

Growth of juvenile abalone under aquaculture conditions

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I declare that

“Growth of juvenile abalone under aquaculture conditions”

is my own work, that it has not been submitted for any degree or examination in any other university, and that all the sources I have used or quoted have been indicated and acknowledged by complete references.



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1.1 Aquaculture

Aquaculture is the aquatic counterpart of agriculture (Reay 1979, Kautsky *et al.* 2001) used to refer to the farming of aquatic plants and animals (De Silva and Anderson 1994, Middleden and Redding 1998). This implies that the natural ecosystem processes are manipulated and modified in order to favor the production of selected species (Folke and Kautsky 1989). The term 'mariculture' has often been interchanged with aquaculture (Reay 1979). Mariculture may be more specifically defined as the cultivation, management and harvesting of marine organisms in their natural habitat on specially constructed channels or tanks within a controlled environment (Lincoln *et al.* 1998).

Aquaculture originated some 4000 years ago in the Asian region, mainly by the Chinese (Reay 1979, Hecht and Britz 1990, Joseph 1998); it was initiated firstly through the use of freshwater aquaculture before the more recent practice of marine aquaculture developed (Lee 1998). The practice of aquaculture has since increased (Kautsky *et al.* 1997, 2001, Qian 2001) by almost five times the global population rate between the years 1980 and 1990 (Lee 1998). Troell *et al.* (2004) suggests that currently, aquaculture accounts for approximately 30% of total fish food production globally and is also growing rapidly at a rate of about 4-11% annually (Troell *et al.* 1999).

The main reasons behind the rapid increase in aquaculture are as a result of an increase in direct human consumption. Aquaculture has thus been regarded as a reliable means of increasing food security for people worldwide (Reay 1979, Naylor *et al.* 2000, Bryceson 2002). Decreased global commercial yields from wild catches (Kautsky *et al.* 1997), as well as the high market demand for aquaculture products, have intensified the practice of aquaculture (Tseng and Fei 1987, Jensen 1993, Bautista-Teruel and Millamena 1999,

Bautista-Teruel 2002). Furthermore aquaculture has also increased as a result of increased biological research (Reay 1979) and increased seed supply for fishery enhancement (Nelson *et al.* 2002).

The success and optimal growth of species in an aquaculture system is influenced by numerous external factors. Such factors include water flow (Kautsky and Folke 1989), salinity (Langdon *et al.* 2004), the tank system and stocking density (Gowen *et al.* 1990), water temperature (Nelson *et al.* 2002, Steinarsson and Imsland 2003, Langdon *et al.* 2004), as well as food quality and quantity (Gowen *et al.* 1990, Bautista-Teruel *et al.* 2002).

1.2 Types of Aquaculture

Aquaculture is a diverse activity whereby the degree of intensity, through intervention and manipulation of the cultured animals' life cycle, is determined by the purpose and scale at which organisms are cultured (Kautsky *et al.* 2000). This degree of manipulation may either be extensive, semi-intensive, or intensive (Naylor *et al.* 2000, Troell *et al.* 2004). Extensive aquaculture is traditional farming (Troell *et al.* 1999) through maintaining the natural processes (Kautsky and Folke 1989, Troell *et al.* 2004) whereby there is little or no manipulation of the natural ecosystem (Folke and Kautsky 1989). Such farming is usually practiced in small-scales, mainly performed by family members and relatives (Furey *et al.* 2003, Nils Kautsky, pers. comm.) and involves mainly the exclusion of predators and reduction of competitors for the cultured organism (Naylor *et al.* 2000).

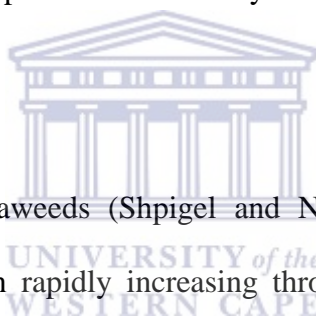
Semi-intensive aquaculture is a transition between extensive aquaculture and intensive aquaculture (Furey *et al.* 2003). In semi-intensive aquaculture there is an inclusion of external inputs such as an enhancement of food supply added to the aquaculture system (Naylor *et al.*

2000). This type of aquaculture differs from extensive aquaculture in that the natural food is enhanced by fertilization (Kautsky *et al.* 2001). It differs from intensive aquaculture in that there is lower production and less technology involved (Furey *et al.* 2003). Semi-intensive aquaculture is seen as a throughput system (Furey *et al.* 2003) since feed may be added to the system resulting in accumulation of wastes (Troell *et al.* 1999).

Intensive aquaculture relies on auxiliary inputs in order to ensure the optimal growth conditions for the cultured organism(s) (Folke and Kautsky 1989). Such inputs involve the addition of nutritionally balanced diets, chemicals, as well as increasing the organism density in each of the culture tanks (Naylor *et al.* 1998, 2000). Intensive aquaculture uses formulated feeds (Kautsky *et al.* 2001) of which the cultured organism(s) consume only a small proportion while the rest is discharged to the surrounding environment (Folke *et al.* 1997). Approximately 25%-30% of the supplied feed is consumed while about 70% of it is discharged to the environment (Folke and Kautsky 1989, Kautsky and Folke 1989). Intensive aquaculture, therefore, have much more dramatic effects on the local natural enrichment than either extensive or semi-intensive aquaculture.

Types of aquaculture include monoculture, co-culture and polyculture. Monoculture is a traditional practice (Folke *et al.* 1997) whereby a single species or crop is cultured, sometimes year after year without rotation (Kautsky and Folke 1989). This results in considerable environmental impacts, such as eutrophication and development of harmful algal blooms (Gowen *et al.* 1990). The culture of a single species in monoculture may also reduce the resilience of the system thus making it more susceptible to pests and disease. This, in turn, may result in species with little genetic variation and thus most vulnerable to catastrophic epidemics.

The terms polyculture and co-culture have been interchanged to refer to the culture of more than one species in a single culture system. Polyculture is a traditional aquaculture system that mainly aims to increase crop diversity within the farmed area (Neori *et al.* 2004). In such a system there does not necessarily have to be a mutually beneficial process between the cultured species, as is the case with co-culture (Langdon *et al.* 2004). According to Langdon *et al.* (2004), co-culture occurs when two or more organisms are grown together in the same culture medium and depends on selection of component organisms that are complementary, resulting in processes that are mutually beneficial to all the co-cultured organisms. Co-culture systems promote nutrient cycling within a system, since waste of one component is utilized by another to enhance the overall production of the system (Kautsky *et al.* 2001, Langdon *et al.* 2004, Neori *et al.* 2004).



Co-culture uses animals and seaweeds (Shpigel and Neori 1996), and as a result, the cultivation of seaweeds has been rapidly increasing throughout the world (Stirk and van Staden 2004). Many nations, globally, consume seaweeds as a staple diet. The decline and inability of natural stocks to supply the growing human demands has been cited as the driving force behind increased seaweed cultivation (Molloy and Bolton 1996, Troell *et al.* 1997). Seaweeds are cultured for abalone feed (Wakibia *et al.* 2001), for agar [alginates and carrageenans] (Rebello *et al.* 1996, Neori *et al.* 2004) and also for soil fertilizers and animal fodder (Stirk and van Staden 2004). Cultivated seaweeds include the gracilarioids [*Gracilaria gracilis* (Anderson *et al.* 1996, Rebello *et al.* 1996, Wakibia *et al.* 2001), *G. chilensis* (Buschmann *et al.* 1996, Troell *et al.* 1997) and *G. lemaneiformis* (Neori *et al.* 2004)]. *Ulva* species [*U. lactuca* (Schuenhoff *et al.* 2003) and *U. rigida* (Boarder and Shpigel 2001)] as

well as *Laminaria japonica*, *Undaria pinnatifida* (Tseng and Fei 1987, Wikfors and Ohno 2001) and *Palmaria mollis* (Langdon *et al.* 2004) are other cultivated seaweeds.

Animals used in aquaculture include crustaceans, finfish, and mollusks (Reay 1979, De Silva 1998). Crustacean culture is dominated with shrimps (De Silva 1998, Neori *et al.* 2004), finfish by milkfish, trout, and carp (Reay 1979, Troell *et al.* 1997, 1999) while mollusk culture has been dominated by oysters, clams, and scallops (Joseph 1998). Abalone have only recently been recognized as a group with great potential to the aquaculture industry (Naylor *et al.* 2000) and approximately 15 abalone species are globally exploited for commercial purposes (Bester *et al.* 2004, Sales and Janssens 2004).

1.3 Abalone and their culture

Abalone are marine gastropods that belong to the family *Haliotidae* of the phylum Mollusca (Barkai and Griffiths 1986, Fallu 1991, White 1995, Huchette *et al.* 2003). They bear only a single shell, which reflects an ancient evolutionary trait, characteristic of the subclass *Archaeogastropoda* (Barkai and Griffiths 1986, Mai *et al.* 2001, Sale and Britz 2002, Galindo *et al.* 2003). While most authors believe that there are approximately one hundred (100) *Haliotis* species globally (Hahn 1989, Fallu 1991, Simpson 1994, Lyon 1995, Elliott 2000), it has been reported that some of these are subspecies (White 1995). The latest figures are that there are currently about 90 *Haliotis* species worldwide (Sales and Britz 2001, Bester *et al.* 2004, Sales and Janssens 2004).

The genus *Haliotis* has a worldwide distribution (Bester *et al.* 2004, Najmudeen and Victor 2004), found mainly along rocky shores of almost all continental shelves (Hahn 1989). They are usually found between the intertidal and the littoral zone (Hecht 1994, Knauer 1994,

White 1995). Factors such as seawater temperature (Steinarsson and Imsland 2003), type of substratum (de Waal *et al.* 2003) and food availability (Hecht and Britz 1990, Cook 1991) may contribute in determining the distribution of abalone.

Temperate abalone species are generally larger compared to tropical species (Jarayabhand and Paphavasit 1996). This has been attributed to a number of factors. Not only does colder, temperate water have a relatively high primary production rate compared to warm nutrient-poor tropical water (Hecht and Britz 1990), but the relatively high abundance of food and oxygen in temperate water is assumed as the main factors resulting in the larger temperate abalone. Abalone have also been observed to have some digestive problems in warmer water (Fleming *et al.* 1996) and this could also explain the smaller abalone in tropical waters. However, such digestive problems have been observed mainly when abalone are fed artificial diets at high temperatures (Kevin Ruck, pers. comm.). The suggested reason is that warmer water increase fermentation within the abalone (Fleming *et al.* 1996) resulting in a condition referred to as bloating (Macey and Coyne 2005).

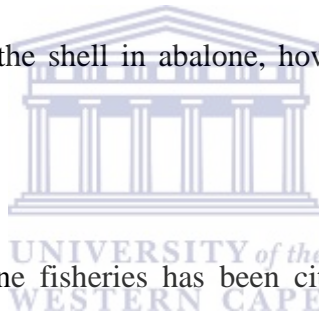
Abalone are herbivorous invertebrates that naturally feed on macroalgae (Elliott 2000, Sales and Britz 2001, Nelson *et al.* 2002, Tanaka *et al.* 2003) and are sometimes even referred to as algivorous animals (Shpigel *et al.* 1999, Mai *et al.* 2001). The macroalgal preference of different abalone species varies worldwide depending on habitat and the availability of macroalgal species (Dunstan *et al.* 1996, Nelson *et al.* 2002). Although a variety of macroalgae are naturally consumed (Britz 1991), kelp, by far, forms the primary food source for these herbivores (Hahn 1989, Rosen *et al.* 2000).

The natural feeding patterns of abalone change during different stages of their life cycle (de Waal *et al.* 2003). This is attributed not only to the increased mouth size (Fleming *et al.* 1996), but also to morphological changes of the radula as the abalone grows (Kawamura *et al.* 2001, Daume and Ryan 2004, Onitsuka *et al.* 2004). Changes in diet could also be due to transformations in the gut micro-organisms, such as bacteria and enzymes within the digestive system, of abalone as the abalone grows and thus enabling them to digest macroalgae (Erasmus *et al.* 1997, Tanaka *et al.* 2003).

At the larval stage, abalone feed mainly on yolk (Fallu 1991). Post-larval juveniles prefer to consume benthic diatoms (Fleming *et al.* 1996, Knauer *et al.* 1996, Najmudeen and Victor 2004) and crustose coralline red algae (White 1995, Dunstan *et al.* 1996, de Waal *et al.* 2003) while juvenile abalone feed on microscopic algae (de Waal *et al.* 2003). As the juveniles grow up, they are weaned onto macroalgae (Cook 1991, Dunstan *et al.* 1996). Weaning is a process whereby live food organisms are replaced with formulated diets (Southgate and Partridge 1998), and in the aquaculture industry it is the juvenile stage that is weaned onto formulated (artificial) feeds.

The juvenile abalone are weaned onto formulated feeds in order to achieve higher growth rates, which would be difficult to achieve when feeding on macroalgae (Evans and Langdon 2000). This is because of the low protein composition of marine algae, approximately 10-20% of dry weight, that is inadequate to fulfill the protein requirements of abalone (Fleming *et al.* 1996, Johnston *et al.* 2005). Kelp, for example, has a relatively low nutritional value (protein ca 15%) and an unbalanced amino acid profile (Simpson 1994, Britz 1996a, Erasmus *et al.* 1997, Rosen *et al.* 2000). Wild *Ulva lactuca* also has a low protein content ranging from approximately 3.7-19.9% (Robertson-Anderson 2003, Naidoo *et al.* 2005).

The relatively slow growth rate of abalone fed on macroalgae (Britz 1996a, Cook 1991, Bautista-Teruel *et al.* 1999, Tan and Mai 2001, Muriyama and Kawauchi 2004) has prompted the development and usage of formulated feeds in the aquaculture industry (Britz 1996a, b, Sale and Britz 2001). Southgate and Partridge (1998) classify these feeds as either microencapsulated diets (with ingredients enclosed in a capsule or membrane) or microbound diets (with ingredients held in a matrix or binder). These feeds are formulated so as to fulfil the nutritional requirements of the abalone. Such requirements include carbohydrates (Nelson *et al.* 2002) and protein (Fleming *et al.* 1996, Guzman and Viana 1998) for tissue growth, and lipids as a source of energy (Durazo-Beltran *et al.* 2003) and the regeneration of damaged shells (Nelson *et al.* 2002). Tan and Mai (2001) even suggested that vitamin K possibly contributed to mineralization of the shell in abalone, however, more studies are needed to confirm this.



The rapid decline in wild abalone fisheries has been cited as one of the reasons for the increased practice of abalone aquaculture (Gordon and Cook 2001, de Waal *et al.* 2003). However, the major reason behind the rapid increase in abalone aquaculture is the lucrative market value for their large adductor muscle or foot (Elliott 2000). Although abalone are sold either live, frozen, canned or in dried form, abalone shells are also sold mainly for decoration purposes (Gordon and Cook 2001). The major market for abalone is concentrated in the Far East, although Mexico, USA and Europe also do consume abalone (Oakes and Ponte 1996, Robertson-Anderson 2003).

Successful and lucrative as it may seem, abalone aquaculture is facing numerous challenges ranging from biological, environmental and economical implications. The biological

implications include the spreading of diseases in and around the industry. Environmental issues include ecological impacts through waste discharge and biological pollution to the surrounding environment. Economic challenges are mainly due to financial expenses of which abalone feed accounts most.

The major biological challenge facing the abalone industry is pest control. Abalone shells are susceptible to infestation by sabellid polychaetes (Culver and Kuris 1997, Leighton 1997, Colorni 1998, Finley *et al.* 2000), shell boring sponges (Oakes and Fields 1996) and clams, (Alvarez-Tinajero *et al.* 2000). According to Oakes and Fields (1996), older abalone are more likely to be infested by the sabellids than are the younger ones. This is because smaller abalone grow faster than older ones and are therefore able to produce more shell layers that prevent the spread of the polychaete larvae. Shell infestation usually leads to shell deformation and this results in reduced growth in the abalone (Ruck and Cook 1997, Day *et al.* 2000). Oakes and Fields (1996) attribute the reduced growth to the thickening and downward growth of the leading edge of the infested shell. However, Colorni (1998) suggests that the reduced growth in infested abalone could be due to the abalones investing more energy in shell repairing than they normally do. The impact of the sabellids in the South African abalone (*Haliotis midae*) is likely to be significant only in cultured abalone (Ruck and Cook 1997, Ruck *et al.* 1997). This could be due to the feeding of farmed abalone on the high protein formulated feed Abfeed® resulting in relatively high nitrogenous waste that is conducive to polychaete generation (Simon *et al.* 2004). However, in California, where the sabellids were accidentally introduced from South Africa (Culver and Kuris 1997, Day 2000, Finley *et al.* 2000), numerous fatalities of the Californian red abalone (*H. rufescens*) have been associated with sabellid infestation.

Different treatment measures have been attempted as means of eradicating or controlling the spread of sabellid polychaetes. Oakes and Fields (1996) suggested the use of food poisoning and the development of means to interfere with the sabellids' reproductive cycle. This led to the test of toxin-containing microcapsules such as the microencapsulated liposomes (Fields 1997, Ruck and Cook 1997, Ruck *et al.* 1997). Leighton (1997) observed that exposing infested abalones to temperatures near but below their upper thermal limits, killed all life stages of the sabellids. This may be due to the fact that the life cycle and generation time of the sabellids are highly temperature dependant (Finley *et al.* 2000). Culver and Kuris (1997) suggested isolation of infested stocks as a means to prevent the transfer of shell infestation from infested abalones to uninfected abalone.

However, relatively little success has been achieved with the use of the above-mentioned treatments as a means of controlling the sabellid polychaetes infestation (Culver & Kuris 1997, Shields *et al.* 1997). This is because some of the treatments used tended to also affect the host abalone (Fields 1997). A treatment that would be able to prevent the rapid spread and self-fertilization of the sabellids, without harming the host abalone, would thus be the ideal treatment for such polychaetes. Thus far, isolation of infested abalone stocks seems to be the most popular means employed to prevent the spread of these polychaetes.

Yet another biological challenge facing the abalone industry is Withering Syndrome (WS). WS is a disease found in both terrestrial animals (such as cows and rodents) and marine animals (Friedman *et al.* 2003). In abalone, WS has been described as a chronic (Friedman *et al.* 2000b), progressive (Moore *et al.* 2000, Braid *et al.* 2005), epizotic (Gardner *et al.* 1995) disease affecting both wild and cultured abalone (Friedman *et al.* 2000a, 2003). The impact of this disease has been catastrophic in the California black abalone, *Haliotis rocherodii*,

since its first detection in the mid-80s (Ruediger *et al.* 1997, Shields *et al.* 1997, Taniguchi *et al.* 1997, Moore *et al.* 2000, Friedman *et al.* 2003).

WS is caused by a rickettsia-like prokaryote (RLP) bacterium that has been identified as *Canidatus xenohaliotis californiensis* (Finley and Friedman 2000, Friedman *et al.* 2000a, b, Moore *et al.* 2000, Friedman *et al.* 2003, Braid *et al.* 2005). The RLP bacterium lines and infects the epithelial cells of the digestive tract of the host abalone (Caceres-Matinez *et al.* 2000, Gardner *et al.* 1995). Such an infection results in the degeneration of the digestive gland (Friedman *et al.* 1997, 2000b, 2003, Gardner 1995) and this leads to reduced appetite and metabolic efficiency, causing starvation that probably leads to death (Shields *et al.* 1997). Friedman *et al.* (1997, 2003), Moore *et al.* (2000) and Braid *et al.* (2005) have observed that elevated water temperatures tend to enhance the expression of WS in abalone.

Friedman *et al.* (2000a) suggested the use of Oxytetracycline (OTC) as a treatment for WS infected abalone. In a follow-up research, Friedman *et al.* (2003) found that oral and injection administration of OTC increase the survival rate of WS infected abalone. However, persistence of OTC residues in the gut of treated abalone (Friedman *et al.* 2003) may affect consumers of abalone and thus hinder OTC as an effective treatment for WS. Reducing the water temperature has instead been suggested as an effective treatment that reduces the expression of WS in infected abalones (Friedman *et al.* 1997). However, reducing water temperatures may have a negative effect on some abalone species that may have higher optimal temperature ranges.

Aquaculture, in general, contributes to the degradation of the surrounding environment (Troell *et al.* 1997, Kautsky *et al.* 2000). This is because most aquaculture systems are

throughput systems (Folke *et al.* 1994) whereby resources that were introduced to the system are later released to the environment as waste discharge (Gowen *et al.* 1990, Folke *et al.* 1994, 1997). Such discharge waste may include chemicals, microorganisms, as well as parasites (Kautsky *et al.* 2001). However, the most common form of aquaculture waste generated may either be dissolved or suspended particulate matter (Troell *et al.* 1999). While dissolved waste includes the release of nitrogen and phosphorus from abalone faeces (Troell *et al.* 1997, Kautsky *et al.* 2000), particulate wastes include any organic matter (Folke *et al.* 1994) such as excretory materials (Gowen *et al.* 1990) and uneaten food pellets (Kautsky *et al.* 2000). The release of aquaculture wastes results in changes to the surrounding ecosystem through e.g. water eutrophication (Folke and Kautsky 1992, Folke *et al.* 1997). This may, in turn, promote the occurrences of harmful algal blooms (HABs) that are increasingly threatening the aquaculture industry worldwide (Buschmann *et al.* 1996, Botes *et al.* 2003).

Integrated aquaculture has therefore been proposed and is regarded as an environmentally friendly means of recycling aquaculture wastes (Troell *et al.* 1997, 1999). Integrated aquaculture may be vaguely defined as the cultivation and rearing of plants and animals from both the aquatic and terrestrial environments (Max Troell pers. comm.). This implies that one can culture aquatic plants with terrestrial animals, terrestrial plants with aquatic plants, and/or aquatic plants with aquatic animals. This system tries to mimic the natural ecosystem functions, whereby an output from one subsystem, which may have been wasted, becomes input to another subsystem resulting in a greater efficiency of output (Neori *et al.* 2004). Integrated aquaculture utilizes other cultured organisms (e.g. seaweeds, filter feeding mussels, oysters) to serve as biofilters (Neori *et al.* 2004).

However, aquaculture has not successfully managed to reduce pressure on wild fisheries stocks but only diverted the fish consumption from humans, to aquaculture (Folke *et al.* 1998). Most animals, including abalone, require high levels of protein in their diets so as to achieve high growth rates and the main source of protein used is fishmeal (Folke *et al.* 1998, Guzman and Viana 1998). The high demand for fishmeal (Folke *et al.* 1998) and fish oil extracts (Kautsky *et al.* 2001) as a protein source puts pressure on the wild fisheries (Larsson *et al.* 1994) as more fish are captured in the wild to support the ever-growing demand for fishmeal (Naylor *et al.* 2000). Such fish are usually the smaller pelagic fish stocks, which are regarded as of low or no economic value (Kautsky *et al.* 2001). Presently 2-2.8 Kg wild pelagic fish is needed to feed and produce 1Kg of cultured salmon, trout or marine shrimp while approximately 4.5Kg of wild pelagic fish is required to feed to produce 1Kg of marine finfish and eel (Kautsky *et al.* 2001). This in turn reduces the availability of food to marine predators who rely on these pelagic fishes for food (Naylor *et al.* 2000). As a result, changes in the structure of the food web will no doubt lead to a reduction in biodiversity and aquatic productivity.

Fishmeal-based feed is the most expensive component of the aquaculture industry (Britz *et al.* 1997, Sales and Janssens 2004). Finding an alternative protein source(s) is therefore one of the industry's major challenges (Kautsky *et al.* 1997). Extensive researches on replacement of fishmeal protein (e.g. Britz 1996b, Shipton and Britz 2001, Kratrachue *et al.* 2004) have been conducted thus far. Casein, soybean (Guzman and Viana 1998, Kratrachue *et al.* 2004) and sunflower meal (Shipton and Britz 2001) have been found to be suitable protein sources for abalone. However, casein is too expensive while the other sources of protein generally have to be used in combination with fishmeal or other protein sources (Guzman and Viana 1998).

1.4 Statement of the problem

Numerous research conducted thus far have clearly demonstrated the significant role played by formulated feeds in enhancing growth of cultured abalone (Fleming *et al.* 1996, Sales and Britz 2001). Although fresh seaweed have been found to be good supplementary diets to formulated feeds (e.g. Stepto and Cook 1996, Simpson and Cook 1998, Shpigel *et al.* 1999, Naidoo *et al.* in press.), most of this research has been focused on older, grow-out abalone. Very few studies (e.g. Britz 1996a, Knauer *et al.* 1996) have been conducted on post-weaning *H. midae*, and such studies have generally compared growth of abalone fed either fresh seaweed or formulated feeds. Thus far, insufficient research has been devoted to the role of formulated feeds fortified with fresh seaweed.

Furthermore, previous research has studied the growth of cultured organisms in various systems such as ponds (e.g. Azim *et al.* 2004, Lamoureux *et al.* 2005), cages (e.g. Fredriksson *et al.* 2004, Chambers and Ernst 2005), on longline ropes (e.g. Duckworth and Battershill 2003) and in tanks (e.g. Summerfelt and Pene 2005). However, no research has been devoted to simultaneously comparing different water flow-systems. Also, since most farmers tend to use their own conventional culture baskets, no studies have been undertaken to compare various basket designs and test how they affect growth of cultured abalone. Recently, there has been a call by the Marine and Coastal Management branch of the Department of Environmental Affairs and Tourism, in a frontier programme for mariculture research and development, to conduct research on optimization of abalone tank and basket design in farms.

1.5 Aims of study

Chapter 1: General Introduction

The aim of this research was two fold. The first aim was to test the suitability of different seaweeds and formulated feeds as food for post-weaning juvenile South African abalone (*Haliotis midae* Linnaeus). The second aim was to test the effects of basket design on the growth of grow-out juvenile abalone in both flow-through and re-circulation systems.



CHAPTER 2

The growth of post-weaning abalone (*Haliotis midae* Linnaeus) on various formulated feeds fortified with fresh wild seaweed

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2.1 Abstract

The effects of five formulated (artificial) feeds fortified with fresh, wild seaweed on the growth of post-weaning juvenile abalone, *Haliotis midae* (Linnaeus) were investigated. Growth was monitored on a commercial abalone farm over a period of 11-months in an experiment consisting of 11 treatments with 2 replicates (n = 50 individuals per replicate). The first 5 diet treatments comprised the fishmeal-based protein feeds Abfeed, Adam & Amos 'a', Adam & Amos 'b' and Adam & Amos 'c', and an all-seaweed-based protein FeedX. An additional 5 diet treatments comprised the formulated feeds above, all fortified with fresh, wild seaweeds (the kelp, *Ecklonia maxima* - ca 15% protein and *Ulva lactuca* - 3.7-19.9% protein). The final diet comprised a mixture of seaweeds (*Ulva*, kelp and red seaweeds); this diet was incorporated so as to compensate for nutrients that may be lacking in a single seaweed diet. Results showed that all fishmeal-based protein feeds performed better than the all-seaweed-based protein feed, FeedX (0.80 ± 0.02 BW $\% \cdot \text{day}^{-1}$ SGR; 0.864 g $\cdot \text{mm}^{-1}$ final CF). Abfeed (1.00 ± 0.02 BW $\% \cdot \text{day}^{-1}$ SGR; 1.312 g $\cdot \text{mm}^{-1}$ final CF) performed best of all formulated feeds. Fortification with fresh, wild seaweed significantly improved growth of all abalone. Even more striking was that the condition factors of those feeds that performed particularly poorly in the growth trials were dramatically increased by fortification. In contrast to a recent study, the mixed diet performed poorly against most of the feeds tested. Possible reasons for this are discussed. This study showed that feeding trials based solely on formulated feeds would benefit greatly by the incorporation of fresh seaweed into the diet.

Key words: Abfeed, diet, formulated feed, fortification, growth, *Haliotis midae*, seaweed.

2.2 Introduction

While the abalone fishery in South Africa dates back to 1949 (Tarr 1992), the history of abalone aquaculture is relatively recent with major developments only occurring during the 1990s (Sales and Britz 2001, Simon *et al.* 2004). The successful spawning of *Haliotis midae* in the late 1980s marked the inception of abalone farming in South Africa (Hecht and Britz 1990, Cook 1998). Currently there are approximately twelve (12) abalone farms operating in South Africa (Sales and Britz 2001; Simon *et al.* 2004), with the industry expected to expand (Tony Bennet, Abalone Farmers Association of Southern Africa, pers. comm.).

Of the six (6) *Haliotis* species endemic to Southern African waters (Barkai and Griffiths 1986, Hahn 1989; Hecht 1994), only one (*Haliotis midae*) is exploited commercially (Lyon 1995, Sales and Britz 2001). The relatively large size of *H. midae* makes this species financially lucrative (Oakes and Ponte 1996) with current international market prices ranging between US\$34 - US\$36 per Kg (Macey and Coyne 2005). By far the largest current export market for *H. midae* is concentrated in the Far East where it is treated as part of traditional cuisine and ceremony (Lyon 1995, Sales and Britz 2001).

However, abalone farming is both expensive and time consuming since *Haliotis* species are relatively slow growing (Britz 1996a). Wild *H. midae*, for example, may only reach their maximum growth of 200mm shell length in about 30 years (Tarr 1995, Sales and Britz 2001). Under aquaculture conditions, *H. midae* have been recorded to have a relatively faster growth rate (Cook 1991, Tarr 1995, Naidoo *et al.* in press) taking approximately 4-5 years to attain 100mm shell length, which is regarded as the standard international market size (Macey and Coyne 2005). There is, however, a growing market demand for smaller 'cocktail abalone' of

about 40-70mm shell length (Cook 1991, Jarayabhand and Paphavasit 1996, Najmudeen and Victor 2004).

Diet is important in abalone and has been cited as a contributing factor in the control of growth between wild and farmed abalone (Fleming *et al.* 1996, Bautista-Teruel *et al.* 2003). In the wild, kelp (*Ecklonia maxima* and *Laminaria pallida*) provides the primary food source for *H. midae* (Sales and Britz 2001, de Waal *et al.* 2003, Etheridge *et al.* 2003). However, wild abalone have to consume a variety of seaweeds (Barkai and Griffiths 1986), in large quantities, to fulfill their nutritional requirements (Britz 1996, Viera *et al.* 2005). This is because kelp has a relatively low protein content (ca 15%), has an unbalanced amino acid profile (Troell *et al.* in press) and is thus of limited nutritional value (Hahn 1989).

Kelp is either absent, scarce or insufficient to sustain the current commercial production of abalone (Hecht and Britz 1990, Anderson *et al.* 2003, Viera *et al.* 2005, Johnston *et al.* 2005, Anderson *et al.* in press) and this has prompted research into the production and use of artificially formulated diets. Research into the development of nutritionally balanced formulated feeds for *H. midae* have been undertaken since the early 1990s (e.g. Britz 1991, Britz *et al.* 1994, Britz 1996a, b). These artificial feeds are formulated so as to supply the necessary protein and carbohydrate balance ratios required to produce optimum growth in abalone (Middlen and Redding 1998, Bautista-Teruel *et al.* 2002). It has been ascertained that the protein requirement for *H. midae* is up to 47% (Britz 1996b, Viera *et al.* 2005) while the carbohydrate requirements for most *Haliotis* species ranges between 43% - 48% (Sales and Janssens 2004).

In South Africa, two formulated (artificial) feeds are currently in production. Of the two feeds only Abfeed® (Marifeed Pty Ltd., South Africa) comes close to meeting the nutritional requirements of *H. midae*. Previous studies have shown that Abfeed® dramatically improves growth over other feeds (Britz *et al.* 1994, Britz 1996a, b, Guzmán and Viana 1998, see also Shipton *et al.* 2002). However, new research has shown that incorporating fresh seaweed combinations into the diet may significantly improve growth over artificial feeds (e.g. Naidoo *et al.* in press). All of these studies have, however, concentrated on grow-out juveniles (>20mm shell length) and none so far, with the exception of Knauer *et al.* (1996), have attempted to test different feed combinations on the growth of post-weaning abalone (>6mm shell length). The aim of the present research was to test various formulated feeds (both locally produced and one international brand) and fresh wild seaweeds on the growth of post-weaning juvenile South African abalone (*Haliotis midae*).



2.3 Material and Methods

2.3.1 Experimental system

The research was conducted on the Jakobsbaai Sea Products (17° 53' 12.5" E, 32° 58' 2.5" S, Western Cape, South Africa) abalone farm. Flow through seawater of $1400 \pm 100 \text{L.h}^{-1}$ was supplied at $14.5 \pm 2.5^\circ\text{C}$ in the holding tanks. The abalone were grown in culture baskets (80 x 57 x 25cm; length, width and depth respectively), subdivided with feeding plates to increase the surface area.

2.3.2 Experimental animals

Post-weaning juvenile abalone were supplied by the Jakobsbaai Sea Products abalone farm. Since the growth of abalone is variable (depending mainly on size and feeding rate) abalone of similar size and the same gene pool were used. All abalone used were spawned in July 2003, and were approximately six months old at commencement of the experiment. Prior to commencement of the experiment, the abalone underwent a 2-week conditioning period and were fed only *Ulva* and kelp. The abalone were then subdivided into two replicate baskets of 1000 individuals per replicate, per diet treatment. The initial weight and shell length of the abalone were measured at $0.296 \pm 0.0053 \text{g}$ and $12.717 \pm 0.0151 \text{mm}$ respectively. Thereafter, both weight and shell length were measured on a monthly basis for 11 months.

2.3.3 Diets

Eleven (11) diet treatments were used during the experiment. Five of these {Abfeed® (Marifeed Pty Ltd, South Africa), Adam & Amos® 'a' (AAa), Adam & Amos® 'b' (AAb), Adam & Amos® 'c' (AAc) (Adam & Amos Abalone Foods Pty Ltd, Australia) and FeedX} comprised formulated feeds. Since Abfeed® is the most widely used formulated feed in

South Africa, it was chosen as the control feed. Of the five formulated feeds, the first four comprised of animal-based protein while the fifth is an all-seaweed-based protein feed. Most formulated feeds are protein enriched, containing fishmeal as the source of protein (Fleming *et al.* 1996, Folke *et al.* 1998, Guzman and Viana 1998). All formulated (artificial) feeds tested were sent away to an independent laboratory (Animal Production Laboratory, Institute for Animal Production, Department of Agriculture: Western Cape, Elsenburg) for compositional analysis (Table 1). The approximate dry dimensions of the various feed pellets are also given for length, width and thickness respectively (see Table 1). An additional five diet treatments comprised the previous five (5) formulated feeds, all fortified with fresh wild seaweeds (*Ecklonia maxima* - ca 15% protein; *Ulva lactuca* - 3.7-19.9% protein). The kelp *E. maxima* was chosen because it is the natural diet of *H. midae*. *Ulva lactuca* was selected because previous studies (e.g. Schoenhoff *et al.* 2003, Najmudeen and Victor 2004, Naidoo *et al.* in press) had shown that, when used as a supplement, it results in significant increases in growth.

Abalone tend to consume a variety of seaweeds as part of their natural diet (Sales and Britz 2001, Viera *et al.* 2005). These seaweeds are selected mainly according to their abundance and availability in the surrounding area (Nelson *et al.* 2002). Bearing this in mind, an additional diet comprising a mixture of seaweeds (*Ulva*, kelp and red seaweeds) was incorporated so as to compensate for nutrients that may be lacking in a single species diet. All experimental animals were fed 2 - 3 times per week and the amount of feed provided was as per the manufacturer's prescription per mean body weight. Tanks and baskets were cleaned every eight days to remove any faeces and uneaten feed.

2.3.4 Sampling and data collection

The experiment was conducted over an 11-month period, beginning in February 2004. Monthly measurements of 50 individuals from each of the 2 replicates were randomly taken. Before all weight measurements, abalone were blotted dry to remove excess water. Weight was recorded to 0.01g while shell length was measured along the longest axis to the nearest 0.01mm.

Daily increment increase in shell length (DISL) was calculated using the formula of Mai *et al.* (2001) and Zhu *et al.* (2002):

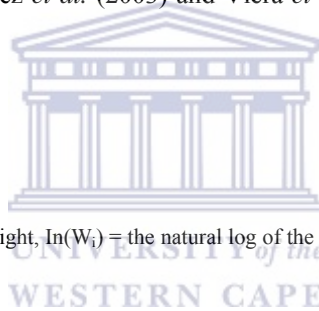
$$\text{DISL } (\mu\text{m/day}) = [(\text{SL}_t - \text{SL}_i)/t] \times 1000$$

Where SL_t = final mean shell length, SL_i = initial mean shell length and t = the feeding trial period in days.

Abalone specific growth rate (SGR in %weight.day⁻¹) was calculated as in Neori *et al.* (2000); Nelson *et al.* (2002); Gomez-Montez *et al.* (2003) and Viera *et al.* (2005) using the formula:

$$\text{SGR} = \frac{\{\ln(W_f) - \ln(W_i)\} \times 100}{t}$$

Where $\ln(W_f)$ = the natural log of the final mean weight, $\ln(W_i)$ = the natural log of the initial mean weight, and t = the feeding trial period in days.



2.3.5 Condition factor

The condition factor is a concept that was developed to account for the relationship between the weight of abalone per unit shell length (see Britz, 1996b).

$$\text{CF (g.mm}^{-1}\text{)} = [\text{BW (g) / SL (mm)}]^{2.99} \times 5575$$

Where CF = the condition factor, BW = the mean body weight and SL = the mean shell length.

2.3.6 Statistical analysis

All data are expressed as means \pm se. Data for all experimental replicates were pooled as no significant differences were found between them. Since one factor (diet type) was tested against one variable (growth), a one-way analysis of variance (ANOVA) was used to compare the differences between the final means of all diet treatments. Ideally, an analysis of co-variance (ANCOVA) should have been used in such a study. However, this analysis was not inappropriate for this study since we did not measure the same individuals from month to month. To test for correlation, the body weight and shell length of abalone from each diet treatment were compared by means of a linear regression test. All results were considered statistically significant at $P < 0.05$.



2.4 Results

2.4.1 Diets

All feeds reflected a positive correlation between body weight gain and growth in shell length (Table 2). The highest SGR was recorded for the fortified formulated feeds (Table 2) although Abfeed® on its own also produced a relatively high SGR. The fortified AAa feed (Table 2) yielded the highest daily growth in shell length ($63.06 \pm 0.52 \mu\text{m}\cdot\text{day}^{-1}$) with FeedX yielding the lowest shell growth ($28.48 \pm 0.19 \mu\text{m}\cdot\text{day}^{-1}$). The growth of abalone fed the animal-based protein feeds fared significantly better ($P=0.0037$) than those fed the all-seaweed-based feed (Figures 1 & 2). There was no difference ($P=0.9469$) in the growth of abalone fed either of the different Adam & Amos® feeds (Figure 1). Fortifying the formulated feeds with fresh wild seaweed (Figure 2) significantly improved the growth ($P=0.005$) of all abalone. The Mixed diet fared poorly against most feeds (Figure 2). The all-seaweed-based protein feed performed the poorest of all feeds (Table 2; Figures 1 & 2) in both weight and length measures. The relative increase in growth (Figure 2) was, however, more dramatic for the Adam & Amos® and FeedX feeds.

2.4.2 Condition factor

Fortification of the feeds with fresh wild seaweed greatly improved the condition factor of all formulated feeds (Table 2). Even more striking was that the condition factor of those feeds that performed particularly poorly, was dramatically increased by fortification with fresh wild seaweed.



2.5 Discussion

The differential growth rates observed in cultured abalone may be due to a number of factors. These could include: the nutrient composition (i.e. the protein and carbohydrate concentrations - Middlen and Redding 1998, Nelson *et al.* 2002, Bautista-Teruel *et al.* 2003, Viera *et al.* 2005) and digestibility (Sale and Britz 2001, 2002, Gomez-Montes *et al.* 2003); processing techniques (Booth *et al.* 2002, Sales and Britz 2002); diet particle size (Southgate and Partridge 1998); feed pellet size (Fleming *et al.* 1996); the presence of attractants (Fleming *et al.* 1996, Sales and Janssens 2004); and palatability (Kautsky *et al.* 2001).

The major nutritional requirements for optimum growth in abalone include the necessary carbohydrate (Nelson *et al.* 2002) and protein ratios (Fleming *et al.* 1996, Guzman and Viana 1998). While the carbohydrate content of the best performing formulated feed (Abfeed®, 57.3%) and the worst performing FeedX (47.3%) both seem optimal for abalone requirements (i.e. 43-48%, Sales and Janssens 2004), differences in their performance could be explained by the differences in their protein content. Although the optimum protein requirements of *Haliotis midae* is about 47% (Britz 1996b, Sales *et al.* 2003), abalone fed protein diets of 36.5 - 39.4% have been found to be acceptable (Guzmán and Viana 1998). Not only did FeedX (19.2%) have a substantially lower protein content than Abfeed® (34.7%), but animal-based proteins were more readily digested than plant-based proteins (Durazo-Beltrán *et al.* 2003). FeedX consist of dry kelp and Britz (1996b) showed that growth of abalone fed only dry kelp was very poor. It can therefore be suggested that Abfeed® contributed more to *H. midae*'s protein requirements than FeedX did. The protein and carbohydrate contents of the AA feeds were relatively similar to those of Abfeed® so these factors could not have been responsible for the poor growth obtained with the former feeds. However, the variable differences in the

protein content of the different AA feeds probably accounts for the different growth rates obtained with these feeds.

While this research showed that Abfeed® out-performed the Adam & Amos® feeds, Boarder and Shpigel (2001) reported better yields with the latter feed compared to Abfeed®, when used on *H. roei*. Although none of these diets were specifically formulated for *H. roei*, all Adam & Amos® feeds were generally formulated for the nutrient requirements of Australian abalone, while Abfeed® was formulated for the South African *H. midae*. The use of different *Haliotis* species in the two studies could have accounted for the conflicting results, as different *Haliotis* species often have different food preferences and feed conversion efficiencies. Also, Boarder and Shpigel (2001) used grow-out abalone (initial size 6.17±2.28g) while in this study, post-weaning abalone (0.296±0.0053g) were used. The different size groups used in these two studies could also account for the observed differences as nutrient requirements of smaller abalone often differ from that of older ones; it is now known that younger abalone need higher dietary protein feed compared to older ones (Shipton and Britz 2001).

The different processing techniques used when formulated feeds are manufactured could also affect the digestibility of various feeds (Booth *et al.* 2002). Sales and Britz (2002) state that cooked starches result in highly digestible carbohydrate, which could increase feed conversion efficiency. Booth *et al.* (2002) also observed that pellet extrusion significantly increases apparent digestibility, compared to cold pelleted and steam pelleted techniques. While there is no way to ascertain with any degree of certainty whether the different processing techniques could have affected growth rates, this still remains a relevant factor.

Diet particle size has also been reported to affect the digestibility of a formulated feed (Southgate and Partridge 1998). Smaller diet particles increased surface area making it easier for abalone to efficiently assimilate the feed. Sales and Britz (2002) and Sales and Janssens (2004) reported that reducing the diet particle size to between 150-450 μm significantly increased apparent digestibility in *H. midae*; particle sizes smaller than 450 μm causing no significant difference in digestibility. Although feed manufacturers do not divulge such information, particle size could have played a significant role in the digestibility of the feeds tested. Similarly, the binders used in these feeds could also have affected growth by influencing the digestibility of each feed. Southgate and Partridge (1998) suggested that microbound feeds (compared to microencapsulated feeds) were more easily digested since the nutrients were embedded on a matrix. However, depending on the binder used, microbound feeds tend to be highly soluble compared to microencapsulated feeds. Boarder and Shpigel (2001) observed that the Adam & Amos® feeds disintegrated faster than Abfeed®. This suggests that the Adam & Amos® feeds were made of a different type of binder than Abfeed®.

Formulated feed pellet size has also been cited as a likely cause of differences in abalone growth rates (Fleming *et al.* 1996). Although there have been no studies on the effects of pellet size on the intake or digestibility of various feeds, Ruscoe *et al.* (2005) observed that the redclaw crayfish preferred to feed on larger pellets. This may also apply to abalone, as both these animals are sedentary and therefore prefer to trap their food. All the AA diet pellets were relatively small compared to the Abfeed® pellets; the latter no doubt has a larger surface area for the rasping action of the abalone. Stewart and Grant (2002) also observed that smaller pellets disintegrated faster, losing 30% of their mass after only 120 hours in seawater, thus limiting the duration for feeding on such pellets. This would suggest that the

abalone could have had more time to feed on the larger Abfeed® pellets, therefore increasing their feed intake.

The feed pellet colour could also have affected abalone growth by determining the feeding rate of abalone on each diet. Although there is currently insufficient knowledge pertaining to the effects of pellet colour, Fleming *et al.* (1996) suggested that, since abalone are generally nocturnal animals, darker feeds may be more attractive to abalone as they simulate their natural feed colours. Abfeed® was darker than all the Adam & Amos® feeds and colour may have influenced the consumption rates of these feeds. However, pellet colour could not solely be used to explain the poor growth rates obtained with the FeedX pellets, as these pellets were the darkest of all.

The shape of the pellets could also have affected the growth of abalone on the various formulated diets. Fleming *et al.* (1996) suggested that abalone prefer to consume flat, broad pellets that have an increased surface area. Not only were the Abfeed® pellets relatively broad, but they were roughly half the thickness of the other formulated feeds. Knauer *et al.* (1996) suggested that breaking the pellets into smaller fragments (but not too small) increased the feed consumption rates in abalone as this reduced hard edge effects. It is highly likely that the thin Abfeed® pellets would be more susceptible to breakage and thus easily consumed by abalone.

Although wild abalone are typically sedentary animals relying on drift seaweed, farmed abalone tend to be more mobile, roaming for their food. The presence, or absence, of attractants in the various diets could have affected abalone growth rates as attractants stimulated abalone feed intake and thus enhancing growth rates. However, Fleming *et al.*

(1996) cautioned that although the presence of attractants in artificial feeds drew abalone, feed consumption varied and no significant growth changes have yet been recorded. This could be due to the composition of the various feed attractants used, since most attractants used were usually formulated or extracted from seaweeds (Sales and Janssens 2004). On the other hand, feeds comprising fishmeal have been found to be more attractive to abalone (Lee *et al.* 2004) and this could also have contributed to the higher growth rates recorded for all the animal-based protein feeds relative to the all-seaweed based FeedX.

Low palatability could also have accounted for the poor growth rates obtained with FeedX. This is because the less palatable feeds become wasted, as feed intake is low and thus resulting in low growth rates (Kautsky *et al.* 2001, Lee *et al.* 2004). When the baskets were cleaned, more un-eaten bits of FeedX (relative to other feeds) were observed, suggesting that the abalone consumed less of this feed. The low palatability of this feed could therefore be due to the absence, or reduced effects of, attractants (Fleming *et al.* 1996, Sales and Janssens 2004).

Fortifying the formulated feeds with fresh wild seaweed significantly improved the growth of abalone on all feeds. This was probably due to the contribution of fresh seaweeds toward the nutrients that may be lacking in the formulated feeds. Although fresh wild seaweed typically had low protein contents, (Fleming *et al.* 1996, Robertson-Anderson 2003, Johnston *et al.* 2005, Naidoo *et al.* in press), both the kelp *E. maxima* (Simpson 1994, Stepto and Cook 1996) and *Ulva* (Schoenhoff *et al.* 2003, Najmudeen and Victor 2004) were valuable supplementary feeds that promoted good growth in *H. midae*. When fed fresh seaweeds, abalone consume approximately 7-10% of their body weight daily (Rotmann 1999, Neori *et*

al. 2000). This relatively high feed consumption efficiency, together with the high nutritional value of the formulated feeds, could have resulted in the enhanced growth recorded.

The use of mixed diets has been reported to enhance abalone growth since a variety of seaweeds may compensate for the nutrients that may be lacking in other seaweeds (Schneider *et al.* 2005, Naidoo *et al.* in press). Nelson *et al.* (2002) found that a mixed diet yielded relatively higher growth compared to single seaweed diets. Seaweed feeds generally tended to yield lower growth rates compared to formulated feeds (Guzman and Viana 1998, Bautista-Teruel *et al.* 2003, Lee 2004, Lee *et al.* 2004). In comparison, this study also showed that the mixed diet produced lower growth compared to most formulated feeds. In contrast, however, Naidoo *et al.* (in press) showed that a mixed diet produced better growth than the formulated Abfeed® diet. Such contrasting results could again be due to the different abalone size classes as well as the seaweed used in the two studies. While post-weaning abalone (14.57 ± 0.227 mm) were used in the current study, older grow-out juveniles (34.7 ± 5.8 mm) were used in the Naidoo *et al.* (in press) study. Furthermore, fresh wild *U. lactuca* (3.7-19.9 % protein, Robertson-Anderson 2003) was used in the current study while farm-grown protein enriched *U. lactuca* (33.4%-36.6 % protein) was used in Naidoo *et al.* (in press). In comparison, the Naidoo *et al.* (in press) mixed diet could then be equated to the fortified Abfeed® (Abfeed® + Kelp + *Ulva*) diet used in the current study.

The condition factor (CF), the relationship between the weight of abalone per unit shell length, accounts for the amount of feed invested in developing both the body weight and shell length. This factor is important for farmers as it is used during the abalone grading process for market purposes. Abalone that have high CF values tend to be fat and possess relatively short shells, reflecting that nutrients were invested more into body weight than into shell

growth. In this study, fortifying the formulated feeds with fresh seaweed resulted in higher CF values. Considering that Boarder and Shpigel (2001) reported that feeding on *Ulva* produced relatively low CF values in *H. midae*, fortifying with fresh wild *U. lactuca* alone could not have accounted for the recorded higher CF values in this study. On the other hand, Naidoo *et al.* (in press) showed that fortifying Abfeed® with fresh kelp produced a significantly higher bodyweight to shell length ratio relative to Abfeed® on its own. The high CF values observed in the fortified feeds could therefore be due to compensation of nutrients lacking in *Ulva* by those present in both fresh kelp and the formulated feeds.

Generally, most abalone farmers are satisfied with an average growth rate of 67-100µm/day (Fleming *et al.* 1996; Mai *et al.* 2001). However, such farmers are culturing mostly smaller tropical abalone that tend to grow faster than the bigger temperate species. The current results show that the growth of *H. midae* on all diet treatments failed to achieve such growth rates. This is understandable since *H. midae* is the second largest (and thus relatively slow growing) abalone globally (White 1995). The growth rates obtained with Abfeed® appears to be adequate for achieving the general market size within the usual 4-5 year period. The other formulated feeds will no doubt take longer to achieve the market size. All of the abalone fed the fortified feeds, however, will reach the market size relatively earlier than those fed only the formulated and mixed feed diets.

2.6 Conclusion

This study is consistent with previous work (e.g. Hahn 1989, Guzman and Viana 1998, Bautista-Teruel *et al.* 2003, Lee 2004) showing that animal-based protein feeds yield better growth rates than seaweed-based protein feeds. The control feed (Abfeed®) outperformed all other formulated feeds, reflecting it as the best feed for the South African abalone, *H. midae*. However, fortifying the formulated feeds with fresh wild seaweed result in better growth. Additional studies are needed to determine the relative importance of the various factors that may have contributed to the different performances of the various feeds. While no one factor (any and/or all may have contributed) can be singled out as the major factor contributing to increased growth in post-weaning *H. midae*, the results of this study clearly show the importance of supplementing existing formulated feeds with fresh seaweed in the culture of juvenile South African post-weaning abalone. *H. midae*.



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2.9 Tables

Table 1. Approximate dimensions of pellets and nutritional analysis of the various formulated feeds used. Abfeed®, Adam & Amos® ‘a’ (AAa), Adam & Amos® ‘b’ (AAb), Adam & Amos® ‘c’ (AAc) and FeedX.

Feed	Moisture (%)	Ash (%)	Protein (%)	Fibre (%)	Fat (%)	Carbohydrate (%)	Dimensions (length:width:thickness in mm)
Abfeed®	~10	5.6	34.7	1.6	2.4	57.3	17: 9: 1
AAa	~10	7.8	34.2	1.8	2.3	55.2	6: 6: 2
AAb	~10	7.8	28.8	2.1	3.2	60.2	6: 6: 2
AAc	~10	7.6	28.1	2.3	3.3	61.0	6: 6: 2
FeedX	~10	32.9	19.2	10.9	0.7	47.3	40: 10: 2

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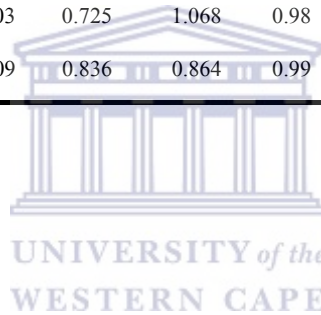
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Table 2. Growth parameters of post-weaning abalone grown on eleven diet treatments. Specific growth rate (SGR), daily increment increase in shell length (DISL), condition factor (CF), Adam & Amos® 'a' (AAa), Adam & Amos® 'b' (Aab), Adam & Amos® 'c' (AAc), Ulva and kelp (U/K).

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Diet	SGR	DISL	Initial CF	Final CF	Correlation		Rank		
	(%weight.day ⁻¹)	(µm.day ⁻¹)	(g.mm ⁻¹)	(g.mm ⁻¹)	r	r ²	SGR	DISL	CF
Abfeed®+U/K	1.05±0.02	61.89±0.03	0.719	1.447	0.95	0.90	1	3	1
Abfeed®	1.00±0.02	61.44±0.09	0.863	1.312	0.97	0.95	3	4	2
AAc+U/K	0.97±0.02	63.05±0.04	0.980	1.189	0.96	0.92	4	2	3
AAc	0.82±0.02	44.66±0.08	0.740	1.178	0.98	0.97	8	9	4
AAa+U/K	1.02±0.02	65.30±0.02	0.709	1.167	0.97	0.94	2	1	5
FeedX+U/K	0.80±0.02	43.57±0.06	0.712	1.134	0.97	0.94	9	10	6
AAa	0.84±0.02	50.08±0.09	0.882	1.131	0.98	0.96	7	7	7
Mixed diet	0.79±0.02	45.86±0.06	0.901	1.115	0.97	0.94	10	8	8
AAb+U/K	0.93±0.02	60.71±0.03	0.948	1.110	0.97	0.94	5	5	9
AAb	0.85±0.02	50.12±0.03	0.725	1.068	0.98	0.96	6	6	10
FeedX	0.49±0.02	27.40±0.09	0.836	0.864	0.99	0.98	11	11	11



2.10 Figure Captions

Figure 1. Growth in body weight (means \pm se) of post-weaning abalone grown on formulated feeds.

Figure 2. Growth in body weight (means \pm se) of post-weaning abalone grown on all diet treatments.



2.11 Figures

Figure 1.

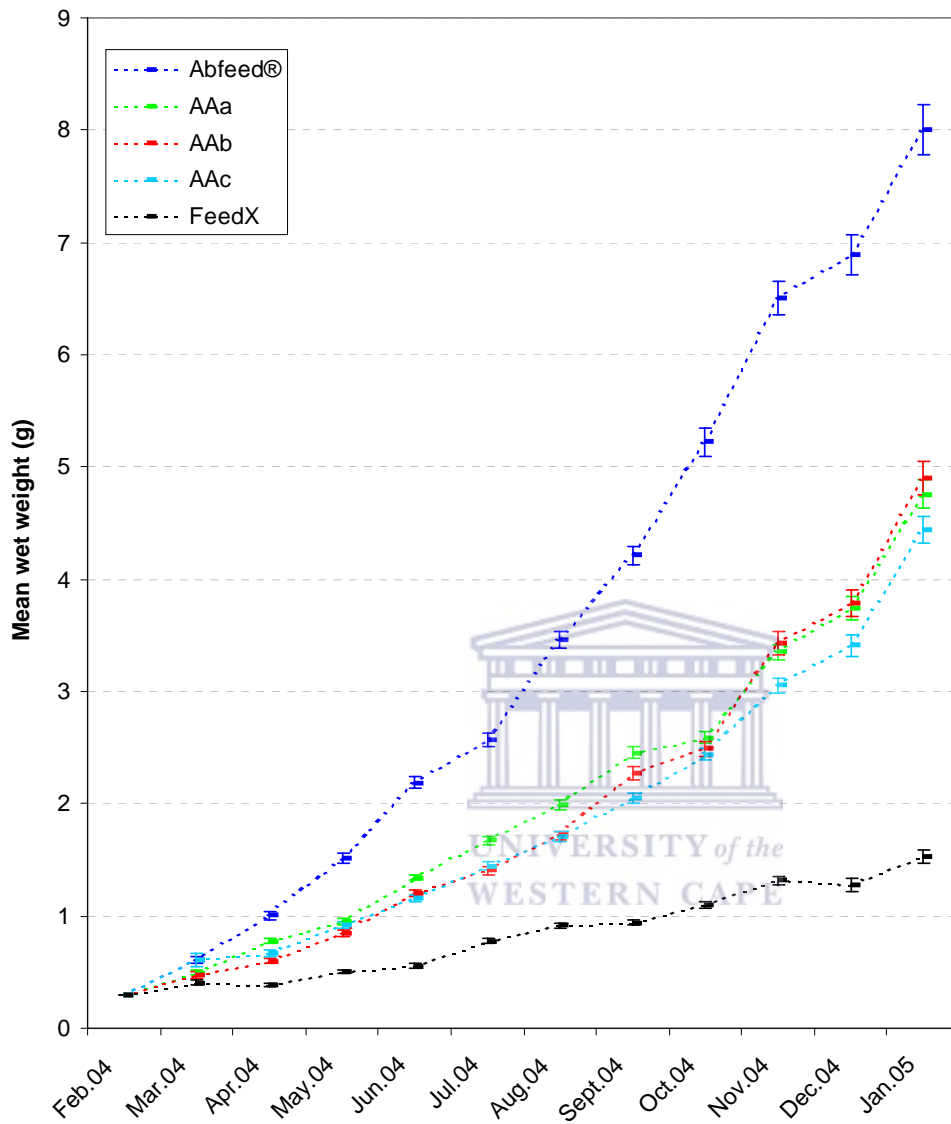
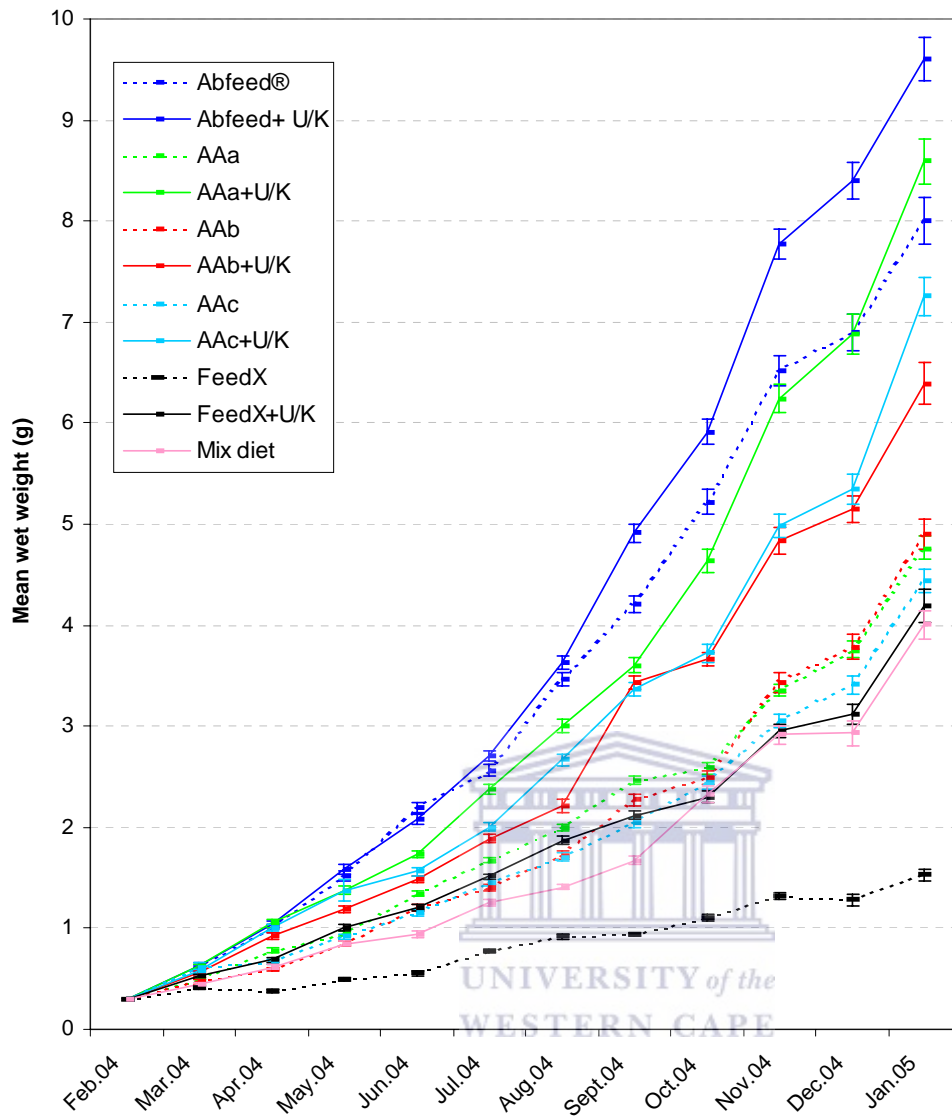


Figure 2.



CHAPTER 3

The effects of differences in basket design and water flow-systems in the culture of the South African abalone, *Haliotis midae* Linnaeus, using various diet treatments.

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3.1 Abstract

The effects of differences in four basket designs [Global Ocean (GO), Ivey Blue down (Idn), Ivey blue up (Iup) and Jakobsbaai Sea Products (JSP)] currently used in the culture of the South African abalone, *Haliotis midae* (Linnaeus), were investigated under both a flow-through and a re-circulation water system. Growth of grow-out abalone was monitored on a commercial abalone farm over a period of 7-months in an experiment consisting of 4 diet treatments [1) the formulated fishmeal-based protein feed Abfeed®, 2) a formulated all-seaweed-based protein FeedX, 3) fresh kelp (*Ecklonia maxima*) and 4) a mixture of fresh wild seaweeds {the kelp, *Ecklonia maxima*, *Ulva lactuca* and *Gracilaria gracilis*} in combination with Abfeed®] with 2 replicates per basket design (N = 350 individuals per replicate). The results showed that basket design did indeed affect abalone growth. In the re-circulation system, the Idn baskets produced relatively higher growth in abalone with the Mixed diet (SGR = $0.218 \pm 0.07\% \cdot \text{day}^{-1}$) and kelp (final CF = $1.274 \text{g} \cdot \text{mm}^{-1}$). In the flow-through system, however, diet determines the effectiveness of any one basket design with the GO basket producing best results (SGR = $0.115 \pm 0.04\% \cdot \text{day}^{-1}$, final CF = $0.708 \text{g} \cdot \text{mm}^{-1}$) when Abfeed® was used as the test feed. Similarly, water flow-systems also produced differences in growth with higher growth rates being recorded in the re-circulation system. This study shows that besides feed, factors such as basket design and water flow-system may invariably influence the success and/or failure of a culture system for the South African abalone, *Haliotis midae*.

Key words: Basket design, flow-through, growth, *Haliotis midae*, re-circulation, water flow-systems.

3.2 Introduction

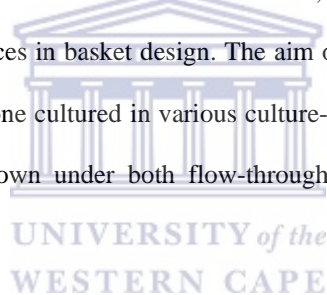
While the abalone fishery in South Africa dates back to 1949 (Tarr 1992), the history of abalone aquaculture is relatively recent with major developments only occurring during the 1990s (Sales and Britz 2001, Simon *et al.* 2004). The successful spawning of *Haliotis midae* in the late 1980s marked the inception of abalone farming in South Africa (Hecht and Britz 1990, Cook 1998). Currently there are approximately twelve (12) abalone farms operating in South Africa (Sales and Britz 2001; Simon *et al.* 2004), with the industry expected to expand (Tony Bennet, Abalone Farmers Association of Southern Africa, pers. comm.).

In the aquaculture industry, water is a valuable component that serves mainly as a medium for transporting suspended organic particles (Folke and Kautsky 1989, 1992), dissolved oxygen (Kautsky *et al.* 2001, Huchette *et al.* 2003), pollutants (Neori *et al.* 2004) and aquaculture wastes (Gowen *et al.* 1990, Kautsky *et al.* 2001). Any, or all, of the above factors could have far reaching consequences (such as anoxia, eutrophication, increased parasites) should there be an imbalance in the aquaculture system. It is therefore vital to monitor factors such as water temperature (Tarr 1995, Guzman and Viana 1998), quality (Kautsky and Folke 1989, Naylor *et al.* 2000), and flow rate (Wells *et al.* 1998).

Aquaculture water systems are either based on a flow-through system, a re-circulation system, or a combination of both (Furey *et al.* 2003). The flow-through system relies on an unlimited water source for sustainability. According to Furey *et al.* (2003) and Troell *et al.* (1999), such water must travel fast (depending on aquaculture intensity and stocking density) so as to remove waste and oxygenate the tanks. The re-circulation system reuses water and therefore reduces dependence on water replacement to maintain water quality (Losordo *et al.* 1998). Re-circulation water systems are more water-sustaining since they only use a fraction

of what flow-through systems use and only a small proportion of the water may be lost (Losordo *et al.* 1998). Fewer water resources are therefore needed for this water system as the water replacement rate is reduced by 30-50 times compared to the flow-through system (Deville *et al.* 2005). Re-circulation systems are, however, more dependent on biofilters and protein skimmers to combat excess wastes. This system is usually employed in intensive or integrated aquaculture, where wastewater is reused as feed for the organisms that serve as biofilters in such systems (Naylor *et al.* 2000).

Most growth studies on abalone have so far concentrated on growth in either flow-through (e.g. Bautista-Teruel and Millamena 1999, Boarder and Shpigel 2001) or re-circulation (e.g. Britz *et al.* 1997, Shipton *et al.* 2002) systems. None have examined the comparative growth of abalone from both systems. Furthermore, based on the individual needs and farm design, most abalone farms utilize only a single basket design to stock their abalone. These baskets, however, often differ in their construction from farm to farm and to date, no one (that we are aware of) has tested the effects of differences in basket design. The aim of this research was therefore to 1) monitor the growth of abalone cultured in various culture-basket designs, and 2) to compare the growth of abalone grown under both flow-through and re-circulation systems.



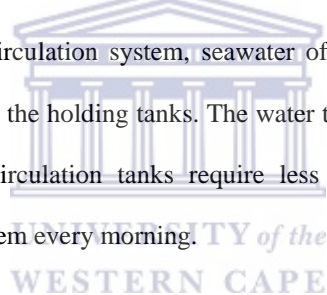
3.3 Material and Methods

3.3.1 Experimental system

This research was conducted on the Jakobsbaai Sea Products (17° 53' 12.5" E, 32° 58' 2.5" S, Western Cape, South Africa) abalone farm. On this farm, a flow-through water system as well as a re-circulation system are employed. The flow-through system is a unidirectional open-ended system where water from the ocean enters the system at one end and exits through the other. The re-circulation system is a semi-enclosed system with only a small fraction of the water leaving the system.

Flow-through system: In the flow-through system, seawater of $900\pm 100\text{L}\cdot\text{h}^{-1}$ was supplied at $14.5\pm 1.4^\circ\text{C}$ in the holding tanks. The flow direction within each tank was changed on a weekly basis to compensate for end effects. Because of the high flow rate, waste products were simply expelled through the open end. Nonetheless, the flow-through tanks were cleaned at least three times a month.

Re-circulation system: In the re-circulation system, seawater of $220\pm 100\text{L}\cdot\text{h}^{-1}$ was supplied at approximately 16.9°C in the holding tanks. The water turnover period was maintained at 85 hours. The re-circulation tanks require less cleaning as waste products were drained from the system every morning.



3.3.2 Experimental animals

Since abalone have a very heterogeneous growth rate (Lee 2004), genetically similar grow-out juvenile ($49.54\pm 0.15\text{mm}$; $27.81\pm 0.07\text{g}$) abalone were used for the experiment. All abalone were selected using a 52mm radius grading ring. Abalone were subdivided into two replicate baskets of 350 individuals per basket, per diet treatment.

3.3.3 The baskets

Four basket designs namely GO (Global Ocean Pty Ltd, South Africa), Iup [Ivey Blue up] and Idn [Ivey Blue down] (Ivey Blue Pty Ltd, South Africa), and JSP (Jakobsbaai Sea Products, Jakobsbaai, South Africa) were used in the research. The approximate dimensions of the GO (690 x 520 x 550 mm), the Idn (1037 x 517 x 540 mm) Iup (1037 x 517 x 540 mm) and JSP (1200 x 500 x 500 mm) baskets are given for length, width and height respectively. The GO baskets (Figure 1) have holes drilled only in their bottoms. The Iup baskets (Figure 2) have holes throughout the basket, forcing water to circulate *up* through the basket. In contrast, the Idn baskets (Figure 3) have only two rows of holes along the base of the sides of the baskets, forcing water to circulate *down* into the baskets. The JSP baskets (Figure 4) are made of 16mm radius mesh.

3.3.4 Diets and experimental treatments

Four (4) diet treatments were variably incorporated into the experiment comprising 1) the formulated fishmeal-based protein feed, Abfeed®, 2) a formulated all-seaweed-based protein feed, FeedX, 3) fresh kelp (*Ecklonia maxima*) and 4) a mixture of fresh wild seaweeds (*Ecklonia maxima*, *Ulva lactuca* and *Gracilaria gracilis*) in combination with Abfeed®. The diets were incorporated solely to test the effects of basket design and flow-systems, and not to compare the different diets.

3.3.4a Testing basket design:

To determine the possible effects of basket design on growth of juvenile abalone, two experimental treatments were run.

1). In the flow-through system: Only 2 diets (Abfeed® and kelp) were incorporated into all 4 basket designs.

2). In the re-circulation system: All 4 diet treatments were incorporated into only 2 basket (GO and Idn) designs. Due to design constraints of the re-circulation system, the remaining baskets (Iup and JSP) could not be tested.

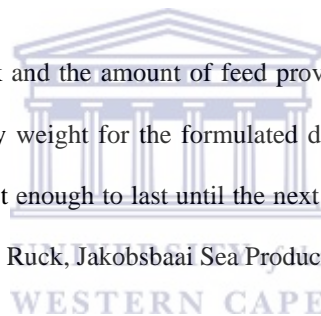
3.3.4b Testing flow-systems:

To determine the possible effects of flow-systems (Flow-through vs re-circulation) on the growth of juvenile abalone, again two experimental treatments were run. This was limited by the basket designs that could be incorporated into the re-circulation system (see above).

1) GO basket design: All 4 diet treatments were incorporated into the GO baskets for both flow-systems.

2) Idn basket design: Only 2 diets (Abfeed® and kelp) were incorporated into the Idn baskets for both flow-systems.

All animals were fed three times per week and the amount of feed provided was as per the manufacturer's prescription per mean body weight for the formulated diets. The amount of fresh seaweed given to the animals was just enough to last until the next feeding period; this ascertained from previous farm data (Kevin Ruck, Jakobsbaai Sea Products, pers. comm.).



3.3.5 Sampling and data collection

The experiment was conducted over a six-month (6) period. Monthly measurements of 60 individuals from each of the two replicates were randomly sampled. Before all weight measurements, abalone were blotted dry to remove excess water. Abalone body weight was recorded to the nearest 0.01g, while shell length was measured along the longest axis to the nearest 0.01mm.

Abalone specific growth rate (SGR in %weight.day⁻¹) was calculated using the formula (see Evans and Langdon 2000, Neori *et al.* 2000, Nelson *et al.* 2002 and Gomez-Montez *et al.* 2003):

$$\text{SGR} = \frac{\{\ln(W_f) - \ln(W_i)\} \times 100}{t}$$

Where $\ln(W_f)$ = the natural log of the final mean weight, $\ln(W_i)$ = the natural log of the initial mean weight, and t = the feeding trial period in days.

The condition factor (CF), a concept that was developed to account for the relationship between the weight of abalone per unit shell length, was calculated using a formula of Britz (1996b):

$$\text{CF (g.mm}^{-1}\text{)} = [\text{BW (g) / SL (mm)}^{2.99}] \times 5575$$

Where CF = the condition factor, BW = the mean body weight and SL = the mean shell length.

3.3.6 Statistical analysis

All data are expressed as means \pm standard errors. Data for all experimental replicates were pooled as no significant differences were found between them. The analysis for this study was done using SAS V9.1 (SAS® Institute Inc., Cary, NC, USA). An initial analysis of covariance was first tested with the baseline value of the outcome (either length or weight) used as a covariate. This was done to account for any differences in starting values. To test for actual differences resulting from differences in basket design and flow-systems, Mixed Procedure (3-way ANOVA) was tested on the final moth's data. The factors used in the analysis were basket type, flow-system type and diet type. All data were regarded as significant at $P < 0.05$.

3.4 Results

3.4.1 Testing basket design

Growth of grow-out juvenile abalone was clearly affected by both basket design and flow-system.

1) *Flow-through system* (Figure 5):

While there was no significant difference ($P>0.05$) between growth of abalone in the four different basket designs when testing with kelp, growth results with Abfeed® ($P=0.0002$) showed clear differences between basket designs. When testing with Abfeed®, the GO basket produced the best results, followed by the Iup basket, then the Idn basket, and followed lastly by the JSP basket design.

2) *Re-circulation system* (Figure 6):

Although abalone growth on some diet treatments (i.e. kelp and FeedX) showed no significant differences ($P=0.8493$ – kelp and $P=0.7595$ – FeedX) in abalone stocked in the Idn and GO baskets, abalone grown in the Idn baskets fared better overall. The data clearly show that abalone grown on Abfeed® ($P=0.0326$) and the Mixed diet ($P=0.0463$) do better in the Idn baskets. Overall the Idn baskets yielded the highest SGR and CF values (Table 1) when used in the re-circulation system.

3.4.2 Testing water flow-systems

With the exception of a poor-performing FeedX diet (Figure 7), higher growth rates were recorded in the re-circulation system compared to those in the flow-through system when either GO (Figure 7) or Idn (Figure 8) baskets were used in the growth trials. This data is supported by the generally higher SGR and CF values (Table 1) obtained for baskets used in the re-circulation system.

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While different diets were used only as a measure to determine the effects of basket design and water flow-systems, some interesting deductions can be made. No matter the basket design, FeedX (formulated all-seaweed-based feed) always yielded the comparatively poorest growth (Figures 6 & 7). The JSP basket has clearly not been made to incorporate formulated feeds (Figures 4 & 5, Table 1) and has produced the greatest variation in growth rates. Abalone growth showed the least variation in the GO baskets (Figures 5 & 6).



3.5 Discussion

This study clearly demonstrated that the basket design and water flow-systems influenced the growth of abalone. The major factors known to generally affect abalone growth rates include water temperature (Tarr 1995, Guzman and Viana 1998, Steinarsson and Imsland 2003), water flow rate (Wells *et al.* 1998), water quality (Kautsky and Folke 1989, Naylor *et al.* 2000) and food availability (Huchette *et al.* 2003). Higher temperatures generally yield higher growth rates in abalone (Fielding 1995, Britz *et al.* 1997, Neori *et al.* 1998). Since the recirculation system in this study operated at a comparatively higher temperature (16.9°C), this probably accounted for the better abalone growth obtained from this system.

Although differences in water temperature seemed a likely factor resulting in the variable growth rates recorded in the two flow-systems compared, Tarr (1995) cautioned that although temperature affected abalone growth, the notion that warmer water results in relatively faster growth rates doesn't always hold true. When abalone are, for example, fed formulated diets, relatively high temperatures often cause "bloating" that may result in mortality (Fleming *et al.* 1996, Macey and Coyne 2005). However, this was not the case in this study as no temperature mortalities were recorded in either system. Furthermore, the temperatures experienced in both systems were well within the abalone's normal temperature range of a 12°C minimum on the West Coast to a 21°C maximum on the East Coast (Britz *et al.* 1997). Abalone only cease to grow when temperatures are extremely high and well beyond the abalone's natural range (Harris *et al.* 2005).

The variable growth between the two systems could also have been due to differences in water flow-rates, since water flow-rate has been advocated to produce differences in abalone

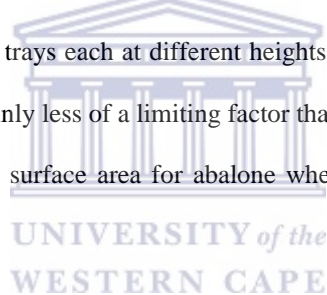
growth rates (Wells *et al.* 1998). This is due to the variable generation of nitrogenous waste in the form of ammonia, large quantities of which can deplete the available oxygen and thus reduce abalone growth rates (Troell *et al.* 1999, Harris *et al.* 2005). Maintaining a steady water flow-rate is therefore important in the system so as to sufficiently aerate the water (Hecht 1994, Shpigel *et al.* 1999, Demetropoulos and Langdon 2004, Langdon *et al.* 2004) and in particular, a relatively rapid water flow-rate is necessary to flush out all metabolic and other wastes from the culture system (Kautsky and Folke 1989, Evans and Langdon 2000, Deviller *et al.* 2005). While the flow-through system in this study appears to provide the better flow-rate requirements, best growth results were still obtained from the re-circulation system. Wells *et al.* (1998) showed that when exposed to rapid flow rates, abalone tend to incur higher metabolic costs, as they have to increase their attachment strengths. This could account for the comparatively low CF values obtained in the flow-through system.

Like water temperature and flow rate, differences in water quality could also have accounted for the variable growth, since water quality has also been advocated as being important in culture systems (Kautsky and Folke 1989, Naylor *et al.* 2000). Low water quality could have resulted in reduced dissolved oxygen which, in turn, compromised abalone growth. Although the water quality may have deteriorated for a number of reasons, it was most likely due to the generation of waste beyond the culture system's ability to assimilate such waste (Folke *et al.* 1994). While the Iup basket had the least number of holes, creating potential for stagnation spots, design was clearly not a negating factor (Figure 5, Table 1) when used in the flow-through system.

This study has demonstrated that diet type affected abalone growth. In this study, it was observed that more uneaten pellets of FeedX were collected during cleaning. This could be

attributed to low digestibility and/or palatability of this feed. This is because less digestible and/or less palatable feeds tended to end up deteriorating and generating waste (Kautsky *et al.* 2001), since the type of feed given to cultured abalone influenced the waste output in an abalone aquaculture system (Gowen *et al.* 1990). The highly soluble feed pellets affected water quality more than did those that were less soluble. The solubility rate of different feed pellets therefore determined just how long these pellets remained in water without dissolving (Sales and Britz 2001). The Abfeed® pellets have been recorded to have a relatively low solubility rate (Boarder and Shpigel 2001) and would therefore not affect water quality.

In the wild, food availability has also been reported as a factor that affects abalone growth (Huchette *et al.* 2003). The interior design of the various baskets affected feed accessibility to abalone. This was reflected by the low CF and SGR values obtained using Abfeed® in the JSP baskets that had broad mesh sides and only one feeding tray. The abalone were always clustered on the feeding tray (pers. obs.) and this limited feed access for some abalone. The GO, Idn and Iup baskets had three feeding trays each at different heights within the baskets, so access to feed in these baskets was certainly less of a limiting factor than in the JSP basket. Having more feeding trays provided more surface area for abalone when formulated feeds were used.



All these factors considered, basket design clearly also influenced growth in abalone. With the exception of a poor performing FeedX, the Idn baskets produced better growth in abalone grown in the re-circulation system. This was surprising, as the GO baskets had specifically been designed for the re-circulation system employed on the Jakobsbaai Sea Products farm. In the flow-through, basket design was relatively variable with feed-type being the main factor determining the effectiveness of the basket design. Nonetheless, even in the flow-

through system, basket design was especially critical when especially formulated (pelleted) feeds were used.



3.6 Conclusion

In conclusion, basket design and water flow-systems clearly influenced the growth of the abalone, *Haliotis midae*. While abalone grew comparatively faster in the re-circulation system than in flow-through system, basket designs produced variable results. In the re-circulation system, the Idn basket design produced relatively high growth in abalone. In the flow-through system, however, diet determined the effectiveness of any one basket design.

3.7 Recommendations

In flow-through system:

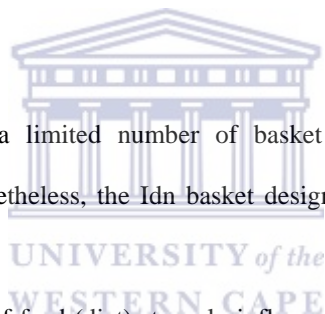
If kelp is used as a feed, any of the 4 basket designs can be incorporated as they all produce comparable results.

If formulated feeds are used, the order of basket preference should be; GO, Iup, Idn and lastly JSP.

In re-circulation system:

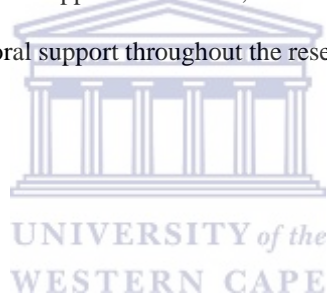
Due to design constraints, only a limited number of basket designs could be incorporated into this system. Nonetheless, the Idn basket design should be chosen over the GO basket.

It should be noted though that the choice of feed (diet) strongly influences the effectiveness of a basket design so that the above recommendations should be viewed only as a guide.



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3.10 Tables

Table 1. Growth of grow-out juvenile abalone in the various basket designs and water flow-systems. Specific growth rate (SGR), body weight (BW), condition factor (CF), Global Ocean basket (GO), Ivey Blue down basket (Idn), Ivey Blue up basket (Iup), Jakobsbaai Sea Products basket (JSP), re-circulation flow-system (R¹) and flow-through flow-system (F²).

Feeds	Initial weight (g±se)	Final weight (g±se)	Initial length (mm±se)	Final length (mm±se)	Initial CF (g.mm ⁻¹)	Final CF(g.mm ⁻¹)	SGR (BW%.day ⁻¹)
Idn R ¹ Mix	27.90±0.89	57.23±1.11	49.64±0.28	64.66±0.46	1.322	1.230	0.218±0.07
Idn R ¹ Abfeed	28.11±0.50	55.83±1.09	50.09±0.18	63.28±0.43	1.296	1.280	0.208±0.24
Idn R ¹ Kelp	27.24±0.61	52.33±1.12	49.92±0.25	62.02±0.44	1.269	1.274	0.198±0.19
GO R ¹ Kelp	27.77±0.44	52.78±0.84	49.98±0.13	63.91±0.33	1.289	1.175	0.195±0.19
GO R ¹ Abfeed	28.02±0.42	50.94±1.35	50.08±0.11	63.55±0.52	1.293	1.153	0.181±0.36
GO R ¹ Mix	27.90±0.51	49.94±1.02	50.01±0.16	63.03±0.56	1.293	1.159	0.176±0.21
Idn R ¹ FeedX	27.36±0.65	44.64±0.78	49.97±0.31	60.05±0.38	1.271	1.197	0.148±0.05
Idn F ² Kelp	27.48±0.57	42.66±0.95	49.74±0.27	58.17±0.41	1.295	1.258	0.133±0.15
GO R ¹ FeedX	27.85±0.46	42.92±0.73	50.13±0.08	60.05±0.40	1.281	1.151	0.131±0.14
GO F ² Mix	27.54±0.51	41.86±0.85	50.15±0.24	58.59±0.39	1.266	1.208	0.127±0.16
JSP F ² Kelp	27.83±0.61	40.95±0.97	49.37±0.33	55.99±0.64	1.340	1.354	0.117±0.14
Iup F ² Kelp	27.61±0.66	40.60±0.81	49.91±0.21	57.33±0.48	1.287	1.251	0.117±0.06
GO F ² Abfeed	28.10±0.51	41.03±0.83	49.99±0.15	57.95±0.44	1.304	1.224	0.115±0.04
GO F ² Kelp	28.39±0.47	40.90±0.87	50.06±0.18	59.25±0.48	1.313	1.142	0.111±0.18
Iup F ² Abfeed	27.89±0.64	39.07±0.76	50.02±0.21	58.64±0.52	1.292	1.125	0.102±0.05
Idn F ² Abfeed	27.81±0.56	37.27±0.71	49.89±0.25	57.15±0.40	1.298	1.159	0.089±0.08
GO F ² FeedX	27.90±0.48	36.88±0.68	49.99±0.23	57.94±0.40	1.295	1.100	0.085±0.10
JSP F ² Abfeed	27.89±0.60	32.40±0.62	49.59±0.32	54.02±0.31	1.325	1.192	0.045±0.01

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3.11 Figure Captions

Figure 1: The three-dimensional view of the GO basket

Figure 2: The three-dimensional view of the Iup basket

Figure 3: The three-dimensional view of the Idn basket

Figure 4: The three-dimensional view of the JSP

Figure 5: The effects of basket design on growth of juvenile abalone within the flow-through system incorporating only two feeds.

Figure 6: The effects of the Idn and GO basket designs on growth of juvenile abalone within the re-circulation system.

Figure 7: The effects of flow-through vs. re-circulation water systems on growth of abalone using the GO basket design.

Figure 8: The effects of flow-through vs. re-circulation water systems on growth of abalone using the Idn basket design.



3.12 Figures

Figure 1.



Figure 2.



Figure 3.



Figure 4.



Figure 5.

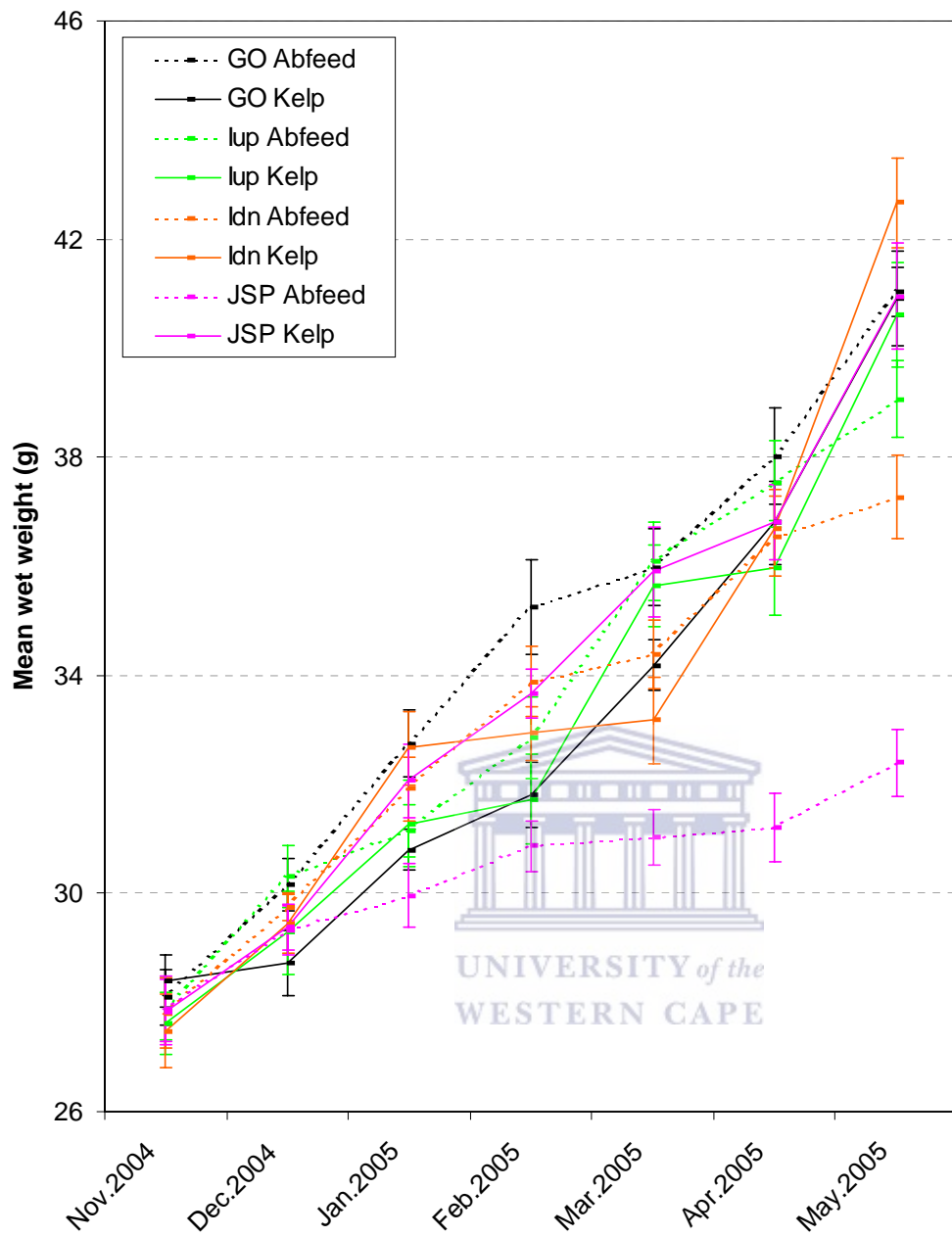


Figure 6.

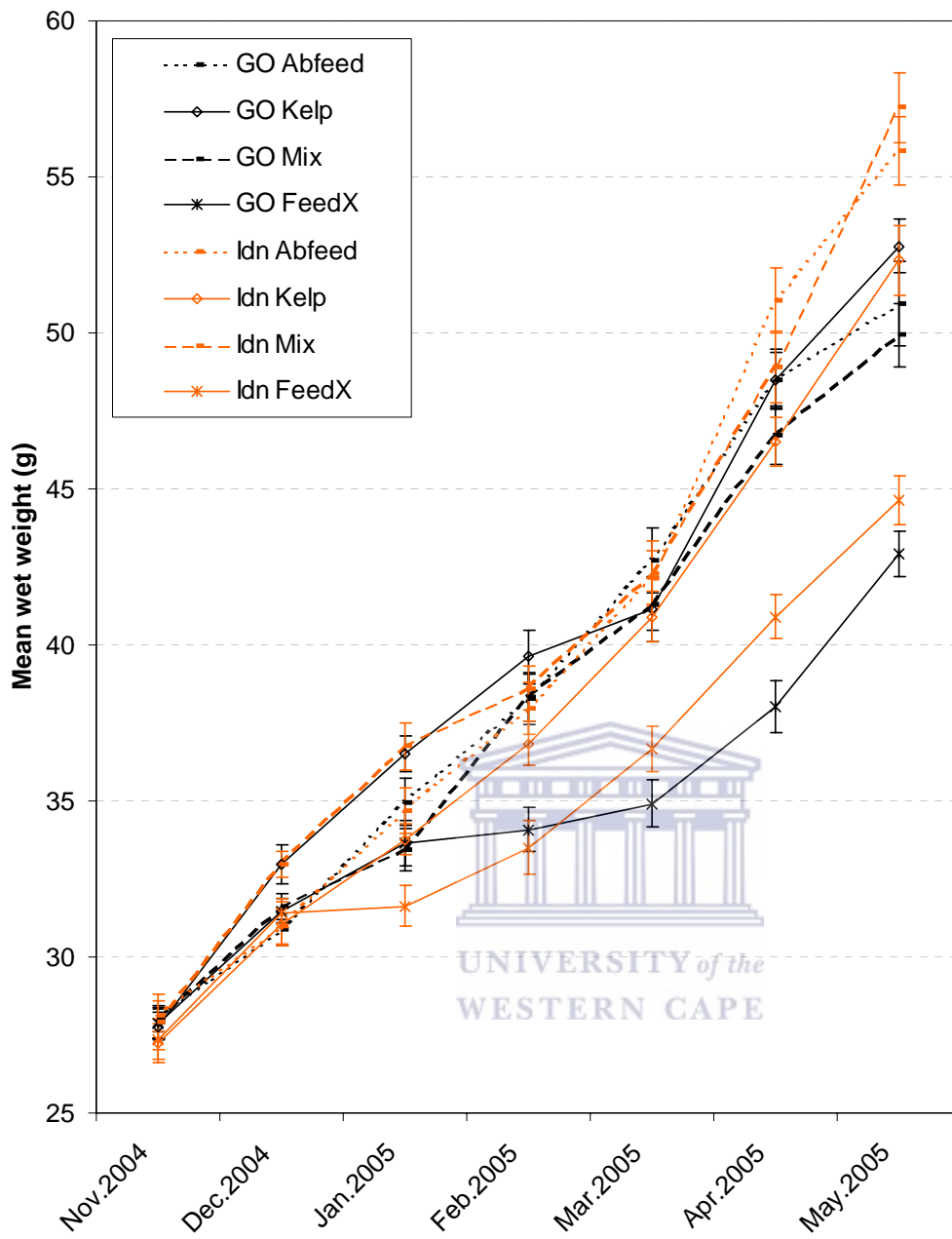


Figure 7.

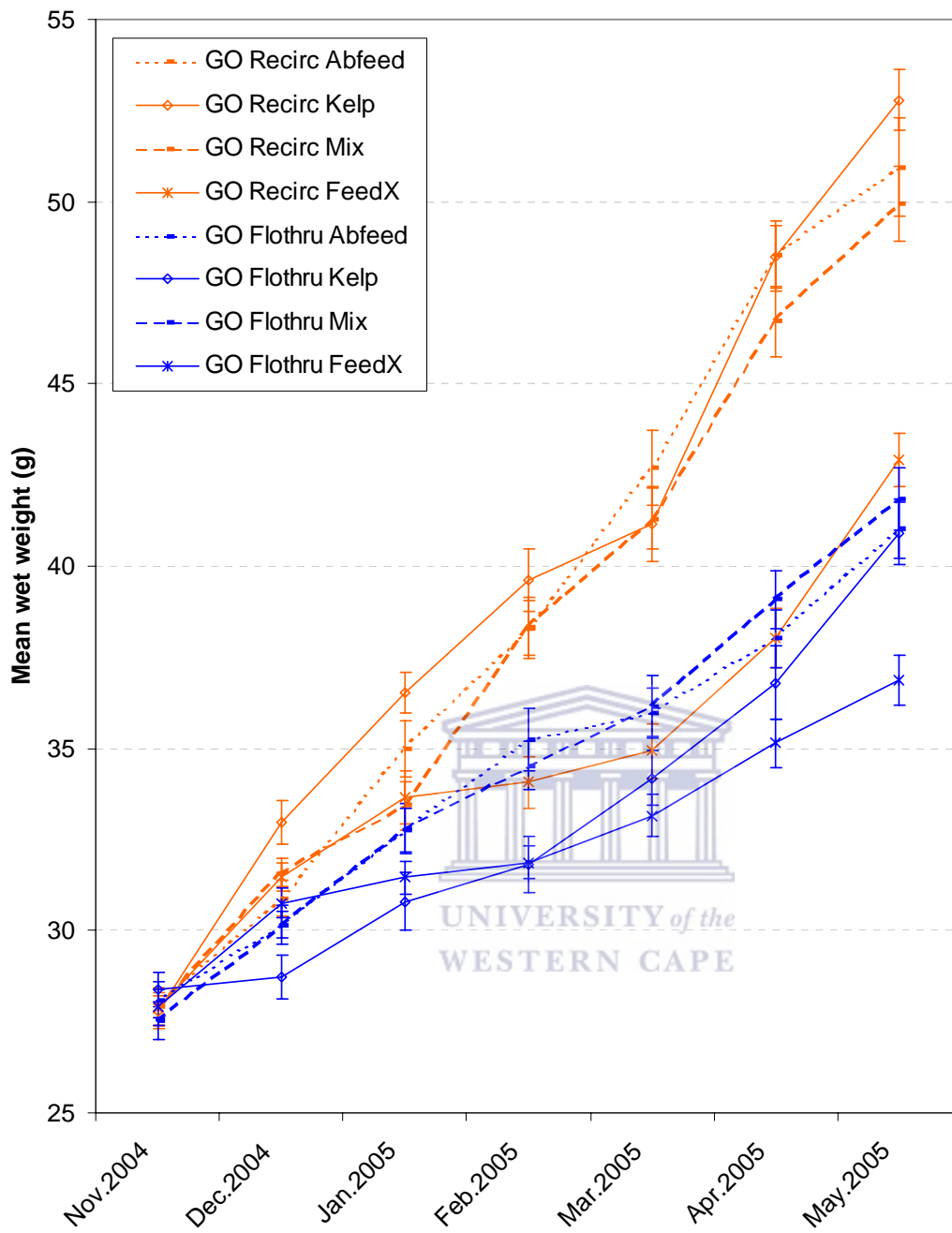
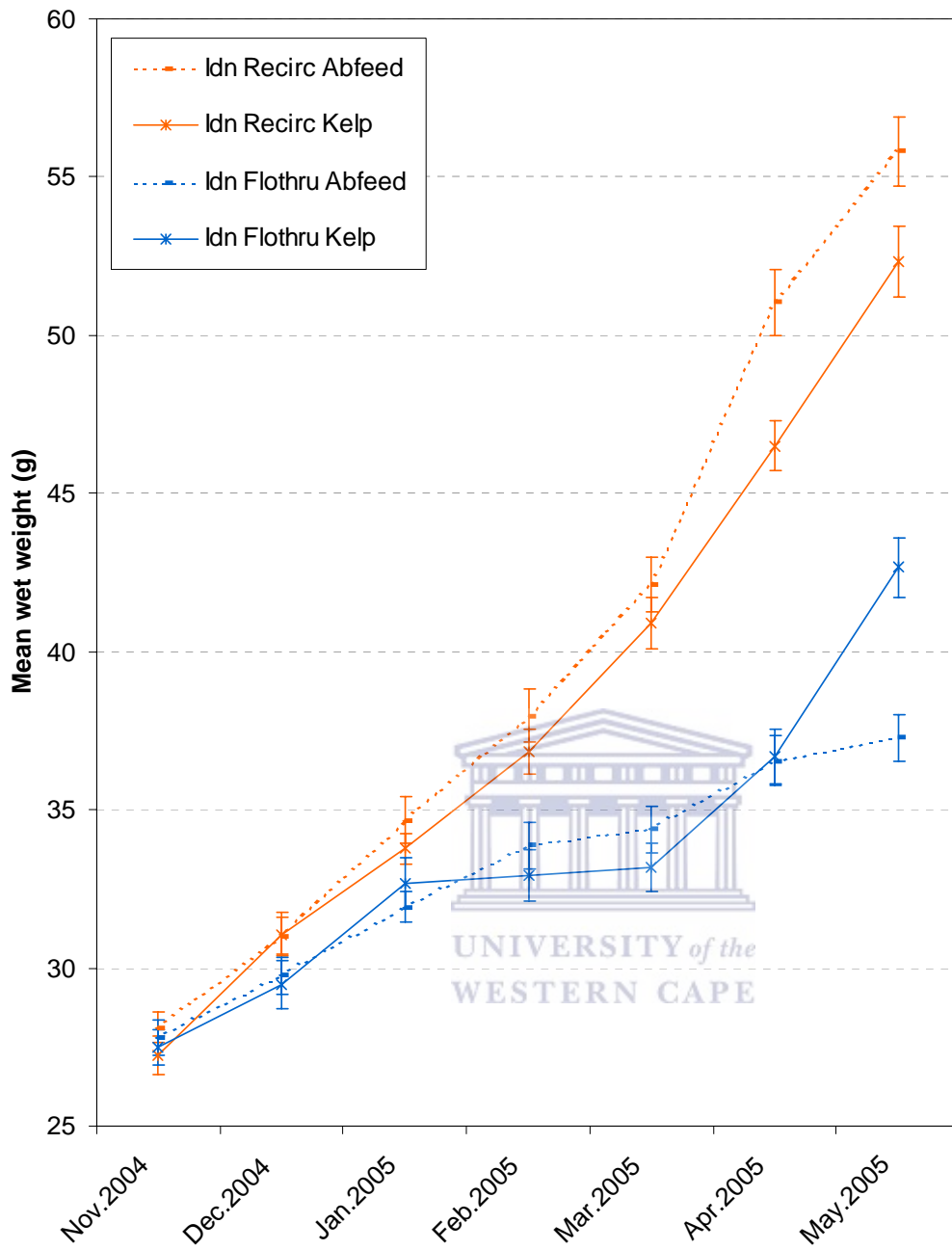


Figure 8.



4.1 General Discussion

The outcomes of this study have demonstrated that diet type is indeed an important component affecting growth of cultured abalone. Although formulated feeds generally outperform fresh seaweed diets (e.g. Bautista-Teruel *et al.* 2002), the results from this research have demonstrated the importance of fortification with fresh seaweed on the growth of post-weaning abalone. While most formulated feeds satisfy the nutritional requirements of abalone, the attractiveness of fresh seaweed as a natural diet have resulted in the enhanced growth recorded in this research.

Although no one factor could be singled out as the sole causative of the variable growth rates obtained using various formulated diets, differences in protein content and protein sources accounted for differences in the growth of abalone. This is consistent with previous studies that have shown fishmeal-based formulated feeds to perform better than the all-seaweed-based feeds (Fleming *et al.* 1996, Knauer *et al.* 1996, Bautista-Teruel *et al.* 2003). Our results could also reflect the effects of different methods used to process the fishmeal incorporated in the various fishmeal-based diets used, since Sales and Britz (2002) demonstrated that different processing methods also affect the digestibility of a diet.

Not only has this research demonstrated the effects of diet choice and supplementation (fortification) on abalone growth, but it has also highlighted the importance of external factors such as water flow-system and basket design on abalone growth. The results of this study have demonstrated that flow-systems do affect abalone growth. Although numerous external factors could be responsible, differences in water temperature seemed a likely factor resulting in the variable growth rates recorded in the two flow-systems compared. However,

the effects of temperature difference between these systems could depend on the geographical location of the farm. A flow-through system on the West Coast pumps water from the colder Atlantic Ocean whereas a similar system pumping water from the warmer Indian Ocean on the East Coast, would probably have elevated temperatures similar to those recorded in our re-circulation system.

In conclusion, all the animal-protein based diets yielded better growth rates compared to the all-seaweed based feed. Fortifying formulated diets with fresh seaweed further enhanced abalone growth. Although formulated diets increase abalone growth, using fortified diets is therefore a more commercially sensible option, since the high growth of abalone fed fortified diets shortens the normal harvest period and thus reducing farm expenses on abalone feed. However, further research is needed to investigate the effects of feed pellet dimensions and colour on abalone growth. The nutritional effects of different fishmeal sources, and the processing techniques, used in fishmeal incorporated in abalone diets also need further attention.

The re-circulation water flow-system yielded better growth rates compared to the flow-through flow-system. Basket design also affected abalone growth rates, as the interior design dictated feed access to abalone. The orientation of wholes around the different baskets determined the water circulation within each basket type. Conclusive subjective rating of basket design could not be done since there were at least two interactions between the three variables at any given time, such that basket design and diet type also affected abalone growth in both flow-systems. Further research is therefore necessary to find means of increasing basket surface area without compromising water quality.

4.2 General references

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