweek van tilapia in sulke water.*

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Thanks Lord Jesus.

..........in memory of my beloved father, Sana Asgodom, who passed away three years ago.

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Chapter 1: Literature review

1. Background

Tilapia is one of the most widely farmed fish in the world. The raising of Tilapia in earthen ponds is thought to have a long history that dates back to thousands of years BC (Knud-Hansen, 1998). Of the cichlids, commonly known as tilapia, *Oreochromis mossambicus* (Peters, 1852)[*] is one of commercially important species.

Distinct advantages (Popma and Masser, 1999) of tilapia culture include their tolerance to poor water quality and the fact that they feed on a wide range of natural food organisms, (Beveridge et al., 1991). In addition to being low feeding trophic level, they also exhibit characteristics such as fast growth, large size at reproduction and low production costs that make them attractive species for aquaculture (Costa-pierce and Rakocy, 1997). Popma and Masser, 1999, included the biological constraints in tilapia aquaculture as their inability to withstand sustained water temperatures below 10 to 12 ºC and early sexual maturity that results in spawning before fish reach market size.

The maximum total weight of fish which can be produced in a pond largely depends on the quantity of suitable food available (Hepher, 1988); source or supply of the later is technically used to classify aquaculture systems as extensive, semi-intensive and intensive. In semi-intensive systems, fertilization (a means of increasing nutrients available to the plants growing in them; FAO and UNEP, 1987) of production units plays a role in increasing the natural food availability (Brunson et al., 1999), which in turn increases fish survival and growth (Ludwig et al., 1998) and the yield (Diana et al., 1991).

Tilapia are considered to be filter feeders that harvest and ingest a wide variety of natural food organisms, including plankton, some aquatic macrophytes, planktonic and benthic invertebrates, larval fish, detritus, and decomposing organic matter (Hofer and Schiemer, 1983; Popma and Masser, 1999). Adults exhibit herbivorous habits (FAO and UNEP,

[*] Species content included in the site: http://nis.gsmfc.org/nis_factsheet.php?toc_id=195 (09 Feb. 2004)

1987) depending mainly on plant matter or detritus of plant origin such as blue-green and green algae, diatoms, macrophytes and amorphous detritus (Jauncey and Ross, 1982).

2. Feeding and Nutrition

A. Nature and Size of Food Ingested by Tilapia

Natural food organisms account for a considerable percentage of tilapia growth. The Nile or Blue tilapia, for example, digest 30 to 60 percent of the protein in filamentous and planktonic algae (Popma and Masser, 1999) dominated by the green and blue-green taxa. Many authors indicate that blue-green algae (BGA) are ingested and digested more efficiently than green algae (GA). McDonald (1985) found that *Anabaena flos-aquae*, filamentous BGA, is the easiest to be assimilated by Blue tilapia (*Sarotherodon aurea*) in comparison to three GA's, *Chlamydomonas sp* and *Ankistrodesmus falcatus*.

Turker et al. (2002; 2003) examined the rates of filtration in a Partitioned Aquaculture System, where dissolved and particulate organic matter are largely assimilated by BGA (i.e. *Microcystis* and *Merismopedia*) and GA (i.e. *Scenedesmus*, *Ankistrodesmus* and *Tetraedron*); which are then ingested and assimilated by filter-feeders. They found that Nile tilapia was successful at filtering larger particle size phytoplankton from both groups of algae (i.e. *Scenedesmus*; *Microcystis*), but more efficient at filtering cynobacteria at a higher filtration rate. Ingestion rates were even higher for *Oreochromis niloticus* when fed the larger *Anabaen cylinderica* (Northcott et al., 1991) and the *Oscillatoria sp* dominated periphytic community (Dempster et al., 1993) than the above-indicated larger cynobacteria- *Microcystis aeruginosa*. On the other hand, Turker and colleagues (2002) reported that Nile tilapia filtration rate (FR) of cynobacteria was lower, but showed a higher FR for GA, than for silver carp (*Hypophthalmichthys molitrix*) in their comparative study. Silver carp consumes primarily phytoplankton of particle size (8-100 μm) which is smaller than large quantities of zooplankton and detritus (17-3000 μm) ingested by bighead carp (Cremer and Smitherman, 1980). Fish biomass and the presence or absence of silver carp were seen to suppress the phytoplankton community (bloom of *Anabaena flos-aquae*) during seasonal experiments in summer and autumn, respectively, reported by Fukushima et al. (1999). Therefore, silver carp may have higher ingestion and/or filtration rates feeding on larger particle size cynobacteria than *O. niloticus*. Congo tilapia, Tilapia rendalli, compared to silver carp is only efficient to filter a wide range of phytoplankton as small as 5 µm (Popma, 1982) and as big as 15 µm diameter (Starling and Rocha, 1990). *O. niloticus* can sieve a range of particle sizes less than 50 µm; however, particles in this range are too small to be retained by branchial arches of *O. esculentus* (Goodrich et al., 2000). Most suspension-feeding fishes including (*O. esculentus* and) Ngege tilapia (news and views, 2001) consume much larger particles ranging from 40-1000 µm in size.

B. Suspension Feeding in Tilapia

Fish of the genus *Oreochromis* feed mainly on microscopic organisms. Adult *Oreochromis mossambicus* is primarily an omnivorous bottom feeder (Jauncey and Ross, 1982; Lovell, 1989; Bocek, 1996), showing a wide range of feeding habits consiting mainly of detritus and plant material (Hay, 1974; Global Invasive Species Database, 2003; De Silva et al., 1984). Unlike the Nile or Blue tilapia, it is less efficient at harvesting planktonic algae (Popma and Masser, 1999). But, compared to other phytoplanktivorous fishes, tilapia in general are efficient in filtering smaller size particles. They are known to remove 1 μm non particle-bound bacteria (Beveridge et al., 1989; 1991); and plankton as small as the solitary coccoid green algae, *Nannochloris*, which is less than 5 μm in diameter (Lovell, 1989).

The commercially important *Oreochromis* can utilize 30-60 % of the protein in algae (Popma and Lovshin, 1994), with blue green-algae like Microcystis sp (Colman and Edwards, 1987) being digested more efficiently than green algae (Turker et al., 2002; 2003). In ponds receiving fertilizers, natural foods greatly play a role in sustaining crops of tilapia, even in pond systems with inputs of protein enriched supplemental feeds. Schroeder (1983) found out those natural food organisms contributed 50 to 70 % of tilapia growth, in tilapia-carp polyculture ponds supplied with manures and feeds.

Of the taxonomic groups blue-green and green algae, particles with longer dimensions have recently been shown to be more suitable for ingestion by tilapia. This is greatly because the gill rackers (Jauncey and Ross, 1982) of tilapia are short and widely spaced.

Examples of Cyanobacteria and green algae species include *Microcystis, Anabaena*, *Oscillatoria, Spirulina*, *Chlorella, Ankistrodesmus* and *Scenedesmus*.

Although tilapia are often considered to be ‘filter feeders’, due to their ability to efficiently harvest plankton from the water, they do not physically filter the water through gill rakers as efficiently as true filter-feeders such as gizzard shad and silver carp (Popma and Masser, 1999). Dempster et al. (1993) suggest that the relative ‘filter feeding’ may not be an important method of ingesting algae by *O. niloticus*; and that this ingestion of algae may be achieved by other means (Dempster et al., 1995) akin to particulate feeding on aggregations of algae in the water column or flocculent surface scums of cynobacteria or by grazing on periphytic mats. *O. esculentus* does not use branchial arches to retain food particles by sieving or by mucus entrapment (Goodrich et al., 2000); specially when the particle sizes are less than 50 µm.

Suspension feeding fishes such as herring, mackerel, gizzard shad, goldfish and tilapia (UC DAVIS NEWS, 2001) have a similar physical mechanism of retaining food particles which is not yet fully understood; that is, these fish don’t swallow much water with their food and so, somehow, food and water are separated. Sanderson et al.(2001) report that the gizzard shad, ngege tilapia and goldfish use a ‘cross-flow filtration’, a method widely used in beverage manufacture to minimize clogging, concentrate and eject particles from the system in solution, to remove small zooplankton and phytoplankton from the water.

C. Supplementary feeding

The cost of Finfish aquaculture is rated according to the level of production per farm. It typically includes factors such as feed costs and the capital outlay of facilities and other fixed expenses related to economic interest and depreciation (Lipton and Harell, 1985). Feed represents the largest expenditure item in semi-intensive and intensive aquaculture systems. Protein is the most expensive macro-nutrient in fish feeds (Gitonga et al., 2003). Nutritionally balanced tilapia diets are generally expensive and comprise the largest production cost item in commercial production of stocks. However, when feeds with containing high-quality macro and micro-nutrient content are used to get increased fish yields, tilapia culture can generate large profits. Costs can be cut through application of fertilizers in pond cultures to increase natural food organisms available for fish as noted previously.

Relevance of supplementary feeding in semi-intensive tilapia ponds can be explained by the compensation fish receive for nutrient deficiencies found in natural food organisms in fertilized ponds. Besides, protein supplements are necessary to increase fish yield results for a culture (Li and Yakupitiyage, 2003). In the case of pond fertilization to increase natural food productivity; Fixed-input, PONDCLASS©, and Algal Bioassay fertilization approaches (Knud-Hanson et al., 2003) are all suggested strategies able to result in higher survival values of more than 75%. With the use of fertilizers and low cost feeding in ponds, periphyton substrate application in Bangladesh has supported fish production in shallow freshwater ponds (Azim et al., 2002). Should one consider reducing the use of artificial supplementation, there are several options to complementing the natural food availability in ponds that support culture fish. Some more techniques tried include bamboo substrate and supplemental feeding (Keshavanath et al., 2004) and biofilter media (Ridha and Cruz, 2001).

3. Aspects of Salinity Tolerance

Cichlids are secondary freshwater fish in their evolution. They have two advantages over primary freshwater species: they exhibit good tolerance to large amounts of dissolved minerals in water and they exploit wide variety of food organisms in their feeding habits because of the adaptive morphology of their pharyngeal apparatus (Wilkins, 2001). In the case of tolerance to salinity, many of them are euryhaline for their ability to tolerate certain ranges of brackish and/or salt waters. Temperature is another important environmental factor. Tilapia can only perform considerable growth enhancement in salt water under their temperature tolerance range. Likongwe et al.(1996) suggested that growth rates of juvenile *Oreochromis niloticus* may be high with higher temperature and lower salinity regimes.

Successful fish growth can be attained from marine pond cultures after receiving proper acclimation (Suresh and Lin, 1992). Researchers including Stickney (1986), Darryl (2000), Al-Amoudi (1987) and Villegas (1990) have indicated the surpassing tolerance of

O. mossambicus and its hybrids to various salinity strengths as well as potential production of cultures in salt water compared to other euryhaline tilapia species. However, most of those culture trials were conducted through giving fish the necessary acclimation until salinity regimes under demonstration had been reached. Literature on response of *O. mossambicus* that have been directly transferred to shock salinity is very limited. This is especially a case when trials have been considered as far as employing tilapia fry.

4. Prospectus on the Use of Spirulina

Spirulina has been used as a dietary supplement / food source by different populations around the world. In humans it has been sought after to play roles in health and well-being because of its well-balanced and highly nutritious content (Asia Pacific Bio-Tech, 2003). The growing interest in the use of Spirulina for its nutritional properties is clear. Spirulina Contains protein as high as 70%, varying between 50% and 70%; has a good nutritional profile in amino acids, vitamins, non-saturated fatty acids, and essential mineral content (Falquet, 1997).

Spirulina has the potential to contribute to the supply of nutrient requirements in fish.. Besides to the nutritional importance, *Spirulina* is being used as effective pigmentation source. Antimicrobial drugs have frequently been used in animal production for therapeutic purposes as well as non-therapeutic purposes primarily to promote growth of fish and land animals (ASM News, 2002).

It is thought that positive economics can be implied by the use of drugs through promoting growth or improving productive efficiency and avoiding or reducing mortality and morbidity of organisms. In aquaculture for example, improved growth and/or feed efficiency can reduce associated costs of feed and time as well as provide quick returns. However, antimicrobial resistance is an important issue on the other hand, as the growth and proliferation of undesirable micro-organisms may soon outpace the ability to control and mitigate their effect on health and environment. Resistant micro-organisms emerge and proliferate through selective pressure in the environment, exerted by antimicrobials of which prime purpose consideration is prophylaxis or growth promotion (Witte, 2000).

Spirulina contains natural ingredients, carotenoid and vitamins for example; those that enhance reproduction, immune function and an increase in growth (Lorenz, 1999). It is therefore a new option to develop the fish feed production and nutrition industry towards the use of natural ingredients contained in *Spirulina* that potentially eliminate use of antibiotics, synthetic pigments, and other chemicals.

Fish larvae die in their early stages due to infections of opportunistic pathogens. Problems of reduced survival and quality of larvae may be curbed through complementing nutrition with the use of *Spirulina* to help improve the immune function of newly hatched fish. The natural astaxanthin is an essential vitamin present in *Spirulina* shown to improve larval quality and survival of shrimp (Lorenz, 1998).

5.Conclusion

Oreochromis mossambicus has been regarded as one of the commercially important culture species. It has become clear that attention on culture aspects of this species is growing due to its tolerance of a wide range of environmental conditions. The cichlids are generally warm water fish (*the* PennState Agricultural Sciences, 1995); therefore, generally constrained with their level and consistency of survival in temperatures below their tolerance range. Surpassing tolerance of *Oreochromis mossambicus* to salinity as well as its low trophic feeding habit viz. the advantage of exploiting natural food organisms triggers the need to investigate culture that is concurred with marine *Spirulina*. *Spirulina* can be fed to fish to improve biological performance of fries as well as help mitigate economic problems associated with production costs involved in the use of expensive artificial feed. However, to best benefit from *Spirulina's* nutritional properties, there is a need to compliment meals that appropriately fix associated nutrient deficiencies.

Chapter 2: Laboratory Experiment

An Investigation of the Survival Level of *Oreochromis mossambicus* fry variably kept in a Closed System.

Chapter Summary

Three independent trials were performed to demonstrate effects of salinity and ration sizes with the use of dried Spirulina *on survival of* Oreochromis mossambicus *fry. The experiments were done in a closed laboratory system at the Welgevallen Experimental Farm of the Division of Aquaculture of the University Stellenbosch. In Phase 1, freshwater-hatched fries were transferred to eight salinity concentartions - 0, 5, 10, 15, 20, 25, 30 and 35 parts per thousand (ppt) - to evaluate the tolerance level of starved fries after transfer from freshwater. The following two trials were done to demonstrate effects of types and ration sizes of preserved food as well as complement rate supplementation of feed and* Spirulina *on performance of fries in the closed system. Fries received three types of dried* Spirulina platensis *at 0, 2.5, 5, 10, 20, 40, 80, 160, 320, 640 and 1280 percent of bodyweight in the second experiment. In the last trial the commercial pre-starter (AquaNutro, Malmesbury) to complement* Spirulina *in five different ways at 0, 5 and 20 % of body weight. Input percent of feed complement were in the ratio 0:100, 25:75, 50:50, 75:25 and 100:0 supplement to* Spirulina*. Salinity tolerance level of starved fries and ration size results of this paper had been expected to guide the use of live* Spirulina *natural food source for* Oreochromis mossambicus *fry.*

Introduction

Aquaculture is receiving growing attention for wild fisheries are declining while, at the same time, an increasing demand for fish and seafood keeps on creating void in the market (Michael, 2001). In the production of fish high levels of consistency in survival rate and growth need to be maintained to get the desired increase in fish yield. Improvements in those two important factors of production can be made through giving important attention towards optimizing critical environmental factors such as temperature, salinity and nutrient concentration (Boyd, 1990).

Unlike agriculture and land animal production, wild genetic resources for aquaculture are scarce. Their availability and quality depend on biological factors such as those related to season. The shrimp industry for example is facing constraints with uncertainties, inefficiencies, and economic loss continuously contributed by lack of reliable supply of disease-resistant post-larvae (Treece, 2000). This implies the need to solve problems in aquaculture production that are associated with prolonged seasonal availability of fingerlings for stock. For hatcheries to ensure enough production of quality larvae as well as subsequent production performance in grow out ponds, factors affecting growth and survival need to be controlled within limits suitable for the specific organism. The following website contains a manual on the presence of species specific tolerance range for environmental fluctuations specifically regarding salinity tolerance and survival potential of both stenohaline (narrow range of salt tolerance) and euryhaline (wide range of salt tolerance) species in ponds directly connected to the ocean as variation is very low, if other factors can be controlled https://www.denix.osd.mil/denix/Public/Library/Watershed/wqmsec4.html (visited on July 21, 2003).

Nowadays, the literature is swelling with results of determination of survival and growth relating environmental factors and nutritional status or maintaining availability of nutrients mainly through fertilizer application. Such findings include for Yi and Lin (2002); and Abdelghany and Ahmad (2002). It is also well documented that variations in environmental factors like temperature and salinity affect survival and production performance through interfering with physiological well-being of organisms (Imsland et al., 2002; Morgan et al., 1997; Nakano et al., 1998). Juvenile fish such as black sea bream that are acclimated to hyposmotic environments with reduced ration sizes may face a degree of osmoregulatory problems as a result of reductions in chloride cell morphometrics (Kelly et al., 1999).

Aquaculture nutrition has the aim of enhancing yield and economic returns of crops at reduced cost of production. Nevertheless, cost of feeding fish still holds the biggest percent of production cost and feared to be more limiting than fry cost in the near future, for example, in the production of Malaysian prawn (Ung, 1988). In that case commercial feeding becomes an important aspect of artificial breeding of fish and shell-fish requiring important considerations[†] of nutrient ingredients and expenses involved in commercial diet production. Tacon and De Silva (1997) emphasized increasing availability of natural food organisms in semi-intensive fish farming systems to optimise nutrient dynamics of ponds and reducing feeding costs through use of improved fertilizer application, feed formulation and preparation as well as feed and water management techniques.

In the current paper tolerance of newly hatched *Oreochromis mossambicus* fry to salinity and effects of nutrition manipulation on survival and growth in a closed laboratory system are covered. Egg hatchability of Nile tilapia as well as growth of hatched fries can be increased with increasing levels of dietary crude protein fed to brood fish reared at 7 and 14 parts per thousand (ppt) water salinities (El-Sayed *et al*., 2003).

Three independent experiments were conducted to give ways for use of live marine *Spirulina* for tilapia fry. One of these involved survival determination of freshwater hatched *Oreochromis mossambicus* kept starved in ranges of salinity strengths prepared in a closed laboratory system (figure1) in containers. The other two were determination of fry survival as water quality and nutrient level is impacted with applications of *Spirulina* and supplemented *Spirulina* respectively. These were tried in freshwater but in the same laboratory system arrangement. Microsoft Excel ® 2002 and MINITAB 13™ (MINITAB.Inc, 2000) were used to sketch values on graphs and for analysis of variation (ANOVA) respectively. Survival and growth data as well as ANOVA tables are contained in Appendix at the end of this paper.

[†] http://www.tcru.ttu.edu/tcru//kc/pubs/parker/p11eval/11eval.htm

Results showed good survival of starved fry to salinity regimes up to 15 g/lt after direct transfer from freshwater. Fry can only survive with low input levels of *Spirulina* or supplemented *Spirulina*. Their survival performance was highly affected with increased percents of input.

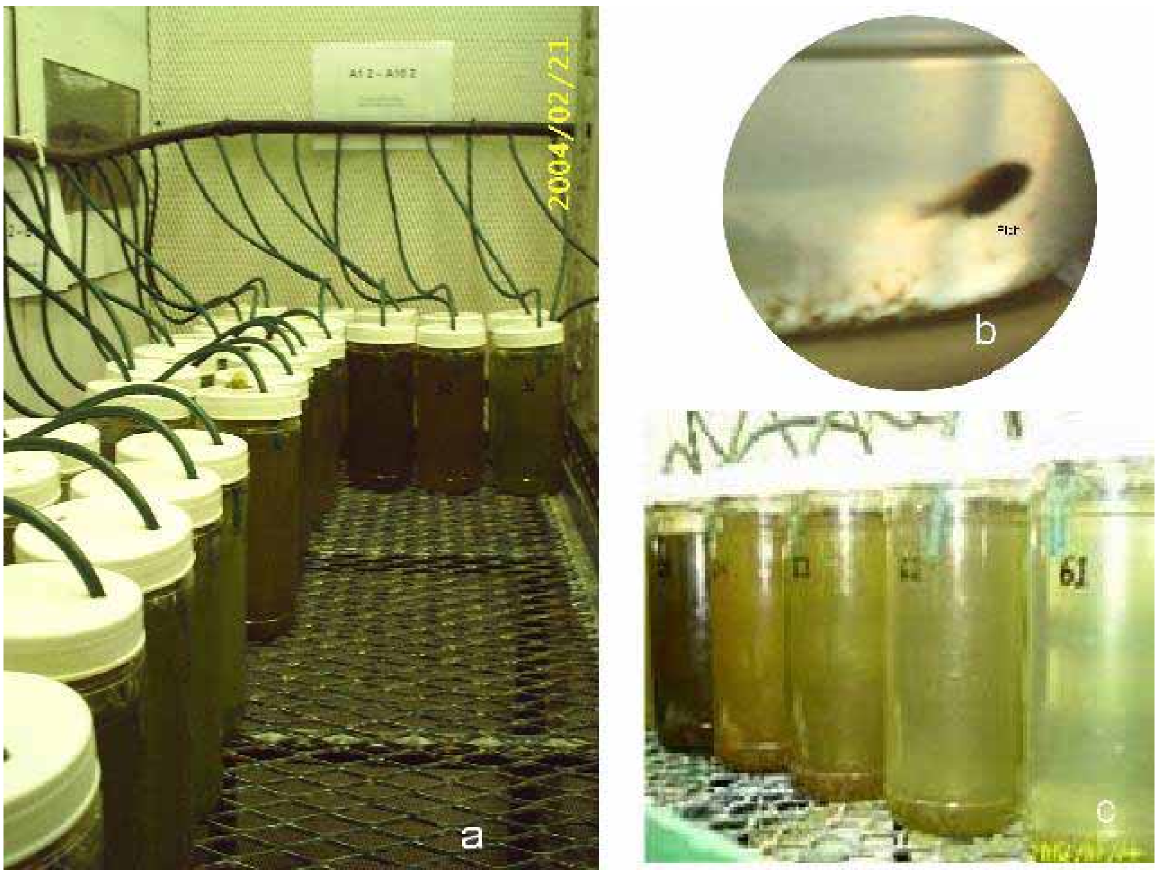


Figure 1. Picture of the system. a) Arrangement of fish containers and oxygen supply in the closed laboratory. b) An enlarged view of tilapia fry treated in this experiment. c) Differing degrees of turbidity and coloration due to varying levels of input.

Chapter 2.1. Tolerance of Tilapia Fry to shock salinity

In aquaculture production the *Oreochromis mossambicus* is preferable for saltwater culture as compared to other commercially important tilapia because it can tolerate wider range of salinity extended near full strength seawater. However, it could be important to determine the maximum tolerable shock salinities to begin with and ensure production of acclimated cultures with enhanced level and precision of survival performance. Previous research done by Fitwi (2003) fully demonstrated mature *O. mossambicus* to have tolerated shock salinities from 0-25 g/lt (measurement identical to parts per thousand-‰). Other *Oreochromis* varieties have less affinity to shock salinity; and, some acclimated cultures of these types have been found to have exhibited good survival merely up to 20 ‰ salinity (Robert, 2003).

A nine-day laboratory trial was conducted at the Welgevallen Experimental Farm, Stellenbosch, to investigate the survival of freshwater-hatched tilapia fry along a gradient of salinity from 0 up to 35 grams per litre (g/lt) at interval of 5, and each replicated to 12. The trial was undertaken in temperature insulated room with containers, where 0.06g sized fry were introduced into each. Significance of the result had been expected to direct and enable successive trials that are aimed at evaluating survival and production performance of tilapia in a reasonable salinity range; through concurred use of varying density and type of *Spirulina* for its nutritional value.

Materials and Methods

Newly freshwater hatched *Oreochromis mossambicus* fry (0.06g weight) were used in this laboratory study. The trial was performed to determine the effects of shock salinity on survival of fish fry. The strengths of salinity included were 0, 5, 10, 15, 20, 25, 30 and 35g/lt (‰). Tilapia fry were not fed during the experimental time. This trial involved twelve replicates for each salinity treatment (0 g/lt was the control measure).

The experiment was conducted in laboratory at the Welgevallen Experimental Farm, Stellenbosch. To keep temperature constant at around 29 ^{0}C fish containers were placed in insulated room at all times. Water temperature required for optimum growth of tilapia need not be less than 29 ^{0}C but may reach up to 31 ^{0}C (Popma and Lovshin, 1994).

Fry mortality count was made daily. Cumulated number of dead fries for a day was divided to the initial amount in each container, multiplied by 100, to get percentage mortality for the inclusive time elapsed. The analysis was subjected to one-way Analysis of Variance (ANOVA).

Results and Discussion

The results revealed that salinity has affected survival of *O. mossambicus* fry (figure 2). They turned out rare to moderate mortalities up to salinity regime of 15 g/lt (‰) over the trial time. For included regimes above 20 ‰ (i.e. 25, 30 and 35 ‰) the mortalities were 100% starting from the first day of the experimental time. The later cases, but those with lower strengths specifically as being seen in the earlier stages, were significantly different from the control treatment (0 ‰).

Fry mortality increased significantly ($P<0.05$) for the most part with increasing level of salinity viz. 100% mortality at 25 ppt starting from Day 1, at 20 ppt after Day 5, at 15 ppt after Day 7 and at 10 ppt after Day 8. From Day 1 until Day 5 mortalities were found to be 83-92%, 8-54% and 0-20% at 20, 15 and 10 ppt salinities respectively. Mortalities at lower salinities as well as the up going trend after some days were to be expected for fish were kept starved during the experimental period. The trend salinity has affected fry survival and significance of the results for the period is depicted in the ANOVA table (1), on daily bases. It is important to note down again here that: because fish were not being offered any food, mortalities after Day 6 as influenced by low-ranged salinity strengths thus were merely aggravated.

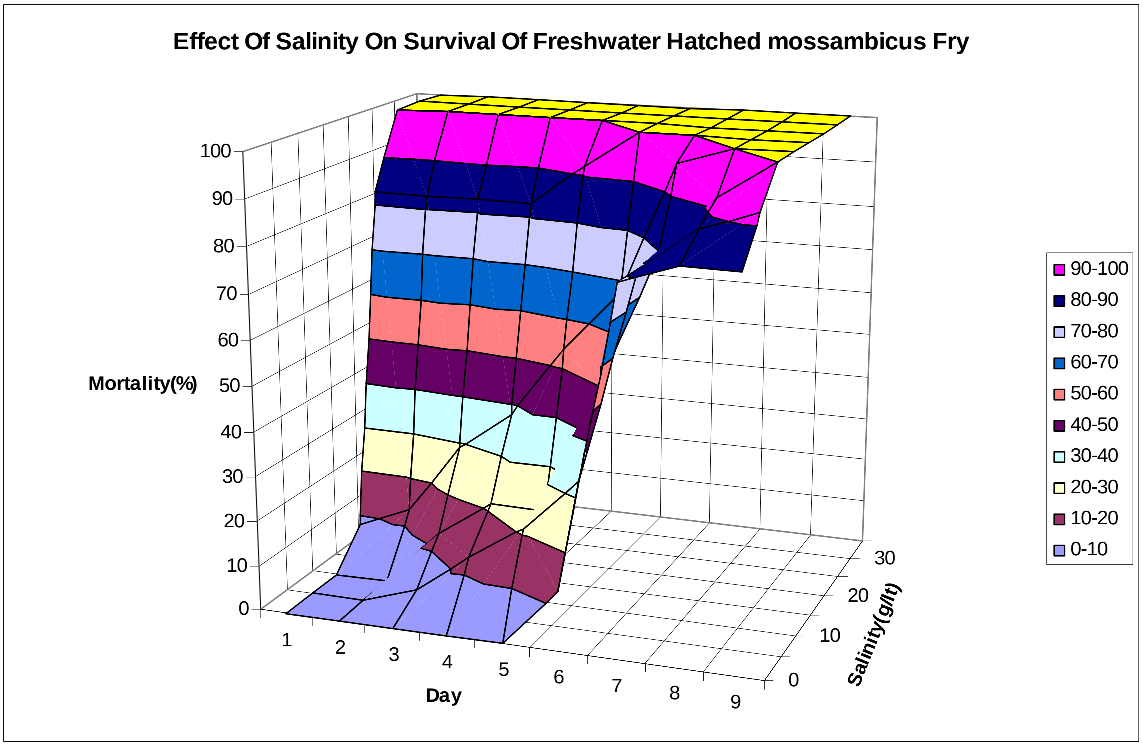


Figure 2 Effect of salinity on freshwater hatched *Oreochromis mossambicus* fry

Table 1 One-way ANOVA table: a summary of Tukey's Pair wise Comparison.

	Salinity (g/lt)							
Days	0	5	10	15	20	25	30	35
Day 1	0.00±0.00a	0.00±0.00a	0.00±0.00a	8.33±19.46a	83.33±32.57b	100±0.00b	100±0.00b	100±0.00b
Day 2	0.00±0.00a	0.00±0.00a	0.00±0.00a	12.50±22.61a	83.33±32.57b	100±0.00b	100±0.00b	100±0.00b
Day 3	0.00±0.00a	4.17±14.43ac	12.50±22.61ac	29.17±39.65c	83.33±32.57b	100±0.00b	100±0.00b	100±0.00b
Day 4	0.00±0.00a	12.50±22.61ac	20.83±25.75ac	37.50±37.69c	83.33±32.57b	100±0.00b	100±0.00b	100±0.00b
Day 5	0.00±0.00a	20.83±25.75a	20.83±25.75a	54.17±45.02c	91.67±19.46b	100±0.00b	100±0.00b	100±0.00b
Day 6	12.50±22.61a	33.33±24.61ad	50.00±30.15de	66.67±44.38ce	100±0.00b	100±0.00b	100±0.00b	100±0.00b
Day 7	79.17±33.43a	79.17±33.43a	79.17±25.75a	95.83±14.43a	100±0.00a	100±0.00a	100±0.00a	100±0.00a
Day 8	83.33±24.62a	87.50±31.08a	91.67±19.46a	100±0.00a	100±0.00a	100±0.00a	100±0.00a	100±0.00a
Day 9	83.33±24.62a	91.67±28.87a	100±0.00a	100±0.00a	100±0.00a	100±0.00a	100±0.00a	100±0.00a

Mean values with any dissimilar superscripts in each row, differ significantly ($p<0.05$)

The results show *O. mossambicus* fry can tolerate shock salinities almost up to a level of 15 g/lt(‰). However, optimum survival can only be attained at salinities below 15 ‰. Significant mortality resulted after Day 6 in the control salinity regime-0 ‰ (i.e. freshwater). This can be explained due to the exhaustion of fish for they were not fed. Food deprivation also exacerbates mortalities of tilapia in salinity regimes up to 15 ‰ through affecting fish adaptability to other characteristics of the medium or in the acclimation process (Mathilakath *et al*., 1996) of tilapia in the salt water.

The results in this trial are expected to guide successive experiments that will employ live *Spirulina sp*. as live food for tilapia fry. Most marine micro algae are extremely tolerant to salinity changes and grow optimally at salinities from 20-24 ‰(g/lt)- that is to some extent lower than their native habitat (FAO Fisheries Technical Paper, 1996). Although *Spirulina sp.* is believed to be tolerant to a wide range of salinity, it is important to investigate its strength with the salinity regimes included in this trial to best fit findings that allow optimal growth and benefit from them in using live *Spirulina sp.* as food source for tilapia.

Chapter 2.2. Effects of Inputs of Three Forms of Spirulina Platensis

Production systems can be fertilized to increase availability of nutritious natural food organisms like *Spirulina* to reduce use of artificial feed. *Spirulina* is thought to serve fish as source of important nutrients, best desired to replace some expensive artificial feed complements for cheaper protein supplemental as well as indispensable pigment provision. Its use as vitamin resources supply becomes clear in the case of nursing young-age fish business to attain quality fingerlings for stock that are highly resistant to disease with improved production performance. However, knowledge in the physical property of dry *Spirulina* and preferred input rate for optimal performance by tilapia fry is limited. This may hinder or lessen its use by farmers who aim to get the cheapest source of nutrient for their fish (references?).

In a case where concurring culture of live *Spirulina* in fish nursery systems becomes unaffordable, use of dried *Spirulina* is another option. Different input rates of three dried forms of *Spirulina platensis* types were used in this experiment to investigate their impacts on newly hatched *Oreochromis mossambicus* fry, as described thoroughly in the Materials and Methods section. Consistently high fry mortality was observed in the closed system, without variation of effects for each type. It is discussed that enhanced level of fry survival can only be attained by applying low rates of input that ensure maintained water quality.

Materials & Methods

Freshwater hatched fry with a sample average weight of 0.01g were used in this experiment. This phase employed use of three types of dry *Spirulina platensis* to demonstrate growth and survival performance of *Oreochromis mossambicus* fry in fresh water. The three types of preserved algae tried were labeled as A, B and C to represent *Spirulina* powder, granulated *Spirulina* and *Spirulina* flake respectively (figure 3). A, B and C are dry *Spirulina platensis* that had been achieved with different drying processes.. 90 containers were used where five to seven fish fry randomly stocked to each.

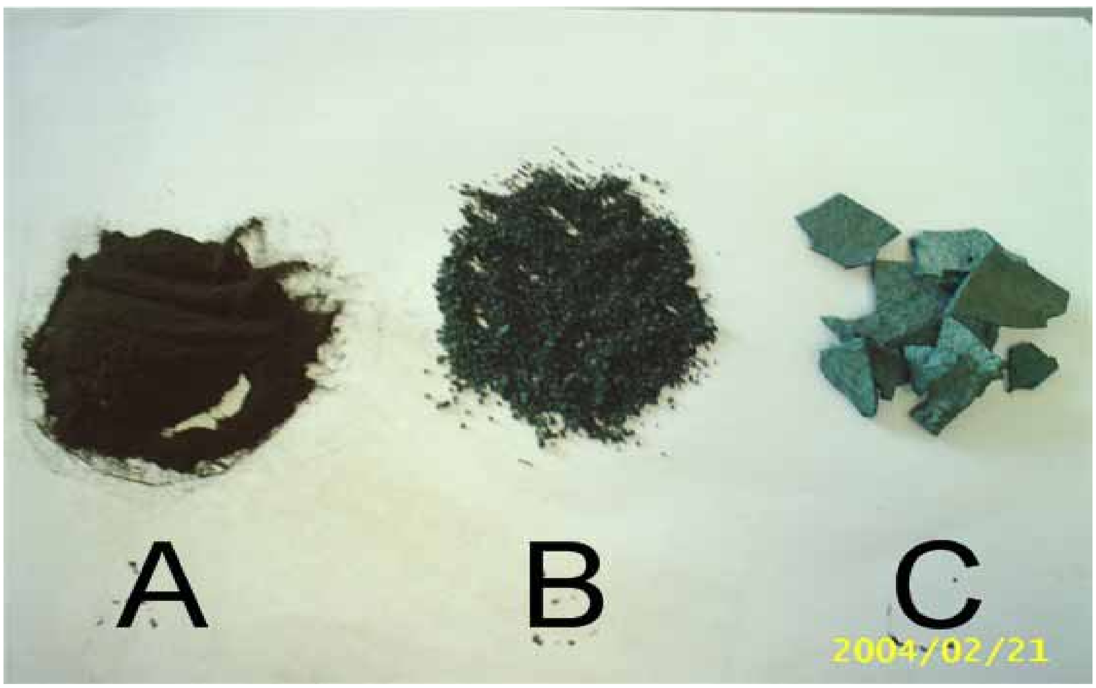


Figure 3. Picture of the three preserved *Spirulina platensis*.

The experimental design had 9 replicates for algae input level and three for each type of the dry *Spirulina platensis*. Input levels were 2.5%, 5%, 10%, 20%, 40%, 80%, 160%, 320%, 640% and 1280% of bodyweight per day with a control regime of 0%. Fish received each of the three dry Spirulina for each regime on a basis of three times a day feeding practice. However, because of limited technique of weighing very low amount of algae; calculated amounts of the input regimes were given as weekly ration at the beginning of the experiment and then likely for the rest of the experimental time.

In this (temperature insulated, no water discharge) closed laboratory system; environmental factors that affect fry survival and growth, were kept constant throughout the time. Temperature control was at the same level like in the above trial to allow optimum growth and survival. Fish containers were not replaced with new water in need of determining the degree to which fry can resist reduced quality of water that might have been exacerbated by amount of *Spirulina* input. However, oxygen was supplied (as illustrated in figure 1-a) in an attempt to reduce the consequences.

Fish growth (in grams) data collection was made every second day after the first day of feeding fry in the laboratory system. Average growth rate was calculated each day growth was recorded. Likewise dead fish were counted every second day, after the first day of feeding fry in the system, to get cumulated mortality as percent of initial amount of fish in each container. Finally growth and mortality data received two-way and one-way analysis of variance (ANOVA) to investigate significant effects of type and input treatments.

Results and discussion

Two-way analysis results showed no significant effects of *Spirulina* types on fish growth (with $P>0.05$). Besides to the type treatment variable, the input amount variable resulted in significant ($P<0.05$) effects on growth only on days 3,4,5,6 and 8. These effects are made clear in the univariate analysis where growth on days 3, 4,5,6,7, and 8 were significantly different ($P<0.05$). These results didn't imply growth was enhanced with certain input levels included. However, maximum growth achieved were recorded with type C viz. 66.7% growth of fish (0.01 to 0.03g) at 10%, 75%(to 0.04g) at 20 and 80%, 80%(to 0.05g) at 40% input rates respectively. With type A and B fish grew up by 75% at 640% and 80% at 320% input rates.

Growth values due to the various input treatments fit polynomial lines (figure 4) except level 10% input that best fit linear function. The relationships amongst the inputs and growth for all the three types were

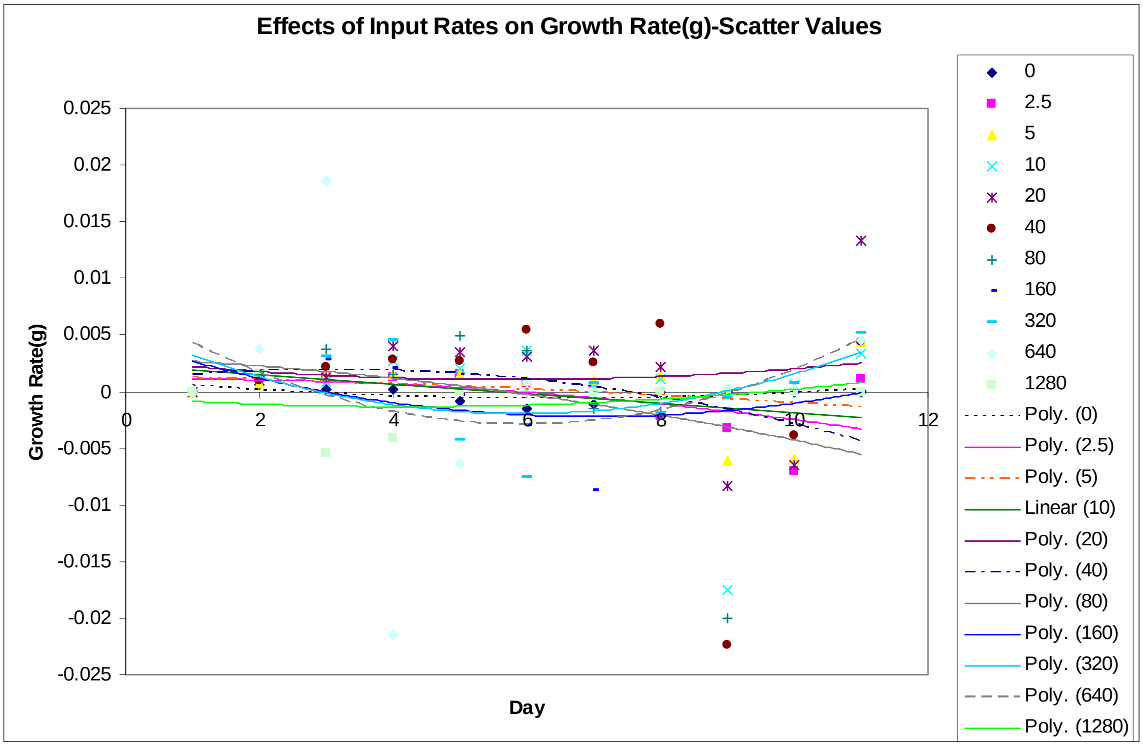
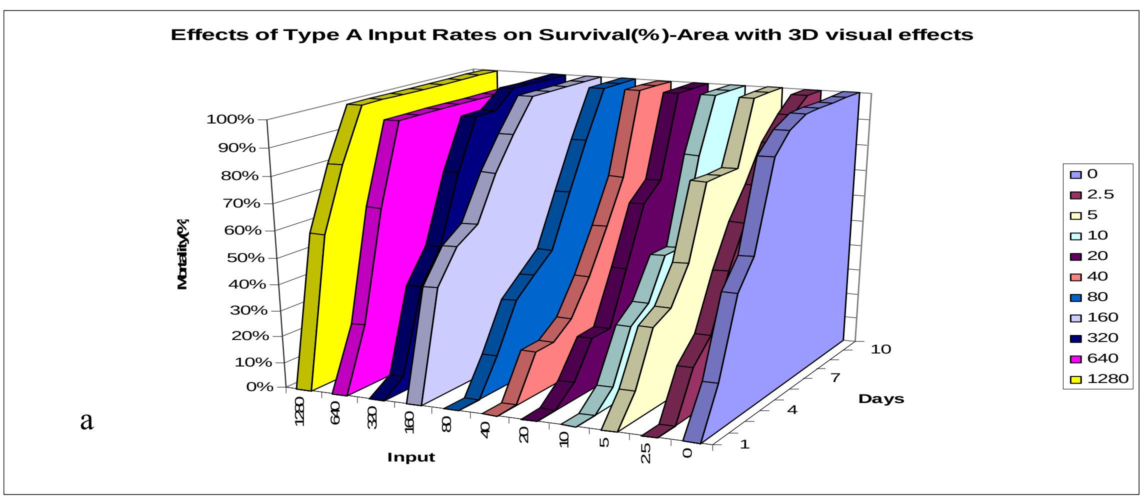


Figure 4. Regression lines drawn to show relationships of growth values with each input level used. Equations below correspond for input levels on the legend in left-right with top-bottom order. (Growth rate data in Appendix B1.1)

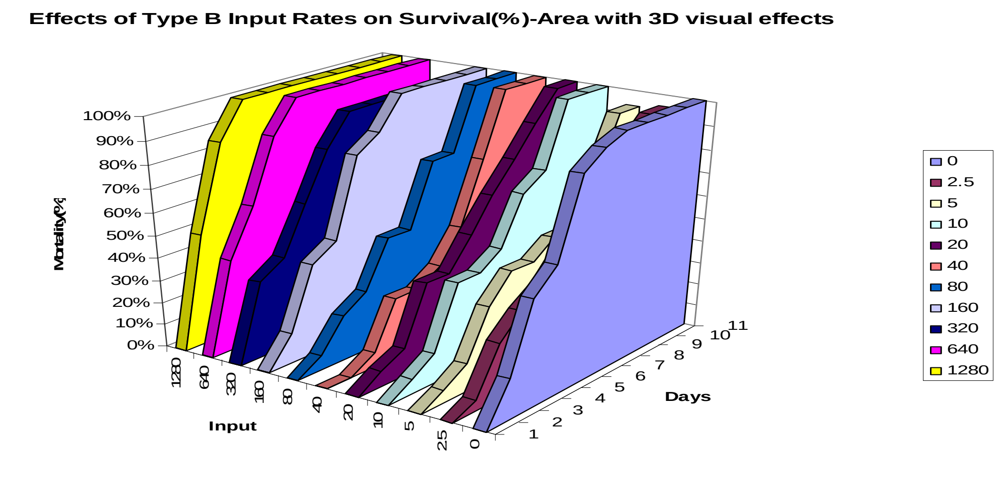
y = 4E-05x2 - 0.0005x + 0.001, r^2 = *0.371*; y = -4E-05x2 + 6E-05x + 0.0011, r^2 = *0.3196*;
y = -1E-05x2 - 0.0001x + 0.0013, r^2 = *0.0717*; y = -0.0004x + 0.0023, r^2 = *0.0553*;
y = 5E-05x2 - 0.0006x + 0.0027, r^2 = *0.0075*; y = -0.0001x2 + 0.0006x + 0.001, r^2 = *0.0742*;
y = -5E-05x2 - 0.0002x + 0.0029, r^2 = *0.1659*; y = 0.0001x2 - 0.0019x + 0.0044, r^2 = *0.2457*;
y = 0.0002x2 - 0.0025x + 0.0055, r^2 = *0.2747*; y = 0.0003x2 - 0.0035x + 0.0075, r^2 = *0.0836*;
y = 5E-05x2 - 0.0004x - 0.0005, r^2 = *0.1125*

weak (with r^2 close to zero) and negative correlation coefficients for most of the equations. In those containers with high inputs, irregularity of trends can be explained by observation on water quality. Poor quality effects might have negated regular growth of fish. The tendencies of mortality also have affected the proportionate amounts of fish in the design after time (effects will be followed down); which together with the limited sensitivity of growth record technique could have added to the irregularity of fry growth values. Fries were not seen to grow otherwise when they didn't receive inputs of either A, B or C type *Spirulina*.

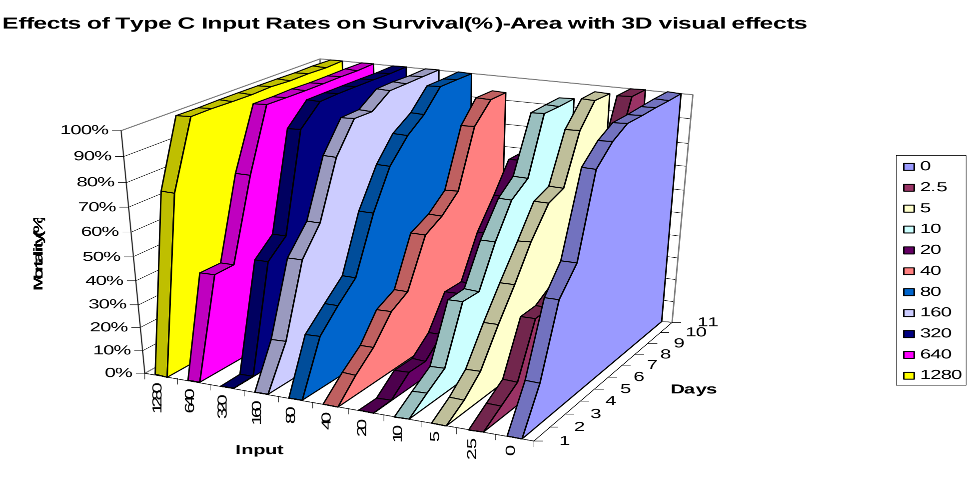
Two-way ANOVA showed significant ($p<0.05$) effects of input levels on mortality throughout the time except for days 9 and 10, and no effects of type on mortality except for day 8 (where $p<0.05$). Percent of fry mortality results are sketched in graphs a, b, and c (figure 5) for type A, B and C input amounts, respectively.



a



b



c

Figure 5-a, b, c. Effects of input rates on fry survival (%). The control level (0% input) and levels above 80% accounted for large mortality values than the rest. (Mortality Data in Appendix B2.1)

One-way analysis of the effect of input amount indicates significant differences on mortality for days 1 to 8. Summary of one-way input ANOVA and Tukey's Pairwise Comparison of mortality mean values are presented in table 2. Input levels 2.5-80% resulted in moderate mortalities and were significantly different from the control regime (no input) throughout days 2-7. Levels above 80% greatly affected fish survival and were not significantly different from the control for most of the experimental time though they reviled high mortality results at some time. Very high input of *Spirulina* could have reduced quality of water; as the available amount of oxygen falls down the capacity of water to support fish will also decline. Fish could also die due to rise up of bottom sedimentation observed, which might have given rise to unsuitable water chemistry but suitable for growth of harmful microorganisms.

Table 2. One Way ANOVA for Fry Mortality: a Summary of Tukey's Pair wise Comparison. Mean values with any dissimilar superscripts in each row, differ significantly ($p<0.05$)

	Spirulina Input Level(As Percent of Fish Body Weight)										
Day	0%	2.5%	5%	10%	20%	40%	80%	160%	320%	640%	1280%
1	17.67± 14.40^{ab}	3.44± 6.88^{a}	7.44± 12.08^{a}	4.11± 8.19^{a}	2.22± 6.67^{a}	2.78± 8.33^{a}	9.56± 14.82^{ab}	24.00± 19.67^{ab}	13.00± 22.27^{ab}	35.22± 25.86^{bc}	59.44± 25.79^{c}
2	46.67± 7.21^{a}	17.56± 13.21^{b}	18.56± 13.08^{b}	10.44± 9.98^{b}	8.56± 10.19^{b}	12.78± 12.91^{b}	21.22± 13.92^{bc}	47.00± 13.73^{a}	41.56± 16.82^{ac}	55.56± 17.41^{a}	88.33± 20.62^{d}
3	56.67± 11.27^{a}	30.78± 9.85^{b}	31.56± 10.32^{bc}	33.44± 10.22^{bcd}	21.11± 19.00^{b}	23.11± 12.32^{b}	31.89± 12.18^{be}	54.89± 14.42^{ad}	53.56± 19.37^{ace}	85.67± 24.42^{f}	100.00± 0.00^{f}
4	89.33± 9.54^{ae}	39.89± 9.13^{bcd}	43.78± 15.67^{bd}	35.33± 11.61^{bcd}	23.33± 20.00^{c}	26.22± 14.53^{bc}	48.56± 12.98^{d}	79.22± 14.92^{a}	82.00± 15.80^{ae}	98.44± 4.67^{ae}	100.00± 0.00^{e}
5	96.33± 5.50^{a}	50.22± 16.63^{b}	57.44± 18.93^{b}	48.22± 15.21^{b}	38.89± 21.47^{b}	39.56± 14.98^{b}	56.00± 19.04^{b}	89.56± 9.98^{a}	95.56± 8.82^{a}	98.44± 4.67^{a}	100.00± 0.00^{a}
6	100.00± 0.00^{a}	62.11± 17.22^{bc}	63.78± 19.43^{bc}	59.33± 11.69^{bc}	50.78± 20.57^{b}	51.11± 18.19^{b}	74.56± 18.63^{cd}	98.11± 5.67^{a}	95.56± 8.82^{ad}	98.44± 4.67^{a}	100.00± 0.00^{a}
7	100.00± 0.00^{a}	68.78± 21.56^{bc}	64.67± 20.17^{bc}	74.22± 9.43^{bc}	63.00± 18.93^{bc}	67.67± 14.74^{bc}	82.44± 14.75^{ac}	100.00± 0.00^{a}	97.78± 6.67^{a}	98.44± 4.67^{a}	100.00± 0.00^{a}
8	100.00± 0.00^{a}	85.89± 15.93^{ab}	86.22± 16.47^{ab}	97.78± 6.67^{a}	81.44± 21.48^{b}	95.00± 10.00^{ab}	100.00± 0.00^{a}	100.00± 0.00^{a}	97.78± 6.67^{a}	98.44± 4.67^{a}	100.00± 0.00^{a}
9	100.00± 0.00^{a}	98.44± 4.67^{a}	96.89± 6.86^{a}	97.78± 6.67^{a}	89.67± 20.02^{a}	97.78± 6.67^{a}	100.00± 0.00^{a}	100.00± 0.00^{a}	97.78± 6.67^{a}	98.44± 4.67^{a}	100.00± 0.00^{a}
10	100.00± 0.00^{a}	98.44± 4.67^{a}	96.89± 6.86^{a}	97.78± 6.67^{a}	89.67± 20.02^{a}	97.78± 6.67^{a}	100.00± 0.00^{a}	100.00± 0.00^{a}	97.78± 6.67^{a}	98.44± 4.67^{a}	100.00± 0.00^{a}

Regression lines are drawn to illustrate the relationship of mortality values for each input regime tried (figure 6). The logarismic relationships amongst mortality levels and input rates for all types used in each container were strong (r^2 much close to 1) indicating majority effects on fry survival could be described by the level of input. Mortality positively and logarismically increased with time. Generally fry mortality was high with 0% input and levels above 80% of body weight.

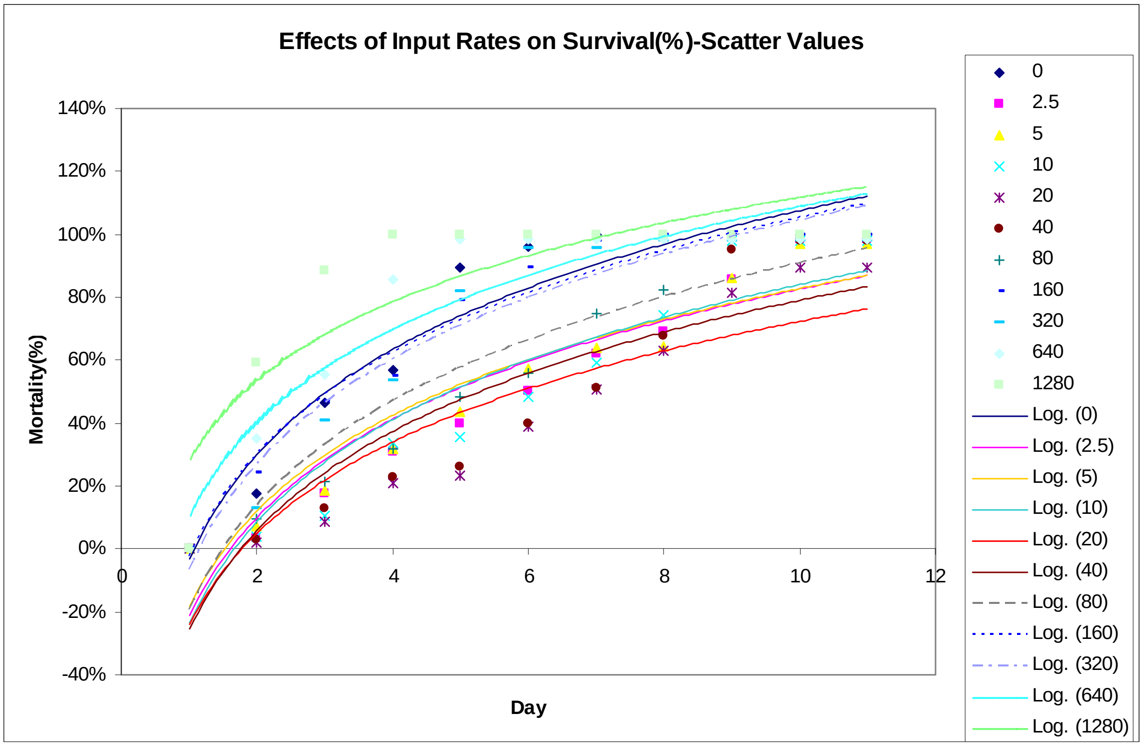


Figure 6. Effects of Spirulina input levels on fry survival(%) in closed system. Regression lines are drawn to show relationships of scattered mortality values. Equations below correspond for 0-1280% levels of input in left-right with top-bottom order. (Mortality Data in Appendix B 2.2)

$y = 0.4812\mathrm{Ln}(x) - 0.0323$, $r^2 = 0.9322$; $y = 0.4502\mathrm{Ln}(x) - 0.2109$, $r^2 = 0.8884$;
$y = 0.4387\mathrm{Ln}(x) - 0.1826$, $r^2 = 0.9042$; $y = 0.4677\mathrm{Ln}(x) - 0.2366$, $r^2 = 0.8563$;
$y = 0.4177\mathrm{Ln}(x) - 0.2388$, $r^2 = 0.8163$; $y = 0.4541\mathrm{Ln}(x) - 0.2554$, $r^2 = 0.8008$;
$y = 0.4784\mathrm{Ln}(x) - 0.1936$, $r^2 = 0.9094$; $y = 0.4674\mathrm{Ln}(x) - 0.0229$, $r^2 = 0.9641$;
$y = 0.4817\mathrm{Ln}(x) - 0.0653$, $r^2 = 0.9317$; $y = 0.4271\mathrm{Ln}(x) + 0.1071$, $r^2 = 0.8888$;
$y = 0.3625\mathrm{Ln}(x) + 0.2848$, $r^2 = 0.7543$

These results indicate the need to provide fries with low rates of *Spirulina* somewhere between 2.5 and 40% of their weight. Any of the A-B-C dry *Spirulina* fed to newly-hatched fries at very low rate of input can improve survival level and consistency. Input rates much higher than 40% of body weight in the closed system were observed to further reduce water quality and rather increase mortality of fries.

Chapter 2.3. Effects of Inputs of Supplemented Spirulina platensis

For larvae of *Oreochromis mossambicus* exhibit carnivorous feeding habits depending mainly on zooplankton as well as problems associated with nutrient deficiencies of a single food ingredient, it is important considering complementary supplementation on the use of natural *Spirulina* algae. Protein supplementation improves survival values and increases fry yield. Here five ways of feed-*Spirulina* complement percentages were tried at low levels (0, 5 and 20) of input calculated as percent of body weight. Supplement percentages didn't result in significant growth differences. The closed system manipulation of ration has resulted in reduced survival performance. In such a system input rate of less than or equal to 5% body weight and feed-*Spirulina* complement ratio of less than 50:50 percent would be preferable.

Materials and Methods

This trial was made to demonstrate the use of *Spirulina platensis* for fish fry that is supplemented with powdered fish feed. It was tried in the same laboratory where the above trials were undertaken and factors prepared like in trial two. Fish with a sample average weight of 0.01g were fed at 5 and 20 % of body weight. Here the control treatment was 'no-input' of Spirulina and feed (that is fish were fed at 0% of body weight). For each treatment fish feed was supplemented with Spirulina in five different proportions viz. 0:100, 25:75, 50:50, 75:25 and 100:0 feed to Spirulina (F:Sp) percent ratios, respectively. Each proportion treatment was then made by five replicates where a single container was stocked with five fish. However, the same as the case in trial two, weekly rations were cumulative amounts of inputs fed to fish at the beginning of the trial. Fish growth (in grams) data was taken second day after the first day of feeding fry in the laboratory system. Average growth rate was calculated each day growth was recorded. Each container representing a replicate of each treatment was looked over every day for dead tilapia. Daily fry mortality record was expressed in percent increase by dividing the number of dead fish that were being totaled until the count day to the initial input amount. Analyses of data collected on mortality and growth were performed to see significant effects of varying input level and feed to Spirulina proportion variables.

Results and discussion

The analyses of variance indicated no clear effect of ration sizes or percentage supplement treatments on fish growth ($P>0.05$) during the time-dimension of the experiment. And, only Day1 ANOVA had indicated significant effects of the input rate treatment variable on fry growth. Nevertheless, *Oreochromis mossambicus* fries of the size treated in this experiment showed maximum growth of 61.5% (fed 25:75 feed-*Spirulina* complement) and 64.1% (with 0:100) at 20 percent of body weight input level at the end of the experiment. At 5 percent input rate fish grew by 52.7% (fed with 50:50, 25:75 and 0:100 ration sizes). Fry growth rates resulted at each application rate are best described by corresponding logarismic scale with growth as illustrated in figure 7.

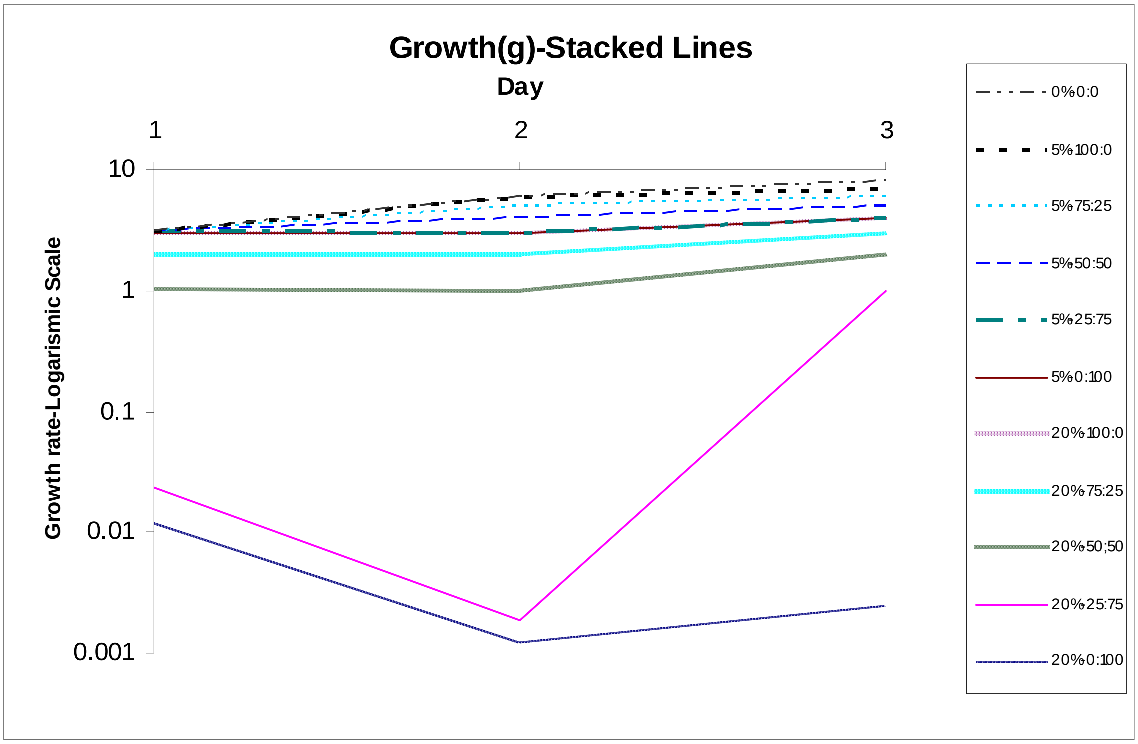


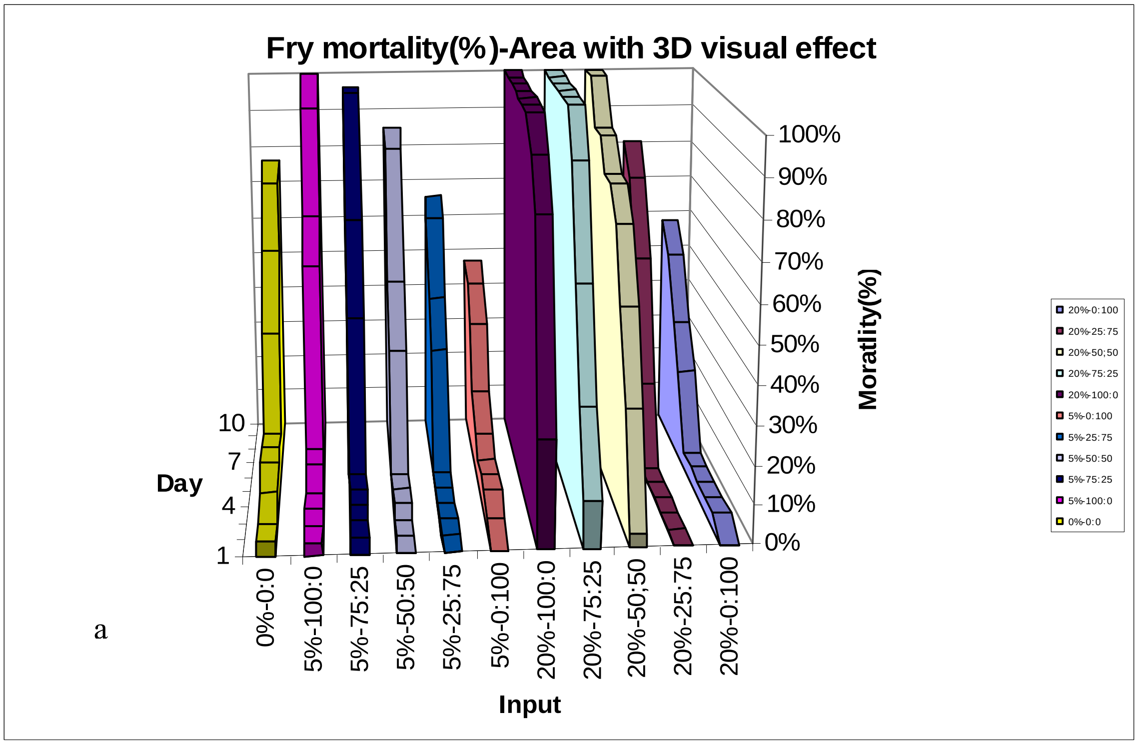
Figure 7. Fry growth rate of fries fed with different proportions of larval feed and Spirulina at 0, 5 and 20% of body weight. Logarismic scale is used to avoid negative values after the eighth day (Day 2 in this graph). Growth in Day 3 was rated at 10th day of experiment from Day 2. Corresponding values don't imply fry growth were weaker than rated at Day 2. Lower rates simply were associated with shorted time elapsed. However, it is clear that containers with 100% mortality after Day 2 represent bigger negative growth rates; hence, larger logarismic values. (Growth rate data in Appendix C 1.1)

Logarismic growth rate generally increases both with decreasing input rate and reduced inclusion of *Spirulina*. In other words, fry growth rate follows increasing trend with increasing input amount and higher inclusion of *Spirulina* for larger logarism corresponds to smaller or negative growth rate. However it should be noted that the above description avoids inclusion of treatments that resulted in 100% fry mortality after Day 2 representing larger logarisms.

Input level, described as percent of body weight, affected fry survival significantly ($p<0.05$) in days 2-8. During this time mortality was higher at 20% than at 5% rate and when fish received no input(table 3). However the degree of mortality similarly increased when the inclusion of fish meal becomes heavier (figure 8-a, b).

Table 3. One-way ANOVA for mortality of fish fed at 0, 5 and 20%. Mean values with any dissimilar superscripts in each row, differ significantly (p<0.05).

Day	0	5	20
1	4.00 ± 8.16ab	0.68 ± 3.40a	8.68 ± 16.34b
2	4.00 ± 8.16a	1.48 ± 5.14a	29.48 ± 36.61b
3	8.00 ± 16.33a	2.16 ± 5.99a	42.00 ± 44.72b
4	12.00 ± 16.33a	2.16 ± 5.99a	52.80 ± 47.22b
5	12.00 ± 16.33a	2.96 ± 6.95a	56.80 ± 49.89b
6	12.00 ± 16.33a	3.60 ± 8.79a	56.80 ± 49.89b
7	36.00 ± 23.80a	34.76 ± 25.46a	66.28 ± 41.26b
8	56.00 ± 15.28a	50.52 ± 22.88a	74.80 ± 34.05b
9	72.00 ± 20.82a	74.52 ± 22.39a	84.40 ± 23.82a
10	76.00 ± 23.80a	78.52 ± 21.92a	87.48 ± 20.38a



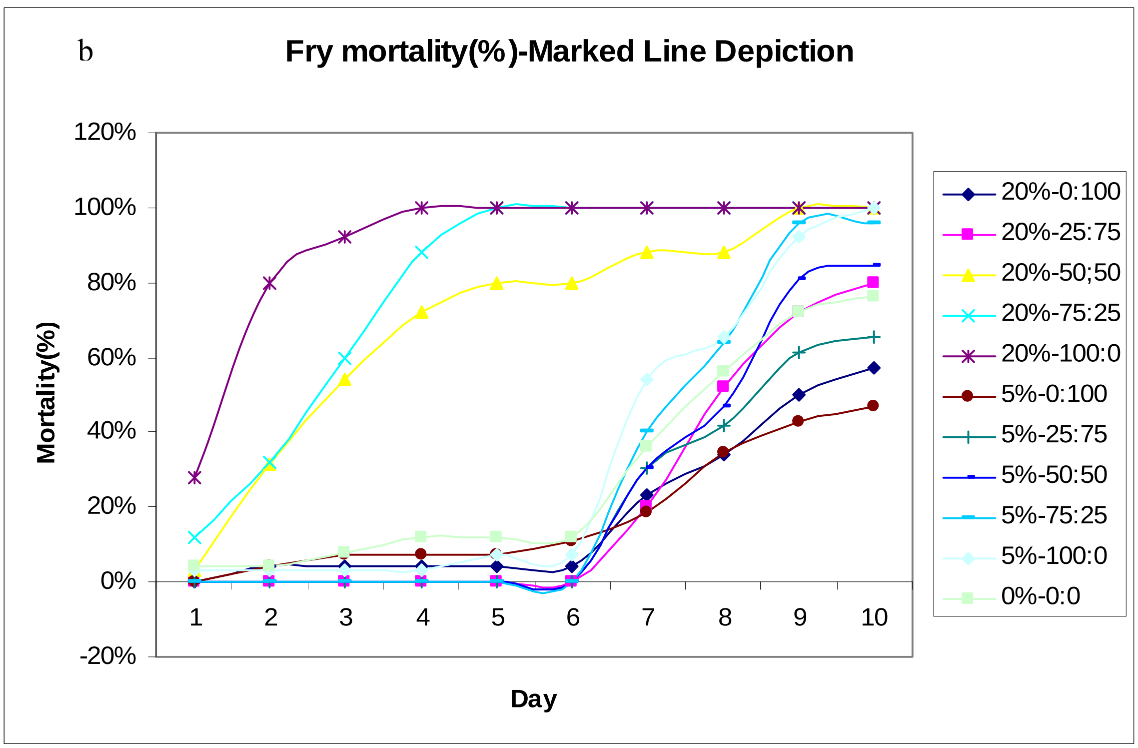


Figure 8-a, b. Fry mortality (%) due to input of *Spirulina* and feed generally increased as the amount of Spirulina supplementation decreases. Ratios are given in percent of feed to *Spirulina* (F:Sp) supplementation at 5 and 20% of body weight. (Mortality Data in Appendix C 2.1)

For all the replicates, there were strong polynomial relationship amongst input rates and percent of fry survival (with r^2 close to 1) as illustrated in figure 9. In a system with no discharge of water, low input rates of artificial feed complement can help guarantee improved quality of water that consequently enable enhanced survival and production performance of tilapia fry with *Spirulina* nutritional replacement. Although tilapia are highly resistant to reduced quality of water, they result in well enhanced survival and increased yield where good water quality is kept through mechanisms as is a performance parameter in closed design recirculation system evaluated by Shnel et al. (2002). In ponds water quality parameters can be improved through decreasing inputs of commercial feeds and letting natural productivity increase by fertilizer application (Green et al., 1995).
Reduced percent of fry mortality in the current paper is implied by increased percent of *Spirulina* inclusion; hence, the importance of natural pigment supplement.

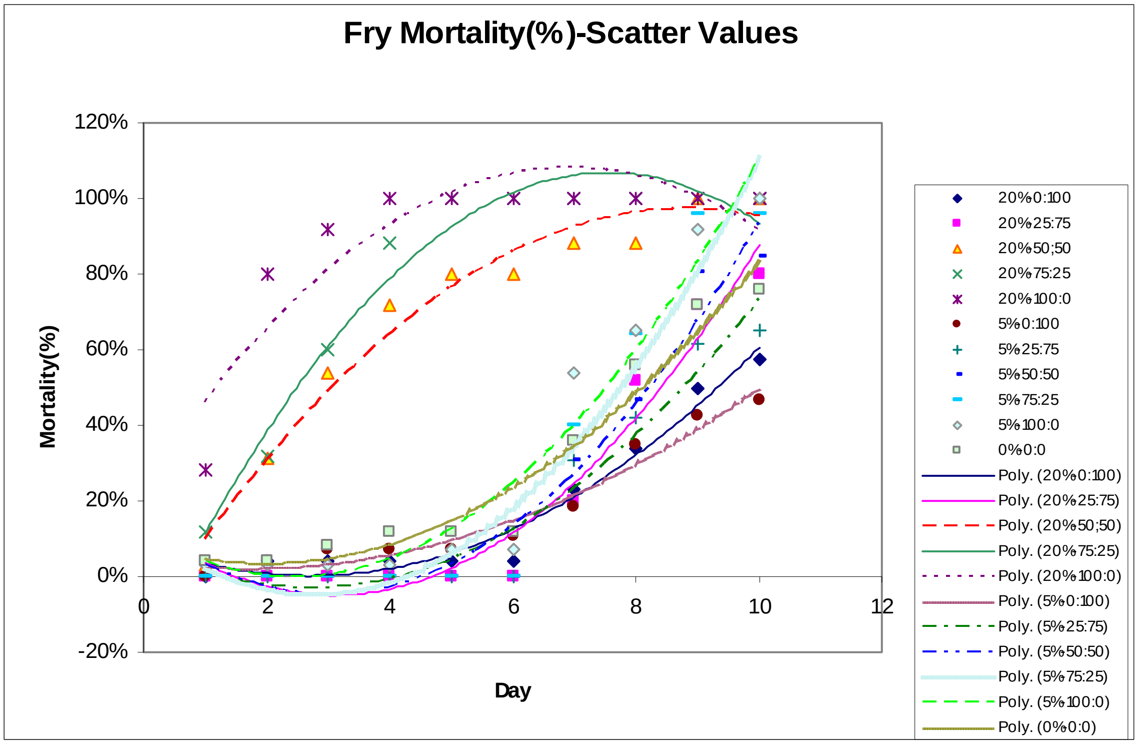


Figure 9. Regression lines fit polynomial equations. R-square values (close to 1) show great relationship of mortality values resulted at 0, 5 and 20% input rates. Equations below correspond for ration sizes on the legend in left-right with top-bottom order. (Mortality Data in Appendix C 2.1)

y = 0.0114x2 - 0.0616x + 0.086, r^2 = *0.9615*; y = 0.0194x2 - 0.1198x + 0.136, r^2 = *0.9524*;
y = -0.0144x2 + 0.2531x - 0.1423, r^2 = *0.9661*; y = -0.0223x2 + 0.3362x - 0.1993, r^2 = *0.9681*;
y = -0.0176x2 + 0.2435x + 0.2373, r^2 = *0.7979*; y = 0.0069x2 - 0.0232x + 0.0422, r^2 = *0.9585*;
y = 0.0144x2 - 0.0787x + 0.0768, r^2 = *0.9425*; y = 0.0201x2 - 0.121x + 0.1342, r^2 = *0.954*;
y = 0.0221x2 - 0.1236x + 0.124, r^2 = *0.9381*; y = 0.0194x2 - 0.0947x + 0.1146, r^2 = *0.9475*;
y = 0.0124x2 - 0.0496x + 0.0867, r^2 = *0.9554*

Conclusion

Salinity and feed manipulation results have implications in the closed system fry laboratory arrangement. Good tolerance of confined *Oreochromis mossambicus* fries to shock salinity contributes beyond their surpassing gradual-acclimation capacity. This makes them more preferable for salt water grow-out. However their survival and growth is also influenced by nutritional status and water parameters. Vijayan et al (1996) discussed the difficulties in regulating plasma Cl^- of food-deprived tilapia in seawater acclimation. That implies acclimation of freshwater hatched fries in saltwater would require good nutritional support to undergo minimal loss in osmoregulation and maintenance retaining much energy for growth. Fasting tilapia are thought to switch their course of metabolic energy from growth to basal metabolism including maintenance of ion and water balance (Uchida et al., 2003). Determination of specific ration size is also important because of impacts on somatic and otolith (Massou et al., 2002) growth besides to survival, yield and net profits of crops. In the course of artificial larvae propagation and nutrition, other problems of prime consideration include predation, cannibalism and first feeding problems (Rice et al., 1994). These can result in high percentages of mortality.

In a closed system confinement reduced amount of input would be preferable to keep good quality of water to achieve sustained levels of survival and yield. Newly hatched tilapia fries can depend on natural food organisms after stocking and therefore do not need to receive supplement feed for a while. In a work done by Brown et al. (2001) delaying onset of supplement feeding has resulted on reduced use of input amount yet improved net value of *Oreochromis niloticus* crops. It is evidenced in the current paper that less than 5-10 (as % of fish weight) input value of *Spirulina* and less than 50% feed complement ratio when supplemented assure reduced mortality. Growth can also be consistent with improved quality of water. Previously, Olvera-Novoa et al. (1998) supported purely up to 40% *Spirulina Platensis* replacement of fish meal at 6% of body weight for improved growth performance in closed-recirculating system and higher sizes with phosphorus mineral supplement. The current findings can be used to guide use of live *Spirulina* and its productive availability that can be made to support nutritional requirements of tilapia fries in salt water culture.

Recommendation

To help fit the current findings and to make use of salt water concurred cultures of *O. mossambicus* fry and *Spirulina platensis*, it will be necessary to find the tolerance level of live *Spirulina* to the salinity regimes where *mossambicus* fry showed good survival performance. Improved productivity of *Spirulina* in ponds can greatly support culture of tilapia. This may also reduce impacts of reduced water quality on mortality due to high levels of dry particulate matter accumulation. *Spirulina* growth and productivity can be aided through fertilizer application. However availability and survival requires good knowledge of application levels that also need to be made to keep water medium parameters that are optimum requirements for enhanced survival and growth performances achievable by culture of *O. mossambicus* fry in saltwater.

With changes in technique there could be possibility of investigating the optimum *Spirulina* supplement size at those very low input rates described as percent of body weight. For example, a technique that enables investigation of growth rate of newly-hatched fry on a daily basis is sought after with ration manipulation that allow moderate mortality level in a closed system. Finally to go with the use of inputs in closed systems and towards the use of live *Spirulina* nutrient concurred to salt water *mossambicus* culture, analyses of important water quality parameters such as alkalinity, dissolved oxygen (DO), biochemical oxygen demand (BOD) and related to nutrient dynamics are of prime factors of consideration.

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Appendix

A1:- Mortality data resulted with salinity treatments

Salinity	Replicate	Mortality (%)								
Treatment 1 (0 g/lt)	**0**	0%	0%	0%	0%	0%	50%	100%	100%	100%
	0	0%	0%	0%	0%	0%	0%	50%	50%	50%
	0	0%	0%	0%	0%	0%	0%	0%	50%	50%
	0	0%	0%	0%	0%	0%	0%	50%	50%	50%
	0	0%	0%	0%	0%	0%	50%	50%	50%	50%
	0	0%	0%	0%	0%	0%	0%	100%	100%	100%
	0	0%	0%	0%	0%	0%	0%	100%	100%	100%
	0	0%	0%	0%	0%	0%	0%	100%	100%	100%
	0	0%	0%	0%	0%	0%	0%	100%	100%	100%
	0	0%	0%	0%	0%	0%	0%	100%	100%	100%
	0	0%	0%	0%	0%	0%	50%	100%	100%	100%
	0	0%	0%	0%	0%	0%	0%	100%	100%	100%
Treatment 2 (5 g/lt)	**5**	0%	0%	0%	50%	50%	50%	100%	100%	100%
	5	0%	0%	0%	50%	50%	50%	100%	100%	100%
	5	0%	0%	0%	0%	50%	50%	50%	100%	100%
	5	0%	0%	0%	0%	0%	0%	50%	50%	100%
	5	0%	0%	0%	0%	0%	50%	100%	100%	100%
	5	0%	0%	0%	0%	0%	50%	100%	100%	100%
	5	0%	0%	50%	50%	50%	50%	100%	100%	100%
	5	0%	0%	0%	0%	50%	50%	100%	100%	100%
	5	0%	0%	0%	0%	0%	50%	50%	100%	100%
	5	0%	0%	0%	0%	0%	0%	100%	100%	100%
	5	0%	0%	0%	0%	0%	0%	0%	0%	0%
	5	0%	0%	0%	0%	0%	0%	100%	100%	100%
Treatment 3 (10 g/lt)	**10**	0%	0%	0%	0%	0%	50%	100%	100%	100%
	10	0%	0%	0%	50%	50%	50%	50%	50%	100%
	10	0%	0%	50%	50%	50%	50%	50%	50%	100%
	10	0%	0%	0%	0%	0%	50%	100%	100%	100%
	10	0%	0%	50%	50%	50%	100%	100%	100%	100%
	10	0%	0%	0%	0%	0%	50%	50%	100%	100%
	10	0%	0%	0%	0%	0%	0%	100%	100%	100%
	10	0%	0%	0%	0%	0%	50%	50%	100%	100%
	10	0%	0%	0%	0%	0%	0%	100%	100%	100%
	10	0%	0%	0%	50%	50%	50%	50%	100%	100%
	10	0%	0%	0%	0%	0%	100%	100%	100%	100%
	10	0%	0%	50%	50%	50%	50%	100%	100%	100%
Treatment 4 (15 g/lt)	**15**	0%	0%	0%	50%	100%	100%	100%	100%	100%
	15	0%	0%	100%	100%	100%	100%	100%	100%	100%
	15	0%	0%	0%	0%	0%	0%	50%	100%	100%
	15	0%	50%	50%	50%	50%	50%	100%	100%	100%
	15	50%	50%	50%	50%	100%	100%	100%	100%	100%
	15	50%	50%	50%	50%	100%	100%	100%	100%	100%
	15	0%	0%	0%	0%	0%	0%	100%	100%	100%
	15	0%	0%	0%	50%	50%	100%	100%	100%	100%
	15	0%	0%	0%	0%	50%	100%	100%	100%	100%
	15	0%	0%	100%	100%	100%	100%	100%	100%	100%

	15	0%	0%	0%	0%	0%	50%	100%	100%	100%
	15	0%	0%	0%	0%	0%	0%	100%	100%	100%
Treatment 5 (20 g/lt)	**20**	100%	100%	100%	100%	100%	100%	100%	100%	100%
	20	100%	100%	100%	100%	100%	100%	100%	100%	100%
	20	100%	100%	100%	100%	100%	100%	100%	100%	100%
	20	100%	100%	100%	100%	100%	100%	100%	100%	100%
	20	100%	100%	100%	100%	100%	100%	100%	100%	100%
	20	50%	50%	50%	50%	100%	100%	100%	100%	100%
	20	100%	100%	100%	100%	100%	100%	100%	100%	100%
	20	0%	0%	0%	0%	50%	100%	100%	100%	100%
	20	50%	50%	50%	50%	50%	100%	100%	100%	100%
	20	100%	100%	100%	100%	100%	100%	100%	100%	100%
	20	100%	100%	100%	100%	100%	100%	100%	100%	100%
	20	100%	100%	100%	100%	100%	100%	100%	100%	100%
Treatment 6 (25 g/lt)	**25**	100%	100%	100%	100%	100%	100%	100%	100%	100%
	25	100%	100%	100%	100%	100%	100%	100%	100%	100%
	25	100%	100%	100%	100%	100%	100%	100%	100%	100%
	25	100%	100%	100%	100%	100%	100%	100%	100%	100%
	25	100%	100%	100%	100%	100%	100%	100%	100%	100%
	25	100%	100%	100%	100%	100%	100%	100%	100%	100%
	25	100%	100%	100%	100%	100%	100%	100%	100%	100%
	25	100%	100%	100%	100%	100%	100%	100%	100%	100%
	25	100%	100%	100%	100%	100%	100%	100%	100%	100%
	25	100%	100%	100%	100%	100%	100%	100%	100%	100%
	25	100%	100%	100%	100%	100%	100%	100%	100%	100%
	25	100%	100%	100%	100%	100%	100%	100%	100%	100%
Treatment 7 (30 g/lt)	**30**	100%	100%	100%	100%	100%	100%	100%	100%	100%
	30	100%	100%	100%	100%	100%	100%	100%	100%	100%
	30	100%	100%	100%	100%	100%	100%	100%	100%	100%
	30	100%	100%	100%	100%	100%	100%	100%	100%	100%
	30	100%	100%	100%	100%	100%	100%	100%	100%	100%
	30	100%	100%	100%	100%	100%	100%	100%	100%	100%
	30	100%	100%	100%	100%	100%	100%	100%	100%	100%
	30	100%	100%	100%	100%	100%	100%	100%	100%	100%
	30	100%	100%	100%	100%	100%	100%	100%	100%	100%
	30	100%	100%	100%	100%	100%	100%	100%	100%	100%
	30	100%	100%	100%	100%	100%	100%	100%	100%	100%
	30	100%	100%	100%	100%	100%	100%	100%	100%	100%
Treatment 8 (35 g/lt)	**35**	100%	100%	100%	100%	100%	100%	100%	100%	100%
	35	100%	100%	100%	100%	100%	100%	100%	100%	100%
	35	100%	100%	100%	100%	100%	100%	100%	100%	100%
	35	100%	100%	100%	100%	100%	100%	100%	100%	100%
	35	100%	100%	100%	100%	100%	100%	100%	100%	100%
	35	100%	100%	100%	100%	100%	100%	100%	100%	100%
	35	100%	100%	100%	100%	100%	100%	100%	100%	100%
	35	100%	100%	100%	100%	100%	100%	100%	100%	100%
	35	100%	100%	100%	100%	100%	100%	100%	100%	100%
	35	100%	100%	100%	100%	100%	100%	100%	100%	100%
	35	100%	100%	100%	100%	100%	100%	100%	100%	100%
	35	100%	100%	100%	100%	100%	100%	100%	100%	100%

A2:- ANOVA summary of salinity effects

One Way Analysis of Variance of Mortality Data

One-way ANOVA: Day1 versus Salinity

Analysis of Variance for Day1

Source	DF	SS	MS	F	P
Salinity	7	214063	30580	169.96	0.000
Error	88	15833	180		
Total	95	229896			

One-way ANOVA: Day2 versus Salinity

Analysis of Variance for Day2

Source	DF	SS	MS	F	P
Salinity	7	210182	30026	152.81	0.000
Error	88	17292	196		
Total	95	227474			

One-way ANOVA: Day3 versus Salinity

Analysis of Variance for Day3

Source	DF	SS	MS	F	P
Salinity	7	179349	25621	61.14	0.000
Error	88	36875	419		
Total	95	216224			

One-way ANOVA: Day4 versus Salinity

Analysis of Variance for Day4

Source	DF	SS	MS	F	P
Salinity	7	157891	22556	49.37	0.000
Error	88	40208	457		
Total	95	198099			

One-way ANOVA: Day5 versus Salinity

Analysis of Variance for Day5

Source	DF	SS	MS	F	P
Salinity	7	149974	21425	45.94	0.000
Error	88	41042	466		
Total	95	191016			

One-way ANOVA: Day6 versus Salinity

Analysis of Variance for Day6

Source	DF	SS	MS	F	P
Salinity	7	103932	14847	29.72	0.000
Error	88	43958	500		
Total	95	147891			

One-way ANOVA: Day7 versus Salinity

Analysis of Variance for Day7

Source	DF	SS	MS	F	P
Salinity	7	9167	1310	3.37	0.003
Error	88	34167	388		
Total	95	43333			

One-way ANOVA: Day8 versus Salinity

Analysis of Variance for Day8

Source	DF	SS	MS	F	P
Salinity	7	3932	562	2.30	0.033
Error	88	21458	244		
Total	95	25391			

One-way ANOVA: Day9 versus Salinity

Analysis of Variance for Day9

Source	DF	SS	MS	F	P
Salinity	7	3229	461	2.56	0.019
Error	88	15833	180		
Total	95	19063			

B1:- Growth data resulted with *Spirulina* type and rate treatments

Input	Replicates	Growth(g)										
		Initial Wt	Day1	Day2	Day3	Day4	Day5	Day6	Day7	Day8	Day9	Day10
2.5% Body Weight	A11	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.00	0.00	0.00	0.00
	A12	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.00	0.00
	A13	0.01	0.01	0.01	0.01	0.02	0.01	0.01	0.01	0.00	0.00	0.00
	B11	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.01	0.00	0.00	0.00
	B12	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.00	0.00	0.00
	B13	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.01
	C11	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.00	0.00
	C12	0.01	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.00	0.00
	C13	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.00	0.00
5% Body Weight	A21	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.00	0.00	0.00
	A22	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.00	0.00	0.00
	A23	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.02	0.00	0.00	0.00
	B21	0.01	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.02
	B22	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.02	0.00	0.00
	B23	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.00	0.00
	C21	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.00	0.00	0.00
	C22	0.01	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.00	0.00
	C23	0.01	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.02
10% Body Weight	A31	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.00	0.00	0.00
	A32	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.02	0.00	0.00	0.00
	A33	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.00	0.00	0.00
	B31	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.00	0.00	0.00
	B32	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.00	0.00	0.00
	B33	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.00	0.00	0.00
	C31	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.03	0.03
	C32	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.00	0.00	0.00
	C33	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.00	0.00	0.00
20% Body Weight	A41	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.03	0.00	0.00	0.00
	A42	0.01	0.01	0.01	0.01	0.02	0.02	0.03	0.03	0.00	0.00	0.00
	A43	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.03	0.00	0.00	0.00
	B41	0.01	0.01	0.01	0.02	0.02	0.02	0.03	0.02	0.03	0.00	0.00
	B42	0.01	0.01	0.01	0.02	0.02	0.02	0.03	0.03	0.00	0.00	0.00
	B43	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.03	0.03	0.00	0.00

	C41	0.01	0.01	0.01	0.01	0.02	0.03	0.03	0.03	0.04	0.04	0.04
	C42	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.03	0.04	0.03	0.04
	C43	0.01	0.01	0.01	0.02	0.01	0.02	0.03	0.03	0.04	0.04	0.04
40% Body Weight	**A51**	0.01	0.01	0.01	0.01	0.02	0.02	0.03	0.03	0.00	0.00	0.00
	A52	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.02	0.00	0.00	0.00
	A53	0.01	0.01	0.01	0.01	0.02	0.02	0.03	0.03	0.00	0.00	0.00
	B51	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.03	0.00	0.00	0.00
	B52	0.01	0.01	0.01	0.01	0.02	0.03	0.03	0.03	0.00	0.00	0.00
	B53	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.03	0.00	0.00	0.00
	C51	0.01	0.01	0.02	0.02	0.02	0.02	0.02	0.03	0.00	0.00	0.00
	C52	0.01	0.01	0.01	0.01	0.02	0.02	0.03	0.04	0.05	0.00	0.00
	C53	0.01	0.01	0.01	0.02	0.01	0.03	0.04	0.04	0.03	0.04	0.04
80% Body Weight	**A61**	0.01	0.01	0.01	0.02	0.02	0.02	0.03	0.03	0.00	0.00	0.00
	A62	0.01	0.01	0.02	0.02	0.02	0.03	0.03	0.00	0.00	0.00	0.00
	A63	0.01	0.01	0.01	0.02	0.02	0.03	0.02	0.03	0.00	0.00	0.00
	B61	0.01	0.01	0.01	0.01	0.02	0.03	0.03	0.03	0.00	0.00	0.00
	B62	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.00	0.00	0.00
	B63	0.01	0.01	0.01	0.02	0.02	0.02	0.03	0.03	0.00	0.00	0.00
	C61	0.01	0.01	0.02	0.01	0.02	0.03	0.02	0.00	0.00	0.00	0.00
	C62	0.01	0.01	0.02	0.02	0.02	0.03	0.03	0.04	0.00	0.00	0.00
	C63	0.01	0.01	0.01	0.02	0.02	0.02	0.00	0.00	0.00	0.00	0.00
160% Body Weight	**A71**	0.01	0.01	0.02	0.02	0.02	0.02	0.00	0.00	0.00	0.00	0.00
	A72	0.01	0.01	0.01	0.02	0.02	0.00	0.00	0.00	0.00	0.00	0.00
	A73	0.01	0.01	0.01	0.01	0.02	0.02	0.00	0.00	0.00	0.00	0.00
	B71	0.01	0.01	0.01	0.01	0.02	0.02	0.00	0.00	0.00	0.00	0.00
	B72	0.01	0.01	0.01	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	B73	0.01	0.01	0.02	0.02	0.02	0.02	0.00	0.00	0.00	0.00	0.00
	C71	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.00	0.00	0.00	0.00
	C72	0.01	0.01	0.01	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	C73	0.01	0.01	0.02	0.02	0.02	0.00	0.00	0.00	0.00	0.00	0.00
320% Body Weight	**A81**	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.00	0.00	0.00	0.00
	A82	0.01	0.01	0.01	0.02	0.02	0.00	0.00	0.00	0.00	0.00	0.00
	A83	0.01	0.01	0.01	0.01	0.02	0.00	0.00	0.00	0.00	0.00	0.00
	B81	0.01	0.01	0.01	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00

	B82	0.01	0.01	0.01	0.02	0.02	0.03	0.04	0.04	0.04	0.04	0.05
	B83	0.01	0.02	0.02	0.02	0.02	0.00	0.00	0.00	0.00	0.00	0.00
	C81	0.01	0.01	0.02	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	C82	0.01	0.01	0.01	0.02	0.02	0.00	0.00	0.00	0.00	0.00	0.00
	C83	0.01	0.01	0.02	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00
640% Body Weight	**A91**	0.01	0.01	0.01	0.02	0.02	0.03	0.03	0.03	0.03	0.04	0.04
	A92	0.01	0.01	0.15	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	A93	0.01	0.01	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	B91	0.01	0.01	0.01	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	B92	0.01	0.01	0.02	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	B93	0.01	0.02	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	C91	0.01	0.01	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	C92	0.01	0.01	0.02	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	C93	0.01	0.02	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
1280% Body Weight	**A101**	0.01	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	A102	0.01	0.01	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	A103	0.01	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	B101	0.01	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	B102	0.01	0.01	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	B103	0.01	0.01	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	C101	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	C102	0.01	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	C103	0.01	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control(0%)	**O94**	0.01	0.01	0.01	0.01	0.01	0.01	0.00	0.00	0.00	0.00	0.00
	O95	0.01	0.01	0.01	0.01	0.01	0.00	0.00	0.00	0.00	0.00	0.00
	O91	0.01	0.01	0.01	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00

B1.1. Average growth rate(g) as a result of A-B-C input rates

Input Rate	Day										
	1	2	3	4	5	6	7	8	9	10	11
0	**0**	**0.000549**	**0.00021**	**0.000222**	**-0.00089**	**-0.0015**	**-0.00111**	**0**	**0**	**0**	**0**
2.5	0	0.001082	0.001196	0.001204	0.001574	0.000741	0.000315	-0.00219	-0.00319	-0.00707	0.001111
5	0	0.000704	0.0015	0.001566	0.001489	0.001	0.000963	0.001315	-0.00609	-0.006	0.004444
10	**0**	**0.001241**	**0.000815**	**0.00263**	**0.002037**	**0.003611**	**0.000333**	**0.001111**	**-0.01756**	**0.000556**	**0.003333**
20	0	0.001333	0.001204	0.003944	0.0035	0.00313	0.003611	0.00213	-0.00833	-0.00648	0.013333
40	0	0.000963	0.002148	0.00287	0.002741	0.005407	0.002611	0.005963	-0.02237	-0.00389	0.004444
80	0	0.001463	0.003685	0.001574	0.004907	0.003593	-0.0015	-0.00172	-0.02	0	0
160	**0**	**0.001185**	**0.002852**	**0.002037**	**-0.0003**	**-0.00289**	**-0.00867**	**-0.00222**	**0**	**0**	**0**
320	0	0.001759	0.003019	0.004593	-0.00426	-0.00756	0.000778	-0.00189	-0.00056	0.000778	0.005222
640	0	0.003704	0.01863	-0.02144	-0.00633	0.000889	-0.00033	0.000222	0.000333	0.000222	0.004556
1280	0	0.001556	-0.00548	-0.00407	0	0	0	0	0	0	0

B2:- Mortality data resulted with *Spirulina* type and rate treatments

Input	Replicates	Mortality (%)										
		Day0	Day1	Day2	Day3	Day4	Day5	Day6	Day7	Day8	Day9	Day10
2.5% Body Weight	A11	0%	0%	20%	20%	40%	60%	60%	100%	100%	100%	100%
	A12	0%	0%	33%	33%	50%	50%	83%	83%	83%	100%	100%
	A13	0%	0%	0%	25%	50%	75%	75%	75%	100%	100%	100%
	B11	0%	0%	40%	40%	40%	60%	60%	80%	100%	100%	100%
	B12	0%	0%	20%	20%	40%	40%	80%	80%	100%	100%	100%
	B13	0%	14%	14%	43%	43%	57%	57%	57%	57%	86%	86%
	C11	0%	0%	0%	20%	20%	20%	40%	40%	80%	100%	100%
	C12	0%	17%	17%	33%	33%	33%	33%	33%	67%	100%	100%
	C13	0%	0%	14%	43%	43%	57%	71%	71%	86%	100%	100%
5% Body Weight	A21	0%	0%	20%	20%	60%	80%	80%	80%	100%	100%	100%
	A22	0%	0%	40%	40%	40%	80%	80%	80%	100%	100%	100%
	A23	0%	33%	33%	44%	44%	67%	67%	67%	100%	100%	100%
	B21	0%	0%	20%	40%	40%	40%	60%	60%	80%	80%	80%
	B22	0%	17%	17%	33%	67%	67%	67%	67%	83%	100%	100%
	B23	0%	0%	0%	25%	25%	25%	25%	25%	50%	100%	100%
	C21	0%	0%	20%	20%	20%	60%	80%	80%	100%	100%	100%
	C22	0%	0%	0%	20%	40%	40%	40%	40%	80%	100%	100%
	C23	0%	17%	17%	42%	58%	58%	75%	83%	83%	92%	92%
10% Body Weight	A31	0%	0%	20%	20%	20%	40%	40%	80%	100%	100%	100%
	A32	0%	0%	0%	33%	50%	50%	50%	83%	100%	100%	100%
	A33	0%	0%	0%	25%	25%	50%	50%	75%	100%	100%	100%
	B31	0%	0%	20%	40%	40%	40%	80%	80%	100%	100%	100%
	B32	0%	0%	0%	40%	40%	60%	60%	80%	100%	100%	100%
	B33	0%	20%	20%	40%	40%	40%	60%	60%	100%	100%	100%
	C31	0%	0%	0%	20%	20%	20%	60%	60%	80%	80%	80%
	C32	0%	0%	17%	33%	33%	67%	67%	67%	100%	100%	100%
	C33	0%	17%	17%	50%	50%	67%	67%	83%	100%	100%	100%
20% Body Weight	A41	0%	0%	20%	20%	20%	60%	60%	80%	100%	100%	100%
	A42	0%	0%	0%	40%	40%	60%	60%	60%	100%	100%	100%
	A43	0%	0%	0%	0%	0%	0%	67%	67%	100%	100%	100%
	B41	0%	20%	20%	40%	40%	40%	60%	80%	80%	100%	100%
	B42	0%	0%	0%	20%	20%	60%	80%	80%	100%	100%	100%
	B43	0%	0%	17%	50%	50%	50%	50%	67%	83%	100%	100%

	C41	0%	0%	0%	0%	0%	20%	20%	53%	70%	87%	87%
	C42	0%	0%	0%	0%	0%	20%	20%	20%	40%	40%	40%
	C43	0%	0%	20%	20%	40%	40%	40%	60%	60%	80%	80%
40% Body Weight	**A51**	0%	0%	0%	0%	0%	40%	40%	40%	100%	100%	100%
	A52	0%	0%	33%	33%	33%	33%	50%	83%	100%	100%	100%
	A53	0%	0%	17%	17%	33%	33%	67%	83%	100%	100%	100%
	B51	0%	0%	0%	20%	20%	20%	40%	80%	100%	100%	100%
	B52	0%	0%	20%	40%	40%	60%	80%	80%	100%	100%	100%
	B53	0%	0%	0%	20%	20%	20%	20%	60%	100%	100%	100%
	C51	0%	0%	20%	20%	20%	40%	40%	60%	100%	100%	100%
	C52	0%	25%	25%	38%	50%	50%	63%	63%	75%	100%	100%
	C53	0%	0%	0%	20%	20%	60%	60%	60%	80%	80%	80%
80% Body Weight	**A61**	0%	0%	0%	20%	40%	40%	80%	80%	100%	100%	100%
	A62	0%	0%	40%	40%	40%	60%	80%	100%	100%	100%	100%
	A63	0%	0%	0%	33%	33%	33%	33%	67%	100%	100%	100%
	B61	0%	20%	20%	20%	60%	60%	80%	80%	100%	100%	100%
	B62	0%	0%	20%	20%	40%	40%	80%	80%	100%	100%	100%
	B63	0%	0%	20%	40%	40%	40%	60%	60%	100%	100%	100%
	C61	0%	33%	33%	33%	67%	67%	83%	100%	100%	100%	100%
	C62	0%	0%	25%	25%	50%	75%	75%	75%	100%	100%	100%
	C63	0%	33%	33%	56%	67%	89%	100%	100%	100%	100%	100%
160% Body Weight	**A71**	0%	40%	60%	60%	60%	80%	100%	100%	100%	100%	100%
	A72	0%	50%	50%	67%	83%	100%	100%	100%	100%	100%	100%
	A73	0%	33%	50%	50%	83%	83%	100%	100%	100%	100%	100%
	B71	0%	0%	20%	20%	60%	80%	100%	100%	100%	100%	100%
	B72	0%	40%	40%	60%	100%	100%	100%	100%	100%	100%	100%
	B73	0%	0%	60%	60%	80%	80%	100%	100%	100%	100%	100%
	C71	0%	33%	33%	67%	67%	83%	83%	100%	100%	100%	100%
	C72	0%	0%	50%	50%	100%	100%	100%	100%	100%	100%	100%
	C73	0%	20%	60%	60%	80%	100%	100%	100%	100%	100%	100%
320% Body Weight	**A81**	0%	0%	40%	40%	60%	80%	80%	100%	100%	100%	100%
	A82	0%	17%	50%	67%	83%	100%	100%	100%	100%	100%	100%
	A83	0%	0%	20%	40%	80%	100%	100%	100%	100%	100%	100%
	B81	0%	40%	40%	40%	100%	100%	100%	100%	100%	100%	100%
	B82	0%	0%	20%	60%	60%	80%	80%	80%	80%	80%	80%

	B83	0%	60%	60%	80%	80%	100%	100%	100%	100%	100%	100%
	C81	0%	0%	60%	80%	100%	100%	100%	100%	100%	100%	100%
	C82	0%	0%	25%	25%	75%	100%	100%	100%	100%	100%	100%
	C83	0%	0%	50%	50%	100%	100%	100%	100%	100%	100%	100%
640% Body Weight	**A91**	0%	43%	57%	86%	86%	86%	86%	86%	86%	86%	86%
	A92	0%	29%	71%	100%	100%	100%	100%	100%	100%	100%	100%
	A93	0%	0%	67%	100%	100%	100%	100%	100%	100%	100%	100%
	B91	0%	40%	40%	80%	100%	100%	100%	100%	100%	100%	100%
	B92	0%	0%	60%	80%	100%	100%	100%	100%	100%	100%	100%
	B93	0%	80%	80%	100%	100%	100%	100%	100%	100%	100%	100%
	C91	0%	40%	40%	100%	100%	100%	100%	100%	100%	100%	100%
	C92	0%	25%	25%	25%	100%	100%	100%	100%	100%	100%	100%
	C93	0%	60%	60%	100%	100%	100%	100%	100%	100%	100%	100%
1280% Body Weight	**A101**	0%	80%	100%	100%	100%	100%	100%	100%	100%	100%	100%
	A102	0%	40%	40%	100%	100%	100%	100%	100%	100%	100%	100%
	A103	0%	50%	100%	100%	100%	100%	100%	100%	100%	100%	100%
	B101	0%	80%	100%	100%	100%	100%	100%	100%	100%	100%	100%
	B102	0%	25%	75%	100%	100%	100%	100%	100%	100%	100%	100%
	B103	0%	40%	80%	100%	100%	100%	100%	100%	100%	100%	100%
	C101	0%	100%	100%	100%	100%	100%	100%	100%	100%	100%	100%
	C102	0%	80%	100%	100%	100%	100%	100%	100%	100%	100%	100%
	C103	0%	40%	100%	100%	100%	100%	100%	100%	100%	100%	100%
Control(0%)	**O94**	0%	0%	44%	44%	78%	89%	100%	100%	100%	100%	100%
	O95	0%	20%	40%	70%	90%	100%	100%	100%	100%	100%	100%
	O91	0%	33%	56%	56%	100%	100%	100%	100%	100%	100%	100%

B 2.1 Tables of Average Mortality(%) as result of A, B, and C input rates

A Input Rate	Day										
	1	2	3	4	5	6	7	8	9	10	11
0	**0%**	**18%**	**47%**	**57%**	**89%**	**96%**	**100%**	**100%**	**100%**	**100%**	**100%**
2.5	**0%**	**0%**	**18%**	**26%**	**47%**	**62%**	**73%**	**86%**	**94%**	**100%**	**100%**
5	**0%**	**11%**	**31%**	**35%**	**48%**	**76%**	**76%**	**76%**	**100%**	**100%**	**100%**
10	**0%**	**0%**	**7%**	**26%**	**32%**	**47%**	**47%**	**79%**	**100%**	**100%**	**100%**
20	**0%**	**0%**	**7%**	**20%**	**20%**	**40%**	**62%**	**69%**	**100%**	**100%**	**100%**
40	**0%**	**0%**	**17%**	**17%**	**22%**	**36%**	**52%**	**69%**	**100%**	**100%**	**100%**
80	**0%**	**0%**	**13%**	**31%**	**38%**	**44%**	**64%**	**82%**	**100%**	**100%**	**100%**
160	**0%**	**41%**	**53%**	**59%**	**76%**	**88%**	**100%**	**100%**	**100%**	**100%**	**100%**
320	**0%**	**6%**	**37%**	**49%**	**74%**	**93%**	**93%**	**100%**	**100%**	**100%**	**100%**
640	**0%**	**24%**	**65%**	**95%**	**95%**	**95%**	**95%**	**95%**	**95%**	**95%**	**95%**
1280	**0%**	**57%**	**80%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**

B Input Rate	Day										
	1	2	3	4	5	6	7	8	9	10	11
0	**0%**	**18%**	**47%**	**57%**	**89%**	**96%**	**100%**	**100%**	**100%**	**100%**	**100%**
2.5	**0%**	**5%**	**25%**	**34%**	**41%**	**52%**	**66%**	**72%**	**86%**	**95%**	**95%**
5	**0%**	**6%**	**12%**	**33%**	**44%**	**44%**	**51%**	**51%**	**71%**	**93%**	**93%**
10	**0%**	**7%**	**13%**	**40%**	**40%**	**47%**	**67%**	**73%**	**100%**	**100%**	**100%**
20	**0%**	**7%**	**12%**	**37%**	**37%**	**50%**	**63%**	**76%**	**88%**	**100%**	**100%**
40	**0%**	**0%**	**7%**	**27%**	**27%**	**33%**	**47%**	**73%**	**100%**	**100%**	**100%**
80	**0%**	**7%**	**20%**	**27%**	**47%**	**47%**	**73%**	**73%**	**100%**	**100%**	**100%**
160	**0%**	**13%**	**40%**	**47%**	**80%**	**87%**	**100%**	**100%**	**100%**	**100%**	**100%**
320	**0%**	**33%**	**40%**	**60%**	**80%**	**93%**	**93%**	**93%**	**93%**	**93%**	**93%**
640	**0%**	**40%**	**60%**	**87%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**
1280	**0%**	**48%**	**85%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**

C Input Rate	Day										
	1	2	3	4	5	6	7	8	9	10	11
0	**0%**	**18%**	**47%**	**57%**	**89%**	**96%**	**100%**	**100%**	**100%**	**100%**	**100%**
2.5	**0%**	**6%**	**10%**	**32%**	**32%**	**37%**	**48%**	**48%**	**77%**	**100%**	**100%**
5	**0%**	**6%**	**12%**	**27%**	**39%**	**53%**	**65%**	**68%**	**88%**	**97%**	**97%**
10	**0%**	**6%**	**11%**	**34%**	**34%**	**51%**	**64%**	**70%**	**93%**	**93%**	**93%**
20	**0%**	**0%**	**7%**	**7%**	**13%**	**27%**	**27%**	**44%**	**57%**	**69%**	**69%**
40	**0%**	**8%**	**15%**	**26%**	**30%**	**50%**	**54%**	**61%**	**85%**	**93%**	**93%**
80	**0%**	**22%**	**31%**	**38%**	**61%**	**77%**	**86%**	**92%**	**100%**	**100%**	**100%**
160	**0%**	**18%**	**48%**	**59%**	**82%**	**94%**	**94%**	**100%**	**100%**	**100%**	**100%**
320	**0%**	**0%**	**45%**	**52%**	**92%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**
640	**0%**	**42%**	**42%**	**75%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**
1280	**0%**	**73%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**

B 2.2. Average Mortality as a result of A-B-C Input rates

Input Rate	Day										
	1	2	3	4	5	6	7	8	9	10	11
0	0%	18%	47%	57%	89%	96%	100%	100%	100%	100%	100%
2.5	0%	3%	18%	31%	40%	50%	62%	69%	86%	98%	98%
5	0%	7%	19%	32%	44%	57%	64%	65%	86%	97%	97%
10	0%	4%	10%	34%	35%	48%	59%	74%	98%	98%	98%
20	0%	2%	9%	21%	23%	39%	51%	63%	81%	90%	90%
40	0%	3%	13%	23%	26%	40%	51%	68%	95%	98%	98%
80	0%	10%	21%	32%	49%	56%	75%	82%	100%	100%	100%
160	0%	24%	47%	55%	79%	90%	98%	100%	100%	100%	100%
320	0%	13%	41%	54%	82%	96%	96%	98%	98%	98%	98%
640	0%	35%	56%	86%	98%	98%	98%	98%	98%	98%	98%
1280	0%	59%	88%	100%	100%	100%	100%	100%	100%	100%	100%

B3:- ANOVA summary of Spirulina type and rate effects on growth

Two Way Analysis of Variance of Growth Rate Data

Two-way ANOVA: Day1 versus SP TYPE, INPUT
Analysis of Variance for Day1

Source	DF	SS	MS	F	P
SP TYPE	2	0.0000086	0.0000043	0.82	0.444
INPUT	10	0.0000562	0.0000056	1.07	0.391
Error	86	0.0004496	0.0000052		
Total	98	0.0005144			

Two-way ANOVA: Day2 versus SP TYPE, INPUT
Analysis of Variance for Day2

Source	DF	SS	MS	F	P
SP TYPE	2	0.000411	0.000205	1.00	0.371
INPUT	10	0.003023	0.000302	1.48	0.162
Error	86	0.017592	0.000205		
Total	98	0.021025			

Two-way ANOVA: Day3 versus SP TYPE, INPUT
Analysis of Variance for Day3

Source	DF	SS	MS	F	P
SP TYPE	2	0.000491	0.000245	1.08	0.345
INPUT	10	0.004830	0.000483	2.12	0.031
Error	86	0.019616	0.000228		
Total	98	0.024937			

Two-way ANOVA: Day4 versus SP TYPE, INPUT
Analysis of Variance for Day4

Source	DF	SS	MS	F	P
SP TYPE	2	0.0002011	0.0001005	3.08	0.051
INPUT	10	0.0010573	0.0001057	3.24	0.001
Error	86	0.0028098	0.0000327		
Total	98	0.0040682			

Two-way ANOVA: Day5 versus SP TYPE, INPUT
Analysis of Variance for Day5

Source	DF	SS	MS	F	P
SP TYPE	2	0.0000478	0.0000239	0.69	0.506
INPUT	10	0.0013674	0.0001367	3.93	0.000
Error	86	0.0029906	0.0000348		
Total	98	0.0044058			

Two-way ANOVA: Day6 versus SP TYPE, INPUT
Analysis of Variance for Day6

Source	DF	SS	MS	F	P
SP TYPE	2	0.0000265	0.0000132	0.55	0.577
INPUT	10	0.0009693	0.0000969	4.05	0.000
Error	86	0.0020595	0.0000239		
Total	98	0.0030554			

Two-way ANOVA: Day7 versus SP TYPE, INPUT
Analysis of Variance for Day7

Source	DF	SS	MS	F	P
SP TYPE	2	0.0000611	0.0000306	0.95	0.392
INPUT	10	0.0005281	0.0000528	1.64	0.109
Error	86	0.0027716	0.0000322		
Total	98	0.0033608			

Two-way ANOVA: Day8 versus SP TYPE, INPUT
Analysis of Variance for Day8

Source	DF	SS	MS	F	P
SP TYPE	2	0.0008402	0.0004201	5.42	0.006
INPUT	10	0.0069847	0.0006985	9.01	0.000
Error	86	0.0066669	0.0000775		
Total	98	0.0144918			

Two-way ANOVA: Day9 versus SP TYPE, INPUT
Analysis of Variance for Day9

Source	DF	SS	MS	F	P
SP TYPE	2	0.0001423	0.0000711	1.31	0.274
INPUT	10	0.0009052	0.0000905	1.67	0.100
Error	86	0.0046564	0.0000541		
Total	98	0.0057039			

Two-way ANOVA: Day10 versus SP TYPE, INPUT
Analysis of Variance for Day10

Source	DF	SS	MS	F	P
SP TYPE	2	0.000480	0.000240	2.27	0.110
INPUT	10	0.001412	0.000141	1.33	0.226
Error	86	0.009111	0.000106		
Total	98	0.011003			

One Way Analysis of Variance of Growth Rate Data

One-way ANOVA: Day1 versus INPUT
Analysis of Variance for Day1

Source	DF	SS	MS	F	P
INPUT	10	0.0000187	0.0000019	1.14	0.377
Error	22	0.0000360	0.0000016		
Total	32	0.0000548			

One-way ANOVA: Day2 versus INPUT
Analysis of Variance for Day2

Source	DF	SS	MS	F	P
INPUT	10	0.0010075	0.0001008	1.41	0.240
Error	22	0.0015733	0.0000715		
Total	32	0.0025808			

One-way ANOVA: Day3 versus INPUT
Analysis of Variance for Day3

Source	DF	SS	MS	F	P
INPUT	10	0.0016100	0.0001610	2.30	0.049
Error	22	0.0015383	0.0000699		
Total	32	0.0031483			

One-way ANOVA: Day4 versus INPUT
Analysis of Variance for Day4

Source	DF	SS	MS	F	P
INPUT	10	0.0003524	0.0000352	2.84	0.020
Error	22	0.0002733	0.0000124		
Total	32	0.0006257			

One-way ANOVA: Day5 versus INPUT
Analysis of Variance for Day5

Source	DF	SS	MS	F	P
INPUT	10	0.0004558	0.0000456	5.34	0.001
Error	22	0.0001878	0.0000085		
Total	32	0.0006436			

One-way ANOVA: Day6 versus INPUT
Analysis of Variance for Day6

Source	DF	SS	MS	F	P
INPUT	10	0.0003231	0.0000323	3.34	0.009
Error	22	0.0002130	0.0000097		
Total	32	0.0005361			

One-way ANOVA: Day7 versus INPUT
Analysis of Variance for Day7

Source	DF	SS	MS	F	P
INPUT	10	0.0001760	0.0000176	2.40	0.042
Error	22	0.0001612	0.0000073		
Total	32	0.0003372			

One-way ANOVA: Day8 versus INPUT
Analysis of Variance for Day8

Source	DF	SS	MS	F	P
INPUT	10	0.0023282	0.0002328	4.59	0.001
Error	22	0.0011155	0.0000507		
Total	32	0.0034437			

One-way ANOVA: Day9 versus INPUT
Analysis of Variance for Day9

Source	DF	SS	MS	F	P
INPUT	10	0.0003017	0.0000302	1.15	0.375
Error	22	0.0005796	0.0000263		
Total	32	0.0008814			

One-way ANOVA: Day10 versus INPUT
Analysis of Variance for Day10

Source	DF	SS	MS	F	P
INPUT	10	0.0004707	0.0000471	0.66	0.751
Error	22	0.0015770	0.0000717		
Total	32	0.0020478			

B4:- ANOVA summary of Spirulina type and rate effects on mortality

Two Way Analysis of Variance of Mortality Data

Two-way ANOVA: Day1 versus SP TYPE, INPUT
Analysis of Variance for Day1

Source	DF	SS	MS	F	P
SP TYPE	2	244	122	0.44	0.647
INPUT	10	27993	2799	10.07	0.000
Error	86	23899	278		
Total	98	52135			

Two-way ANOVA: Day2 versus SP TYPE, INPUT
Analysis of Variance for Day2

Source	DF	SS	MS	F	P
SP TYPE	2	34	17	0.08	0.919
INPUT	10	55126	5513	27.48	0.000
Error	86	17254	201		
Total	98	72415			

Two-way ANOVA: Day3 versus SP TYPE, INPUT
Analysis of Variance for Day3

Source	DF	SS	MS	F	P
SP TYPE	2	250	125	0.60	0.551
INPUT	10	59891	5989	28.69	0.000
Error	86	17956	209		
Total	98	78097			

Two-way ANOVA: Day4 versus SP TYPE, INPUT
Analysis of Variance for Day4

Source	DF	SS	MS	F	P
SP TYPE	2	287	144	0.86	0.426
INPUT	10	78128	7813	46.85	0.000
Error	86	14343	167		
Total	98	92758			

Two-way ANOVA: Day5 versus SP TYPE, INPUT
Analysis of Variance for Day5

Source	DF	SS	MS	F	P
SP TYPE	2	189	94	0.48	0.621
INPUT	10	58967	5897	29.90	0.000
Error	86	16960	197		
Total	98	76116			

Two-way ANOVA: Day6 versus SP TYPE, INPUT
Analysis of Variance for Day6

Source	DF	SS	MS	F	P
SP TYPE	2	91	45	0.24	0.787
INPUT	10	39385	3938	20.84	0.000
Error	86	16256	189		
Total	98	55731			

Two-way ANOVA: Day7 versus SP TYPE, INPUT
Analysis of Variance for Day7

Source	DF	SS	MS	F	P
SP TYPE	2	750	375	2.34	0.103
INPUT	10	23160	2316	14.42	0.000
Error	86	13811	161		
Total	98	37721			

Two-way ANOVA: Day8 versus SP TYPE, INPUT
Analysis of Variance for Day8

Source	DF	SS	MS	F	P
SP TYPE	2	1098.0	549.0	5.57	0.005
INPUT	10	4235.1	423.5	4.30	0.000
Error	86	8478.0	98.6		
Total	98	13811.1			

Two-way ANOVA: Day9 versus SP TYPE, INPUT
Analysis of Variance for Day9

Source	DF	SS	MS	F	P
SP TYPE	2	255.5	127.8	2.32	0.105
INPUT	10	783.8	78.4	1.42	0.185
Error	86	4744.5	55.2		
Total	98	5783.8			

Two-way ANOVA: Day10 versus SP TYPE, INPUT
Analysis of Variance for Day10

Source	DF	SS	MS	F	P
SP TYPE	2	255.5	127.8	2.32	0.105
INPUT	10	783.8	78.4	1.42	0.185
Error	86	4744.5	55.2		
Total	98	5783.8			

One Way Analysis Of Variance of Mortality Data

One-way ANOVA: Day1 versus INPUT
Analysis of Variance for Day1

Source	DF	SS	MS	F	P
INPUT	10	27993	2799	10.20	0.000
Error	88	24142	274		
Total	98	52135			

One-way ANOVA: Day2 versus INPUT
Analysis of Variance for Day2

Source	DF	SS	MS	F	P
INPUT	10	55126	5513	28.06	0.000
Error	88	17288	196		
Total	98	72415			

One-way ANOVA: Day3 versus INPUT
Analysis of Variance for Day3

Source	DF	SS	MS	F	P
INPUT	10	59891	5989	28.95	0.000
Error	88	18206	207		
Total	98	78097			

One-way ANOVA: Day4 versus INPUT
Analysis of Variance for Day4

Source	DF	SS	MS	F	P
INPUT	10	78128	7813	46.99	0.000
Error	88	14630	166		
Total	98	92758			

One-way ANOVA: Day5 versus INPUT
Analysis of Variance for Day5

Source	DF	SS	MS	F	P
INPUT	10	58967	5897	30.26	0.000
Error	88	17149	195		
Total	98	76116			

One-way ANOVA: Day6 versus INPUT
Analysis of Variance for Day6

Source	DF	SS	MS	F	P
INPUT	10	39385	3938	21.20	0.000
Error	88	16346	186		
Total	98	55731			

One-way ANOVA: Day7 versus INPUT
Analysis of Variance for Day7

Source	DF	SS	MS	F	P
INPUT	10	23160	2316	14.00	0.000
Error	88	14561	165		
Total	98	37721			

One-way ANOVA: Day8 versus INPUT
Analysis of Variance for Day8

Source	DF	SS	MS	F	P
INPUT	10	4235	424	3.89	0.000
Error	88	9576	109		
Total	98	13811			

One-way ANOVA: Day9 versus INPUT
Analysis of Variance for Day9

Source	DF	SS	MS	F	P
INPUT	10	783.8	78.4	1.38	0.203
Error	88	5000.0	56.8		
Total	98	5783.8			

One-way ANOVA: Day10 versus INPUT
Analysis of Variance for Day10

Source	DF	SS	MS	F	P
INPUT	10	783.8	78.4	1.38	0.203
Error	88	5000.0	56.8		
Total	98	5783.8			

C1:- Growth data resulted with feed-Spirulina ratio and input rate treatments

Input Level	F:Sp Ratio	Replicates	Initial Wt	Day1	Day2	Day3
			Growth(g)			
20%	0:100	1	0.0104	0.024	0.027	0.029
		2	0.0104	0.0196	0.02425	0.027
		3	0.0104	0.02	0.0198	0.026
		4	0.0104	0.026667	0.023667	0.024
		5	0.0104	0.02325	0.025	0.0262
	25:75	1	0.0104	0.018	0.026	0.0265
		2	0.0104	0.0196	0.023333	0
		3	0.0104	0.024	0.021	0
		4	0.0104	0.028	0.023	0.027
		5	0.0104	0.0198	0.0192	0.022
	50:50	1	0.0104	0.028	0.021333	0
		2	0.0104	0	0	0
		3	0.0104	0	0	0
		4	0.0104	0	0	0
		5	0.0104	0	0	0
	75:25	1	0.0104	0	0	0
		2	0.0104	0	0	0
		3	0.0104	0	0	0
		4	0.0104	0	0	0
		5	0.0104	0	0	0
	100:0	1	0.0104	0	0	0
		2	0.0104	0	0	0
		3	0.0104	0	0	0
		4	0.0104	0	0	0
		5	0.0104	0	0	0
5%	0:100	1	0.0104	0.019	0.020667	0.021
		2	0.0104	0.0184	0.019667	0.02
		3	0.0104	0.02225	0.0205	0.021
		4	0.0104	0.0164	0.0215	0.022
		5	0.0104	0.0158	0.020333	0.021
	25:75	1	0.0104	0.0196	0.019	0.02
		2	0.0104	0.022	0.0215	0.022
		3	0.0104	0.0174	0.020667	0.022
		4	0.0104	0.019	0.020333	0.0205
		5	0.0104	0.018	0.0205	0.021
	50:50	1	0.0104	0.0178	0.020667	0.021
		2	0.0104	0.022	0.0205	0.022
		3	0.0104	0.019	0.018	0.02
		4	0.0104	0.021667	0.01975	0.022
		5	0.0104	0.026	0.01975	0
	75:25	1	0.0104	0.0192	0.016	0
		2	0.0104	0.0174	0.014	0
		3	0.0104	0.024	0.0185	0
		4	0.0104	0.0186	0.015	0
		5	0.0104	0.0198	0.0185	0.0198

		1	0.0104	0.0186	0.018	0
		2	0.0104	0.02	0.019	0
	100:0	3	0.0104	0.0172	0.014	0
		4	0.0104	0.02325	0.015	0
		5	0.0104	0.0184	0.019333	0
		1	0.0104	0.014	0.016	0
		2	0.0104	0.0134	0.014333	0.015667
0%	**0:00**	3	0.0104	0.0125	0.012	0
		4	0.0104	0.0112	0.012	0.013
		5	0.0104	0.010667	0.0115	0.0135

C 1.1. Average Growth Rate(g) as result of Input Manipulation

	Day		
Input Rate-Ration	1	2	3
20%-0:100	0.012303	0.00124	0.002497
20%-25:75	0.01148	0.000627	-0.00741
20%-50;50	-0.0048	-0.00133	-0.00427
20%-75:25	-0.0104	0	0
20%-100:0	-0.0104	0	0
5%-0:100	0.00797	0.002163	0.000467
5%-25:75	0.0088	0.0012	0.0007
5%-50:50	0.010893	-0.00156	-0.00273
5%-75:25	0.0094	-0.0034	-0.01244
5%-100:0	0.00909	-0.00242	-0.01707
0%-0:0	0.001953	0.000813	-0.00473

C2:- Mortality data resulted with feed-Spirulina ratio and input rate treatments

Input Level	F:Sp Ratio	Replicates	Mortality (%)									
			Day1	Day2	Day3	Day4	Day5	Day6	Day7	Day8	Day9	Day10
		1	0%	0%	0%	0%	0%	0%	40%	40%	40%	60%
		2	0%	0%	0%	0%	0%	0%	0%	20%	40%	40%
	0:100	3	0%	0%	0%	0%	0%	0%	0%	0%	40%	40%
		4	0%	0%	0%	0%	0%	0%	17%	50%	50%	67%
		5	0%	20%	20%	20%	20%	20%	60%	60%	80%	80%
		1	0%	0%	0%	0%	0%	0%	40%	60%	60%	60%
		2	0%	0%	0%	0%	0%	0%	20%	40%	100%	100%
	25:75	3	0%	0%	0%	0%	0%	0%	0%	80%	80%	100%
		4	0%	0%	0%	0%	0%	0%	40%	80%	80%	80%
		5	0%	0%	0%	0%	0%	0%	0%	0%	40%	60%
		1	0%	0%	0%	0%	0%	0%	40%	40%	100%	100%
		2	0%	40%	80%	80%	100%	100%	100%	100%	100%	100%
20%	50:50	3	17%	17%	50%	100%	100%	100%	100%	100%	100%	100%
		4	0%	40%	80%	80%	100%	100%	100%	100%	100%	100%
		5	0%	60%	60%	100%	100%	100%	100%	100%	100%	100%
		1	0%	0%	0%	80%	100%	100%	100%	100%	100%	100%
		2	40%	40%	100%	100%	100%	100%	100%	100%	100%	100%
	75:25	3	20%	20%	100%	100%	100%	100%	100%	100%	100%	100%
		4	0%	100%	100%	100%	100%	100%	100%	100%	100%	100%
		5	0%	0%	0%	60%	100%	100%	100%	100%	100%	100%
		1	20%	80%	100%	100%	100%	100%	100%	100%	100%	100%
		2	0%	100%	100%	100%	100%	100%	100%	100%	100%	100%
	100:0	3	40%	40%	80%	100%	100%	100%	100%	100%	100%	100%
		4	20%	80%	80%	100%	100%	100%	100%	100%	100%	100%
		5	60%	100%	100%	100%	100%	100%	100%	100%	100%	100%
5%		1	0%	20%	20%	20%	20%	20%	40%	40%	40%	40%
		2	0%	0%	0%	0%	0%	0%	0%	40%	40%	60%
	0:100	3	0%	0%	17%	17%	17%	33%	33%	33%	33%	33%
		4	0%	0%	0%	0%	0%	0%	20%	20%	60%	60%
		5	0%	0%	0%	0%	0%	0%	0%	40%	40%	40%
		1	0%	0%	0%	0%	0%	0%	0%	0%	40%	60%
		2	0%	0%	0%	0%	0%	0%	60%	60%	60%	60%
	25:75	3	0%	0%	0%	0%	0%	0%	0%	40%	80%	80%
		4	0%	0%	0%	0%	0%	0%	33%	50%	67%	67%
		5	0%	0%	0%	0%	0%	0%	60%	60%	60%	60%
		1	0%	0%	0%	0%	0%	0%	40%	40%	80%	80%
		2	0%	0%	0%	0%	0%	0%	0%	60%	80%	80%
	50:50	3	0%	0%	0%	0%	0%	0%	60%	80%	80%	80%
		4	0%	0%	0%	0%	0%	0%	33%	33%	83%	83%
		5	0%	0%	0%	0%	0%	0%	20%	20%	80%	100%
		1	0%	0%	0%	0%	0%	0%	40%	80%	100%	100%
		2	0%	0%	0%	0%	0%	0%	0%	80%	100%	100%
	75:25	3	0%	0%	0%	0%	0%	0%	60%	60%	100%	100%
		4	0%	0%	0%	0%	0%	0%	80%	80%	100%	100%
		5	0%	0%	0%	0%	0%	0%	20%	20%	80%	80%

		1	0%	0%	0%	0%	0%	0%	60%	60%	100%	100%
		2	17%	17%	17%	17%	17%	17%	50%	67%	100%	100%
	100:0	3	0%	0%	0%	0%	0%	0%	40%	80%	100%	100%
		4	0%	0%	0%	0%	20%	20%	80%	80%	80%	100%
		5	0%	0%	0%	0%	0%	0%	40%	40%	80%	100%
		1	0%	0%	0%	0%	0%	0%	60%	60%	100%	100%
		2	0%	0%	0%	0%	0%	0%	0%	40%	40%	40%
0%	**0:00**	3	0%	0%	0%	20%	20%	20%	20%	80%	80%	100%
		4	0%	0%	0%	0%	0%	0%	40%	40%	80%	80%
		5	20%	20%	40%	40%	40%	40%	60%	60%	60%	60%

C 2.1. Average Mortality(%) as result of Input Manipulation

Input Rate-Ration	Day									
	1	2	3	4	5	6	7	8	9	10
20%-0:100	0%	4%	4%	4%	4%	4%	23%	34%	50%	57%
20%-25:75	0%	0%	0%	0%	0%	0%	20%	52%	72%	80%
20%-50;50	3%	31%	54%	72%	80%	80%	88%	88%	100%	100%
20%-75:25	12%	32%	60%	88%	100%	100%	100%	100%	100%	100%
20%-100:0	28%	80%	92%	100%	100%	100%	100%	100%	100%	100%
5%-0:100	0%	4%	7%	7%	7%	11%	19%	35%	43%	47%
5%-25:75	0%	0%	0%	0%	0%	0%	31%	42%	61%	65%
5%-50:50	0%	0%	0%	0%	0%	0%	31%	47%	81%	85%
5%-75:25	0%	0%	0%	0%	0%	0%	40%	64%	96%	96%
5%-100:0	3%	3%	3%	3%	7%	7%	54%	65%	92%	100%
0%-0:0	4%	4%	8%	12%	12%	12%	36%	56%	72%	76%

C3:- ANOVA summary of feed-Spirulina ratio and input rate effects on growth

Two Way Analysis of Variance of Growth Rate Data

Two-way ANOVA: Day1 versus At Level %, F:Sp

Analysis of Variance for Gr 1

Source	DF	SS	MS	F	P
At Level	2	0.0012189	0.0006095	12.12	0.000
F:Sp	4	0.0000683	0.0000171	0.34	0.850
Error	68	0.0034190	0.0000503		
Total	74	0.0047062			

Two-way ANOVA: Day2 versus At Level %, F:Sp

Analysis of Variance for Day2

Source	DF	SS	MS	F	P
At Level	2	0.0000353	0.0000176	2.95	0.059
F:Sp	4	0.0000417	0.0000104	1.74	0.151
Error	68	0.0004070	0.0000060		
Total	74	0.0004840			

Two-way ANOVA: Day3 versus At Level %, F:Sp

Analysis of Variance for Day3

Source	DF	SS	MS	F	P
At Level	2	0.0002527	0.0001263	2.08	0.133
F:Sp	4	0.0006887	0.0001722	2.83	0.031
Error	68	0.0041353	0.0000608		
Total	74	0.0050767			

One Way Analysis of Variance of Growth Rate Data

One-way ANOVA: Day1 versus At Level %

Analysis of Variance for Day1

Source	DF	SS	MS	F	P
At Level	2	0.0012189	0.0006095	12.58	0.000
Error	72	0.0034873	0.0000484		
Total	74	0.0047062			

One-way ANOVA: Day2 versus At Level %

Analysis of Variance for Day2

Source	DF	SS	MS	F	P
At Level	2	0.0000353	0.0000176	2.83	0.066
Error	72	0.0004487	0.0000062		
Total	74	0.0004840			

One-way ANOVA: Day3 versus At Level %

Analysis of Variance for Day3

Source	DF	SS	MS	F	P
At Level	2	0.0002527	0.0001263	1.89	0.159
Error	72	0.0048240	0.0000670		
Total	74	0.0050767			

One Way Analysis of Variance of Growth Rate Data

One-way ANOVA: Day1 versus F:Sp

Analysis of Variance for Day1

Source	DF	SS	MS	F	P
F:Sp	4	0.0000683	0.0000171	0.26	0.904
Error	70	0.0046379	0.0000663		
Total	74	0.0047062			

One-way ANOVA: Day2 versus F:Sp

Analysis of Variance for Day2

Source	DF	SS	MS	F	P
F:Sp	4	0.0000417	0.0000104	1.65	0.171
Error	70	0.0004423	0.0000063		
Total	74	0.0004840			

One-way ANOVA: Day3 versus F:Sp

Analysis of Variance for Day3

Source	DF	SS	MS	F	P
F:Sp	4	0.0006887	0.0001722	2.75	0.035
Error	70	0.0043880	0.0000627		
Total	74	0.0050767			

C4:- ANOVA summary of feed-Spirulina ration and input rate effects on mortality

Two Way Analysis of Variance of Mortality Data

Two-way ANOVA: Day1 versus At Level %, F:Sp

Analysis of Variance for Day1

Source	DF	SS	MS	F	P
At Level	2	808	404	3.71	0.030
F:Sp	4	884	221	2.03	0.100
Error	68	7398	109		
Total	74	9091			

Two-way ANOVA: Day2 versus At Level %, F:Sp

Analysis of Variance for Day2

Source	DF	SS	MS	F	P
At Level	2	11997	5998	12.56	0.000
F:Sp	4	1918	479	1.00	0.412
Error	68	32479	478		
Total	74	46393			

Two-way ANOVA: Day3 versus At Level %, F:Sp

Analysis of Variance for Day3

Source	DF	SS	MS	F	P
At Level	2	23144	11572	14.88	0.000
F:Sp	4	2369	592	0.76	0.554
Error	68	52892	778		
Total	74	78406			

Two-way ANOVA: Day4 versus At Level %, F:Sp

Analysis of Variance for Day4

Source	DF	SS	MS	F	P
At Level	2	36049	18024	21.35	0.000
F:Sp	4	3363	841	1.00	0.416
Error	68	57403	844		
Total	74	96814			

Two-way ANOVA: Day5 versus At Level %, F:Sp

Analysis of Variance for Day5

Source	DF	SS	MS	F	P
At Level	2	41563	20781	22.13	0.000
F:Sp	4	3459	865	0.92	0.457
Error	68	63844	939		
Total	74	108866			

Two-way ANOVA: Day6 versus At Level %, F:Sp

Analysis of Variance for Day6

Source	DF	SS	MS	F	P
At Level	2	40899	20449	21.59	0.000
F:Sp	4	3597	899	0.95	0.441
Error	68	64401	947		
Total	74	108897			

Two-way ANOVA: Day7 versus At Level %, F:Sp

Analysis of Variance for Day7

Source	DF	SS	MS	F	P
At Level	2	15933	7966	8.68	0.000
F:Sp	4	7591	1898	2.07	0.095
Error	68	62433	918		
Total	74	85956			

Two-way ANOVA: Day8 versus At Level %, F:Sp

Analysis of Variance for Day8

Source	DF	SS	MS	F	P
At Level	2	8108	4054	6.33	0.003
F:Sp	4	2403	601	0.94	0.448
Error	68	43581	641		
Total	74	54092			

Two-way ANOVA: Day9 versus At Level %, F:Sp

Analysis of Variance for Day9

Source	DF	SS	MS	F	P
At Level	2	2148	1074	2.21	0.117
F:Sp	4	3058	764	1.58	0.191
Error	68	32995	485		
Total	74	38200			

Two-way ANOVA: Day10 versus At Level %, F:Sp

Analysis of Variance for Day10

Source	DF	SS	MS	F	P
At Level	2	1820	910	2.00	0.143
F:Sp	4	4163	1041	2.29	0.069
Error	68	30940	455		
Total	74	36923			

One Way Analysis of Variance of Mortality Data

One-way ANOVA: Day1 versus At Level %

Analysis of Variance for Day1

Source	DF	SS	MS	F	P
At Level	2	808	404	3.51	0.035
Error	72	8283	115		
Total	74	9091			

One-way ANOVA: Day2 versus At Level %

Analysis of Variance for Day2

Source	DF	SS	MS	F	P
At Level	2	11997	5998	12.56	0.000
Error	72	34396	478		
Total	74	46393			

One-way ANOVA: Day3 versus At Level %

Analysis of Variance for Day3

Source	DF	SS	MS	F	P
At Level	2	23144	11572	15.08	0.000
Error	72	55261	768		
Total	74	78406			

One-way ANOVA: Day4 versus At Level %

Analysis of Variance for Day4

Source	DF	SS	MS	F	P
At Level	2	36049	18024	21.36	0.000
Error	72	60765	844		
Total	74	96814			

One-way ANOVA: Day5 versus At Level %

Analysis of Variance for Day5

Source	DF	SS	MS	F	P
At Level	2	41563	20781	22.23	0.000
Error	72	67303	935		
Total	74	108866			

One-way ANOVA: Day6 versus At Level %

Analysis of Variance for Day6

Source	DF	SS	MS	F	P
At Level	2	40899	20449	21.65	0.000
Error	72	67998	944		
Total	74	108897			

One-way ANOVA: Day7 versus At Level %

Analysis of Variance for Day7

Source	DF	SS	MS	F	P
At Level	2	15933	7966	8.19	0.001
Error	72	70024	973		
Total	74	85956			

One-way ANOVA: Day8 versus At Level %

Analysis of Variance for Day8

Source	DF	SS	MS	F	P
At Level	2	8108	4054	6.35	0.003
Error	72	45984	639		
Total	74	54092			

One-way ANOVA: Day9 versus At Level %

Analysis of Variance for Day9

Source	DF	SS	MS	F	P
At Level	2	2148	1074	2.14	0.125
Error	72	36052	501		
Total	74	38200			

One-way ANOVA: Day10 versus At Level %

Analysis of Variance for Day10

Source	DF	SS	MS	F	P
At Level	2	1820	910	1.87	0.162
Error	72	35102	488		
Total	74	36923			