UNIVERSITY OF CAPE COAST

CROSSBREEDING OF FOUR POPULATIONS OF BLACK-CHINNED TILAPIA (*SAROTHERODON MELANOTHERON*) FROM GHANA TO ENHANCE AQUACULTURE PRODUCTION

BY

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DECLARATION

Candidate's Declaration

I hereby declare that this thesis is the result of my own original research and that no part of it has been presented for another degree in this university or elsewhere.

Candidate's Signature ……Date …….

Name: LAWRENCE ARMAH AHIAH

Supervisors' Declaration

We hereby declare that the preparation and presentation of the thesis were supervised in accordance with the guidelines on the supervision of thesis laid down by the University of Cape Coast.

Principal Supervisor's signature ……..Date ……

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ABSTRACT

The current study evaluated growth performance, survival rate, feed conversion ratio, heterosis, heritability, responses to selection and breeding values of four populations of *Sarotherodon melanotheron* from Fosu Lagoon, Brimsu, Baifikrom and Weija reservoirs in 4 × 4 diallel crosses aimed at producing a hybrid with high growth, reproductive and survival traits to enhance aquaculture production. Triplicates of a single-pair mating design for the diallel crosses yielded 3,840 fingerlings. These were tagged and stocked communally in both freshwater (cages) and brackish water (tanks) for 120 days using a randomized complete block design. Different doses (0, 30, 60, 90 and 120 mg/kg) of 17α-methyltestosterone were fed to black-chinned tilapia fry for 28 days to determine the effective dose for all-male sex reversal. The results indicated that five hybrids (Baifikrom × Fosu (RF), Fosu × Weija (FW), Brimsu × Weija (BW), Baifikrom × Brimsu (RB) and Brimsu × Baifikrom (BR)) were significantly ($P < 0.05$) heavier in harvest body weight compared to their parental stocks and other hybrids. Overall mean heterosis estimates indicated that seven (7) out of the twelve hybrids exhibited positive heterosis ranging from 0.17 ± 1.95 to 22.17 ± 5.52 %. Moderate to high heritability ranging from 0.29 ± 0.05 to 0.63 ± 0.21 were observed for the hybrids. The pooled genetic performance of the hybrids suggested the need to develop two separate lines for fresh and brackish water culture. However, the hybrid of Weija × Brimsu (WB) was most suitable for culture in both fresh and brackish water. The 17α-methyltestosterone test indicated that *S. melanotheron* fry fed 90 mg/kg feed for 28 days, had the highest sex ratio of 84.13 % males. For effective sex reversal in *S. melanotheron*, the hormone dosage should not exceed 90 mg/kg.

KEY WORDS

All-male tilapia

Breeding value

Heritability

Heterosis

Response to selection

Sarotherodon melanotheron

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DEDICATION

To my wife, Mrs. Jackline Ahia-Armah and children, Morishita, Monalisa, Yukero and Yukiko; and in memory of my mother, Grace Martekor Martei.

TABLE OF CONTENTS

LIST OF TABLES

Page

LIST OF FIGURES

Page

CHAPTER ONE

INTRODUCTION

Background to the study

The fisheries industry in Ghana plays an important role in the economy of the country, reducing poverty through job creation and increasing food and nutritional security. In 2017, it contributed about 6.6 % of Agriculture Gross Domestic Product (AGDP), and 1.2 % of Gross Domestic Product (GDP) (Ghana Statistical Service [GSS], 2018). In Ghana, fish accounts for over 60 % of the national dietary animal protein in diets. The fisheries sector provides direct and indirect livelihoods for about 2.7 million Ghanaians (Fisheries Commission [FC], 2015). However, fish production mainly from the marine and freshwater capture fisheries have been on the decline in recent years. Average annual fish productions from both freshwater and marine covering 2013 – 2017 (FC, 2018) have stabilized around 400,000 metric tonnes. This forms about 35 % of the annual national fish requirements during those periods (2013 – 2017), which stood at 1,153,345.16 metric tonnes in 2017. It is therefore, apparent that Ghana's capture fisheries cannot sustainably supply fish resources to meet the increasing demand as the national population increases steadily. The fish deficit is made up for through fish imports annually, which in 2017 cost over US$ 146 million (FC, 2018). On the other, annual aquaculture productions consistently increased from 32,512 tonnes in 2013 (FC, 2014) to 57,405 tonnes in 2017 (FC, 2018). Aquaculture is considered an important potential to bridging the gap between fish demand and supply in the short and long-term production. In 2017 fish culture production was just about 12 % of the total national fish output. Thus, efforts to enhance fish culture such as culture of *S. melanotheron* in salt

waters of the country will contribute to fish or tilapia production, which Béné and Heck (2005) suggested, has the potential to alleviate malnutrition, poverty and food insecurity.

Although Ghana has more than 90 brackish water bodies along 550 km coastline (Koranteng, Entsua-Mensah & Ofori-Danson, 1997), brackish water culture is rarely practiced in the country except a few cage and pond culture activities along the Ada estuary (personal observation). A major challenge to brackish water aquaculture in Ghana is absence of tested local fish species that could grow very well in these environments to generate enough profit for fish farmers.

Black-chinned tilapia (*Sarotherodon melanotheron*), a species endemic to brackish waters of West Africa has been identified by researchers as a good candidate for brackish water aquaculture. Black-chinned tilapia has been cultured in brush parks, "acadjas", in some African lagoons (Welcomme, 1972). Its potential for pond and intensive culture has also been reported (Egonifgh, Deekae & Marioghae, 1996; Pauly, 1976). However, little work has been done on the intensive culture of the species in West Africa, particularly in the area of crossbreeding and selective breeding, which could enhance its production as observed in *Oreochromis niloticus* (Hamzah et al., 2014; Ponzoni et al., 2011).

Tilapia is one of the most important fish species for freshwater aquaculture (Siddiqui & Al-Harbi, 1995) and mariculture (Hopkins, Ridha, Leclercq, Al-Ameeri, & Ahmad, 1989; Watanabe, Clark, Dunhkam, Wicklund, & Olla, 1990). According to Conte, Gammerdinger, Bartie, Penman and Kocher (2017), tilapias are the second most farmed fish species in the world and serve

as sustainable source of food for millions of people. Tilapias are suitable for various fish farming systems due to their ease of reproduction, resistance to environmental stress, disease, ability to reproduce in captivity and their acceptance of both natural and artificial feeds at all developmental stages. They are hardy (to stress and diseases) and can tolerate a wide range of environmental conditions (El-Sayed, 2006).

It has been reported (Brummett, Angoni & Pouomogne, 2004; Lind, Brummett & Ponzoni, 2012) that Africa's main challenge in fish production has been poor fisheries management and genetic erosion, and presently most aquaculture stocks in use on the continent are genetically inferior to natural stocks. It is generally acknowledged that successful aquaculture development in Africa would require an effective use and management of fish genetic resources as well as improvements in feed quality, local technical skills and marketing strategies (Lind et al., 2012; Ponzoni et al., 2011). Genetically improved strains of tilapia that are fast growing, resistant to disease and suitable for culture in a variety of fish farming environments would go a long way to meet the growing demand for fish protein in the African continent (Greer & Harvey, 2004).

According to Lutz (2006), genetic improvement is one of the most powerful and least expensive means of increasing the efficiency of organisms in culture including aquaculture. Traditional and science-based quantitative genetic approaches have been adopted to improve tilapia phenotypes. Other genetic improvement methods include chromosomal manipulations, gene transfer, physiological alteration of sex determination and genetic marker-assisted breeding (Lutz, 2006; Poompuang & Hallerman, 1997). In most

countries, traditional animal breeding approaches are still the most practical means of improving tilapia stocks for small-scale producers, using additive and non-additive gene effects (Lutz, 2006).

The most common traditional approach to genetic improvement is selective breeding, which is based on the underlying principle that some phenotypic variation is due to individual genotypes, and that a component of these genotypic influences are directly transmissible from parent to offspring. According to Lutz (2006), even in cases of high heritability, a measureable amount of phenotypic variation is needed to enhance traits through selection. Inbreeding and random genetic drift result in lower heritability due to reduced genetic variation. Therefore, selection is the best approach for tilapia genetic improvement where wide genetic variation exists (Lutz, 2006). According to Ponzoni, Nguyen and Khaw (2007), selective breeding has certain advantages over other genetic approaches, such that continuous genetic gains can be transmitted from one generation to the next, and gains in the basic stock (e.g. in a hatchery) can be multiplied and expressed in millions of individuals in the production sector.

Due to the growing importance of tilapia in global aquaculture, the attention and diversity of efforts to improve the genetic baseline of species have intensified (Ponzoni, et al., 2007). These have resulted in global aquaculture production of tilapia increasing from 28,000 to more than 3 million metric tons from 1970 to 2010 (Fitzsimmons, 2010), with Africa the major source of the fish contributing only 19 %. According to Kobayashi et al. (2015), global aquaculture production has been increasing continually for the past three decades. Available records show that global aquaculture production expanded

at an average annual rate of over 8 percent with production increasing from 5.2 million metric tonnes in 1981 to 73.8 million metric tonnes in 2014 (FAO, 2016). On the other hand, the global capture fisheries production has remained stable around 93.4 million tonnes (FAO, 2016) and it is no more capable of sustainably providing fisheries resources to meet increasing demand.

Problem statement and justification

Fish farming started in Ghana in the 1950s (MacPherson & Agyenim-Boateng, 1991). This has been limited to ponds, pens and cages of freshwater fish with very little attention by government to brackish water and marine aquaculture. The main challenges to the development of brackish water culture have been lack of tested local fish species, quality fish feed and management skills. Other constraints limiting the growth of aquaculture industry in Africa and Ghana in particular are poor growth performance and low survival rates of several cultured species (Brummett & Ponzoni, 2009). It is therefore, important to develop genetic improvement programs for brackish water fish species such as the endemic black-chinned tilapia for coastal aquaculture.

Despite the numerous coastal water bodies in Ghana, culture of brackish water fish has not been demonstrated as feasible in the country. There is therefore, the need to assess the suitability of black-chinned tilapia for aquaculture and to establish suitable environments and culture systems that will support hatchery production of black-chinned tilapia in Ghana. Ekau and Blay (2000) reported on black-chinned tilapia as important fish species in the catch of coastal water bodies in Ghana comprising 59 – 90 % of fish landings from lagoons (Blay & Asabere-Ameyaw, 1993; Mensah, 1979; Pauly, 1976;

Welcomme, 1972). It has the potential for aquaculture in brackish water environments. Arizi, Obodai and Aggrey-Fynn (2015) reported size of 19.8 cm total length for *S. melanotheron* sampled from the Dominli lagoon in Ghana. In freshwater, the same species measuring 17.6 cm standard length was reported from the samples harvested from the Brimsu reservoir (Mireku, Blay & Yankson, 2016). It also spawns all year-round, making it a good candidate for aquaculture (Trewevas, 1983).

Fish production can be increased by improving the quality of the culture environment, and growing genetically improved fish (Tave, 1995). One of the principal means of improving fish growth performance is through crossbreeding to produce hybrids with improved growth characteristics (Kirpichnikov, 1971; Zaki, Essa, Soliman & Kosba, 1987). Development of the Akosombo strain of *O. niloticus* (Akosombo strain) by selective breeding (Attipoe, 2006) has resulted in an increase in aquaculture production in Ghana in recent times. However, most fish farmers at Ada, in the Greater Accra Region, using the estuary (brackish water) for cage farming recorded more than 30 % mortality in the first two weeks of stocking the Akosombo strain in their cages (personal observation). There is therefore, the need to generate adequate information (culture approach, species diversification, more variety that is productive, commercial culture, etc.) through research to promote the alternative of black-chinned tilapia in addition to *O. niloticus*.

Objectives of the study

The main goal of the study is to produce a hybrid of *S. melanotheron* (using strains from Brimsu, Baifikrom, Weija and Fosu water bodies) with high reproductive capacity, higher growth and survival rates. The specific objectives include: (i) To assess Feed Conversion Ratio (FCR) of the hybrids under study. (ii) To determine magnitude of heterosis (hybrid-vigour) in a complete diallel cross of the four populations. (iii) To evaluate the genetic parameters (response to selection, heritability and breeding values) of the hybrids under culture. (iv) To determine fecundity of the hybrids under study. (v) To determine the concentration of 17α-Methyltestosterone for production of all-male individuals of the species.

Significance of the Study

The results of the study would benefit coastal communities and Ghana in general as the culturing of *S. melanotheron* varieties with better growth and reproductive performance would enhance yields in production systems.

Organization of the Study

This thesis consists of six chapters. Chapter One introduced the background of the study and outlined its objectives. Chapter Two is a review of literature in context of the study. It discusses previous studies on crossbreeding and selective breeding in relation to the current study. Chapter Three focuses on research methodology and design. It gives the materials and methods employed in the study to achieve the set objectives. The chapter also throws light on

different methods used in sampling, recording and analyzing the collected data. Chapter Four presents the results of the experiments carried out in figures and tables. Interpretation and discussion of the results are placed in Chapter five of this thesis. Conclusions and recommendations are covered in Chapter Six. Finally, references cited in the thesis and appendices are placed at the end of this thesis.

CHAPTER TWO

LITERATURE REVIEW

This chapter covers the theoretical basis for the study on crossbreeding of four populations of black-chinned tilapia (*Sarotherodon melanotheron*) from Ghana**.** It is crossbreeding and selection response research aimed at developing an improved strain of the black-chinned tilapia for culture in both brackish water and freshwater systems. The chapter reviews previous studies on crossbreeding and selective breeding by other researchers and their relation to the current study. It also looks at key concepts and theories of crossbreeding of fish, and how they impact the objectives of this study.

Distribution of *Sarotherodon melanotheron*

Trewavas (1983) described five subspecies of *S. melanotheron* based on morphological characteristics, and these are distributed in West Africa as follows: *S. m. paludinosus* (Trewavas, 1983) found in freshwater bodies in Dakar; S. *m. heudelotii* (Dumeril, 1859) found from Senegal to Guinea; *S. m. leonensis* (Thys van den Audenaerde, 1971) located from Sierra Leone to Liberia; S. *m. melanotheron* (Rüppel, 1852) from Cote d'Ivoire to Cameroon and S. *m. nigripinnis* from Equatorial Guinea to the mouth of the Congo River (Falk, Teugels & Abban, 2000). Further morphological and genetic analyses (Adépo-Gourène, Pouyaud, Teugels, Hansen & Agnèse, 1998; Pouyaud & Agnèse, 1995; Teugels & Hanssens, 1995) had however, questioned the validity of some of the taxa and indicated the necessity to refine the recommended intraspecific classification by Trewavas (1983). Falk, Teugels and Abban (2003) used morphometric, allozyme, globin chain and cytochrome b analyses

to revise the subspecies complex of the black-chinned tilapia. They validated and recognized three subspecies as: *S. m. melanotheron* (Ivory Coast to Benin), *S. m. heudelotii* (Senegal to Guinea) and *S. m. leonensis* (Sierra Leone to Liberia). The subspecies *S. m. nigripinnis* (Trewavas, 1983) was raised to the species level as *S. nigripinnis* (Falk, Teugels, & Abban, 2003), whereas *S. m. paludinosus* and *S. m. heudelotii* subspecies were merged into one subspecies, *S. m. heudelotii*. The current study used the subspecies *Sarotherodon melanotheron melanotheron* harvested from Brimsu, Baifikrom and Weija reservoirs and Fosu Lagoon.

Reproductive biology of the species

Aspects of the biology and fishery of black-chinned tilapia in brackish water and coastal reservoirs in West Africa have been reported (Blay & Asabere-Ameyaw, 1993; Eyeson, 1992; Mireku, Blay & Yankson, 2016; Oribhabor & Adisa-Bolanta, 2009; Ugwumba, 1988). Although the species is said to be predominantly a paternal oral brooder, occasional residual oral brooding has been reported (Eyeson, 1992). According to Trewavas (1983) *S. melanotheron* matures at a very small body size (5.1 – 8.8 cm total body length), which is a trait of an opportunistic invasive species. However, Mireku, Blay and Yankson (2016) observed that *S. melanotheron* matures at a much bigger size in freshwater compared to brackish water. inimid

Under favourable environmental conditions, black-chinned tilapia was reported to spawn once every 22 days (Eyeson, 1979); the species, however, spawns throughout the year (Trewavas, 1983). The female fish produce about 200 – 900 eggs in batches but the number of eggs usually incubated by the male

ranges from 20 – 700 depending on the size of the brooder (Eyeson, 1979; Trewavas, 1983).

Crossbreeding

Crossbreeding is a process of mating different species (interspecific hybridization) or strains of the same species (intraspecific hybridization) to produce offspring with superior traits than their parents. Crossbreeding or hybridization has been used to improve some traits, which were considered desirable in some aquaculture species. Ayles and Baker (1983) investigated genetic differences in growth and survival between strains and hybrids of rainbow trout (*Salmo gairdneri*) and reported significant hybrid vigour for both growth and survival in some of the hybrids. Essa and Haroun (1998) reported enhanced growth performance and survival rates in hybrids of *Oreochromis* species produced through interspecific hybridization. They evaluated the performance of the hybrids produced by the crossbreeding of a) Nile tilapia (*Oreochromis niloticus*), which was considered a fast growing species (Bardach, Ryther & Mclarney, 1972), b) Blue tilapia (*Oreochromis aureus*), considered as cold-tolerant species (Behrends & Smitherman, 1984) and c) Red tilapia, a hybrid produced by the mating of *O. niloticus* and *Oreochromis mossambicus* (Liao & Chang, 1983). Their offspring cultured in freshwater ponds exhibited improved growth and survival rates compared to their parents (positive heterosis). The researchers also demonstrated that backcrossing red tilapia with males of *O. aureus* led to the transmission of its cold-tolerant trait to red tilapia. Suburamanian, Heo and Chellam (2015) conducted genetic assessment for growth performance in diallel cross of wild and cultured giant

freshwater prawn, *Macrobrachium rosenbergii*. They reported additive genetic effect with increased body weight in the hybrid when compared to the purebreed.

Interspecific hybridization has been implicated in improved growth and other desirable characters in a variety of species. The hybrid striped bass, female white bass (*Morone chrysop*) crossed with male striped bass (*Morone saxatilis*), was reported to have exhibited faster growth, aggressive feeding behaviour and wider environmental tolerance compared to the parental species (Bosworth, Libey, & Notter, 1997). Dunham and Brummett (1999) worked on channel catfish and reported that growth improvement through interspecific hybridization was three times better than through mass selection. They further observed that growth rate of the channel catfish, *Ictalurus punctatus* female crossed with Blue Catfish, *Ictalurus furcatus* male, was 35 % faster than the control, and better than all three groups of channel catfish used in the experiment.

Cai, Li and Ma (2004) investigated resistance to disease caused by *Aeromonas sobria* in Nile tilapia (*Oreochromis niloticus*), Blue tilapia (*Oreochromis aureus*) and their hybrids. They reported that the hybrid, produced through the mating of female *O. niloticus* and male *O. aureus* exhibited higher resistance to the disease causing bacteria, *A. sobria* as compared to any of the parental species.

All-male tilapia production through hybridization was reviewed by Wohlfarth (1994). The review covered publications from 1960 – 1993, involving the hybridization of *Oreochromis* species - *O. mossambicus* × *O. hornorum*; *O. niloticus* × *O. hornorum*; *O. niloticus* × *O. macrochir* and *O.*

niloticus × *O. aureus*. The hybrids from all these crosses were all-male but could not be propagated commercially due to either difficulty in producing large quantities, darker in colour or low percentage male ratio.

In general, crossbreeding is used to produce superior fish level of certainty of F1 generation for grow-out (production fish) whereas selective breeding is used to create superior brood fish. In the current study, crossbreeding was used to transfer desirable growth traits into indigenous *S. melanotheron* strains.

Heterosis

Heterosis or hybrid vigour is a phenomenon in which the first generation (F_1) progeny of different species or strains of the same species exhibit greater or lesser trait performance (in terms of length, weight, fecundity, fertility or development) than the best of the two parents or the mean of the two parents or the purebreeds of the cross combination (Falconer & Mackay, 1996; Granier, Audet & Bernatchez, 2011; Nielsen et al., 2010; Nguenga, Teugels & Ollevier, 2000). Reif, Hahn and Melchinger (2012) reported that heterosis results from dominance, over dominance and epistasis effects. They observed that heterosis depends on dominance such that loci without dominance cannot exhibit heterosis. Earlier researchers, Birchler, Auger and Riddle (2003) explained that dominance comes into play when heterosis is contributed by favourable alleles of both parents, whereas over-dominance is a condition where loci in the heterozygous state are superior to that of the parents (Hochholdinger & Hoecker, 2007). Reif et al. (2012) further observed that heterosis depends also on the genetic distance between the breeds. The level of heterosis, therefore,

becomes higher if the genetic distance between the breeds becomes greater. According to Nuruzzaman et al. (2002), the magnitude of heterosis may be positive or negative, and further explained that positive heterosis is desired for yield and negative heterosis for early maturity.

Most studies on heterosis in tilapia have focused on growth (Bentsen et al., 1998; Maluwa & Gjerde, 2006; Nguyen, Pongthana & Ponzoni, 2009; Rezk et al., 2009). El-Zaeem, Ahmed, Salam and Darwish (2012) reported that the hybrid (female Red tilapia × male Nile tilapia) reared at 16 and 32 ppt of salinity exhibited higher positive heterosis compared to the reciprocal hybrid (female Nile tilapia × male Red tilapia) for weight gain and specific growth rate at both salinities. Essa and Haroun (1998) investigated the breeding characteristics, growth performance, survival rate and feed conversion ratio of Nile tilapia (*Oreochromis niloticus*), Blue tilapia (*Oreochromis aureus*), Red tilapia (*Oreochromis sp*.) and their crosses. They reported that the hybrids of *O. niloticus* females × *O. aureus* males; red tilapia females × *O. niloticus* males, *O. aureus* females × *O. niloticus* males, and red tilapia females × *O. aureus* males exhibited positive heterosis values in growth parameters and survival rates. Based on the positive heterosis recorded, these hybrids were adjudged to be of high potential commercial value for fish farming. On the other hand, Nguyen, Pongthana and Ponzoni (2009) investigated heterosis, direct additive genetic and general reciprocal effects from a complete diallel cross involving four strains of red tilapia, *Oreochromis spp* from Malaysia, Stirling, Taiwan and Thailand cultured in both freshwater and brackish water environments. They found out that the average heterosis for body weight across the testing environments was low (4.2 %) and the average of all crossbreeds was not

statistically different from the mean of pure strains. They concluded that the low level of heterosis for harvest weight in their study suggests that performance improvement of red tilapia could be effectively based on the exploitation of additive genetic variation (i.e. through selective breeding rather than crossbreeding).

Selective breeding

Selective breeding is a process of choosing for mating fish that exhibit the best desirable traits (e.g. fast growth performance, high fecundity, salt-tolerance, etc.) expected to be transferred to the next generation to improve the production performance, quality of fish and the breeding value of that population (Tave, 1995; WorldFish, 2004). Lind (2015) studied strain development and improvement in tilapia and showed that there are several selective breeding techniques with increasing complexities. These vary from individual (mass) selection, cohort selection, within-family selection, between-family selection, combined selection and genomic selection. These techniques have been applied in the culture of Coho salmon, *Oncorhynchus kisutch* (Hershberger, Myers, Iwamoto, Mcauley & Saxton, 1990; Nair, Salin, Rajul & Sebastian, 2006), Atlantic salmon, *Salmo salar* (Gjedrem, 2000; Quinton, McMillan & Glebe, 2005) and Nile tilapia, *Oreochromis niloticus* (Bentsen et al., 1998; Eknath et al., 2007).

Within-family selection is a phenomenon where each family is considered a temporary sub-population and selection occurs independently within each family. Bolivar and Newkirk (2000) evaluated response to selection for body weight of Nile Tilapia (*Oreochromis niloticus*) in different culture

environments using the within-family selection approach for 12 generations. They reported that the selected group produced using within-family selection, led to improved growth performance and selection response in tanks, hapas and pond environments. Uraiwan and Doyle (1986) assessed the different selection procedures for tilapia (*O. niloticus*) and observed that within-family selection eliminates large non-genetic components from the variation, and economizes breeding space.

Heritability

Heritability is defined as the proportion of total variance that is genetic and transferrable to offspring (Burton, 1987). According to Wray and Visscher (2008), heritability refers to how much of variation is due to genetic variation of the material relating between a parent and its offspring. They further categorized heritability into narrow sense and broad sense: when the proportion of phenotypic variance occurs as a result of additive genetic variation then it is referred to as narrow sense heritability, whereas broad sense heritability occurs when the proportion of phenotypic variation is due to total genetic variation including both dominance and epistasis effects.

Heritability of growth rate has been studied in several fish species of interest to aquaculture. Saillant, Dupont-Nivet, Haffray and Chatain (2006) worked on Sea bass (*Dicentrarchus labrax*) and reported that heritability estimates tended to increase with the age of the fish. Vandeputte et al. (2004) studied heritability of growth-rated traits in juvenile common carp (*Cyprinus carpio* L.) and observed that heritability for increased weight gain was successful in juvenile carp, using indirect selection for length. Gjedrem (2000)

evaluated heritability in salmonid fish and observed that it was possible to obtain large responses from selection for growth rate. Ma, Saillant, Gatlin III and Gold (2008) worked on heritability estimates on growth traits in larval and early juvenile of Red drum (*Sciaenops ocellatus*) and indicated that selection for faster growth in red drum would be successful at both larval and juvenile stages. According to Saillant, Dupont-Nivet, Haffray and Chatain (2006) the optimization of selection requires knowledge of genetic parameters of characters because best selection strategies depend mainly on heritability of individual characters and genetic correlations between these characters.

Breeding value

Breeding value of an animal is simply the value of the animal as a breeder or parent. According to the United States Department of Agriculture (2014), breeding value is the estimated value expressing the ability of a parent to pass on superior traits to its offspring and it is used for ranking breeding performance of the parent relative to the population average. Conner and Hartl (2004) defined breeding value as the effect of individual genes in a parent on the value of the trait in its offspring. They further explained that the breeding value for a given sire is usually estimated as two times the deviation of the mean of its offspring from the population mean in a paternal half-sib mating design. The breeding value is usually twice the deviation from the mean because the sire only contributes half of the offspring's alleles, and the other half comes from several randomly chosen dams in the population. According to Amari (2016), estimated breeding values provide the most reliable information on how an animal might breed, and this estimated breeding value enables the

comparison of individuals of the same breed from different populations; they are usually expressed in the units of measurement.

Luan, Olesen, Odegard, Kolstad and Nguyen (2008) investigated genotype by environment interaction for harvest body weight and survival of *O. niloticus* in brackish and freshwater ponds. They used the ranking of breeding values for selection of best performing strains for F_1 – F_3 generations and recommended that separate breeding programs should be considered for *O. niloticus* in fresh and brackish water farming. Khaw et al. (2010) used estimated breeding values for live weight to create the select (high breeding values) and control (average breeding values) lines in the experiment conducted for the genetic analysis of the GIFT strain (*Oreochromis niloticus*) in Malaysia.

Maternal and common environmental effect

Wolf and Wade (2009) defined maternal effects as the causal influence of the maternal genotype or phenotype on the offspring's phenotype. Studies of maternal effects on growth in other fishes indicate that these effects occur primarily during early life stages and tend to disappear within few months of growth (Garcia de Leon, Canonne, Quillet, Bonhomme & Chatain, 1998; Herbinger & Cook, 1995).

Common environmental effect is defined as the causal influence of environmental factors or non-heritable factors on the phenotypic expression of the offspring (Landy & Travis, 2018; Tosh, Garber, Trippel & Robinson, 2010). According to Saillant et al. (2006) early common environmental effects seem to limit the potential for selection for increased growth rate in several fish species of interest for aquaculture (Knibb, 1998; Gjedrem, 2000) because it can be

confused with genetic values thus preventing accurate evaluation of these values unless large numbers of replicate are used (Vandeputte et al., 2001).

There are several methods designed to minimize environmental and maternal effects on phenotypic expression so that the genetic effects can be estimated accurately (Crispo, 2008; Kawecki & Ebert, 2004). Garcia de Leon et al. (1998) used microsatellite markers for breed identification in breeding of Sea bass (*Dicentrarchus labrax*) and showed that microsatellites are valuable tools that greatly reduce the impact of common environmental effects and number of replicate ponds needed for selective breeding programs. Haffray et al. (2012) conducted an experiment to quantify and to minimize maternal effects on growth in rainbow trout. All the specimens were raised communally from the eyed stage and DNA parentage assignment was used to determine the families. They reported that the procedure efficiently reduced maternal effects, and increased the heritability for growth substantially. According to WorldFish (2004) the common environmental effects can be reduced by tagging the families and rear them communally in ponds or cages. Falconer and Mackay (1996) pointed out that the genetic and environmental sources of resemblance between families can be separated statistically by comparing the covariance between selected groups of individuals or incorporating the environmental effects in the animal model for the estimation of the genetic parameters.

Fecundity

Fecundity of fish is one of the essential components of fishery biology, which has direct bearing on fish production, stock recruitment and stock management (Qadri, et al., 2015). According to Snyder (1983), fecundity is

often referred to as total, absolute or individual fecundity, and usually defined as the number of ripening oocytes and mature ova or eggs just prior to spawning.

Fecundity of fish is reported to be affected by environmental factors such as availability and quality of food, intensity of predation and other density-dependent factors (Hartmann & Quoss, 1993), the size of the inhabited water body, length of the breeding season and number of broods produced per year (Noakes & Balon, 1982). The time or season of spawning (Jobling, 1995; Kazakov, 1981) and distance of migration to the spawning ground (Beacham & Murray, 1993) also affect fecundity and egg size.

Asaad, Traifalgar and Serrano (2013) studied maternal size effect on fecundity of *Oreochromis mossambicus* (Peters) in freshwater. They reported that fecundity increased with fish size from a mean of 1,465 eggs in small-sized female broodstock (average = 23.28 g) to 4,105 eggs for large-sized female broodstock (average = 350.15 g) but the relative fecundity was significantly higher in bigger-sized *O. mossambicus* female broodstock. They further observed that absolute fecundity and relative fecundity of saline-tolerant *O. mossambicus* were influenced by the maternal body size, body weight and gonad weight, and were linearly correlated. Bone, Marshall and Blaxter (1995) reported that in general fecundity increases with age and size of the fish. Legendre and Ecoutin (1989) evaluated the suitability of brackish water tilapia species (*Tilapia guineensis* and *Sarotherodon melanotheron*) for lagoon aquaculture by comparing the reproductive biology of both natural and cultural populations in the Ebrie lagoon (Ivory Coast). They reported that in rearing enclosures, both species attained sexual maturity at a smaller size, and produce smaller size but more numerous oocytes than in the wild. Legendre and Ecoutin

(1989) observed that in *S. melanotheron*, which is a male mouthbreeder, the number of brooded eggs or fry is positively related to the male body weight.

There was paucity of information on the effect of crossbreeding on fecundity of *S. melanotheron*.

Sex reversal

Sex reversal is a technique of changing sexes of fish from one sex to another (e.g. female to male or vice versa) by administering synthetic steroid hormones (e.g. 17α-methyltestosterone) before or during the period of sexual differentiation (Ferdous & Ali, 2011). According to Phelps and Popma (2000) hormone treatment does not modify the genetic make-up of the fish but directs the expression of the phenotype, and therefore defined sex reversal as the alteration of the phenotype (sex) by administration of synthetic sex hormones. According to Gale, Fitzpatrick, Lucero, Contreras-Sanchez and Schreck (1999) the hormones act as sex-inversion agents by functionally masculinizing or feminizing individual fish in the population. Andersen, Holbech, Gessbo, Norrgen and Petersen (2003) observed that at the time of hatching Zebra fish (*Danio rerio*) fry were sexually undeveloped and during the early period of gonadal differentiation, changes in sex hormone concentration affects the phenotypic sex irrespective of the genetic sex.

Ferdous and Ali (2011) evaluated the optimum dose rate of 17α-methyltestosterone (MT) treatment for sex reversal and its effect on growth performance of Nile tilapia (*Oreochromis niloticus*). They treated the fry with MT dosages of 0 mg/kg (control), 40 mg/kg, 50 mg/kg, 60 mg/kg and 70 mg/kg *ad libitum* for 28 days. Their results showed that each hormone-treated group

gave a significantly higher male to female ratio, while the control group showed normal male to female ratio. The results established that 17α-MT had substantial effect in directing gonadal sex differentiation of *O. niloticus* towards males depending on the dosage involved in the treatment. The researchers observed that the maximum male population (94.28 %) of *O. niloticus* was obtained at the dose of 60 mg MT/kg of feed for 28 days and concluded that 60 mg/kg MT was the optimum effective dose for sex reversal in *O. niloticus.* However, in a similar assessment carried out on *O. niloticus* by Okoko (1996) reported the optimum dosage of 30 mg/kg (99.3 % males), while 60 mg/kg recorded 97 % males. In another comparative study on *Oreochromis niloticus*, Shepperd (1984) reported an equal performance of 98 % males for both 30 and 60 mg/kg MT for the 28 days treatment. In the same studies, Shepperd (1984) reported 94 % and 91 % males respectively for 30 and 60 mg/kg MT for Red tilapia (Hybrid, *O. mossambicus* × *O. hornorum*). Green and Teichert-Coddington (1994) worked on *O. niloticus* using 60 mg/kg of 17α-MT and had 96.8 % males. Phelps, Cole and Katz (1992) reported similar results (97.8 % males) when 60 mg/kg 17α-MT was administered to *O. niloticus*. Guerrero (1975) evaluated the effect of the hormone, 17α-MT on the treatment of *Oreochromis aureus* for sex reversal. He administered doses of 15, 30 and 60 mg/kg for 18 days, and the 30 mg/kg had the highest percentage conversion of 98 % males. Nakamura (1975) demonstrated that at high doses the efficacy of 17α-MT in reversing the genotypic female fish to a phenotypic male was lowered (and vice versa) as demonstrated in the study with *Oreochromis mossambicus.* Nakamura (1975) reported that 50 mg/kg MT treatment of *O. mossambicus* gave 100 % males conversion, whereas 1000 mg/kg gave 61 %

males in the same species. The results from McGeachin, Robinson and Neil (1987) on the same *O. mossambicus* species when treated with 60, 90 and 120 mg/kg MT for 22 days gave 99, 98, and 96 % males respectively. On the other hand, Varadaraj and Pandian (1989) reported that doses of 5, 10, 20, 30 and 40 mg/kg MT when administered *ad libitum* to *O. mossambicus* for 19 days gave 100 % male conversion in each case.

There are few reports on the use of other androgen-base hormones for the treatment of tilapia species for sex reversal. Guerrero (1975) examined the effect of the hormone, 1-dehydrotestosterone in the treatment of *Oreochromis aureus* at 15, 30 and 60 mg/kg concentrations for sex inversion, and the results were 69, 59 and 44 % males respectively. Guerrero (1975) again in the same experiment tested the hormone, ethynyltestosterone on *O. aureus* at 15, 30 and 60 mg/kg doses for sex reversal, the results were 85, 98 and 100 % male conversion respectively. Phelps, Cole and Katz (1992) evaluated fluoxymesterone at doses of 1, 5 and 25 mg/kg on *O. niloticus* for sex inversion, and obtained 87.3, 100 and 100 % males respectively for that particular androgen. The hormone, mestanolone was also tested on *O. niloticus* at 5, 10 and 20 mg/kg for sex reversal; the results were 99.5, 97.0 and 99.0 % males (Soto, 1992). Guerrero and Guerrero (1993) worked on *O. mossambicus* using the hormone, mibolerone at levels of 1.5, 1.75 and 2.0 mg/kg, and the results were 84, 88 and 94 % males respectively. Varadaraj (1990) also tested the androgen-based hormone, 19-norethisterone acetate for sex reversal in *O. mossambicus* at a concentration of 1 mg/kg and the outcome was 52 % males. Galvez, Morrison and Phelps (1996) studied sex reversal in *Oreochromis*

aureus using 25, 50 and 100 mg/kg of trenbolone acetate, and the outcome was 98.3, 99.3 and 99.0 % males respectively.

Studies on the use of synthetic androgens in sex reversal of tilapia show that the steroid usually have both androgenic and anabolic effect on fish (Dan & Little, 2000; Little, Bhujel & Pham, 2003; Mair, Abucay, Beardmore & Skibinski, 1995; Phelps & Popma, 2000). El-Greisy and El-Gamal (2012) worked on *Oreochromis niloticus* using 17α-methyltestosterone at 40, 60 and 80 mg/kg to produce sex reversed population and evaluated them for growth performance, survival, sex ratio and sex stability after 15, 35, 75 and 365 days of culture in freshwater. The results indicated that the maximum sex ratios of males (97 %) and highest mean harvest body weight (215.0 ± 12.8 g) were recorded at 60 mg of 17α-MT/kg of diet after 365 days. Generally, the values of growth parameters recorded were always higher for those fish treated with the 17α-MT/kg diet compared to the control. They attributed the high growth performance of the hormone-treated fish to anabolic effect of the steroid.

However, according to Phelps and Popma (2000) any enhanced growth of sex reversed tilapia, as a result of androgen treatment, is more related to the superior growth of males (naturally metabolic energy is channelled towards growth) than the usual anabolic effect related to enhanced protein synthesis and increase in muscle mass. Hanson, Smitherman, Shelton and Dunham (1983) worked on *O. niloticus* and evaluated the growth performance of sex-reversed males, hybridized males and hand-sexed males (using the genital papilla). Both sets of sex-reversed and hybridized male fish (99.4% and 100% male respectively) grew larger than the hand-sexed fish (93.8 % male), but the hand-sexed male population had 6.2 % females which could have lowered the mean

final body weight. Green and Teichert-Coddington (1994) assessed growth and survival performance of 17α-MT treated and untreated Nile tilapia (*Oreochromis niloticus*) in the fry treatment, nursery and grow-out stages under semi-intensive conditions in earthen ponds. They found no significant difference in growth in any of the stages. Tuan, Little and Mair (1998) also found no difference in growth between hybridized all male population (96.2 % male) and 17α-MT sex reversed male (99.3 % male) populations. However, in a second experiment, Tuan et al. (1998) found out that the sex-reversed males (100 % males) grew larger compared to the hybridized all males, which were made up of only 82.6 % males.

There is paucity of information on sex reversal of *S. melanotheron* regarding the optimum dosage requirement. Apenuvor (2014) assessed 30, 60 and 120 mg 17α-MT/kg feed on *S. melanotheron* strain from the Fosu Lagoon. The results indicated that 120 mg 17α-MT gave the highest percentage male of 92.7. However, the methodology used by Apenuvor (2014) left more questions unanswered. For example, how pure were the fry of *S. melanotheron* harvested from the midst of several other species in that Lagoon where they were harvested for sex reversal experiment?

Effects of water quality on growth performance of *S. melanotheron*

According to Richie and Garling (2003), fish are sensitive to water quality and therefore, feeding should be reduced or stopped if water quality falls below certain optimum levels. Some of the critical water quality parameters that must be monitored in any aquaculture system include pH, temperature, dissolved oxygen (DO), salinity among others.

Effects of pH

The pH of water is one of the most common environmental tests that are usually conducted on fish ponds. This measures the hydrogen ion concentration in the water body. Boyd (1998) defined pH as a measure of whether water is acidic or basic. Chakroff (1976) recommended pH level of 6.5 – 9.0 in fish ponds to ensure best growth performance of *O. niloticus*. According to Schofield (1976), acidic water destroys the gill tissues of *O. niloticus*, causes inflammation and increases mucus secretion in the gills and at pH below 4, the pond water becomes more acidic and fish can die as a result. Ingthamjitr (2003) observed that, when the pond water pH drops to 4 – 5, there were practically no reproduction and poor growth performance, which may be due to poor feeding in fish, and fish kill can occur when the pond pH goes above 11.0. However, Ross (2000) reported that tilapias can tolerate a pH range of 3.7 to 11 for short periods, but best growth rates are achieved between pH of 7 to 9. Popma and Masser (1999) also observed that tilapia can survive pH ranging from 5 to 10 but best growth performance can be achieved at pH range of 6 to 9.

Effects of dissolved oxygen

Dissolved oxygen (DO) is very essential for growth and survival of a fish because it affects respiration as well as nitrite and ammonia toxicity (Nehemia, Maganira & Rumisha, 2012). Mallya (2007) observed that as DO level drops below 5 mg/l, respiration and feeding activities in fish decreases. According to Tom (1998), the low level of DO can lead to decrease in growth rate and resistance to disease in fish. Additionally, fish are unable to assimilate

food consumed when dissolved oxygen level is low. According to Teichert-Coddington and Green (1993) one of the aquacultural attributes of tilapia is their ability to tolerate routine dawn dissolved oxygen concentrations of less than 0.3 mg/l, which is considerably below the tolerance limits for most other cultured fish species. It has been reported that *Sarotherodon melanotheron* can tolerate dissolved oxygen levels as low as 0.1 mg/l and salinity range of 0 ppt to 45 ppt (Pullin & Lowe-McConnell, 1982; Trewavas, 1983). Chakroff (1976) recommended that for best growth performance in *O. niloticus* the DO concentration should be above 5 mg/l. According to Kramer and McClure (1982), tilapia survives short-term anoxia by rising to the water surface to gulp oxygen-rich water, a common response to hypoxia in tropical freshwater fish. Peer and Kutty (1981) observed that tilapia also conserves energy by reducing activity in response to hypoxia. However, extended periods of hypoxia may reduce growth in *O. niloticus* (Chervinski, 1982). Parker and Davis (1981) reported that at DO concentrations below 2 mg/l, fish cease to feed, reduce locomotion and use the available oxygen to support other metabolic systems rather than growth, while fish mortality sets in at DO less than 1 mg/l level (Ayinla, Oladosu, Ajiboye & Ansa, 1994).

Effects of temperature

According to Barrows and Hardy (2001) water temperature is the single most important factor affecting fish growth. This is because fish are cold-blooded and their body temperatures fluctuate with the environmental water temperatures. Martinez, Cristina and Ross (1996) investigated the effects of temperature on food intake, growth and body composition of *Cichlasoma*

urophthalmus (Guter) juveniles and reported that temperature is the most important abiotic factor that affects growth, food intake and feed conversion in fish. Huet (1994) reported that growth was retarded in *O. niloticus* at temperatures above 32 °C. According to Woiwode and Adelman (1991), temperature influences both ingestion and metabolism, which also affect growth rates of the hybrid, Striped bass × White bass. Trewavas (1983) observed that *Sarotherodon melanotheron* is stenothermic and cannot survive in temperatures that vary greatly from 18 – 33 °C, and virtually no breeding occurs below 20 to 23°C. Parker and Davis (1981) reported that in the tropics fish grow best at temperatures between 25 °C and 32 °C, whereas for reproduction it ranged from 22 to 36 °C.

Salinity tolerance

According to Chervinski (1982) many freshwater tilapias are moderately salt tolerant. However, *S. melanotheron* and *Oreochromis mossambicus* are euryhaline inhabiting both freshwater and salt-water environments (Jennings & Williams, 1992; Page & Burr, 1991; Payne, 1983; Shafland, 1996; Trewavas, 1983). Salt tolerance has often been associated with survival (Watanabe, Kuo & Huang, 1985) but in a commercial production system, survival, reproduction and good growth performance should be the criteria for measuring salt tolerance in fish (Payne, 1983; Stickney, 1986). Therefore, tilapias, which do not reproduce or grow under saline conditions, may not be considered for commercial aquaculture.

Several studies have been conducted on the salinity tolerance of tilapias and their hybrids in the past years (Watanabe, Kuo & Huang, 1985

[*Oreochromis aureus*, *O. niloticus* and *O. mossambicus* × *O. niloticus* hybrid]; McGeachin, Wicklund, Olla & Winton, 1987 [*Tilapia aurea*]; Perschbacher & McGeachin, 1988 [*Oreochromis mossambicus* × *Oreochromis urolepis hornorum*]). The Nile tilapia, *Oreochromis niloticus* (Linnaeus) has limited salinity tolerance (Lutz, Armas-Rosales & Saxton, 2010). Lutz et al. (2010) observed that almost all salt-tolerant tilapia being cultured are as a result of crossbreeding to combine growth and other desirable traits from other species or strains.

The salinity tolerance and optimum range of *S. melanotheron* have not been studied. However, Pauly (1976) observed that *S. melanotheron* was the predominant fish in a closed lagoon where the salinity fluctuated from 0 to over 45 ppt. Trewavas (1983) reported that there was 50 % mortality when *S. melanotheron* was transferred directly from freshwater to 30.7 ppt. However, there were no casualties when they were transferred from high salinities to freshwater. For coastal and estuarine communities, developing a commercial tilapia strain with high salt-tolerance and superior growth characteristics might lead to economically profitable productions.

Chapter summary

The current literature review indicated that the black-chinned tilapia, *Sarotherodon melanotheron* were found in both brackish and freshwater environments, and it has the potential to support aquaculture industry when taken through the necessary fish yield improvement techniques such as crossbreeding, selective breeding, all-male sex-reversal, etc. There are currently three main subspecies of the black-chinned tilapia, namely *S. m. melanotheron*,

S. m. heudelotii and *S. m. leonensis*, and those found in Ghanaian waters are of the *S. m. melanotheron* sub-species.

The principles of crossbreeding and selective breeding have been successful in improving growth performance, resistance to diseases, temperature and salinity tolerance in many fish species for aquaculture. It is therefore, most probable that when the principles of crossbreeding and selective breeding are applied to *S. melanotheron* in the current study, desirable results may be achieved.

Maternal and common environmental effects can influence the accuracy of genetic parameter estimates and therefore, the research methodology should be designed to minimize their effects on heritability, response to selection and breeding values.

Poor water quality parameters, pH, DO, temperature and salinity can affect the growth performance of tilapia, hence, must be regulated to mitigate their impact on the growth performance of the experimental fish.

CHAPTER THREE

MATERIALS AND METHODS

This chapter describes the materials and methods used in the study of crosses among four populations of Black-chinned tilapia (*Sarotherodon melanotheron*) to develop a hybrid of the species with improved growth performance traits to enhance aquaculture production in Ghana.

Study Site

The study was conducted at the Aquaculture Demonstration Centre at Ashaiman near Tema, Ghana (Fig. 1). The Centre functions as a fish hatchery for the Fisheries Commission of the Ministry of Fisheries and Aquaculture Development, and specializes in producing tilapia, *Oreochromis niloticus* and catfish, *Clarias gariepinus* fingerlings. It also serves as a demonstration and training centre for newly employed staff of the Fisheries Commission, and prospective fish farmers. It has four 0.15 ha and a 0.20 ha grow-out earthen fish ponds, sixteen 50 m^2 concrete ponds, and three 30 m^2 earthen ponds (Fig. 2). The other facilities include four 2 m^2 fibre-glass tanks, four 0.4 m^2 aquarium tanks and fifteen 6 m^3 hapas. It has a feed mill and a pelletizer for feed preparation.

The Centre was created under the Government's Policy on irrigation projects, which requires three to five percent of irrigable lands to be reserved for fish farming activities.

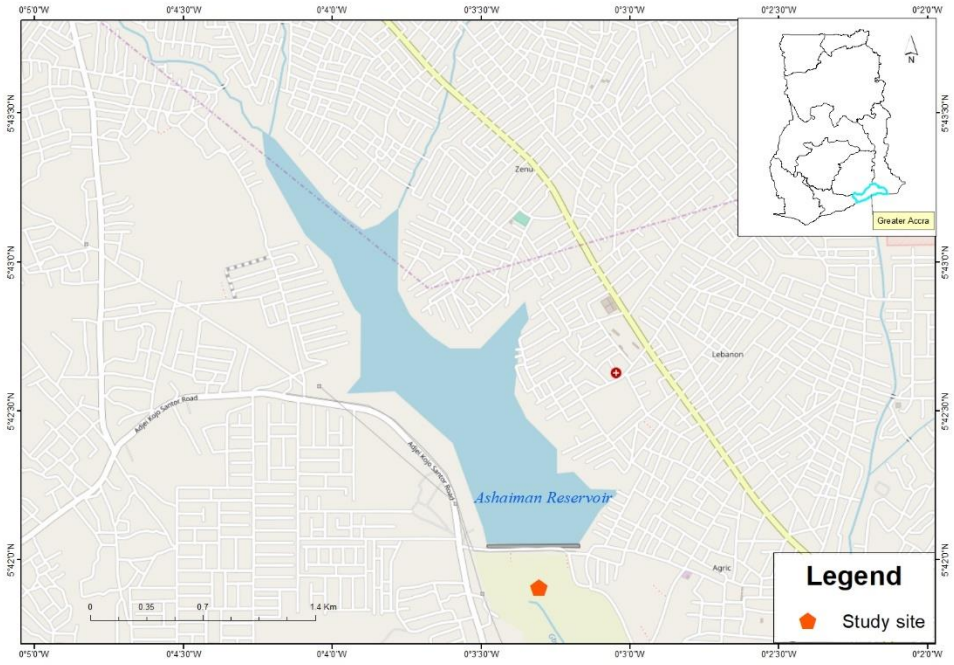


***Figure 1*: Map of Ghana showing the Ashaiman reservoir and the study site**

***Figure 2*: Arial view of the Aquaculture Demonstration Centre at Ashaiman**

Characteristics of sources of fish for study

Broodstocks of *S. melanotheron* were obtained between 8th February, 2016 and 10th March, 2016 from Brimsu reservoir (B), Fosu lagoon (F), Baifikrom reservoir (R) and the Weija reservoir (W).

Brimsu reservoir

The Brimsu reservoir is located on 5°11'N and 1°16'W (Fig. 3A) about 15 km away from Cape Coast in the Central Region of Ghana. It was constructed in 1928 purposely for the supply of potable water for Cape Coast and its environs. The reservoir has a catchment area of about 867 km^2 and surface area of about 278 ha (Bosque-Hamilton, Nana-Amankwah & Karikari, 2004). The Kakum River serves as the main source of water supply for the Brimsu reservoir. At full capacity, the reservoir has a storage volume of about 2.3 x 10^6 m^3 and maximum depth of about 7 m. According to Bosque-Hamilton et. al. (2004) the water level of the reservoir rises in April, at the beginning of the rainy season, begins to fall from October, and reaches its lowest level in March. The reservoir is freshwater with salinity of 0.00 ppt, and serves as habitat for various fish species including *Clarias gariepinus*, *Oreochromis niloticus*, *Tilapia zilli* and *Sarotherodon melanotheron*. Fishermen in the nearby communities depend on this reservoir for their livelihood.

Fosu Lagoon

The Fosu lagoon is located on 5°6'17.87" N, 1°15' 18.68" W (Fig. 3B) in the Cape Coast metropolis of the Central Region of Ghana. The lagoon has a surface area of about 0.61 km^2 (61 ha) with an average depth of 1.5 m and

therefore, considered as a shallow lagoon (Dankwa, Quarcoopome, Owiredu & Amedorme, 2016), surrounded by transport garages, mechanical workshops, educational institutions, hospital and residential facilities. About 120 fishermen depend solely on this Fosu Lagoon for their livelihoods. The black-chinned tilapia is the main fish species landed by these fishermen. Most of the landings are stunted due to the poor environmental condition and intense fishing pressure on the lagoon. According to Armah, Yawson, Pappoe and Afrifa (2010), the water at the bottom of the lagoon has become anoxic with very low concentrations of dissolved oxygen. It has been documented that the Fosu lagoon contains high levels of suspended solids and high concentrations of heavy metals like zinc and copper (Dodoo & Adjei, 1995; Gilbert, Dodoo, Okai-Sam, Essuman, and Quagraine, 2006).

Baifikrom reservoir

Baifikrom is a town, which is The Baifikrom reservoir is located on latitude 5°18’ 52.08”N and longitude 1º 01’45.08”W (Fig. 3C), about 3 km from Mankessim in the Central Region of Ghana. The Apprapong River feeds the Brimsu reservoir. The construction of this reservoir in 1978 was in response to the need for irrigation of farmlands in Mankessim and its environs. The dam has a height of about 10 m and a storage volume of about 5.2 x 10^6 m^3. Fish species found in the reservoir include *T. zilli*, *S. melanotheron* and *C. gariepinus*. Approximately 45 fishermen depend on this reservoir for their livelihoods (personal communication).

Weija reservoir

The Weija reservoir was constructed in 1977 to support the main water treatment plant for Accra and its environs. The reservoir is located on 5˚ 34' 11''N and 0˚ 20'39''W (Fig. 3D), and it supplies about 80 percent of the potable water for the entire city of Accra. According to Vanden-Bossche and Bernacsek (1990), the reservoir is about 14 km long and 2.2 km wide, with a mean depth of about 5 m covering approximately 3,361 ha at its maximum water level. The water in the Weija reservoir is slightly alkaline with high dissolved oxygen levels (Ameka, de-Graft Johnson & Akuamoah, 2000). The reservoir supplies potable water to western parts of the Accra metropolis, and is used for irrigation and fishing purposes. The people in the catchment area are mainly fishermen and farmers.

Fish specimens were weighed with an electronic balance to the nearest 0.01 g. The total and standard lengths were measured with a measuring board to the nearest 0.1 cm. Males and females from each population were held separately in 6 m^3 hapas mounted in a 30 m^2 earthen pond.

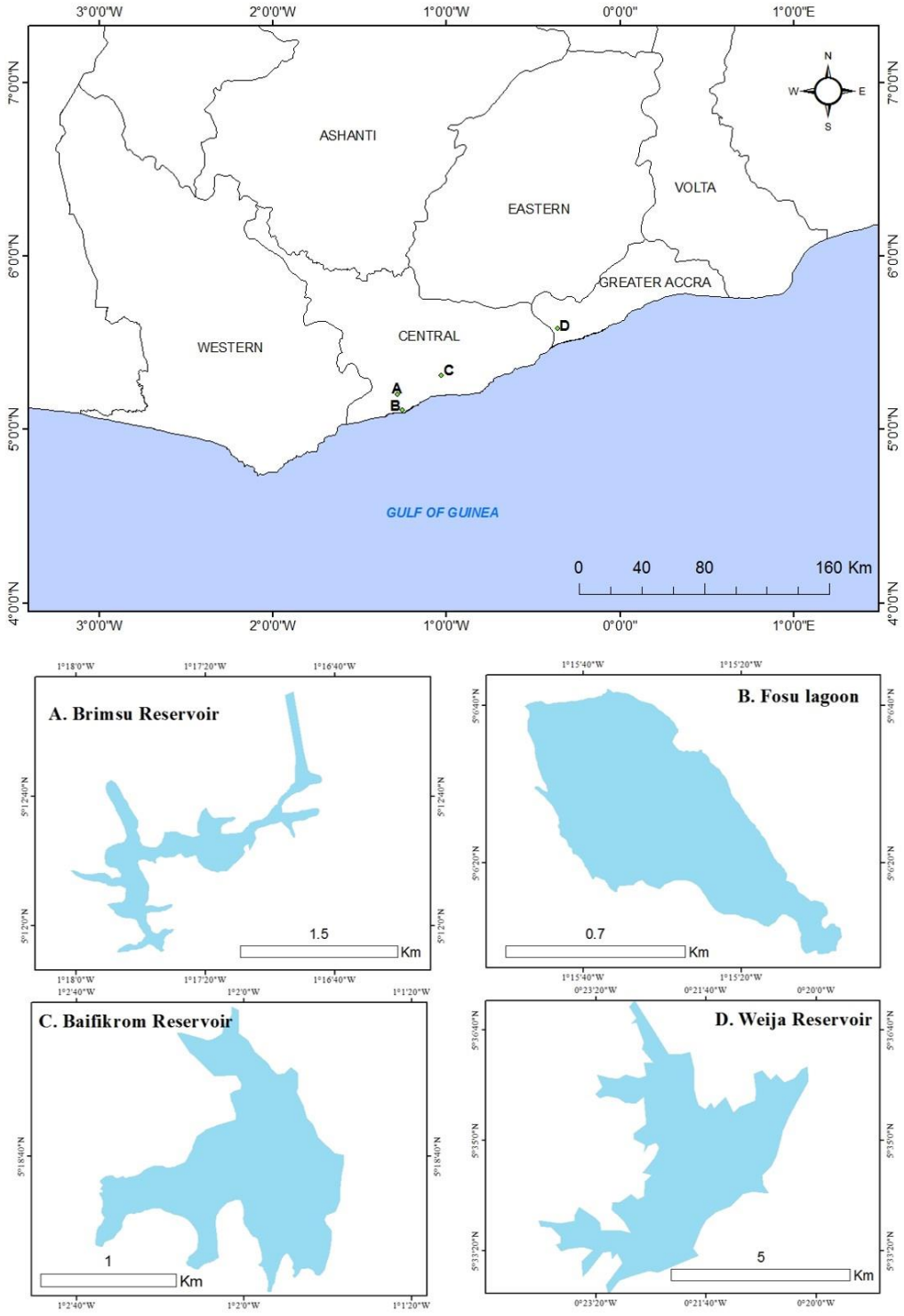


***Figure 3*: Map of Ghana showing Brimsu reservoir (A), Fosu Lagoon (B), Baifikrom reservoir (C) and Weija reservoir (D)**

Pond preparation

A pond of 1,500 m^2 was drained and the bottom limed at a rate of 10 kg lime per 100 m^2. Dried chicken manure was applied at 3 kg per 100 m^2 and the pond filled with water to about 1.2 m deep. The basal liming and manuring were to disinfect the pond, balance the water pH and to stimulate production of algae, the natural food of tilapia.

Prior to flooding the pond with water, forty-eight 6 m^3 hapas were installed in rows in the pond, leaving 20 cm space between the base of the hapa and the bottom of the pond, and one-metre intervals were also left between adjacent hapas to enable water circulation (Fig. 4).

Fry production from diallel crosses of broodstocks

The broodstocks were grouped by sexes in hapas and fed *ad libitum* with 40 % crude protein feed (Ranaan) trice daily at 9:00, 12:00 and 15:00 hours local time for two weeks. Sixteen diallel crosses as illustrated in Table 1 were conducted among the four populations to produce strains of four purebreeds and twelve hybrids following the procedure in the GIFT technology manual (De Vera, 1988; Puttaraksar, 2004). Each cross was conducted in triplicate in 6 m^3 hapas stocked with 2 males and 2 females. The hapas were randomly distributed within the 1,500 m^2 earthen fish pond.

Table 1: ***An illustration of diallel crosses involving four populations of S. melanotheron***

×	Brimsu (B♂)	Weija (W♂)	Baifikrom (R♂)	Fosu (F♂)
Brimsu (B♀)	BB	BW	BR	BF
Weija (W♀)	WB	WW	WR	WF
Baifikrom (R♀)	RB	RW	RR	RF
Fosu (F ♀)	FB	FW	FR	FF

♀ = female; ♂ = male; B, W, R and F represent genes of Brimsu, Weija, Baifikrom and Fosu strains respectively.

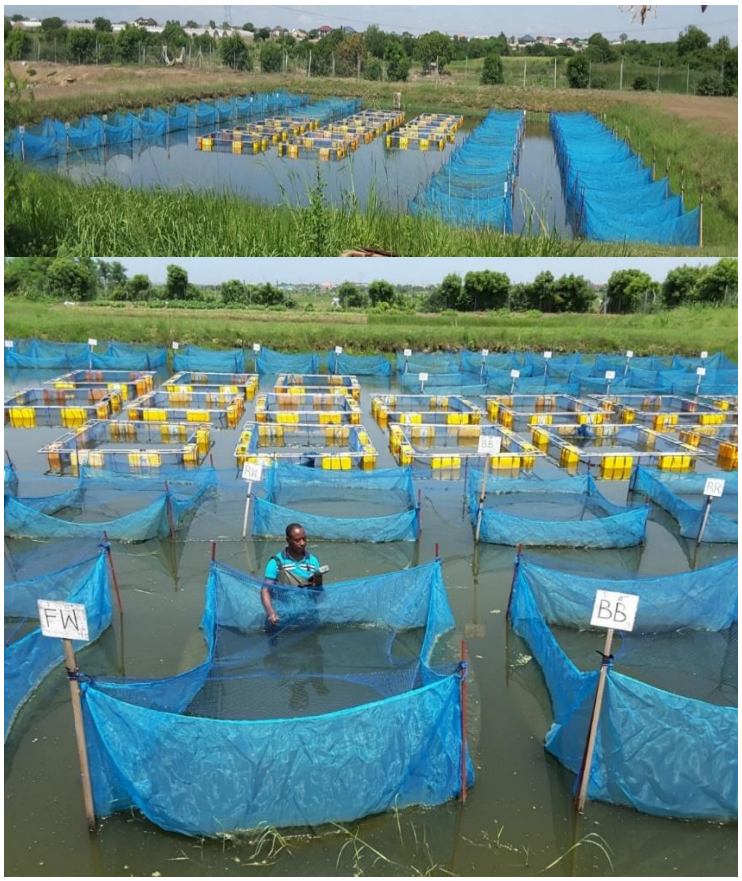


***Figure 4*: Hapas and cages in a 1,500 m^2 earthen fish pond**

Fry harvest and rearing

After one month of stocking, swim-up fry (F_1) were harvested, counted and separated from the parent stocks using a scoop net. The body weight of fry was taken to nearest 0.01 g and the total body length was measured to the nearest 0.1 cm.

Harvested fry were stocked in 6 m^3 hapas at a rate of 200 fry/m^3 to be raised to a size of 5 g. Fry were fed thrice daily at 9:00, 12:00 and 15:00 hours local time to satiation with 40 % crude protein feed. A sample of 30 fish were taken every four weeks, and each individual weighed and measured.

After 8 weeks, 120 fingerlings from each hybrid were transferred into 6 m^3 cages to grow to them for 12 months. Fingerlings were fed thrice daily at 8:00, 12:00 and 16:00 hours local time with 38 % crude protein feed at 5 % body weight for 12 months. The fingerlings were sampled every 2 weeks to take records of body weight, total and standard lengths, and to estimate feed ration. Water quality parameters, pH, temperature and DO within each cage were recorded using a multi-parameter water test kit every fortnight.

Nomenclature of the hybrids

The nomenclature adopted for this study followed the design used by Crespel, Audet, Bernatchez and Garant (2012). As indicated in Table 1, BB, WW, RR and FF represent the purebreeds from Brimsu, Weija, Baifikrom and Fosu respectively. In the other crosses, the first letter stands for the female of the population whereas the second letter represents the male of the crossing population.

Experiment 1: Growth performance test

To assess the growth performance and heterosis of the hybrids, the F_1 offspring produced through the diallel cross were stocked in 6 m^3 hapas at density of 20 fry per m^3 and fed three times daily at 9:00, 12:00 and 15:00 hours local time at 10 % biomass with 40 % crude protein feed. Each hybrid set-up was triplicated resulting in 48 hapas, and the fish were monitored for 90 days. Thirty specimens were sampled from each hapa to record the body weight of individuals to the nearest 0.01 g, and total and standard lengths to the nearest 0.1 cm. Dead fish were removed and the number recorded. The total amount of feed given to the fish during the experimental period was also recorded. The water quality parameters, dissolved oxygen (DO), temperature and pH for each hapa were determined every fortnight.

Computations and statistical analysis of growth performance

Absolute Growth Rate

Absolute growth rate (AGR) of the fish was calculated as the change in weight over a known time interval (Hopkins, 1992). That is:

$$AGR = \frac{W_2 - W_1}{t_2 - t_1}$$

Where, AGR is the absolute growth rate, W_2 and W_1 are final and initial weights in grams respectively, t_2 and t_1 are final and initial time in days respectively.

Specific Growth Rate

Specific growth rate (SGR) was calculate as:

$SGR = \frac{\ln W_2 - \ln W_1}{t_2 - t_1} \times 100$ (Wootton, 1998),

Where, SGR is the specific growth rate, W_2 and W_1 are final and initial weights of the fish, and t_2 and t_1 are the final and initial time in days.

Feed Conversion Ratio

Feed conversion ratio (FCR) of the fish was determined as:

$FCR = \frac{\text{Total feed consumed by fish (g)}}{\text{Total weight gained by fish (g)}}$ (Ridha, 2006)

Survival Rate

The survival rate (SR) of the experimental fish was computed as:

$SR = \frac{N_2}{N_1} \times 100$ (Ridha, 2006)

Where, N_1 is the total number of stocked fish and N_2 is the total number of fish surviving at the end of the observation period.

Condition Factor

The condition factor (K) which indicates the state of well-being or fatness of fish was assessed from the equation (Bagenal, 1978) given as:

$$K = \frac{W}{L^3} \times 100$$

Where, W is the final weight and L is the standard body length in cm.

Estimation of heterosis

Heterosis (H) was determined using the formula of Nguenga, Teugels and Ollevier (2000):

$$H = \frac{\left[C - \left(\frac{P1+P2}{2}\right)\right]}{\left[\left(\frac{P1+P2}{2}\right)\right]} \times 100$$

Where, C is the final mean weight, gain in weight, AGR or SGR of the hybrids, and P1 and P2 are final mean weight, gain in weight, AGR or SGR of the purebreeds in the cross combination. The differences in means of the final weights of the hybrids were determined at 5 % level of significance using one-way ANOVA, and Fisher's multiple range test when differences were indicated.

Experiment 2: Determination of breeding values of the hybrids

Individual adult F_1 fish from each hybrid were weighed to select the heaviest females and males to constitute the select line, and those with average weight to form the control line for the production of second filial generation (F_2). This was to facilitate a reliable assessment of the selection of the broodstocks. The characteristics considered in the selection of the breeders were fast growth rate, normal body shape and colour, absence of skeletal deformities, absence of large wounds and infections.

In each hybrid stock, a male (F_1) was crossed with two females (F_1) in succession in a nested mating design to produce paternal full-sib and half-sib families (F_2). From each hybrid, a male and a female were stocked into 1 m^3 hapas. After two weeks, fry were collected and immediately the female was replaced with another female to mate with the original male fish. The hapas in which fry could not be seen were monitored periodically for fry production. The

harvested fry were transferred into sixteen separate 6 m^3 hapas and reared to fingerling sizes of 5 g to 10 g. The fish were anaesthetized with Tricaine Methane Sulfonate (MS 222) at 140 mg/l, and body weight and length were recorded to the nearest 0.01 g and 0.1 cm respectively. The different hybrids were marked with floy tags. The fish was then placed in the recovery bowl, containing clean aerated water, to resuscitate (Fig. 5)

The Brimsu strain was tagged white and the Baifikrom, Weija and Fosu strains were assigned blue, green and orange respectively (Fig. 6).

***Figure 5*: Samples of tagged fish in recovery bowl**

***Figure 6*: Floy tag colours assigned to the four black-chinned tilapia populations**

Each fish was therefore, tagged with two colours indicating the contributing parents to its genotype. For instance, an offspring of Fosu (F) female × Weija (W) male (FW) was tagged with orange and green, with the orange tag on the left side of the fish when it is supported to stand on its ventral part and facing the left hand of the researcher. The tag on the left side of the fish always bears the inscription of the full or half-sib numbers in Arabic numerals (e.g. 1 – 1 or 2 – 1) where the first number represents the female and the second represents the male. In other words, the inscription 1 – 1 means a cross between female number 1 against male number 1, whereas 2 – 1 means a cross between a female number 2 against male number 1. Note that, it was only female that changes to 1 or 2 but the male had the same number 1.

The tag fixed on the right hand side of the fish bears the identification number of the fish. The identification numbers were preceded with the letter S or C, where S stands for select line and C stands for control line (Fig. 7). For example, the inscription S1 stands for fish number 1 in the select line. Similarly, the inscription C20 stands for fish number 20 in the control line.

***Figure 7*: Identification labels for the select line**

Communal rearing of tagged fish in brackish water

Brackish water used in the growth performance trials was artificial and had salinity of 6 ppt, similar to what had been measured for Fosu Lagoon. The artificial brackish water was prepared by dissolving 6 Kg of common salt (NaCl) in 1,000 L of water (Holmes-Farley, 2007) to give a salinity of 6 ppt.

For Set-up I, 10 full-sib of the select line and 10 full-sib of the control line from each hybrid offspring of female [one] male [one] were stocked together (total of 320 fingerlings) in a 50 m^2 concrete tank filled with brackish water. This set-up was triplicated and the tanks were labelled P1, P2 and P3.

For Set-up II, each hybrid offspring of female [two] male [one], 10 half-sibs (half-siblings of set-up I) of select and control lines were also stocked in the same manner as described for set-up I above, and the ponds were labelled P4, P5 and P6. The fish in both set-ups were fed *ad libitum* with 30 % crude protein feed for 120 days and the total daily weight of feed given was recorded. Water temperature, pH, DO and salinity were also measured bi-weekly.

Communal rearing of tagged fish in freshwater environment

Based on the recommendations of Eisemann, Cooper and Woodruff (1990) for quantitative evaluations of traits such as growth, the testing should be conducted in two or more environments in order to explore genotype-environment interaction, the full-sibs and half-sibs of the hybrids were also tested in cages in freshwater environment.

For growth performance test in freshwater environment (Set-up III), a total of twelve 6 m^3 cages were mounted in a 1,200 m^2 earthen pond. Ten (10) full-sib of the select line from each hybrid population of female [one] male [one]

were stocked together (total of 160 fingerlings) in a 6 m^3 cage. The set-up was triplicated and labelled A_1, A_2 and A_3. In the same way, the offspring of female [one] male [one] control line were stocked in the same manner as described above and labelled B_1, B_2 and B_3.

The select and control lines of offspring of female [two] male [one] (half-sibs) were also stocked in cages as described above. Each set-up was triplicated and labelled C_1, C_2 and C_3 for the select line, and D_1, D_2 and D_3 for the control line. The fish in all the set-ups were fed with 30 % crude protein feed *ad libitum* for 120 days, and the daily weight of feed administered was recorded. Surface water temperature, pH and DO were monitored fortnightly.

Harvesting of tagged fish for assessment

The entire stocks were harvested at the end of the 120 days. Fish in concrete tanks were harvested by total drainage, and a scoop net was used to harvest those in cages. The weight, total and standard lengths, sex, family and individual identification numbers were recorded.

Data Analysis

The statistical analysis was done separately for each experiment and culture environment. General Linear Model (GLM) in R was used to analyze body weights at harvest using the following model:

$$Y_{ijkl} = u + G_i + R_j + S_k + e_{ijkl}$$

Where,

Y_{ijkl} is the final body weight,

u is the overall mean,

G_i is the fixed effect of group,

R_j is the random effect of replicates,

S_k is the fixed effect of sex,

e_{ijkl} is the residual effect.

The differences in the means of the final weights of the hybrids were determined at 5 % level of significance using one-way ANOVA, followed by Fisher's multiple range test when differences were significant, with the Minitab 16 statistical package.

Phenotypic variation (V_p) in the offspring of the hybrids was calculated as: $V_p = V_g + V_e$ where V_g is variation due to genetic effect and V_e is variation due to environmental effect. These components of variation were estimated by subjecting the data on final body weight to analysis of variance using Microsoft Excel 2013 at a p-value of 0.05. The mean squared (MS) values were then used for the estimation (Appendix A) as:

$$V_g = \frac{MS_1 - MS_2}{r}$$

$$V_e = \frac{MS_2}{r}$$

$$V_p = V_g + V_e$$

$$h^2 = \frac{V_g}{V_p}$$ (Zobel & Talbert, 1991)

Where, V_g = genetic variance, MS_1 = mean squared between body weights, MS_2 = mean squared within body weights, r = number of replicates, V_e = environmental variance, V_p = phenotypic variance, h^2 = narrow sense heritability

Breeding value estimation

Breeding value was estimated as $BV = h^2 \times P$ (Nyquist, 1991). Where, BV = breeding value, h^2 = narrow sense heritability and P = phenotype.

Calculation of Response to Selection

The response to selection, R was calculated using the equation: $R\ (\%) = (S/C - 1) \times 100$ (Ponzoni, 2002). Where, S = mean weight of select line, C = mean weight of control line.

Estimation of maternal and common environmental effect

The maternal and common environmental effect (C^2) was calculated as $C^2 = V_d/V_p$ (Hamzah et al., 2014); where, V_p is the phenotypic variance and V_d is the dam variance.

Experiment 3: Determination of fecundity of the hybrids

After 12 months of culture, the F_1 generation were evaluated for fecundity by sampling 10 females from each hybrid population. The total body length and body weight of each individual were recorded. The gonads were removed and weighed to the nearest 0.01 g for the gonadosomatic index (GSI) calculation. Ripe ovaries (stage IV) (Witte & Van Densen, 1995) were preserved in 10 % formalin pending fecundity estimations. The whole count method (Bagenal & Braum, 1978) was employed to estimate the fecundity of the hybrids.

The gonadosomatic index (GSI) was calculated for each fish as:

$\text{GSI} = {}^{\text{GW}}/_{\text{BW}} \times 100$ (Khallaf & Authman, 2010), where, GW is gonad weight, and BW is the body weight of fish.

Absolute Fecundity (AF) was estimated as the mean number of matured eggs per ovary of fish prior to spawning (Duponchele, Cecchi, Corbin, Nunez & Legendre, 2000; Khallaf, Latif & Alnenaei, 1986).

$$\text{Relative Fecundity} = \frac{\text{Absolute fecundity}}{\text{Eviscerated weight (g)}}$$

Where, eviscerated weight = BW – GW

Experiment 4A: Sex reversal of black-chinned tilapia with 17α-methyltestosterone

Preparation of hormone-feed for all-male tilapia production

Five different concentrations of 0, 30, 60, 90 and 120 mg of 17α-methyltestosterone (17α-MT) per kilogram feeds were prepared were prepared using the ethanol evaporation method (Mair & Santiago, 1994). A commercial (Ranaan) fry feed with crude protein content of 40 % was ground into powder and sieved into very fine particles. One kilogram (1 kg) of the feed was weighed into a plastic container and mixed with the different concentrations of 17α-methyltestosterone solution.

A control diet of 0 mg of 17α-MT was prepared by adding 200 ml of alcohol to one-kilogram feed. The other four experimental feeds were prepared with the addition of 30, 60, 90 and 120 mg 17a-MT/kg of feed. The hormone-

feeds were then air-dried in the Laboratory, packed, labelled and stored in a refrigerator until use.

Fry production for sex reversal assessment

Fry were produced by crossing broodstocks (6 males: 6 females) of the Weija strain in 6 m^3 hapa. After 2 weeks, fry of length less than 11 mm (Fig. 8) were selected and stocked in fifteen 72-litre capacity aquarium glass tanks (Fig. 9).

***Figure 8*: A sample fry of 10 mm length used in the sex reversal experiment**

Each tank was stocked with 100 fry and fed with the various concentrations of 17α-MT feeds. The set-up was triplicated for each feed treatment. The tanks were randomly arranged, per the different feeds, on a table within the laboratory. The stocked fry were fed with the hormone-feeds at 20 % biomass, four times daily at 09:00, 11:00, 13:00 and 15:00 hours local time for 28 days. Excess feed and excreta were siphoned out of the tanks daily, and the water was partially changed to reduce the production of algae. The pH, DO and temperature of the water were monitored daily, and through aquarium aerators, the tanks were constantly supplied with oxygen throughout the treatment period.

***Figure 9*: 72-litre capacity aquaria for sex reversal treatment**

Experiment 4B: Growth performance evaluation of the fish treated with different doses of 17α-MT feeds

After the 28-day hormone treatment, the fry were transferred into fifteen 6 m^3 hapas mounted in 1,200 m^2 earthen pond. The hapas in three replicates for each treatment were randomly mounted. A total of 60 fry were stocked in each hapa and fed at 5 % biomass divided into three times daily at 09:00, 12:00 and 15:00 hours local time.

Thirty fish from each treatment were sampled fortnightly to record weight and length to the nearest 0.01 g and 0.1 cm respectively. The total feed given and fish mortalities were recorded during the two-week period. Temperature, DO and pH were measured on each sampling date.

Sex determination using the gonadal squash method

Thirty fish from each treatment were harvested after 60 days, and each specimen was weighed, body lengths measured and preserved in 10 % formalin for sex determination using the gonadal squash method with a slight modification to the method described by Guerrero and Shelton (1974).

The female ovarian tissue was identified by the presence of pre-vitellogenic and vitellogenic oocytes (Fig. 10A) and the male testicular tissues were identified by their characteristic lobular structures (Fig. 10B). The number of males and females were recorded, and the percentage sex reversal for each dosage of 17α-MT was calculated. Chi-squared analysis was used to test for significant deviation of the sex from the expected 1:1 ratio at 5 % level of significance for each treatment.

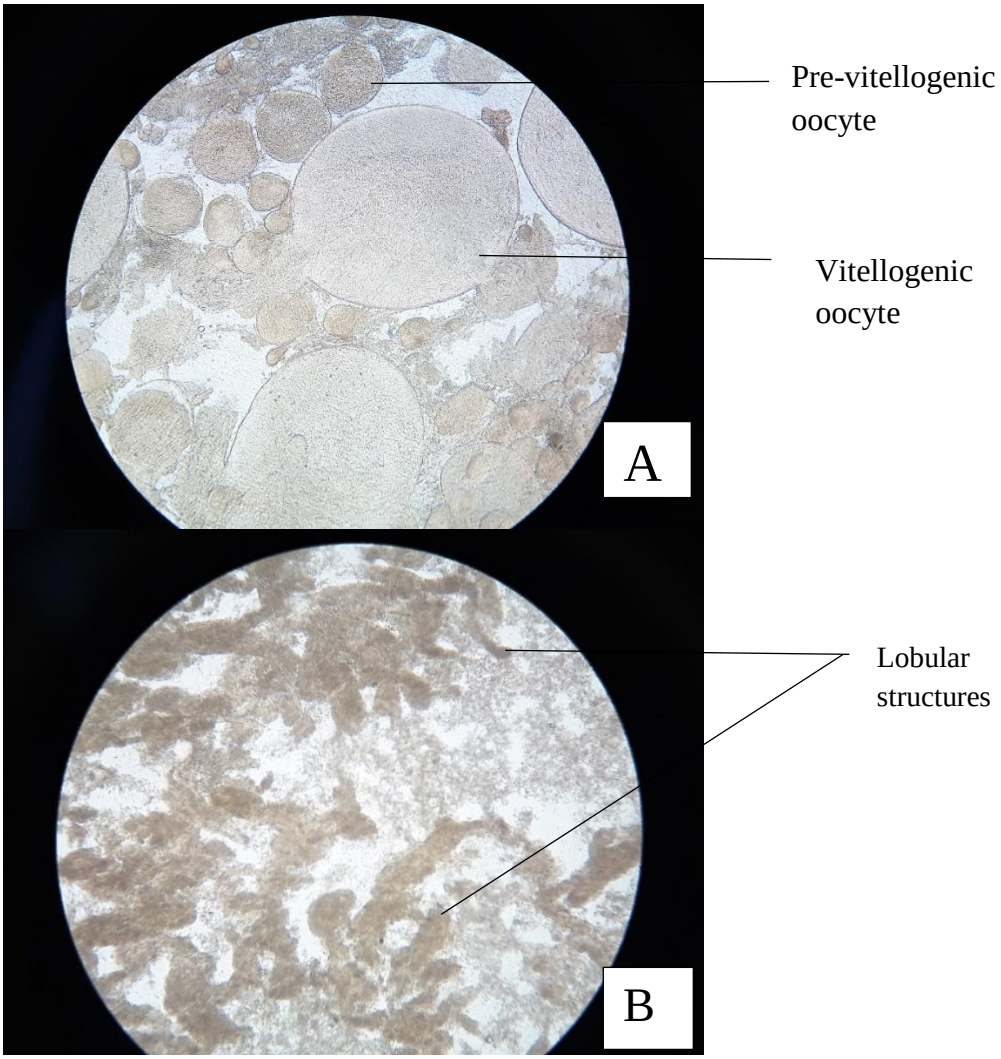


***Figure 10*: Squashed ovarian tissue (A) and testicular tissue (B) of black-chinned tilapia under light microscope (× 10)**

Sex determination using the genital papillae

The remaining treated tilapia in each hapa were further grown for 180 days after which they were harvested for sex determination using the genital papillae. Each fish was physically examined by first smearing the genital papillae with Methylene blue and rinsing it with clean water. The blue stain was either absorbed by the papillae (making the oviduct opening more conspicuous) if the fish was a female or not absorbed by the papilla (leaving no stain on the papilla) if the fish was a male (Fig. 11A).

The female black-chinned tilapia has three openings; urethra, oviduct and anus whereas the male counterpart has only two openings; urethra and anus. It is the oviduct opening that absorbs the blue stain (Fig. 11A), making it easier to identify the female from the male. Figure 11B shows the colour differences between the male and female black-chinned tilapia. The female was identified by its transparent operculum, which appears deep mauve because of the blood that flows through the gills beneath it. The male black-chinned tilapia on the other hand, has a bright golden yellow colour on the operculum (Fig. 11B).

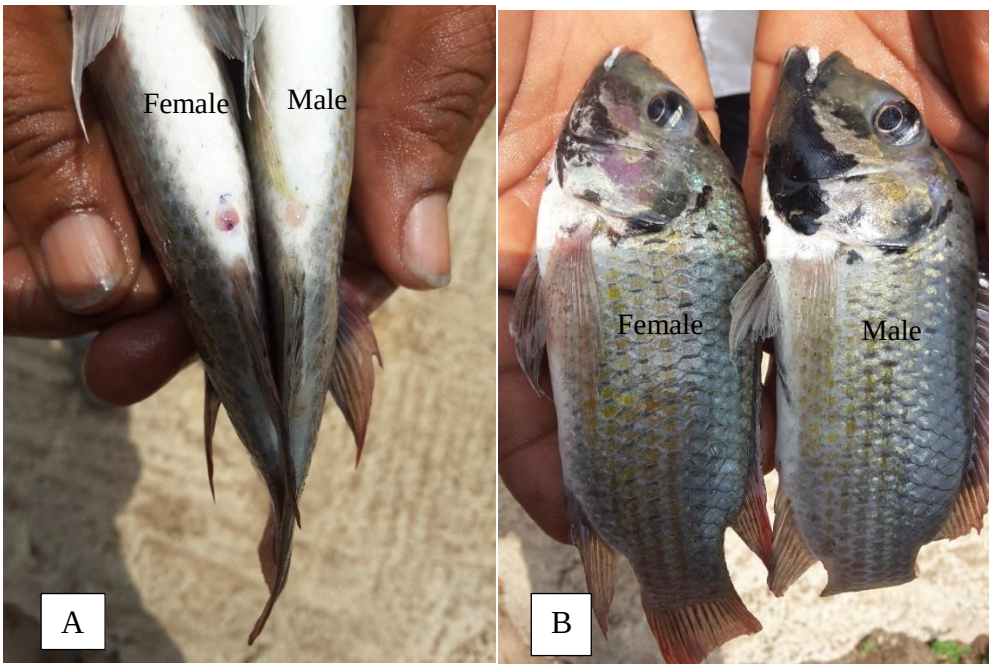


***Figure 11*: Female (left) and male (right) black-chinned tilapia**

CHAPTER FOUR

RESULTS

This chapter outlines the results of the crossbreeding of *S. melanotheron* populations from the reservoirs of Brimsu, Weija, Baifikrom and Fosu Lagoon. It covers results from the evaluations of the hybrids on growth performance, heterosis, genetic improvement, heritability, breeding value, fecundity and all-male production.

Parent stocks

Table 2 gives the total number of parents stocks harvested from the four populations studied, their sexes and mean body weights. A total of 52 parent fish were harvested from Brimsu reservoir, 51 from Weija reservoir, 64 from Baifikrom reservoir and 184 from the Fosu Lagoon. The males from Brimsu had the highest weight of 238.8 ± 12.1 g, whereas the males from Fosu lagoon had the lowest body weight of 18.5 ± 1.9 g.

Table 2*: Number and mean body weights of parent stocks harvested from the various populations*

Population	Number of fish			Weight of fish ± SE (g)	
	Total	Females	Males	Females	Males
Brimsu	52	27	25	226.7±8.2[a]	238.8 ±12.1[a]
Weija	51	25	26	80.0 ±11.5[b]	73.3±6.7[b]
Baifikrom	64	24	40	73.3±12.0[b]	67.5±1.1[b]
Fosu	182	152	30	23.8±1.9[c]	18.5±1.9[c]

Means in the same column with different superscripts are significantly different at the 5 % level of probability

The body weight of the Brimsu strains were significantly heavier ($P < 0.05$) than all the other strains. On the other hand, the body weight of the Weija strains were not significantly different ($P > 0.05$) from the Baifikrom strains.

Experiment 1: Composite fry production

Table 3 shows the number of hybrid fry produced through diallel crosses. The cross between Brimsu female and Brimsu male (BB) produced the highest number of fry (1055 ± 524) whereas the Fosu female and Fosu male (FF) gave the lowest number of fry (142 ± 82.8). Among the hybrids, the cross between Brimsu female and Baifikrom male yielded the highest mean number of fry of 925 ± 333 whereas the lowest mean number of fry (214 ± 62.6) resulted from cross between Brimsu female and Fosu male (BF).

Table 3: ***Number of S. melanotheron hybrid fry produced by diallel crosses***

	Replicates (number of fry)				
Hybrids	**1**	**2**	**3**	**Total number of fry**	**Mean ± SE**
BB	968	194	2004	3166	1055 ± 524^{a}
BF	330	115	197	642	214.0 ± 62.6^{c}
BR	1165	267	1344	2776	925 ± 333^{ab}
BW	1311	134	649	2094	698 ± 341^{abc}
FB	0	1201	323	1524	762 ± 439^{abc}
FF	44	307	76	427	142.3 ± 82.8^{c}
FR	682	622	973	2277	759 ± 108^{abc}
FW	461	427	798	1686	562 ± 118^{abc}
RB	563	0	81	644	322 ± 241^{c}
RF	532	132	379	1043	348 ± 117^{bc}
RR	762	648	799	2209	736.3 ± 45.4^{abc}
RW	30	1236	753	2019	673 ± 350^{abc}
WB	123	311	0	434	217.0 ± 94.0^{c}
WF	287	351	692	1330	443 ± 126^{abc}
WR	955	504	277	1736	579 ± 199^{abc}
WW	545	431	366	1342	447.3 ± 52.3^{abc}

Means with different superscripts are significantly different ($P < 0.05$)

The number of fry produced by BB was significantly higher than that of BF, RB, RF, WB and FF, but not significantly different from the rest of the hybrids under study. Spawning did not occur in three hapas containing FB, RB and WB (Table 3) within the 4 weeks period.

Mean body weight of hybrid fry

The mean body weights of three-week old hybrids are shown in Table 4. The mean body weights ranged from 0.57 ± 0.13 g in the Baifikrom fry to 0.11 ± 0.03 g in the Brimsu female × Fosu male (BF) fry. However, there was no significant difference between the mean body weights of the treatments ($P > 0.05$) as shown in Table 5.

Table 4: ***Mean body weight of three-week old hybrids of S. melanotheron fry from diallel crosses***

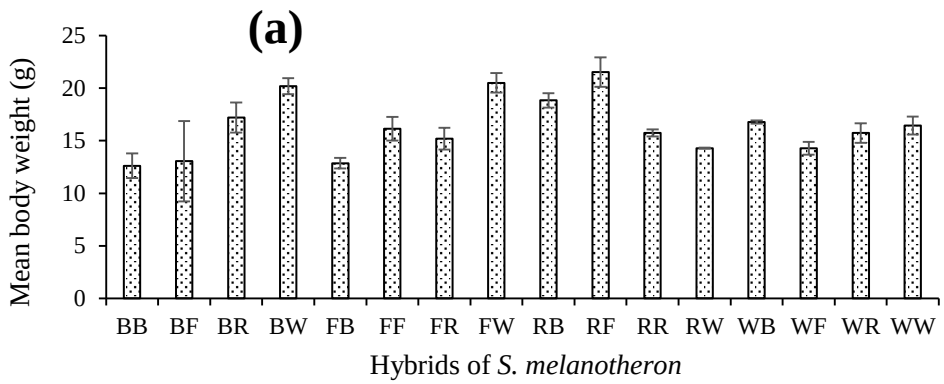
	Replicates (Mean weight (g))			
Hybrids	**1**	**2**	**3**	**Mean Wt. ± SE (g)**
BB	0.26	0.44	0.24	0.31 ± 0.05
BF	0.04	0.18	0.10	0.11 ± 0.03
BR	0.01	0.92	0.43	0.45 ± 0.19
BW	0.35	0.01	0.35	0.24 ± 0.08
FB	0.00	0.14	0.25	0.13 ± 0.05
FF	0.36	0.19	0.55	0.37 ± 0.07
FR	0.26	0.62	0.33	0.40 ± 0.08
FW	0.47	0.61	0.02	0.37 ± 0.13
RB	0.41	0.00	0.05	0.15 ± 0.09
RF	0.32	0.32	0.58	0.41 ± 0.06
RR	0.27	0.56	0.88	0.57 ± 0.13
RW	0.66	0.49	0.46	0.54 ± 0.04
WB	0.97	0.03	0.00	0.33 ± 0.23
WF	0.27	0.22	0.24	0.24 ± 0.01
WR	0.13	0.43	0.15	0.24 ± 0.07
WW	0.09	0.68	0.53	0.43 ± 0.13

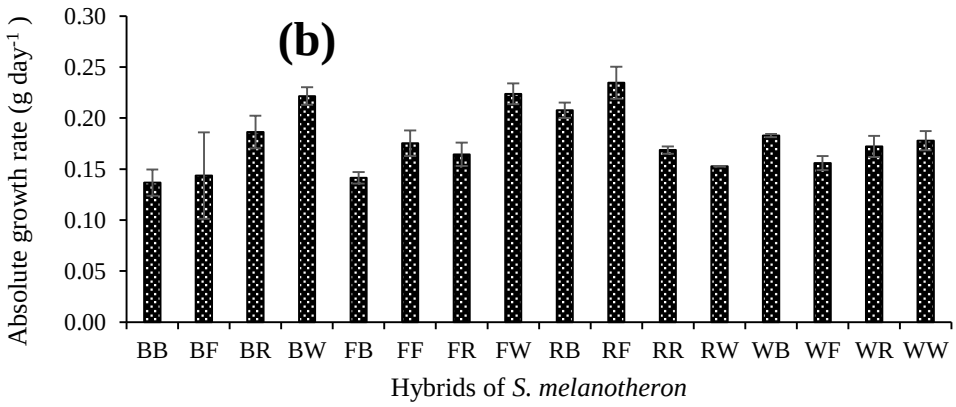
Table 5: ***ANOVA test on mean body weight of three-week old hybrids of S. melanotheron fry***

Source	DF	SS	MS	F	P
Factor	15	0.9115	0.0608	0.99	0.485
Error	32	1.9579	0.0612		
Total	47	2.8694			

Mean body weights, absolute and specific growth rates of *S. melanotheron* pure breeds and hybrids cultured in freshwater for 90 days

Figure 12 shows mean body weights (a), absolute growth rates (b) and specific growth rates (c) of the hybrids cultured in freshwater for 90 days. The mean body weights ranged from 12.62 ± 1.16 g to 21.52 ± 1.40 g. The cross between Baifikrom female and Fosu male (RF) attained the highest mean body weight of 21.52 ± 1.40 g, while the lowest mean body weight of 12.62 ± 1.16 g was recorded for the purebreed from Brimsu (BB). Among the purebreeds, the Weija strain (WW) attained the highest mean body weight of 16.44 ± 0.85 g compared to 16.14 ± 1.13 g, 15.74 ± 0.32 g and 12.62 ± 1.16 g for Fosu (FF), Baifikrom (RR) and Brimsu (BB) respectively. After the 90-day culture period, mean body weights of BW, FW, RB and RF were not significantly different from each other but were significantly heavier ($P < 0.05$) than the rest of the experimental fish.





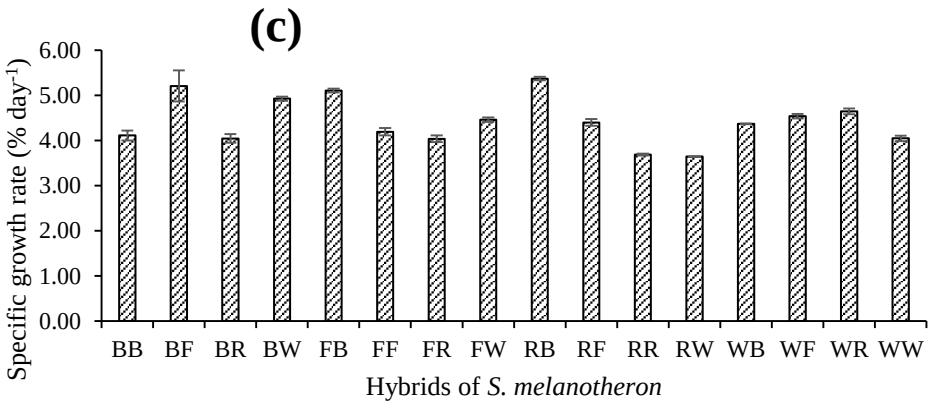


***Figure 12*: (a) Mean body weight, (b) Absolute growth rate and (c) Specific growth rate of hybrids of *S. melanotheron* cultured in freshwater for 90 days. Vertical bars represent standard errors**

The mean absolute growth rate (AGR) recorded for the hybrids ranged from 0.14 ± 0.006 g day^{-1} to 0.23 ± 0.016 g day^{-1} (Fig. 12b). The lowest AGR observed was in the purebreed BB, whereas the highest was recorded for the hybrid RF. The AGR of RF, FW, BW and RB were not significantly different ($P > 0.05$) from each other but were all significantly higher ($P < 0.05$) than the rest of the hybrids.

The hybrid RB exhibited the highest specific growth rate (SGR) of 5.37 ± 0.04 % day^{-1}, while RW had the lowest of 3.64 % day^{-1} (Fig. 12c). The SGR of six (BF, BW, FB, FW, RB, WF and WR) out of the twelve hybrids were significantly higher ($P < 0.05$) than the four purebreeds (BB, FF, RR and WW).

Feed conversion ratio of *S. melanotheron* cultured in freshwater for 90 days

Data on feed conversion ratio (FCR) of purebreeds and hybrids are presented in Table 6. The best FCR was recorded for the mating between Baifikrom female and Brimsu male (RB) with a value of 1.54 ± 0.20, whereas the poorest FCR of 2.78 ± 0.42 was attained by the purebreed, FF. The FCR attained by RB was significantly lower than that of FF, BF and RW.

Table 6: ***Feed conversion ratio of S. melanotheron hybrids cultured in freshwater for 90 days***

	FCR Replicates			
Hybrids	**1**	**2**	**3**	**Mean ± SE**
BB	2.81	2.31	1.41	2.17 ± 0.41^{abcd}
BF	2.67	1.76	2.51	2.31 ± 0.28^{ab}
BR	2.06	1.76	1.35	1.72 ± 0.21^{bcd}
BW	1.73	1.54	1.72	1.66 ± 0.06^{bcd}
FB	2.71	2.29	1.40	2.14 ± 0.39^{abcd}
FF	3.26	3.13	1.93	2.77 ± 0.42^{a}
FR	2.34	1.90	1.57	1.94 ± 0.22^{bcd}
FW	1.72	1.71	1.35	1.59 ± 0.12^{cd}
RB	1.85	1.60	1.16	1.54 ± 0.20^{d}
RF	1.64	1.57	2.05	1.75 ± 0.15^{bcd}
RR	2.28	2.08	2.14	2.17 ± 0.06^{abcd}
RW	2.51	2.30	1.93	2.25 ± 0.17^{abc}
WB	2.10	1.97	1.40	1.83 ± 0.22^{bcd}
WF	2.46	2.05	1.68	2.06 ± 0.23^{bcd}
WR	2.23	1.84	2.17	2.08 ± 0.12^{abcd}
WW	2.16	1.86	1.49	1.84 ± 0.19^{bcd}

Means with different letters in the superscripts are significantly different ($P < 0.05$)

Percentage survival of *S. melanotheron* hybrids cultured for 90 days

The mean percentage survival ranged from 90.0 ± 4.99 % (FF) to 99.96 ± 0.04 % (FB) as shown in Figure 13. The survival of FB was significantly higher ($P < 0.05$) than the other hybrids. The mean survival rate of the

purebreed, FF was significantly lower ($P < 0.05$) than all the fish except the hybrids BF, RF and WB.

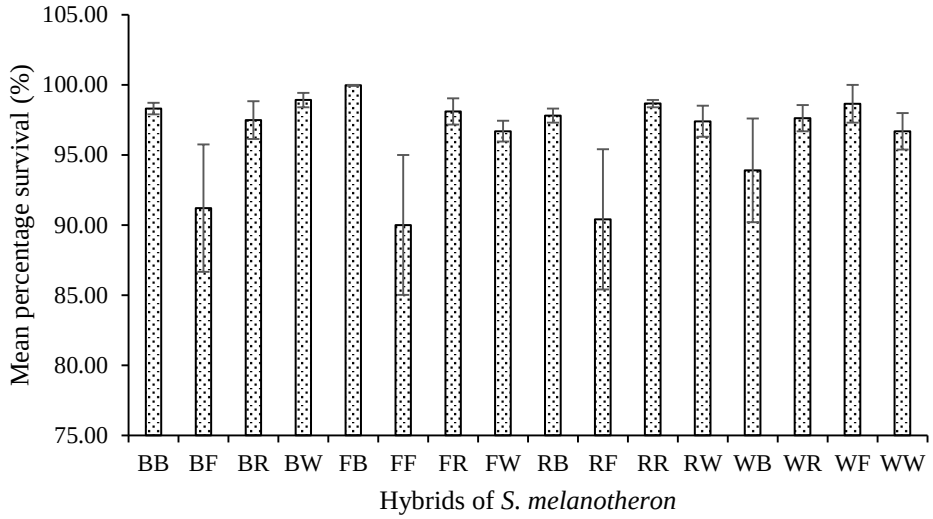


***Figure 13*: Mean percentage survival of hybrid *S. melanotheron* cultured for 90 days. Vertical bars represent standard errors**

Condition factor of hybrids of *S. melanotheron* cultured for 90 days

The condition factor (K) of the various hybrids are shown in Table 7. The hybrid, BF had the highest K of 3.10 ± 0.03 whereas the purebreed, FF exhibited the lowest K of 2.14 ± 0.09. The K factor of BF was significantly higher ($P < 0.05$) than all the experimental fish, except BB, BR, WB and WW.

Table 7: ***Mean condition factors of S. melanotheron hybrids cultured in freshwater for 90 days***

	Replicates of K			
Hybrids	**1**	**2**	**3**	**Mean ± SE**
BB	3.13	3.04	3.06	3.08 ± 0.03^{ab}
BF	3.06	3.15	3.10	3.10 ± 0.03^{a}
BR	2.92	2.99	2.96	2.96 ± 0.02^{abcd}
BW	2.88	2.95	2.88	2.90 ± 0.02^{cd}
FB	2.87	2.85	2.87	2.86 ± 0.01^{cde}
FF	2.10	2.32	2.00	2.14 ± 0.09^{g}
FR	2.96	2.84	2.91	2.90 ± 0.04^{cd}
FW	2.57	2.65	2.88	2.70 ± 0.09^{f}
RB	2.90	2.89	2.93	2.91 ± 0.01^{cd}
RF	2.84	2.88	3.03	2.92 ± 0.06^{cd}
RR	2.66	2.80	2.74	2.73 ± 0.04^{ef}
RW	2.86	2.84	2.87	2.86 ± 0.01^{de}
WB	3.00	3.00	3.03	3.01 ± 0.01^{abc}
WF	2.74	2.65	2.59	2.66 ± 0.04^{f}
WR	2.80	2.84	3.18	2.94 ± 0.12^{bcd}
WW	2.96	2.94	3.07	2.99 ± 0.04^{abcd}

Means with different letters in the superscripts are significantly different ($P < 0.05$)

Heterosis of hybrids of *S. melanotheron* cultured for 90 days

Analysis of heterosis (Table 8) indicated that WB (15.49 %), RB (32.86 %), BW (38.89 %), FW (25.91 %), BR (21.37 %) and RF (35.01 %) exhibited positive heterosis with reference to mid-parent heterosis. Similarly, with respect to best-parent heterosis, the same six hybrids (WB, RB, BW, FW, BR and RF) attained positive heterosis. The hybrids BF (-9.25), FB (-10.57), FR (-4.71), RW

(-11.19), WF (-12.34) and WR (-2.24) on the other hand exhibited negative heterosis. The magnitude of best-parent heterosis calculated for the experimental hybrids ranged from -20.32 % to 33.33 %. When the total length (TL) was used to estimate heterosis, the hybrids which indicated positive heterosis for the earlier parameters (mid-parent and best-parent heterosis) continued to exhibit positive heterosis for total length. However, the magnitudes were lower for final lengths, ranging from -10.48 % to 9.10 %.

Heterosis estimates based on body weight gained (WG) ranged from a minimum of -19.28 % to a maximum of 33.86 % (Table 8). A total of six hybrids (WB, RB, BW, FW, BR and RF) exhibited positive heterosis in terms of body weight gained after 90 days of culture.

In terms of absolute growth rate (AGR), the hybrid, WB had the same performance as its best-parent, and therefore a heterosis value of zero percent. Five of the hybrids exhibited positive heterosis, RB (23.19 %), BW (24.54 %), FW (27.68 %), BR (10.48 %) and RF (33.85 %). It was further observed that the figures obtained for AGR heterosis were from a minimum of -19.23 % to maximum of 33.85 %.

Heterosis based on specific growth rate (SGR) indicated that a total of nine of the twelve hybrids exhibited positive heterosis. Only RW, BR and FR had negative heterosis values of -10.04 %, -1.64 % and -3.66 % respectively.

In terms of survival (SR) only three of the hybrids (FB [1.64 %], BW [0.59 %] and WF [1.99 %]) exhibited positive heterosis. The hybrid FW showed heterosis estimate of zero percent whereas the remaining hybrids exhibited negative heterosis. The overall mean heterosis calculated for each hybrid indicated seven out of the twelve hybrids exhibited positive heterosis ranging

from 0.17 ± 1.95 to 22.17 ± 5.52 %. It was observed that the hybrids, RW and FR exhibited negative heterosis for all the parameters, whereas BW and FW indicated positive heterosis for all parameters. The hybrids WB, RB and RF showed positive heterosis for all the parameters except percentage survival. The overall heterosis estimates for BW, FW, RB and RF were not significantly different from each other but were significantly higher ($P < 0.05$) than all the other hybrids.

Table 8: ***Heterosis estimates for seven parameters in hybrids of S. melanotheron grown for 90 days***

Hybrids	Heterosis (%)							
	MP	**BP**	**TL**	**WG**	**AGR**	**SGR**	**SR**	**Overall Ave.**
BF	-9.25	-19.14	-10.48	-17.95	-17.98	24.22	-7.26	-8.68 ± 4.38^{de}
BR	21.37	9.34	1.39	10.48	10.48	-1.64	-1.20	8.41 ± 2.62^{b}
BW	38.89	22.75	8.52	24.55	24.54	19.83	0.59	21.73 ± 3.90^{a}
FB	-10.57	-20.32	-7.22	-19.28	-19.32	21.80	1.64	-8.37 ± 4.45^{de}
FR	-4.71	-5.89	-3.46	-6.21	-6.21	-3.66	-0.57	-4.83 ± 0.71^{cde}
FW	25.91	24.76	9.10	27.30	27.68	6.43	0.00	19.02 ± 3.70^{a}
RB	32.86	19.70	4.57	23.20	23.19	30.68	-0.84	22.10 ± 4.54^{a}
RF	35.01	33.33	8.21	33.86	33.85	4.93	-8.37	22.17 ± 5.52^{a}
RW	-11.19	-13.08	-3.97	-14.12	-14.15	-10.04	-1.28	-10.94 ± 1.84^{e}
WB	15.49	2.07	0.20	2.75	0.00	6.25	-4.55	3.58 ± 1.86^{bc}
WF	-12.34	-13.14	-2.18	-12.30	-10.99	8.32	1.99	-6.08 ± 2.48^{cde}
WR	-2.24	-4.32	-1.89	-3.25	-3.25	14.79	-1.06	0.17 ± 1.95^{bcd}

MP = Mid-Parent (final body weight); BP=Best Parent (final body weight); TL= Total length; WG= Body weight gained; AGR= Absolute growth rate; SGR= Specific growth rate; SR= Survival rate; Ave. = Average across all parameters

Water quality parameters recorded during the 90-day culture period

Table 9 shows the water quality parameters recorded within the hapas containing the experimental fish. The pH recorded in the hapas ranged between 8.62 ± 0.08 and 8.73 ± 0.08. The minimum pH was recorded in hapas containing the purebreed, RR whereas the maximum was recorded for FF. The mean pH recorded over the period were not significantly different ($P > 0.05$) from each other.

Table 9: ***Mean water quality estimates within hapas containing hybrids of S. melanotheron cultured for 90 days***

Strain	pH	Mean DO (m/l)	Mean temp (°C)
BB	8.64 ± 0.08	5.02 ± 0.95	28.62 ± 0.45
BF	8.69 ± 0.07	4.99 ± 0.90	28.69 ± 0.55
BR	8.67 ± 0.07	5.16 ± 1.11	28.65 ± 0.37
BW	8.70 ± 0.09	4.89 ± 0.86	28.05 ± 0.83
FB	8.64 ± 0.07	4.94 ± 0.92	28.56 ± 0.55
FF	8.73 ± 0.08	5.15 ± 0.95	28.72 ± 0.47
FR	8.68 ± 0.08	4.87 ± 0.80	28.62 ± 0.51
FW	8.64 ± 0.06	4.96 ± 0.91	28.57 ± 0.46
RB	8.63 ± 0.08	5.03 ± 0.94	28.63 ± 0.54
RF	8.69 ± 0.09	4.76 ± 0.66	28.45 ± 0.63
RR	8.62 ± 0.08	5.07 ± 1.08	27.90 ± 0.81
RW	8.63 ± 0.08	4.86 ± 0.84	28.65 ± 0.45
WB	8.69 ± 0.07	4.87 ± 0.79	28.60 ± 0.52
WF	8.65 ± 0.08	5.03 ± 0.94	28.50 ± 0.53
WR	8.64 ± 0.08	4.81 ± 0.82	28.61 ± 0.46
WW	8.65 ± 0.07	5.22 ± 1.00	28.58 ± 0.52

The dissolved oxygen (DO) levels within the various hapas ranged from 4.76 ± 0.66 mg/l for hapas containing BR to 5.22 ± 1.00 mg/l for hapas holding the hybrid, FB. On the other hand, the mean surface water temperature recorded for the culture period ranged from 27.90 ± 0.81 ˚C to 28.72 ± 0.47 ˚C. The

lowest temperature was recorded for the hapas containing RR, whereas the highest was recorded for those holding the purebreed, FF.

Experiments 2: Response to selection and maternal effect on growth of *S. melanotheron* hybrids in brackish water and freshwater

Mean body weight of 8-week old tagged offspring of female [one] × male [one] select and control lines before stocking in brackish water

Figure 14 shows the initial mean body weights of offspring of female [one] × male [one] (a) and female [two] × male [one] (b) select and control lines before stocking in brackish water for growth performance test.

The initial mean body weights for the various experimental fish in the select line of female [one] × male [one] ranged from 4.19 ± 0.24 g to 30.81 ± 3.34 g (Fig. 14a). The lowest initial mean body weight was exhibited by the purebreed BB, while the highest initial mean body weight was attained by FR. The initial mean body weight of FR was significantly higher ($P < 0.05$) compared to all the experimental fish. On the other hand, the initial mean body weight of BB was not significantly different ($P > 0.05$) from BF and WW, but was significantly less than the rest of the experimental fish. The hybrids BR, BW, FB, FF, RF, RR, RW and WF had comparable body weights.

The initial mean body weights for the control line of female [one] × male [one] offspring ranged from 4.37 ± 0.34 g for WW to 17.09 ± 0.59 g for RF (Fig. 14a). The initial mean body weight of WW was not significantly different from BB, BR and WB but was significantly less ($P < 0.05$) than the rest of the hybrids

under study. The initial mean body weight of the hybrid RF was significantly higher ($P < 0.05$) compared to all the other experimental fish. The control lines for FF, FR, FW and WF for Figures 14(a) and 14(b) respectively were lost to rainstorm activities.

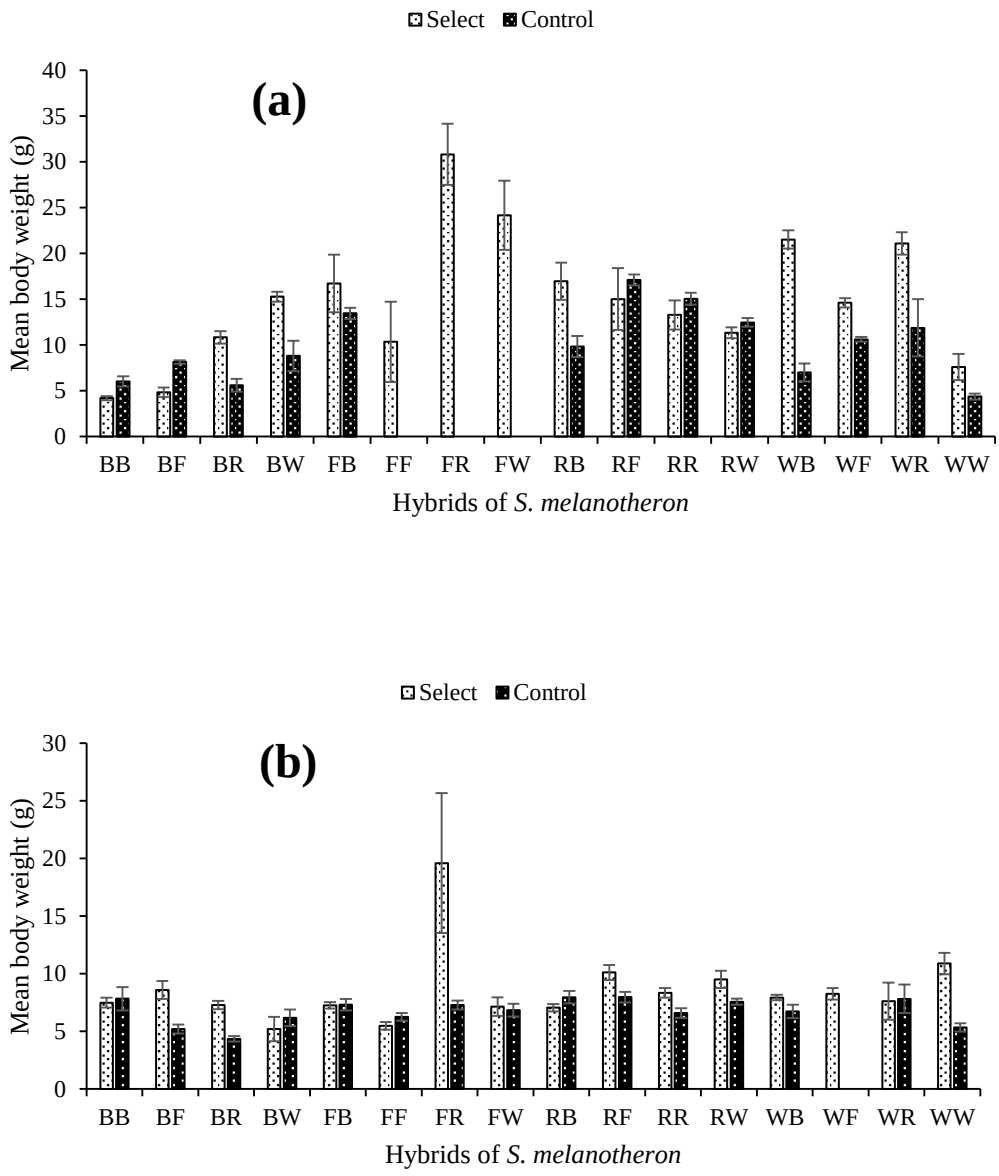


***Figure 14*: Mean body weight of 8-week old offspring of female [one] × male [one] (a) and female [two] × male [one] (b) hybrids of *S. melanotheron* select and control lines before stocking in brackish water. Vertical bars represent standard errors**

The mean body weights for the select line of female [two] × male [one] ranged from 5.20 ± 0.33 g to 19.59 ± 6.06 g for BW and FR respectively (Fig. 14b). The mean body weight of FR was significantly higher ($P < 0.05$) than all the other experimental fish, while the weight of the hybrid BW was not significantly different ($P > 0.05$) from BB, BR, FB, FF, FW, RB and WR.

The lowest mean body weight of 4.35 ± 0.24 g among the offspring of female [two] × male [one] control line was attained by the hybrid BR, while the highest mean body weight of 7.97 ± 0.45 g was exhibited by RF. The body weight of RF was not significantly different ($P > 0.05$) from BB, FB, FR, FW, RB, BW and WB. Similarly, the mean body weight of BR was comparable to BF and WW, but was significantly less ($P < 0.05$) than all the other fish under study.

Mean body weight of 8-week old tagged offspring of female [one] × male [one] select and control lines before stocking in freshwater environment

Figure 15 shows the mean body weights of offspring of female [one] × male [one] (c) and female [two] × male [one] (d) select and control lines before stocking in freshwater for growth performance trials.

The mean body weights recorded for the 8-week old offspring of female [one] × male [one] select line ranged from 7.47 ± 0.33 g to 20.06 ± 1.43 g (Fig. 15c). The lowest mean body weight was exhibited by the hybrid FB, while WR had the highest mean body weight. The body weight of FB was not significantly different ($P > 0.05$) from BB, BF, BR, FB, FF, FW, RF, RR and WW but was significantly lower than the remaining seven hybrids. On the other hand, the

mean body weight of WR was significantly higher ($P < 0.05$) than all the other experimental hybrids, but not significantly different from WB. The control lines for FW in Figure 15(c) and BW and WF in Figure 15(d) were lost due to rainstorm actions.

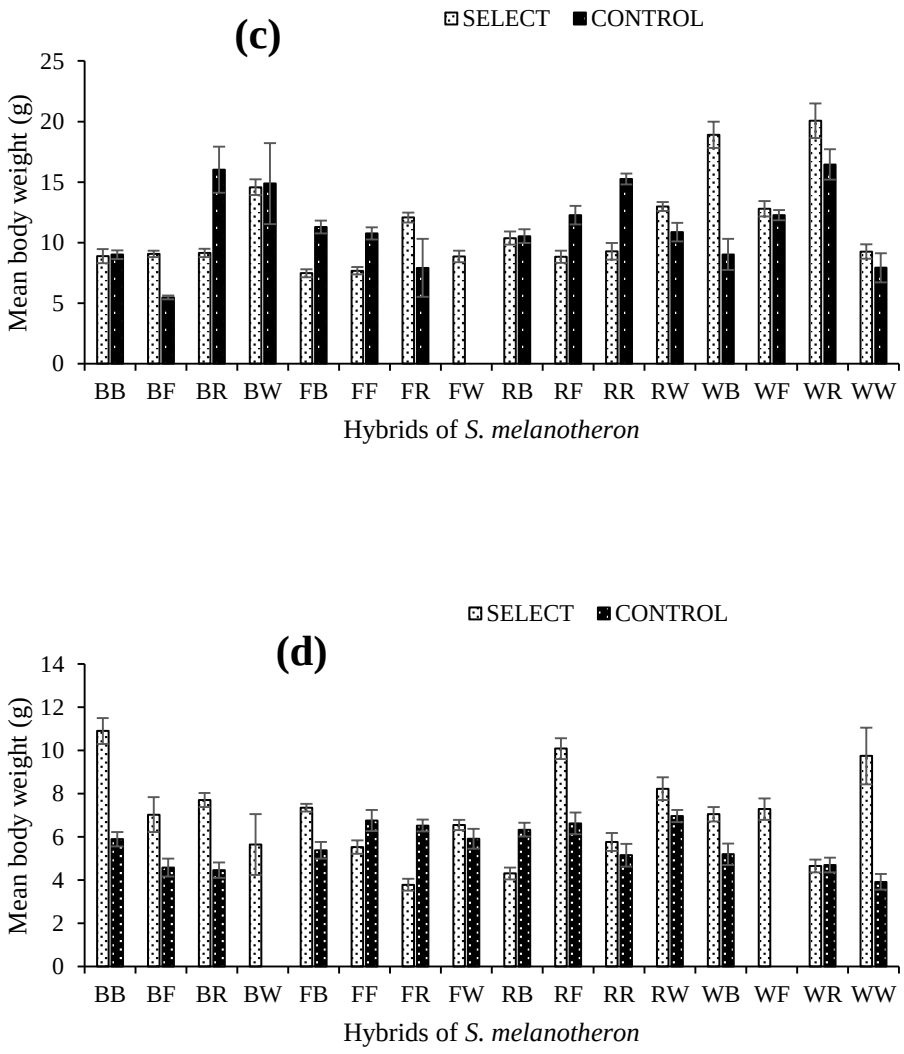


***Figure 15*: Mean body weight of 8-week old tagged offspring of female [one] × male [one] (c) and female [two] × male [one] (d) select and control lines before stocking in freshwater environment. Vertical bars represent standard errors**

Among the control line of female [one] × male [one], the hybrid BF exhibited the lowest mean body weight of 5.46 ± 0.17 g, while the hybrid WR

attained the highest mean body weight of 16.45 ± 1.25 g (Fig. 15c). The mean body weight of WR was not significantly different ($P > 0.05$) from BR, BW and RR, but was significantly ($P < 0.05$) higher than all the remaining experimental fish. The mean body weight of BF was significantly lower ($P < 0.05$) than all the other fish under investigations except WW, which exhibited no significant difference. It is worth noting that the hybrid WR exhibited highest mean body weight in both select and control lines. On the other hand, BF and FB exhibited the lowest mean body weights in the select and control lines respectively.

The mean body weights of offspring of female [two] × male [one] select line before stocking in freshwater ranged from 3.78 ± 0.27 g for the hybrid FR to 10.90 ± 0.60 g for BB (Fig. 15d). The mean body weight of FR was comparable to BW, FF, RB and WR but was significantly lower ($P < 0.05$) than the rest of the experimental fish. The mean body weight recorded for BB was not significantly different from RF and WW, but was significantly higher ($P < 0.05$) than all the remaining hybrids under study.

Mean body weight of tagged offspring of female [one] × male [one] and female [two] × male [one] select and control lines cultured in brackish water for 120 days

The offspring of female [one] male [one] and female [two] × male [one] of the various hybrids of both select and control lines were harvested after 120 days culture in brackish water. The salinity of the pond water during the culture period ranged from 6.11 to 7.25 ppt with a mean of 6.57 ± 0.13 ppt. The surface water temperature ranged between 25.9 ˚C and 28.9 ˚C with a mean of 27.51 ± 0.41 ˚C. The water pH were from 8.62 to 9.90 with a mean of 9.31 ± 0.16. The

dissolved oxygen (DO) level ranged from 2.32 mg/l to 6.82 mg/l, with a mean value of 4.49 ± 0.53 mg/l.

The growth performance of the offspring of female $_{[one]}$ × male $_{[one]}$ and female $_{[two]}$ × male $_{[one]}$ are shown in Figure 16. The final mean body weights of the select line of female $_{[one]}$ male $_{[one]}$ ranged from 26.12 ± 4.09 g to 54.82 ± 2.57 g (Fig. 16a) for WW and FB respectively. The mean body weight of WW was not significantly different from BB, BF, BR, RR and RW. On the other hand, the mean body weight of FB was significantly higher ($P < 0.05$) than all the other experimental fish except WB.

Among the control line of female $_{[one]}$ × male $_{[one]}$, the lowest mean body weight of 17.23 ± 1.03 g was attained by BR, while the highest mean body weight of 38.33 ± 1.42 g was achieved by the hybrid FB. The weight of BR was comparable to BB, but was significantly lower ($P < 0.05$) than all the other hybrids under investigations. On the other hand, mean body weight of FB was significantly higher ($P < 0.05$) than the rest of the fish under study (Fig. 16a).

The mean body weight of the select line of female $_{[two]}$ × male $_{[one]}$ ranged from 18.65 ± 1.34 g to 41.27 ± 8.97 g (Fig. 16b). The lowest mean body weight was attained by RR, while the highest was achieved by FR. The mean body weight of FR was not significantly different from FW and WR, but significantly higher ($P < 0.05$) than all the remaining experimental fish.

For the control line of female $_{[two]}$ × male $_{[one]}$, the hybrid RW exhibited the lowest mean body weight of 15.81 ± 0.94 g, while the highest mean body weight of 20.02 ± 3.04 g was attained by RB (Fig. 16b). The control and select lines of FF, FW and control lines of FR in Figure 16(a), as well as control line of WF in Figure 16(b) were lost due to rainstorm actions.

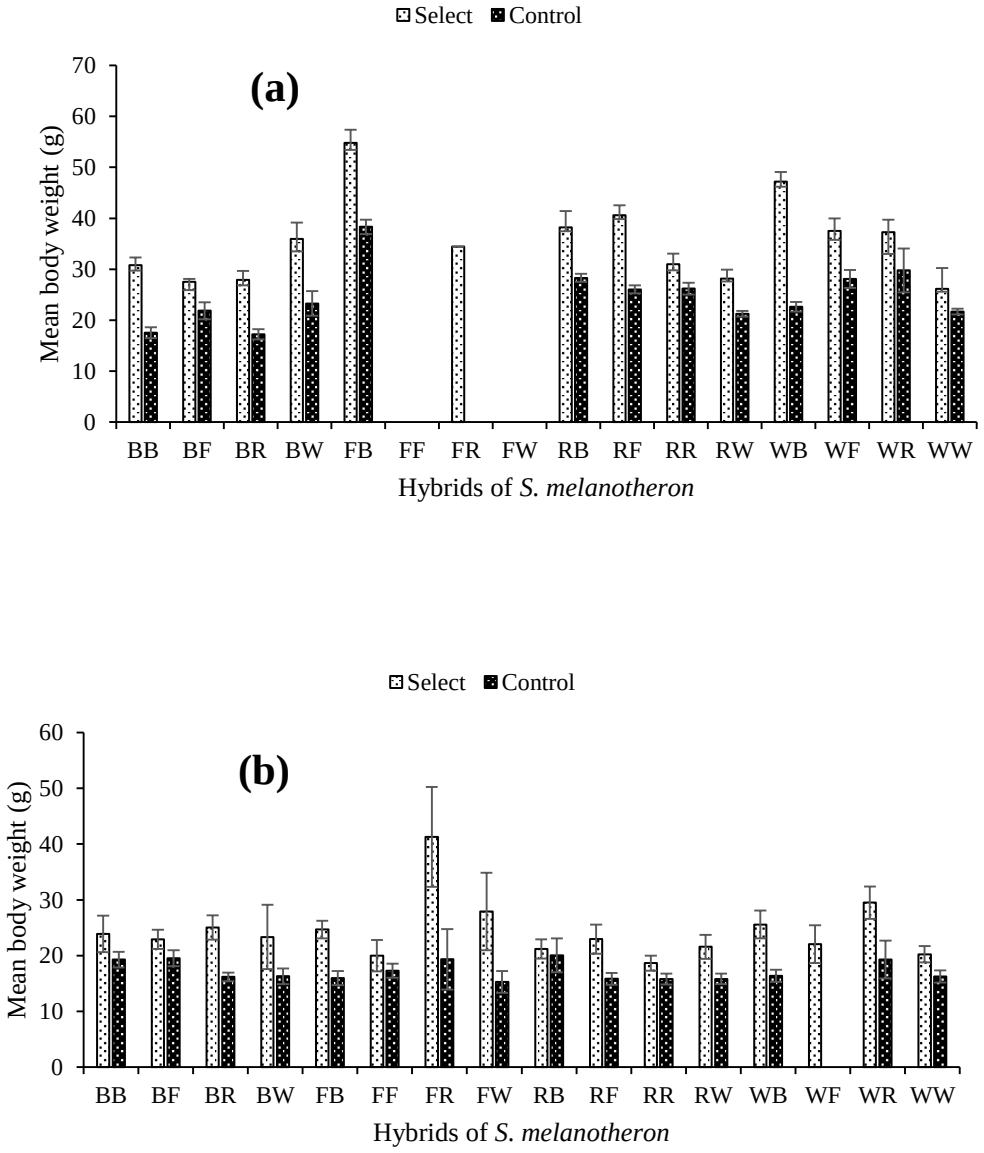


***Figure 16*: Mean body weight of female [one] × male [one] (a) and female [two] × male [one] (b) select and control lines of *S. melanotheron* cultured in brackish water for 120 days. Vertical bars represent standard errors**

Absolute growth rate of offspring of female [one] × male [one] select and control lines cultured in brackish water for 120 days

Mean absolute growth rate (AGR) of the select line of the offspring of female [one] × male [one] ranged from 0.14 ± 0.00 day^{-1} to 0.32 ± 0.00 g day^{-1} (Table 10). The lowest AGR was attained by hybrids BR and RW, while the highest was attained by FB. The mean AGR of FB was significantly ($P < 0.05$) higher than all the other hybrids. Among the sexes, females from FB had the highest AGR, while BR, RB and RW exhibited the lowest AGR. Males from FB exhibited the highest AGR, while those from WW had the lowest.

Table 10: ***Absolute growth rate of offspring of female [one] × male [one] select and control lines cultured in brackish water for 120 days***

	Select line (g day^{-1})				Control line (g day^{-1})		
Hybrids	♀	♂	**Mean ± SE**	**Hybrids**	♀	♂	**Mean ± SE**
BB	0.24	0.20	0.22 ± 0.02^{bc}	BB	0.11	0.09	0.10 ± 0.01^{cde}
BF	0.19	0.18	0.19 ± 0.01^{bcd}	BF	0.17	0.10	0.13 ± 0.03^{bc}
BR	0.14	0.14	0.14 ± 0.00^{cd}	BR	0.10	0.09	0.09 ± 0.01^{de}
BW	0.19	0.10	0.15 ± 0.05^{cd}	BW	*	*	*
FB	0.32	0.32	0.32 ± 0.00^{a}	FB	0.20	0.21	0.21 ± 0.01^{a}
FR	*	*	*	FF	*	*	*
FF	*	*	*	FR	*	*	*
FW	*	*	*	FW	*	*	*
RB	0.14	0.20	0.17 ± 0.03^{bcd}	RB	0.16	0.14	0.15 ± 0.01^{b}
RF	0.22	0.21	0.21 ± 0.00^{bcd}	RF	0.09	0.07	0.08 ± 0.01^{de}
RR	0.15	0.14	0.15 ± 0.01^{cd}	RR	0.10	0.09	0.09 ± 0.00^{de}
RW	0.14	0.14	0.14 ± 0.00^{d}	RW	0.08	0.07	0.07 ± 0.00^{e}
WB	0.20	0.23	0.21 ± 0.01^{bcd}	WB	0.14	0.12	0.13 ± 0.01^{bc}
WF	0.16	0.21	0.19 ± 0.02^{bcd}	WF	0.15	0.14	0.15 ± 0.00^{b}
WR	0.16	0.13	0.15 ± 0.01^{cd}	WR	0.17	0.15	0.16 ± 0.01^{b}
WW	0.21	0.09	0.15 ± 0.06^{cd}	WW	0.15	0.14	0.14 ± 0.00^{b}

Means with different letters in the superscripts are significantly different ($P < 0.05$), * Data not available due mortalities caused by rainstorm activities

In the control line of the offspring of female [one] × male [one], the highest AGR of 0.21 ± 0.01 g day^{-1} was attained by FB, while the lowest of 0.07 ± 0.00 g day^{-1} was achieved by RW (Table 10). The AGR of FB was significantly higher ($P < 0.05$) than all the other experimental fish, while the AGR of RW was not significantly different from BB, BR, RF and RR. In terms of performance based on sex, females from FB had the highest AGR of 0.20 g day^{-1}, whilst the lowest AGR of 0.08 g day^{-1} was recorded for RW after 120 days culture in brackish water. Amongst the males, the highest growth rate of 0.21 g day^{-1} was recorded for hybrid FB, whereas the lowest 0.07 g day^{-1} was attained by RF and RW.

Absolute growth rate of offspring of female [two] × male [one] select and control lines cultured in brackish water for 120 days

Table 11 shows the absolute growth rates of offspring of female [two] × male [one] select and control lines cultured in brackish water. The AGR for the select line was from 0.08 ± 0.00 g day^{-1} (WW) to 0.18 ± 0.00 g day^{-1} (WR). Analysis of variance on the select line indicated no significant difference among the mean AGRs as shown in Table 12. Among the females, the highest AGR of 0.21 g day^{-1} was attained by FR, while the lowest of 0.08 g day^{-1} was achieved by WW. The hybrids FW and WR had the highest AGR of 0.19 g day^{-1} among the males, whereas the lowest of 0.06 g day^{-1} was attained by RR and RW.

Table 11: ***Absolute growth rates of offspring of female [two] × male [one] select and control lines cultured in brackish water for 120 days***

	Select line (g day^{-1})				Control line (g day^{-1})		
Hybrids	♀	♂	**Mean ± SE**	**Hybrids**	♀	♂	**Mean ± SE**
BB	0.16	0.12	0.14 ± 0.02	BB	0.13	0.08	0.11 ± 0.03
BF	0.10	0.13	0.12 ± 0.01	BF	0.13	0.11	0.12 ± 0.01
BR	0.14	0.16	0.15 ± 0.01	BR	0.12	0.10	0.11 ± 0.01
BW	0.16	0.08	0.12 ± 0.04	BW	0.09	0.07	0.08 ± 0.01
FB	0.16	0.13	0.15 ± 0.02	FB	0.07	0.07	0.07 ± 0.00
FF	0.16	0.11	0.13 ± 0.02	FF	0.09	0.09	0.09 ± 0.00
FR	0.21	0.08	0.15 ± 0.06	FR	0.14	0.04	0.09 ± 0.05
FW	0.15	0.19	0.17 ± 0.02	FW	0.08	0.05	0.06 ± 0.02
RB	0.12	0.11	0.11 ± 0.01	RB	0.12	0.07	0.09 ± 0.03
RF	0.10	0.11	0.10 ± 0.00	RF	0.08	0.05	0.07 ± 0.02
RR	0.10	0.06	0.08 ± 0.02	RR	0.06	0.09	0.08 ± 0.01
RW	0.09	0.11	0.10 ± 0.01	RW	0.06	0.08	0.07 ± 0.01
WB	0.17	0.13	0.15 ± 0.03	WB	0.11	0.08	0.09 ± 0.02
WF	0.17	0.06	0.12 ± 0.06	WF	*	*	*
WR	0.18	0.19	0.18 ± 0.00	WR	0.12	0.08	0.10 ± 0.02
WW	0.08	0.08	0.08 ± 0.00	WW	0.09	0.09	0.09 ± 0.00

Table 12: ***ANOVA test on absolute growth rate of hybrids of S. melanotheron (female [two] × male [one]) select line cultured for 120 days in brackish water***

Source	**DF**	**SS**	**MS**	**F**	**P**
Factor	15	0.02629	0.00175	1.16	0.383
Error	16	0.02413	0.00151		
Total	31	0.05042			

The hybrid BF had the highest AGR of 0.12 ± 0.01 g day^{-1} in the control line of female [two] × male [one], while FW exhibited the lowest AGR of 0.06 ± 0.02 g day^{-1} (Table 11). No significant difference was observed among the means of the control line as shown in Table 13. In terms of performance by sexes, the females from FR exhibited the highest growth rate of 0.14 g day^{-1},

whereas the lowest AGR of 0.06 g day^{-1} was attained by RR and RW. Among the males, the BF had the highest AGR of 0.11 g day^{-1}, whilst the lowest of 0.04 g day^{-1} was attained by FR.

Table 13: ***ANOVA test on absolute growth rate of hybrids of S. melanotheron (female [two] × male [one]) control line cultured for 120 days in brackish water***

Source	DF	SS	MS	F	P
Factor	14	0.007569	0.000541	0.73	0.72
Error	15	0.011117	0.000741		
Total	29	0.018686			

Specific growth rate of offspring of female [one] × male [one] select and control lines cultured in brackish water for 120 days

The highest specific growth rate (SGR) of 1.64 ± 0.08 % day^{-1} in the select line of the female [one] × male [one] was attained by BB, whereas the lowest of 0.54 ± 0.07 % day^{-1} was exhibited by WR (Table 14). However, the SGR of BB was not significantly different from BF, but was significantly higher than the rest of hybrids. Amongst the sexes, the female BB had the highest SGR of 1.73 % day^{-1}, while the lowest of 0.61 % day^{-1} was attained by WB and WR. In the males' line, BB exhibited the highest SGR of 1.56, whereas the lowest of 0.47 was attained by WR.

Table 14: ***Specific growth rate of offspring of female [one] × male [one] select and control lines cultured in brackish water for 120 days***

	Select line (% day^{-1})				Control line (% day^{-1})		
Hybrids	**♀**	**♂**	**Mean ± SE**	**Hybrids**	**♀**	**♂**	**Mean ± SE**
BB	1.73	1.56	1.64 ± 0.08^{a}	BB	0.95	0.89	0.92 ± 0.03^{bcd}
BF	1.50	1.49	1.50 ± 0.01^{a}	BF	1.02	0.73	0.88 ± 0.15^{cd}
BR	0.74	0.85	0.80 ± 0.06^{cd}	BR	0.97	1.03	1.00 ± 0.03^{bc}
BW	0.70	0.47	0.58 ± 0.12^{d}	BW	*	*	*
FB	1.02	1.21	1.11 ± 0.09^{b}	FB	0.88	0.87	0.88 ± 0.01^{cd}
FF	*	*	*	FF	*	*	*
FR	*	*	*	FR	*	*	*
FW	*	*	*	FW	*	*	*
RB	0.65	0.74	0.70 ± 0.05^{d}	RB	0.98	0.89	0.94 ± 0.04^{bcd}
RF	1.13	0.86	1.00 ± 0.14^{bc}	RF	0.41	0.35	0.38 ± 0.03^{e}
RR	0.75	0.73	0.74 ± 0.01^{cd}	RR	0.44	0.49	0.47 ± 0.02^{e}
RW	0.73	0.76	0.74 ± 0.02^{cd}	RW	0.48	0.42	0.45 ± 0.03^{e}
WB	0.61	0.71	0.66 ± 0.05^{d}	WB	1.18	1.02	1.10 ± 0.08^{b}
WF	0.74	0.79	0.77 ± 0.02^{cd}	WF	0.80	0.77	0.79 ± 0.01^{d}
WR	0.61	0.47	0.54 ± 0.07^{d}	WR	0.99	0.86	0.92 ± 0.06^{bcd}
WW	1.23	0.80	1.02 ± 0.22^{bc}	WW	1.42	1.26	1.34 ± 0.08^{a}

Means with different letters in the superscripts are significantly different ($P < 0.05$), * Data not available due to mortalities caused by rainstorm activities

Mean SGR of the offspring of female [one] × male [one] control line ranged from 0.38 ± 0.03 % day^{-1} to 1.34 ± 0.08 % day^{-1} for RF and WW respectively (Table 14). The mean SGR of WW was significantly higher than all the hybrids, whilst the specific growth rate of RF was comparable to RR and RW, but was significantly lower than all the remaining hybrids under study. Based on sex, females of WW attained the highest SGR (1.42 % day^{-1}), while the lowest (0.41 % day^{-1}) amongst the females was recorded for RF. The purebreed WW again exhibited the highest SGR (1.26 % day^{-1}) among the males, while RF attained the lowest of 0.35 % day^{-1}.

Specific growth rate of offspring of female [two] × male [one] select and control lines cultured in brackish water for 120 days

The specific growth rate performance of select and control lines of the offspring of female [two] × male [one] are shown in Table 15. For the select line, the SGR ranged between 0.54 ± 0.05 % day^{-1} (WW) and 1.22 ± 0.03 (WR) % day^{-1}. The specific growth rate of WR was comparable to BB, BF, BR, BW, FB, FF, FW, RB and WB but was significantly higher than the rest of the hybrids under investigations. Among the sexes, BW exhibited the highest SGR of 1.25 % day^{-1} among the females, while WW had the lowest of 0.49 % day^{-1}. The hybrid WR exhibited the highest SGR of 1.22 % day^{-1} among the males, whilst WF had the lowest SGR of 0.44 % day^{-1}.

Table 15*: Specific growth rate of offspring of female [two] × male [one] select and control lines cultured in brackish water for 120 days*

	Select line (% day^{-1})				Control line (% day^{-1})		
Hybrids	**♀**	**♂**	**Mean ± SE**	**Hybrids**	**♀**	**♂**	**Mean ± SE**
BB	1.09	0.79	0.94 ± 0.15^{abcdef}	BB	0.96	0.71	0.83 ± 0.13^{bcd}
BF	0.81	0.87	0.84 ± 0.03^{abcdef}	BF	1.16	1.05	1.11 ± 0.06^{a}
BR	1.06	0.97	1.01 ± 0.05^{abcd}	BR	1.10	1.10	1.10 ± 0.00^{ab}
BW	1.25	0.98	1.12 ± 0.12^{ab}	BW	0.84	0.80	0.82 ± 0.02^{cd}
FB	1.08	0.92	1.00 ± 0.08^{abcd}	FB	0.68	0.54	0.61 ± 0.07^{d}
FF	1.22	0.97	1.10 ± 0.13^{abc}	FF	0.83	0.84	0.83 ± 0.01^{bcd}
FR	0.91	0.25	0.58 ± 0.33^{ef}	FR	0.83	0.46	0.64 ± 0.19^{d}
FW	1.07	1.01	1.04 ± 0.03^{abcd}	FW	0.74	0.45	0.59 ± 0.14^{d}
RB	0.93	0.82	0.87 ± 0.05^{abcdef}	RB	0.79	0.61	0.70 ± 0.09^{cd}
RF	0.73	0.64	0.69 ± 0.04^{cdef}	RF	0.66	0.51	0.59 ± 0.08^{d}
RR	0.70	0.57	0.63 ± 0.06^{def}	RR	0.62	0.83	0.73 ± 0.10^{cd}
RW	0.66	0.68	0.67 ± 0.01^{def}	RW	0.56	0.66	0.61 ± 0.05^{d}
WB	1.06	0.87	0.96 ± 0.09^{abcde}	WB	0.72	0.76	0.74 ± 0.02^{cd}
WF	1.07	0.44	0.75 ± 0.32^{bcdef}	WF	*	*	*
WR	1.19	1.25	1.22 ± 0.03^{a}	WR	0.61	0.84	0.73 ± 0.12^{cd}
WW	0.49	0.59	0.54 ± 0.05^{f}	WW	1.00	0.88	0.94 ± 0.06^{abc}

Means with different letters in the superscripts are significantly different ($P < 0.05$), * Data not available due to mortalities caused by rainstorm activities

In the control line of female [two] × male [one], the hybrid BF had the highest SGR of 1.11 ± 0.06 % day^{-1}, whilst RF attained the lowest mean of 0.59 ± 0.08 % day^{-1}. However, the SGR of BF was not significantly different from BR and WW, but was significantly higher than the rest of the hybrids under study. Based on sexes, BF from the female line attained the highest SGR of 1.16 % day^{-1}, whilst the lowest of 0.56 % day^{-1} among the females was attained by RW. Among the males, BR exhibited the highest SGR of 1.10 % day^{-1}, whereas the lowest of 0.45 % day^{-1} was attained by FW.

Mean body weight of offspring of female [one] × male [one] and female [two] × male [one] select and control lines cultured in freshwater for 120 days

Figure 17 shows the mean weights of offspring of female [one] × male [one] and female [two] × male [one] select and control lines cultured in freshwater. The final mean body weights of offspring of female [one] male [one] ranged from 46.48 ± 2.81 g to 73.35 ± 3.80 g (Fig. 17a). The lowest mean body weight was attained by RW, while BW had the highest mean body weight. The mean body weight of RW was not significantly different ($P > 0.05$) from BB, FB, FF, FR, FW, RR and WW but was significantly lower than the rest of the hybrids. On the other hand, the mean body weight of BW was significantly higher ($P < 0.05$) than all the hybrids under study except BF, RB and WF.

In the control line of the offspring of female [one] × male [one], the mean body weight ranged from 37.81 ± 2.44 g for WW to 68.77 ± 3.72 g for BW. The mean body weight of WW was comparable to BB, BF, FB, FF, FR, RR and RW but was significantly lower than the rest of the experimental fish. The mean body weight of BW was significantly higher ($P < 0.05$) than all the hybrids

except RB, WB and WF. Data on the control line of FW in Figure 17(a) and control lines of BW and WF in Figure 17(b) were not available due to mortalities caused by rainstorm activities.

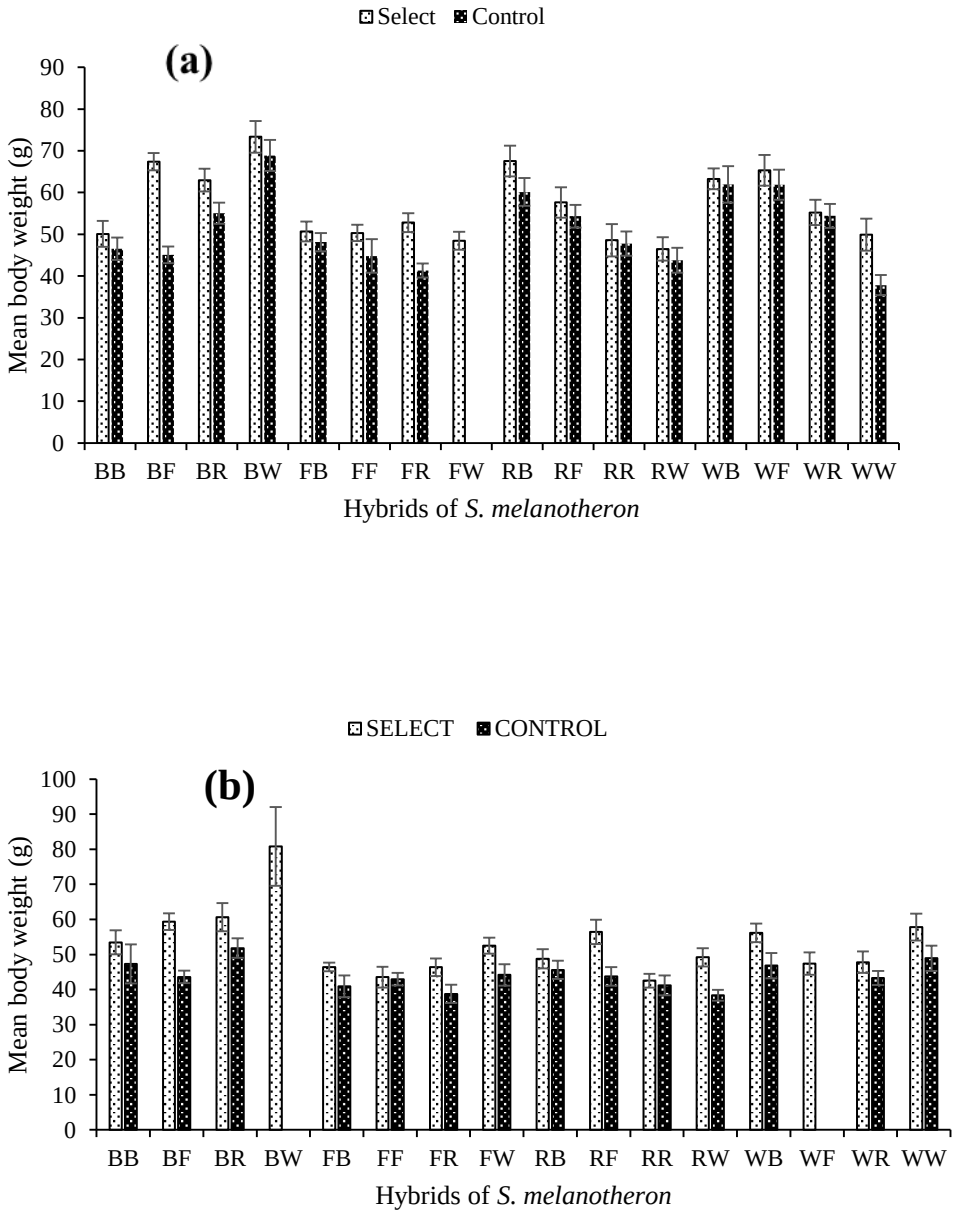


***Figure 17*: Mean body weight of offspring of female [one] × male [one] (a) and female [two] × male [one] (b) select and control lines of *S. melanotheron* cultured in freshwater for 120 days. Vertical bars represent standard errors**

In the select line of offspring of female $_{[two]}$ × male $_{[one]}$, the mean body weights ranged between 42.52 ± 1.96 g (RR) and 80.78 ± 11.20 g (BW) (Fig. 17b). The mean body weight of RR was not significantly different from FB, FF, FR, RB, RW, WF and WR but was significantly less ($P < 0.05$) than the rest of the hybrids. On the other hand, the mean body weight of BW was significantly higher ($P < 0.05$) than all the other hybrids under investigations.

The mean body weights of offspring of female $_{[two]}$ × male $_{[one]}$ control line ranged from 38.37 ± 1.55 g for RW to 51.81 ± 2.78 g for BR. The mean body weight achieved by RW was comparable to BF, FB, FF, FR, FW, RF, RR and WR, but was significantly lower ($P < 0.05$) than the rest of the experimental fish (Fig. 17b).

Absolute growth rate of offspring of female $_{[one]}$ × male $_{[one]}$ select and control lines cultured in freshwater for 120 days

The mean absolute growth rate of the offspring of female $_{[one]}$ × male $_{[one]}$ ranged from 0.28 ± 0.02 g day^{-1} for RW to 0.49 ± 0.02 g day^{-1} for BF (Table 16). However, the AGR of BF was not significantly different from BR, BW, RB, RF and WF but was significantly higher than all the remaining hybrids. Among the females, BF exhibited the highest growth rate of 0.51 g day^{-1}, while the lowest growth rate of 0.21 g day^{-1} was attained by RW. In the males, BW had the highest growth rate of 0.50 g day^{-1}, whilst RR exhibited the lowest of 0.26 g day^{-1}.

In the control line of female $_{[one]}$ × male $_{[one]}$, the mean AGR was between 0.23 ± 0.09 g day^{-1} and 0.45 ± 0.00 g day^{-1} for FW and BW respectively. The

AGR attained by BW was not significantly different from RB, RF, WB and WF but was significantly higher than the rest of the experimental hybrids. Among the sexes, BW exhibited the highest growth rate of 0.45 g day^{-1} amongst the females, while FW had the lowest growth rate of 0.14 g day^{-1}. Among the males, WB had the highest growth rate of 0.49 g day^{-1}, whilst the lowest AGR of 0.22 g day^{-1} was attained by RW.

Table 16: ***Absolute growth rate of offspring of female [one] × male [one] select and control lines cultured in freshwater for 120 days***

	Select line (g day^{-1})				Control line (g day^{-1})		
Hybrids	♀	♂	**Mean ± SE**	**Hybrids**	♀	♂	**Mean ± SE**
BB	0.34	0.32	0.33 ± 0.03^{cde}	BB	0.36	0.27	0.31 ± 0.04^{def}
BF	0.51	0.46	0.49 ± 0.02^{a}	BF	0.33	0.33	0.33 ± 0.00^{bcdef}
BR	0.43	0.44	0.43 ± 0.02^{ab}	BR	0.33	0.28	0.31 ± 0.03^{def}
BW	0.46	0.50	0.48 ± 0.03^{a}	BW	0.45	0.45	0.45 ± 0.00^{a}
FB	0.35	0.36	0.35 ± 0.02^{cde}	FB	0.29	0.32	0.31 ± 0.02^{def}
FF	0.34	0.36	0.35 ± 0.02^{cde}	FF	0.23	0.31	0.27 ± 0.04^{ef}
FR	0.33	0.34	0.33 ± 0.02^{cde}	FR	0.26	0.29	0.28 ± 0.01^{ef}
FW	0.34	0.31	0.33 ± 0.02^{de}	FW	0.14	0.31	0.23 ± 0.09^{f}
RB	0.45	0.43	0.44 ± 0.03^{ab}	RB	0.40	0.42	0.41 ± 0.01^{abcd}
RF	0.41	0.40	0.41 ± 0.03^{abc}	RF	0.36	0.34	0.35 ± 0.01^{abcde}
RR	0.39	0.26	0.31 ± 0.03^{de}	RR	0.30	0.24	0.27 ± 0.03^{ef}
RW	0.21	0.35	0.28 ± 0.02^{e}	RW	0.29	0.22	0.25 ± 0.04^{ef}
WB	0.36	0.37	0.36 ± 0.02^{bcd}	WB	0.39	0.49	0.44 ± 0.05^{ab}
WF	0.43	0.46	0.44 ± 0.03^{ab}	WF	0.37	0.48	0.42 ± 0.06^{abc}
WR	0.29	0.27	0.28 ± 0.03^{e}	WR	0.30	0.33	0.31 ± 0.02^{cdef}
WW	0.34	0.33	0.34 ± 0.03^{cde}	WW	*	*	*

Means with different letters in the superscripts are significantly different ($P < 0.05$), * Data not available due to mortalities caused by rainstorm activities

Absolute growth rate of offspring of female [two] × male [one] select and control lines cultured in freshwater for 120 days

Table 17 shows the absolute growth rates of the select and control lines of the offspring of female [two] × male [one]. In the select line, the AGR ranged from 0.30 ± 0.04 g day^{-1} for RR to 0.65 ± 0.04 g day^{-1} for BW. The AGR of BW was significantly higher than all the experimental fish. Among the sexes, females from BW exhibited the highest AGR of 0.61 g day^{-1}, while RR attained the lowest of 0.26 g day^{-1}. Among the males, BW attained the highest growth rate of 0.70 g day^{-1}, whilst FF had the lowest of 0.33 g day^{-1}.

Table 17: ***Absolute growth rate of offspring of female [two] × male [one] select and control lines cultured in freshwater for 120 days***

	Select line (g day^{-1})				Control line (g day^{-1})		
Hybrids	♀	♂	**Mean ± SE**	**Hybrids**	♀	♂	**Mean ± SE**
BB	0.34	0.36	0.35 ± 0.01^{bc}	BB	0.59	0.31	0.45 ± 0.14
BF	0.40	0.48	0.44 ± 0.04^{b}	BF	0.31	0.34	0.32 ± 0.01
BR	0.40	0.46	0.43 ± 0.03^{b}	BR	0.38	0.40	0.39 ± 0.01
BW	0.61	0.70	0.65 ± 0.04^{a}	BW	*	*	*
FB	0.33	0.38	0.36 ± 0.03^{bc}	FB	0.26	0.34	0.30 ± 0.04
FF	0.30	0.33	0.31 ± 0.02^{bc}	FF	0.31	0.24	0.27 ± 0.04
FR	0.35	0.36	0.36 ± 0.00^{bc}	FR	0.26	0.28	0.27 ± 0.01
FW	0.31	0.42	0.37 ± 0.05^{bc}	FW	0.27	0.39	0.33 ± 0.06
RB	0.29	0.44	0.37 ± 0.08^{bc}	RB	0.33	0.33	0.33 ± 0.00
RF	0.32	0.42	0.37 ± 0.05^{bc}	RF	0.31	0.31	0.31 ± 0.00
RR	0.26	0.34	0.30 ± 0.04^{c}	RR	0.31	0.30	0.30 ± 0.01
RW	0.30	0.38	0.34 ± 0.04^{bc}	RW	0.22	0.31	0.26 ± 0.04
WB	0.37	0.44	0.41 ± 0.03^{bc}	WB	0.28	0.42	0.35 ± 0.07
WF	0.27	0.38	0.33 ± 0.05^{bc}	WF	*	*	*
WR	0.28	0.41	0.35 ± 0.06^{bc}	WR	0.29	0.33	0.31 ± 0.02
WW	0.41	0.40	0.40 ± 0.01^{bc}	WW	0.33	0.41	0.37 ± 0.04

Means with different letters in the superscripts are significantly different ($P < 0.05$), * Data not available due to mortalities caused by rainstorm activities

The purebreed BB had the highest AGR of 0.45 ± 0.14 g day^{-1} in the control line of female [two] × male [one], whereas the lowest growth rate of 0.26 ± 0.04 g day^{-1} was attained by RW. An ANOVA test on the control line

indicated no significant difference among the means as shown in Table 18. In terms of performance by sex, BB achieved the highest AGR of 0.56 g day^{-1} among the females, whilst RW had the lowest growth rate of 0.22 g day^{-1}. In the line of the males, WW exhibited the highest AGR of 0.41 g day^{-1}, while FF attained the lowest of 0.24 g day^{-1}.

Table 18: ***ANOVA test on absolute growth rate of hybrids of S. melanotheron (female [two] × male [one]) control line cultured for 120 days in freshwater***

Source	DF	SS	MS	F	P
Factor	13	0.06805	0.00523	1.09	0.433
Error	14	0.06703	0.00479		
Total	27	0.13508			

Specific growth rate of offspring of female [one] × male [one] select and control lines cultured in freshwater for 120 days

The specific growth rates of the **s**elect and control lines of female [one] × male [one] are shown in Table 19. The SGR for the select line ranged from 0.83 ± 0.02 % day^{-1} to 1.67 ± 0.02 % day^{-1} for WR and BF respectively. However, the SGR of BF was not significantly different from BR, FB, FF and RF, but was significantly higher than the remaining hybrids under examination. Among the females, BF exhibited the highest SGR of 1.69 % day^{-1}, while the WR had the lowest SGR of 0.82 % day^{-1}. The hybrid BF from the males attained the highest SGR of 1.67 % day^{-1}, whilst WR had the lowest performance of 0.85 % day^{-1}.

In the control line, the hybrid BF had the highest specific growth rate of 1.75 ± 0.02 % day^{-1}, while the lowest SGR of 0.83 ± 0.02 % day^{-1} was attained by WR. The SGR of BF was comparable to BR, FB, FF and RF, but was

significantly higher than all the other hybrids. Among the sexes, females from BF exhibited the highest SGR of 1.77 % day^{-1}, whereas FW had the lowest of 0.26 % day^{-1}. In the males, BF had the highest SGR of 1.73 % day^{-1}, while the hybrid FW attained the lowest SGR of 0.80 % day^{-1}.

Table 19: ***Mean specific growth rate of offspring of female*** *[one]* ***× male*** *[one]* ***select and control lines cultured in freshwater for 120 days***

	Select line (% day^{-1})				Control line (% day^{-1})		
Hybrids	♀	♂	**Mean ± SE**	**Hybrids**	♀	♂	**Mean ± SE**
BB	1.34	1.36	1.35 ± 0.01^{bcde}	BB	1.51	1.20	1.35 ± 0.15^{cdef}
BF	1.69	1.65	1.67 ± 0.02^{a}	BF	1.77	1.73	1.75 ± 0.02^{a}
BR	1.46	1.56	1.51 ± 0.05^{abcd}	BR	1.10	0.87	0.99 ± 0.12^{hi}
BW	1.20	1.34	1.27 ± 0.07^{def}	BW	1.45	1.65	1.55 ± 0.10^{abcd}
FB	1.43	1.63	1.53 ± 0.10^{abc}	FB	1.22	1.19	1.21 ± 0.02^{efghi}
FF	1.56	1.50	1.53 ± 0.03^{ab}	FF	1.11	1.15	1.13 ± 0.02^{efghi}
FR	1.15	1.23	1.19 ± 0.04^{efg}	FR	1.61	1.67	1.64 ± 0.03^{abc}
FW	1.41	1.35	1.38 ± 0.03^{bcde}	FW	0.26	0.80	0.53 ± 0.27^{j}
RB	1.51	1.27	1.39 ± 0.12^{bcde}	RB	1.48	1.36	1.42 ± 0.06^{bcde}
RF	1.55	1.56	1.56 ± 0.00^{ab}	RF	1.28	1.21	1.25 ± 0.04^{defgh}
RR	1.46	1.12	1.29 ± 0.17^{cde}	RR	0.98	0.86	0.92 ± 0.06^{i}
RW	0.88	1.19	1.04 ± 0.16^{fgh}	RW	1.11	1.00	1.05 ± 0.06^{fghi}
WB	0.93	1.10	1.01 ± 0.08^{gh}	WB	1.68	1.69	1.68 ± 0.012^{ab}
WF	1.29	1.46	1.37 ± 0.09^{bcde}	WF	1.22	1.45	1.33 ± 0.11^{cdefg}
WR	0.82	0.85	0.83 ± 0.02^{h}	WR	0.90	1.17	1.04 ± 0.14^{ghi}
WW	1.35	1.42	1.39 ± 0.03^{bcde}	WW	*	*	*

Means that do not share a letter in the superscripts are significantly different ($P < 0.05$), * Data not available due to mortalities caused by rainstorm activities

Specific growth rate of offspring of female *[two]* ***× male*** *[one]* ***select and control lines cultured in freshwater for 120 days***

Table 20 shows the specific growth rates of the offspring of female [two] × male [one] select and control lines cultured in freshwater. The specific growth rates in the select line ranged between 1.32 ± 0.05 % day^{-1} (BB) and 2.32 ± 0.13 % day^{-1} (BW). The SGR attained by BW was not significantly different from FR, but was significantly higher than the remaining hybrids under

investigations. Among the sexes, BW had 2.19 % day^{-1}, which was the highest SGR among the females, while BB attained the lowest SGR of 1.26 % day^{-1}. In the males, BW exhibited the highest SGR of 2.44 % day^{-1}, whilst BB had the lowest SGR of 1.37 % day^{-1}.

Table 20: ***Mean specific growth rate of offspring of female [two] × male [one] select and control lines cultured in freshwater for 120 days***

	Select line (% day^{-1})				Control line (% day^{-1})		
Hybrids	♀	♂	**Mean ± SE**	**Hybrids**	♀	♂	**Mean ± SE**
BB	1.26	1.37	1.32 ± 0.05^{h}	BB	1.96	1.59	1.77 ± 0.19^{cde}
BF	1.70	1.96	1.83 ± 0.13^{bcde}	BF	1.88	1.92	1.90 ± 0.02^{abc}
BR	1.65	1.65	1.65 ± 0.00^{defg}	BR	2.07	2.04	2.06 ± 0.02^{ab}
BW	2.19	2.44	2.32 ± 0.13^{a}	BW	*	*	*
FB	1.57	1.50	1.54 ± 0.04^{efgh}	FB	1.65	1.72	1.68 ± 0.03^{cdef}
FF	1.69	1.72	1.70 ± 0.01^{def}	FF	1.61	1.46	1.54 ± 0.07^{efg}
FR	2.03	2.22	2.13 ± 0.09^{ab}	FR	1.49	1.45	1.47 ± 0.02^{fg}
FW	1.55	1.75	1.65 ± 0.10^{defg}	FW	1.60	1.80	1.70 ± 0.10^{cdef}
RB	1.83	2.20	2.01 ± 0.19^{bc}	RB	1.62	1.65	1.63 ± 0.01^{defg}
RF	1.37	1.42	1.39 ± 0.03^{gh}	RF	1.57	1.57	1.57 ± 0.00^{efg}
RR	1.52	1.81	1.67 ± 0.14^{defg}	RR	1.71	1.77	1.74 ± 0.03^{cde}
RW	1.34	1.61	1.47 ± 0.14^{fgh}	RW	1.31	1.55	1.43 ± 0.12^{g}
WB	1.73	1.73	1.73 ± 0.00^{cdef}	WB	1.71	1.98	1.84 ± 0.14^{bcd}
WF	1.50	1.60	1.55 ± 0.05^{defg}	WF	*	*	*
WR	1.70	2.03	1.86 ± 0.17^{bcd}	WR	1.86	1.78	1.82 ± 0.04^{bcd}
WW	1.61	1.47	1.54 ± 0.07^{efgh}	WW	2.11	2.14	2.12 ± 0.02^{a}

Means that do not share a letter in the superscripts are significantly different ($P < 0.05$), * Data not available due to mortalities caused by rainstorm activities

In the control line of female [two] × male [one], the SGR ranged from 1.43 ± 0.12 % day^{-1} for RW to 2.12 ± 0.02 % day^{-1} for WW (Table 20). The SGR attained by WW was significantly higher than all the hybrids except BF and BR. Among the sexes, WW had the highest SGR among both sexes, while the lowest SGR was attained by RW (1.31 % day^{-1}) and FR (1.45 % day^{-1}) for females and males respectively.

Growth performance of offspring of female [one] versus female [two] select lines in freshwater

Figure 18 shows the comparative growth performance of offspring of female [one] and female [two] in freshwater for 120 days. The mean body weights exhibited by the offspring of female [one] ranged from 46.48 ± 2.81 g for RW to 73.35 ± 3.80 g for BW, whereas that of female [two] ranged from 42.52 ± 1.69 g for RR to 80.80 ± 11.20 g for BW. The growth performance of eight hybrids viz BF, FB, FF, FR, RB, WB, WF and WR of female [one] were significantly higher than those recorded for the offspring of female [two]. The hybrid BW had the highest final mean body weight in both female [one] and female [two] lines. This could imply that BW may possess a superior growth gene as compared to the rest of the fish.

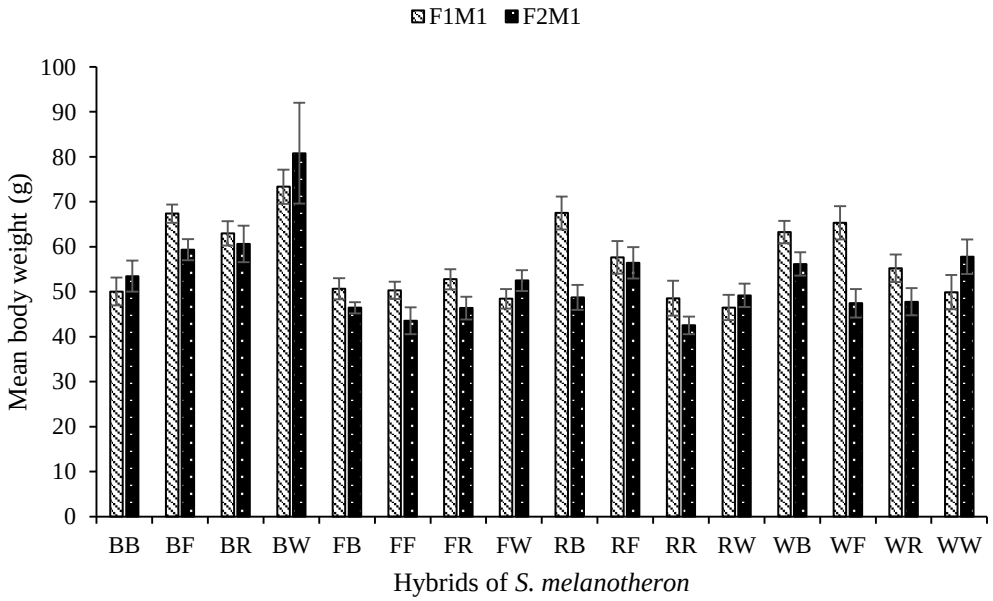


***Figure 18*: Comparison of mean body weights of offspring of female [one] and female [two] select lines of *S. melanotheron* cultured in freshwater for 120 days. Vertical bars represent standard errors**

Growth performance of offspring of female [one] versus female [two] control lines in freshwater

The data on comparative growth performance analysis of the control lines of offspring of female [one] and female [two] cultured in freshwater for 120 days are shown in Figure 19. For female [one], WW had the lowest mean body weight of 37.81 ± 2.44 g, whereas the highest mean body weight of 68.88 ± 3.72 g was attained by BW. Among the offspring of female [two], RW exhibited the lowest mean body weight of 38.37 ± 1.55 g, while the highest of 51.81 ± 2.78 g was attained by BR. Seven hybrids (FB, RB, RF, RR, RW, WB and WR) of female [one] had mean body weights that were significantly higher than that of their counterparts among the female [two] offspring. On the other hand, the purebreed WW of female [two] grew significantly heavier compared to its counterparts in the female [one] line. Data on BW and WF of female [two] as well as FW of female [one] were not available due to mortalities caused by rainstorm activities.

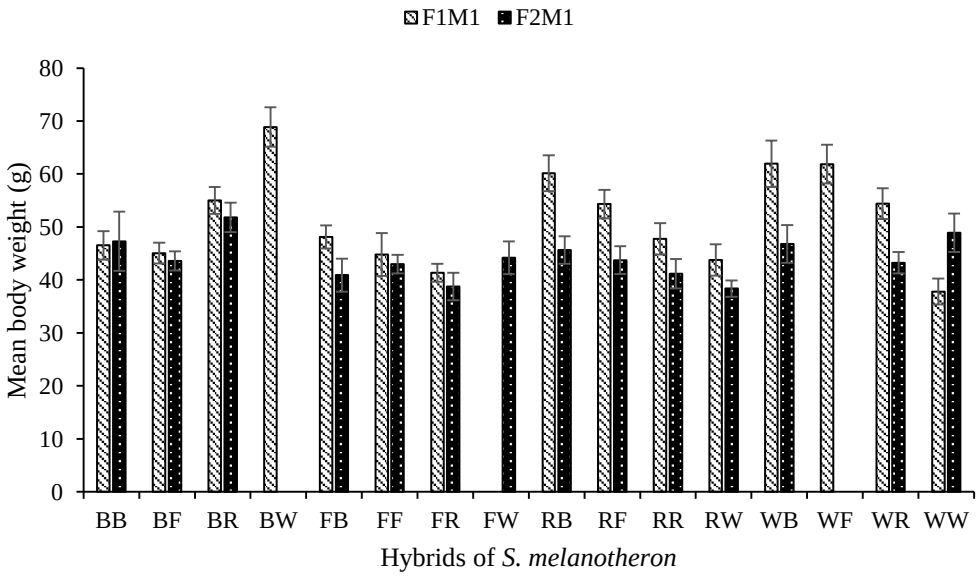


Figure *19*: Comparison of mean body weights of offspring of female [one] and female [two] control lines of *S. melanotheron* cultured in freshwater for 120 days. Vertical bars represent standard errors

Growth performance of offspring of female [one] versus female [two] select lines in brackish water

Figure 20 shows the comparative growth performance of offspring of female [one] and female [two] select lines cultured in brackish water for 120 days. The mean body weights of offspring of female [one] select ranged from 26.12 ± 4.09 g (WW) to 54.82 ± 2.57 g (FB). For the female [two] line, the mean body weights ranged from 18.65 ± 1.34 g for RR to 41.27 ± 8.97 g for FR. The mean body weights of offspring of female [one] select line were always higher than that of female [two] except FR. Data on FF and FW of female [one] were not available due to mortalities caused by rainstorm activities.

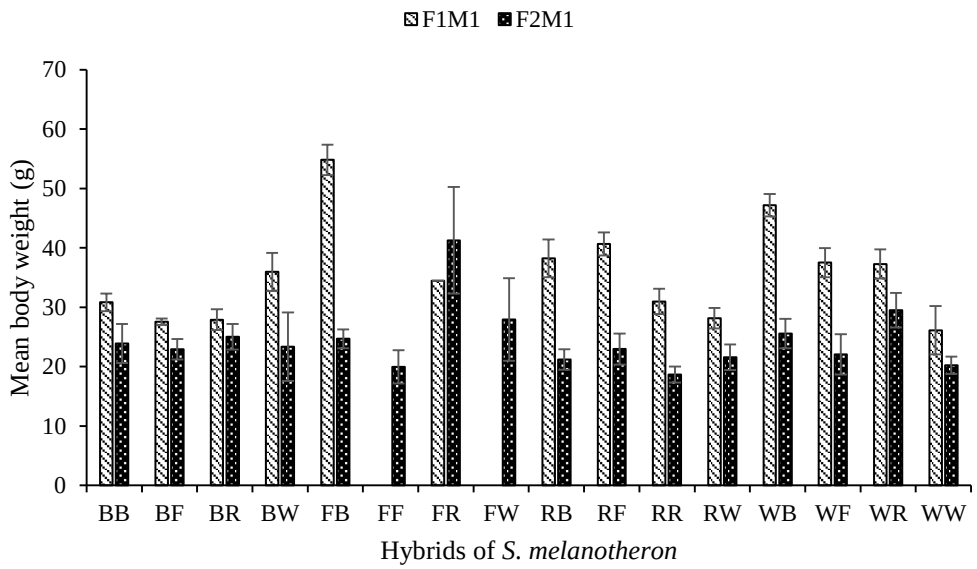


***Figure 20*: Comparison of mean body weight of offspring of female [one] and female [two] select lines of *S. melanotheron* cultured in brackish water for 120 days. Vertical bars represent standard errors**

Growth performance of offspring of female [one] versus female [two] control lines in brackish water

The comparative mean body weights of offspring of female [one] and female [two] control lines cultured in brackish water for 120 days are indicated in Figure 21. The mean body weights of offspring of female [one] ranged from 17.23 ± 1.03 g (BR) to 38.33 ± 1.42 g (FB). On the other hand, the mean body weights of offspring of female [two] ranged from 15.30 ± 1.95 g (FW) to 20.02 ± 3.04 g (RB). The mean body weights of offspring of female [one] were significantly higher compared to offspring of female [two] except BB, BF and BR. Data on FF, FR and FW of female [one] as well as WF of female [two] were not available due to mortalities caused by rainstorm activities.

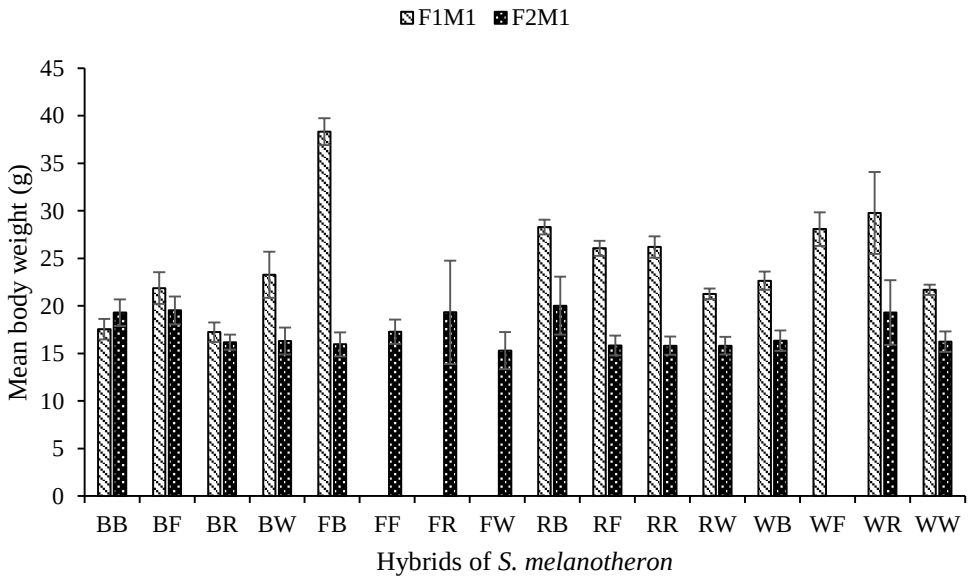


***Figure 21*: Comparison of mean body weights of offspring of female [one] and female [two] control lines of *S. melanotheron* hybrids cultured in brackish water for 120 days. Vertical bars represent standard errors**

Maternal and common environmental effect on hybrids of *S. melanotheron* cultured in fresh and brackish water for 120 days

Table 21 shows the maternal and environmental effect (C^2) estimates of hybrids of *S. melanotheron* cultured in fresh and brackish water for 120 days. The maternal and common environmental effect (C^2) estimates in freshwater ranged from 0.08 ± 0.07 (BB) to 0.29 ± 0.15 (FR), whereas in brackish water it ranged from 0.10 ± 0.07 (BW) to 0.36 ± 0.13 (WR). When the mean C^2 estimates were subjected to ANOVA test it gave P-values of 0.899 and 0.281 for freshwater and brackish water respectively, which were greater than 0.05 (level of statistical significance), indicating no significant differences. It is worth noting that the overall maternal and common environmental effect of the crossbreeds in freshwater amounted to 0.18 ± 0.07, which was lower compared to 0.25 ± 0.06 for brackish water.

Table 21: ***Maternal and common environmental effect of hybrids of S. melanotheron cultured in fresh and brackish water for 120 days***

Maternal and common environmental effect (C^2)		
	Freshwater	**Brackish water**
Hybrid	**Mean ± SE**	**Mean ± SE**
BB	0.10 ± 0.05	0.16 ± 0.07
BF	0.19 ± 0.07	0.33 ± 0.15
BR	0.26 ± 0.15	0.31 ± 0.08
BW	0.24 ± 0.11	0.10 ± 0.07
FB	0.08 ± 0.07	0.21 ± 0.03
FF	0.17 ± 0.07	*
FR	0.29 ± 0.15	*
FW	0.24 ± *	*
RB	0.11 ± 0.04	0.21 ± 0.04
RF	0.14 ± 0.00	0.28 ± 0.01
RR	0.18 ± 0.12	0.32 ± 0.02
RW	0.21± 0.04	0.22 ± 0.03
WB	0.17 ± 0.06	0.36 ± 0.03
WF	0.20 ± 0.00	0.19 ± 0.05
WR	0.22 ± 0.02	0.36 ± 0.13
WW	0.11 ± 0.11	0.20 ± 0.02
Mean ± SE	**0.18 ± 0.07**	**0.25 ± 0.06**

Comparison of growth performance of hybrids of *S. melanotheron* in brackish water and freshwater

Growth performance of full-sib (female [one] × male [one]) hybrids of S. melanotheron select lines in fresh and brackish water

Data on mean body weights of full-sib hybrids of *S. melanotheron* select lines cultured in fresh and brackish water are shown in Figure 22. The mean body weight of the full-sib hybrids cultured in freshwater ranged from 46.48 ± 2.81 g (RW) to 73.35 ± 3.8 g (BW), whereas those in the brackish water was between 26.12 ± 4.09 g (WW) and 54.82 g (FB). The mean weights of hybrids

cultured in freshwater were higher consistently compared to their counterparts in the brackish water except FB, which grew better in brackish water than in freshwater. Data on FF and FW in brackish water were not available due to mortalities caused by rainstorm activities.

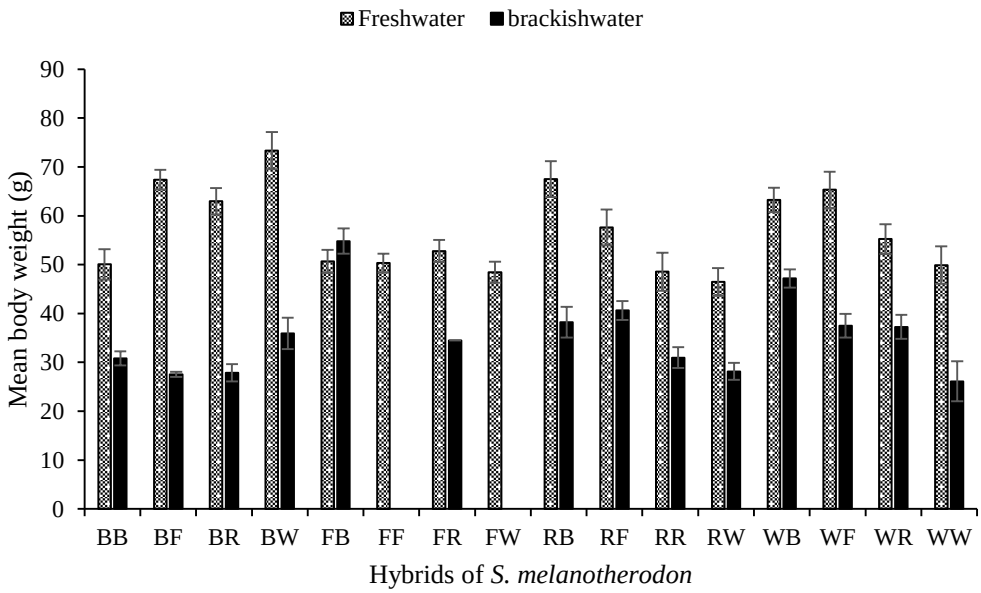


***Figure 22*: Comparison of mean body weights of select lines of full-sib *S. melanotheron* hybrids cultured in brackish and freshwater for 120 days. Vertical bars represent standard errors**

Growth performance of half-sib (female [two] × male [one]) hybrids of S. melanotheron select lines cultured in brackish and freshwater

Figure 23 shows the comparison of the mean body weights of the select lines of female [two] × male [one] cultured in brackish and freshwater for 120 days. The mean body weight of the hybrids in freshwater ranged from 42.52 ± 1.96 g for RR to 80.80 ± 11.2 g for BW, while in brackish water it ranged from 18.65 ± 1.34 g (RR) to 41.27 ± 8.97 g (FR). The final mean weight of the hybrids in freshwater were always significantly higher than their counterparts in brackish water except FR, where there was no significant difference.

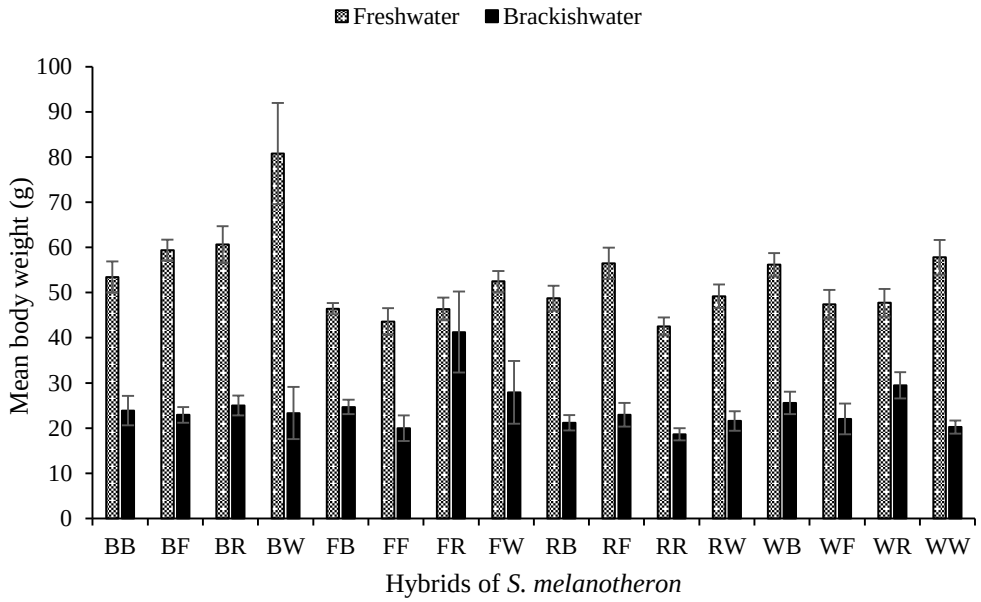


Figure 23: Comparison of mean body weights of select lines of half-sib *S. melanotheron* cultured in brackish and freshwater for 120 days. Vertical bars represent standard errors

Genetic improvement per generation due to selection

Female [one] × Male [one] offspring cultured in freshwater for 120 days

The genetic improvement (GI) of the hybrids in percentages are shown in Table 22. The highest mean genetic improvement of 48.41 ± 7.49 % was attained by the hybrid BF, whereas the lowest GI of 1.78 ± 0.72 % was recorded for the hybrid WR. The genetic improvement of BF was comparable to WW, but was significantly higher than all the other hybrids under investigations. Among the purebreeds, WW had the highest genetic improvement of 30.84 ± 11.9 %, while FF attained the lowest of 14.68 ± 7.46 %.

Table 22: ***Percentage genetic improvement of offspring of female*** $_{[one]}$ ***× male*** $_{[one]}$ ***cultured in freshwater for 120 days***

	Replicates of GI (%)			
Hybrids	**1**	**2**	**3**	**Mean ± SE (%)**
BB	19.55	6.07	26.11	17.24 ± 5.9^{bcde}
BF	48.93	35.18	61.12	48.41 ± 7.49^{a}
BR	19.41	3.60	17.87	13.62 ± 5.03^{bcde}
BW	14.01	10.95	5.44	10.13 ± 2.51^{cde}
FB	41.05	1.85	19.45	20.8 ± 11.3^{bcde}
FF	0.99	26.66	16.38	14.68 ± 7.46^{bcde}
FR	18.14	23.07	45.06	28.76 ± 8.28^{bc}
FW	*	*	*	*
RB	20.09	21.67	0.49	14.08 ± 6.81^{bcde}
RF	13.07	15.59	16.91	15.19 ± 1.13^{bcde}
RR	40.15	5.27	25.60	23.7 ± 10.1^{bcd}
RW	10.33	8.71	8.93	9.32 ± 0.51^{de}
WB	8.94	9.05	10.14	9.38 ± 0.38^{cde}
WF	7.45	0.41	10.27	6.04 ± 2.93^{de}
WR	2.93	0.46	1.94	1.78 ± 0.72^{e}
WW	14.15	24.47	53.88	30.8 ± 11.9^{ab}

Means that do not share a letter in the superscripts are significantly different.
* Data not available due to mortalities caused by rainstorm activities

Female $_{[two]}$ ***× Male*** $_{[one]}$ ***offspring cultured in freshwater for 120 days***

Table 23 shows the genetic improvement estimates of offspring of female $_{[two]}$ × male $_{[one]}$ cultured in freshwater for 120 days. The mean percentage genetic improvement ranged from 9.37 ± 4.81 % for WR to 41.50 ± 16.90 % for WB. The genetic improvement of WB was significantly higher than WR, but not significantly different from the rest of the hybrids under study. Among the purebreeds, WW had the highest mean GI of 23.60 ± 11.40 %, whereas FF attained the lowest GI of 16.30 ± 5.11 %.

Table 23: ***Percentage genetic improvement of offspring of female [two] × male [one] cultured in freshwater for 120 days***

	Replicates of GI (%)			
Hybrids	**1**	**2**	**3**	**Mean ± SE (%)**
BB	19.63	2.53	39.47	20.50 ± 10.70^{ab}
BF	25.91	27.57	28.94	27.47 ± 0.88^{ab}
BR	6.36	48.95	18.49	24.60 ± 12.70^{ab}
BW	*	*	*	*
FB	1.72	15.09	34.28	17.03 ± 9.45^{ab}
FF	34.98	26.41	3.45	21.62 ± 9.41^{ab}
FR	14.45	22.64	35.55	24.21 ± 6.14^{ab}
FW	26.72	*	21.29	24.00 ± 2.71^{ab}
RB	8.50	43.21	6.47	19.40 ± 11.90^{ab}
RF	16.51	54.41	17.85	29.60 ± 12.40^{ab}
RR	24.78	7.11	17.01	16.30 ± 5.11^{ab}
RW	20.75	42.98	23.10	28.94 ± 7.05^{ab}
WB	11.03	69.44	43.91	41.50 ± 16.90^{a}
WF	*	*	*	*
WR	11.85	0.07	16.19	9.37 ± 4.81^{b}
WW	25.40	42.47	2.96	23.60 ± 11.40^{ab}

Means that do not share a letter in the superscripts are significantly different.
* Data not available due to mortalities caused by rainstorm activities

Female [one] × Male [one] offspring cultured in brackish water for 120 days

The mean percentage genetic improvement of offspring of female [one] × male [one] cultured in brackish water ranged from 22.5 ± 10.6 % to 96.81 ± 7.87 % for RR and WB respectively (Table 24). The genetic improvement of WB was comparable to BB, BR, BW and WR, but significantly higher ($P < 0.05$) than all the other hybrids under investigations. Among the purebreeds, BB exhibited the highest GI (79.8 ± 13 %), while RR attained the lowest (22.5 ± 10.6 %).

Table 24: ***Percentage genetic improvement of offspring of female*** *[one]* ***× male*** *[one]* ***cultured in brackish water for 120 days***

	Replicates of GI (%)			
Hybrids	**1**	**2**	**3**	**Mean ± SE (%)**
BB	54.77	98.28	86.33	79.8 ± 13^{ab}
BF	24.25	54.01	38.93	39.06 ± 8.59^{bcd}
BR	92.61	34.77	57.73	61.7 ± 16.8^{abcd}
BW	25.40	81.14	109.78	72.1 ± 24.8^{abc}
FB	36.35	15.20	86.84	46.1 ± 21.3^{bcd}
FF	*	*	*	*
FR	*	*	*	*
FW	*	*	*	*
RB	12.85	23.18	66.19	34.1 ± 16.3^{cd}
RF	56.31	73.24	22.86	50.8 ± 14.8^{bcd}
RR	16.06	43.26	8.24	22.5 ± 10.6^{d}
RW	11.93	41.46	53.46	35.6 ± 12.3^{bcd}
WB	107.96	81.62	100.85	96.81 ± 7.87^{a}
WF	48.22	19.11	32.58	33.31 ± 8.41^{cd}
WR	60.20	93.82	16.71	56.9 ± 22.3^{abcd}
WW	26.53	21.83	46.37	31.58 ± 7.52^{cd}

Means that do not share a letter in the superscripts are significantly different ($P < 0.05$). * Data not available due to mortalities caused by rainstorm activities

Female *[two]* ***× Male*** *[one]* ***offspring cultured in brackish water for 120 days***

Table 25 indicates the percentage genetic improvement of offspring of female [two] × male [one] cultured in brackish water. The purebreed, BB exhibited the lowest mean genetic improvement of 16.6 ± 15.7 %, whereas the highest GI of 101 ± 66.1 % was attained by FW. The mean genetic improvement of FW was significantly higher ($P < 0.05$) than BB, BF, RR and WW but was not significantly different from the rest of the experimental fish.

Table 25: ***Percentage genetic improvement of offspring of female*** $_{[two]}$ ***× male*** $_{[one]}$ ***cultured in brackish water for 120 days***

	Replicates of GI (%)			
Hybrids	**1**	**2**	**3**	**Mean ± SE (%)**
BB	0.90	*	32.29	16.6 ± 15.7^{c}
BF	44.12	12.78	22.47	26.45 ± 9.27^{bc}
BR	85.45	61.20	19.38	55.3 ± 19.3^{abc}
BW	3.05	36.62	85.49	41.7 ± 23.9^{abc}
FB	36.35	33.86	107.22	59.1 ± 24^{abc}
FF	70.77	33.24	87.37	63.8 ± 16^{abc}
FR	*	59.97	68.23	64.1 ± 4.13^{abc}
FW	*	34.93	167.12	101 ± 66.1^{a}
RB	37.09	40.39	68.72	48.7 ± 10^{abc}
RF	61.97	50.84	132.74	81.8 ± 25.6^{ab}
RR	26.26	13.42	30.57	23.42 ± 5.15^{bc}
RW	39.67	34.01	126.99	66.9 ± 30.1^{abc}
WB	22.09	56.70	92.90	57.2 ± 20.4^{abc}
WF	52.47	*	67.15	59.81 ± 7.34^{abc}
WR	84.27	41.43	19.39	48.4 ± 19^{abc}
WW	7.84	38.01	18.73	21.53 ± 8.82^{bc}

Means that do not share a letter in the superscripts are significantly different ($P < 0.05$). * Data not available due to mortalities caused by rainstorm activities

Pooled genetic improvement across select lines of female $_{[one]}$ **and female** $_{[two]}$ **in different environments**

Table 26 shows the pooled genetic improvement (GI) across select lines of the hybrids in fresh and brackish water. The genetic improvement of the hybrids in freshwater ranged from 5.95 ± 2.35 % for RR to 42.14 ± 6.27 % for BF. The genetic improvement of BF was significantly higher than all the hybrids, except FR and WW.

In brackish water, the genetic improvement ranged from 20.62 ± 1.48 % to 82.9 ± 26.9 %. The Lowest GI was exhibited by the purebreed RR, whereas

the highest was attained by the hybrid WB. The genetic improvement of WB was significantly higher ($P < 0.05$) than BF, RB, RR, RW, WR and WW.

Table 26: ***Pooled genetic improvement of S. melanotheron hybrids across select lines of female [one] and female [two] in fresh and brackish water for 120 days***

GI in Freshwater		GI in Brackish water	
Hybrid	**Mean ± SE (%)**	**Hybrid**	**Mean ± SE (%)**
BB	9.2 ± 3.9^{bc}	BB	51.2 ± 25.1^{abc}
BF	42.14 ± 6.27^{a}	BF	25.66 ± 5.73^{bc}
BR	16.93 ± 3.31^{bc}	BR	60.35 ± 4.85^{ab}
FB	11.45 ± 3.85^{bc}	BW	58.85 ± 1.85^{ab}
FF	8.48 ± 5.53^{bc}	FB	52.65 ± 7.85^{abc}
FR	23.84 ± 4.92^{abc}	RB	22.6 ± 11.8^{bc}
RB	10.28 ± 3.48^{bc}	RF	51.28 ± 5.52^{abc}
RF	18.3 ± 12.8^{bc}	RR	20.62 ± 1.48^{c}
RR	5.95 ± 2.35^{c}	RW	34.44 ± 2.96^{bc}
RW	18.3 ± 8.98^{bc}	WB	82.9 ± 26.9^{a}
WB	11.91 ± 8.98^{bc}	WR	44.0 ± 12.1^{bc}
WR	6.02 ± 4.55^{c}	WW	24.45 ± 3.95^{bc}
WW	27.28 ± 3.52^{ab}		

Heritability estimates of hybrids of *S. melanotheron*

Body weight heritability of offspring of female [one] × male [one] in freshwater

Table 27 shows the body weight heritability (h^2) estimates of offspring of female [one] × male [one] select and control lines cultured in freshwater for 120 days. The mean heritability of the select line ranged from 0.16 ± 0.14 (BB) to 0.84 ± 0.06 (FR). The heritability of FR was comparable to BF, BR, WB and WW, but was significantly higher ($P < 0.05$) than the rest of the hybrids. Among the sexes, females from FR had the highest heritability of 0.89, whereas the lowest of 0.02 was attained by FB. In the case of males, the highest h^2 of 0.78 was recorded for FR, whilst BB attained the lowest of 0.02.

In the control line, the mean heritability of female [one] × male [one] ranged between 0.12 ± 0.09 and 0.57 ± 0.06 for BR and FB respectively (Table 27). When the heritability estimates of the control line were subjected to ANOVA test, there was no significant difference as shown in Table 28. For the sexes, BW attained the highest heritability of 0.71 among the females, while BB attained the lowest h^2 of 0.10. The hybrid FB had the highest heritability (0.84) among the males, whilst BR and WF attained the lowest (0.03).

Table 27: ***Body weight heritability estimates of offspring of female [one] × male [one] select and control lines cultured in freshwater for 120 days***

Select line h^2				Control line h^2			
Hybrids	♀	♂	**Mean ± SE**	**Hybrids**	♀	♂	**Mean ± SE**
BB	0.30	0.02	0.16 ± 0.14^{c}	BB	0.10	0.21	0.16 ± 0.06
BF	0.52	0.34	0.43 ± 0.09^{abc}	BF	0.26	0.45	0.36 ± 0.10
BR	0.82	0.35	0.58 ± 0.23^{ab}	BR	0.21	0.03	0.12 ± 0.09
BW	0.26	0.14	0.20 ± 0.06^{bc}	BW	0.71	0.35	0.53 ± 0.18
FB	0.02	0.62	0.32 ± 0.30^{bc}	FB	0.30	0.84	0.57 ± 0.27
FF	0.19	0.27	0.23 ± 0.04^{bc}	FF	0.47	0.28	0.38 ± 0.09
FR	0.89	0.78	0.84 ± 0.06^{a}	FR	0.27	0.38	0.33 ± 0.05
FW	0.48	0.01	0.25 ± 0.24^{bc}	FW	*	*	*
RB	0.15	0.45	0.30 ± 0.15^{bc}	RB	0.29	0.78	0.54 ± 0.24
RF	0.27	0.37	0.32 ± 0.05^{bc}	RF	0.27	0.42	0.34 ± 0.07
RR	0.14	0.43	0.28 ± 0.15^{bc}	RR	0.60	0.23	0.42 ± 0.18
RW	0.35	0.43	0.39 ± 0.04^{bc}	RW	0.49	0.43	0.46 ± 0.03
WB	0.46	0.66	0.56 ± 0.10^{abc}	WB	0.20	0.47	0.34 ± 0.13
WF	0.41	0.37	0.39 ± 0.02^{bc}	WF	0.39	0.03	0.21 ± 0.18
WR	0.39	0.41	0.40 ± 0.0^{bc}	WR	0.48	0.08	0.28 ± 0.20
WW	0.44	0.63	0.54 ± 0.10^{abc}	WW	*	*	*

Means that do not share a letter in the superscripts are significantly different ($P < 0.05$). * Data not available due to mortalities caused by rainstorm activities

Table 28: ***ANOVA test on body weight heritability of offspring of female*** *[one]* ***male*** *[one]* ***control line cultured in freshwater for 120 days***

Source	DF	SS	MS	F	P
Factor	13	0.4953	0.0381	0.83	0.63
Error	14	0.6439	0.046		
Total	27	1.1392			

Body weight heritability of offspring of female *[two]* ***× male*** *[one]* ***in freshwater***

Table 29 shows the heritability estimates of the select and control lines of the offspring of female [two] × male [one] cultured in freshwater for 120 days. The mean heritability of the select line ranged from 0.18 ± 0.14 for WF to 0.81 ± 0.08 for FF. The mean heritability exhibited by FF was not significantly different from FB and BF, but was significantly higher than the rest of the hybrids. Amongst the females, the highest heritability of 0.73 was exhibited by FF, whereas the lowest h^2 (0.04) was attained by WF. The purebreed, FF attained the highest h^2 of 0.89 among the males, whilst RR had the lowest h^2 of 0.11.

Table 29: ***Body weight heritability of offspring of female [two] × male [one] select and control lines cultured in freshwater for 120 days***

Select line h^2				Control line h^2			
Hybrids	♀	♂	**Mean ± SE**	**Hybrids**	♀	♂	**Mean ± SE**
BB	0.17	0.45	0.31 ± 0.14^{cde}	BB	*	0.41	0.41*
BF	0.43	0.68	0.56 ± 0.13^{abc}	BF	0.42	0.02	0.22 ± 0.20
BR	0.28	0.43	0.36 ± 0.08^{bcde}	BR	0.36	0.32	0.34 ± 0.02
BW	0.29	0.13	0.21 ± 0.08^{e}	BW	*	*	*
FB	0.54	0.65	0.60 ± 0.05^{ab}	FB	0.46	0.43	0.45 ± 0.02
FF	0.73	0.89	0.81 ± 0.08^{a}	FF	0.07	0.52	0.30 ± 0.22
FR	0.50	0.28	0.39 ± 0.11^{bcde}	FR	0.50	0.28	0.39 ± 0.11
FW	0.39	0.25	0.32 ± 0.07^{cde}	FW	0.84	0.17	0.50 ± 0.34
RB	0.38	0.45	$0.42 \pm 0.04_{bcde}$	RB	0.46	0.48	0.47 ± 0.01
RF	0.34	0.30	0.32 ± 0.02^{cde}	RF	0.49	0.24	0.36 ± 0.13
RR	0.36	0.11	0.24 ± 0.13^{de}	RR	0.32	0.74	0.53 ± 0.21
RW	0.51	0.55	0.53 ± 0.02^{bc}	RW	0.28	0.10	0.19 ± 0.09
WB	0.51	0.33	0.42 ± 0.09^{bcde}	WB	0.15	0.27	0.21 ± 0.06
WF	0.04	0.32	0.18 ± 0.14^{e}	WF	*	*	*
WR	0.48	0.48	0.48 ± 0.00^{bcd}	WR	0.81	0.48	0.65 ± 0.17
WW	0.47	0.47	0.47 ± 0.00^{bcd}	WW	0.46	0.78	0.62 ± 0.16

Means that do not share a letter in the superscripts are significantly different ($P < 0.05$), * Data not available due to mortalities caused by rainstorm activities

In the control line, the hybrid WR exhibited the highest mean heritability of 0.65 ± 0.17, whereas the lowest heritability of 0.19 ± 0.09 was recorded for RW (Table 29). However, analysis of variance test on heritability of the control line indicated no significant difference ($P > 0.05$) as shown in Table 30. At the sex level, FW had the highest heritability of 0.84 among the females, whereas FF attained the lowest of 0.07. Among the males, the highest heritability of 0.78 was recorded for WW, whereas the lowest h^2 of 0.02 was attained by BF.

Table 30: ***ANOVA test on body weight heritability of offspring of female*** *[two]* ***× male*** *[one]* ***control line cultured in freshwater***

Source	DF	SS	MS	F	P
Factor	13	0.5437	0.0418	0.8	0.65
Error	13	0.676	0.052		
Total	26	1.2197			

Body weight heritability of offspring of female *[one]* ***× male*** *[one]* ***in brackish water***

The mean heritability of the select line of female [one] × male [one] offspring cultured in brackish water for 120 days ranged from 0.19 ± 0.09 (WF) to 0.58 ± 0.39 (WR) (Table 31). There was no significant difference ($P > 0.05$) in the mean heritability estimates as shown in the ANOVA table (Table 32). In terms of sexes, WR exhibited the highest h^2 of 0.98 among the females, whereas the lowest heritability of 0.28 was attained by WF. The purebreed BB from the males had the highest heritability of 0.54, whilst the lowest h^2 (0.09) among the males was attained by FB.

In the control line, BB attained the lowest mean heritability of 0.09 ± 0.08, while RR (Table 31) attained the highest mean heritability of 0.77 ± 0.08. The heritability of RR was comparable to BF, FB, RF, RW, WB, WF, WR and WW, but significantly higher than the remaining experimental hybrids. Among the sexes, the highest heritability of 0.95 amongst the females was attained by BF, while BB had the lowest of 0.17. For the males, RR exhibited the highest h^2 (0.85), whilst BB attained the lowest (0.01).

Table 31: ***Body weight heritability of offspring of female [one] × male [one] select and control lines cultured in brackish water***

	Select line, h^2				Control line, h^2		
Hybrids	♀	♂	**Mean ± SE**	**Hybrids**	♀	♂	**Mean ± SE**
BB	0.46	0.54	0.50 ± 0.04	BB	0.17	0.01	0.09 ± 0.08^{c}
BF	0.36	0.16	0.26 ± 0.10	BF	0.95	0.41	0.68 ± 0.27^{a}
BR	0.47	0.40	0.44 ± 0.03	BR	*	*	*
BW	0.34	0.49	0.42 ± 0.08	BW	*	*	*
FB	0.49	0.09	0.29 ± 0.20	FB	0.37	0.57	0.47 ± 0.10^{ab}
FF	*	*	*	FF	*	*	*
FR	*	*	*	FR	*	*	*
FW	*	*	*	FW	*	*	*
RB	0.49	0.43	0.46 ± 0.03	RB	0.34	0.03	0.19 ± 0.16^{bc}
RF	0.58	0.10	0.34 ± 0.24	RF	0.54	0.43	0.48 ± 0.05^{ab}
RR	0.60	0.35	0.47 ± 0.13	RR	0.69	0.85	0.77 ± 0.08^{a}
RW	0.38	0.31	0.35 ± 0.04	RW	0.50	0.38	0.44 ± 0.06^{abc}
WB	0.79	0.37	0.58 ± 0.21	WB	0.67	0.75	0.71 ± 0.04^{a}
WF	0.28	0.10	0.19 ± 0.09	WF	0.50	0.75	0.62 ± 0.13^{a}
WR	0.98	0.19	0.58 ± 0.39	WR	0.45	0.46	0.46 ± 0.00^{ab}
WW	0.44	0.39	0.42 ± 0.02	WW	0.37	0.46	0.41 ± 0.04^{abc}

Means that do not share a letter in the superscripts are significantly different ($P < 0.05$), * Data not available due to mortalities caused by rainstorm activities

Table 32: ***ANOVA test on body weight heritability of offspring of female [one] × male [one] select line in brackish water***

Source	DF	SS	MS	F	P
Factor	12	0.3339	0.0278	0.54	0.855
Error	13	0.6752	0.0519		
Total	25	1.0092			

Body weight heritability of offspring of female [two] × male [one] in brackish water

Table 33 shows body weight heritability estimates of offspring of female [two] × male [one] select and control lines cultured in brackish water for 120 days. The mean heritability of the select line ranged from 0.14 ± 0.06 to 0.93 ± 0.04.

The lowest heritability was exhibited by BB, whereas the highest was attained by FB. The heritability of FB was significantly higher than BB, WB and WW, but not significantly different ($P > 0.05$) from the rest of the hybrids under study. On the sexes, FB had the highest h^2 of 0.88 among the females, whereas the lowest of 0.05 was attained by WR. Among the males, FB exhibited the highest h^2 of 0.97, while WB attained the lowest of 0.02.

The mean heritability estimates of the control line were from 0.28 ± 0.17 (RR) to 0.89 ± 0.05 (BF). The heritability of BF was significantly higher than RR, but not significantly different from the rest of the hybrids under investigations. The females from BW exhibited the highest heritability of 0.97, while the lowest h^2 (0.23) among the females was attained by BF. In the males, BF had the highest heritability of 0.94, whilst RR attained the lowest h^2 of 0.11.

Table 33: ***Body weight heritability of offspring of female [two] × male [one] select and control lines cultured in brackish water***

Select line, h^2				Control line, h^2			
Hybrid	♀	♂	**Mean ± SE**	**Hybrid**	♀	♂	**Mean ± SE**
BB	0.08	0.21	0.14 ± 0.06^{b}	BB	*	*	*
BF	0.52	0.49	0.50 ± 0.02^{ab}	BF	0.85	0.94	0.89 ± 0.05^{a}
BR	0.42	0.48	0.45 ± 0.03^{ab}	BR	*	*	*
BW	*	*	*	BW	0.97	0.39	0.68 ± 029^{ab}
FB	0.88	0.97	0.93 ± 0.04^{a}	FB	0.38	0.92	0.65 ± 027^{ab}
FF	*	*	*	FF	0.35	0.93	0.64 ± 0.29^{ab}
FR	*	*	*	FR	0.80	0.50	0.65 ± 0.15^{ab}
FW	0.48	0.37	0.42 ± 0.06^{ab}	FW	*	*	*
RB	0.41	0.50	0.45 ± 0.05^{ab}	RB	0.64	0.55	0.59 ± 0.05^{ab}
RF	*	*	*	RF	0.39	0.76	0.58 ± 0.19^{ab}
RR	0.45	0.48	0.46 ± 0.02^{ab}	RR	0.45	0.11	0.28 ± 0.17^{b}
RW	0.31	0.74	0.53 ± 0.21^{ab}	RW	0.24	0.56	0.40 ± 0.16^{ab}
WB	0.44	0.02	0.23 ± 0.21^{b}	WB	*	*	*
WF	0.25	0.71	0.48 ± 0.23^{ab}	WF	*	*	*
WR	0.05	0.92	0.48 ± 0.43^{ab}	WR	0.90	0.52	0.71 ± 0.19^{ab}
WW	0.22	0.09	0.15 ± 0.07^{b}	WW	0.49	0.41	0.45 ± 0.04^{ab}

Means that do not share a letter in the superscripts are significantly different ($P < 0.05$), * Data not available due to mortalities caused by rainstorm activities

Pooled heritability across select lines in fresh and brackish water

Table 34 shows the pooled heritability estimates across the select lines of the hybrids in fresh and brackish water environments. In freshwater, the hybrid FR exhibited the highest heritability of 0.63 ± 0.21, whilst the WR had the lowest h^2 of 0.30 ± 0.09.

In brackish water, the highest pooled heritability of 0.61 ± 0.32 was recorded for FB, whereas the hybrid RF had the lowest value of 0.29 ± 0.05. The mean heritability across the hybrids in freshwater is 0.40 ± 0.03, which is the same for the mean heritability across the hybrids in brackish water.

Table 34*: Heritability estimates on final mean body weight of S. melanotheron hybrids pooled across offspring of female [one] and female [two] lines cultured in fresh and brackish water for 120 days*

Hybrid	h^2 in freshwater Mean ± SE	h^2 in brackish water Mean ± SE
BB	0.49 ± 0.32	0.32 ± 0.18
BF	0.51 ± 0.08	0.38 ± 0.12
BR	0.57 ± 0.01	0.44 ± 0.01
BW	0.37 ± 0.16	0.38 ± 0.04
FB	0.40 ± 0.08	0.61 ± 0.32
FF	0.35 ± 0.12	0.15*
FR	0.63 ± 0.21	0.44*
FW	0.33 ± 0.08	0.42*
RB	0.34 ± 0.05	0.45 ± 0.00
RF	0.34 ± 0.02	0.29 ± 0.05
RR	0.30 ± 0.02	0.47 ± 0.00
RW	0.35 ± 0.04	0.44 ± 0.09
WB	0.44 ± 0.13	0.41 ± 0.17
WF	0.31 ± 0.34	0.34 ± 0.15
WR	0.30 ± 0.09	0.53 ± 0.05
WW	0.36 ± 0.18	0.29 ± 0.13
Mean ± SE	0.40 ± 0.03	0.40 ± 0.03

* Replicates not available due to mortalities caused by storm activities

Breeding value estimates for hybrids of *S. melanotheron*

Mean breeding value and survival rates of offspring of female [one] × male [one] select and control lines cultured in freshwater for 120 day

Table 35 shows the breeding values (BV) and survival rates of offspring of female [one] × male [one] select and control lines cultured in freshwater for 120 days. In the select line, the percentage survival ranged from 53.33 % for FR to 86.67 % for RW. The mean breeding values ranged from 8.32 ± 7.38 g to 44.11 ± 3.43 g. The lowest breeding value was exhibited by BB, whilst the highest was attained by FR. The breeding value of FR was significantly higher ($P < 0.05$) than BB, BW, FB, FF, FW, RB, RF, RR and RW. Amongst the sexes, males from FR had the highest BV of 47.54 g, whilst RR males exhibited the lowest of 7.67 g. The breeding value of WB (40.89 g) was the highest among the females, whereas BB attained the lowest of 0.93 g.

The highest breeding value (36.60 ± 12.70 g) of the control line of female [one] × male [one] in freshwater was exhibited by the hybrid BW, whereas the lowest (6.59 ± 5.03 g) was recorded for BR (Table 35). The entire stocks of FW and WW in the control line were lost to rainstorm action, and were therefore, excluded from the analysis. The breeding value of BW was significantly higher than BB and BR, but not significantly different from the rest of the hybrids under investigation. Among the sexes, males from RB had the highest BV of 48.95 g, whereas BR males had the lowest value of 1.56 g. The hybrid BW exhibited the highest breeding values of 49.30 g among the females, whereas BR had the lowest breeding value. The survival rates of the control line of female [one] male [one] in freshwater ranged from 40.00 % (BR) to 83.33 % (RF).

Table 35: ***Mean breeding values and survival rates of offspring of female [one] × male [one] select and control lines cultured in freshwater for 120 days***

Select line BV (g)					Control line BV (g)				
Hybrids	♀	♂	**Mean ± SE**	**SR (%)**	**Hybrids**	♀	♂	**Mean ± SE**	**SR (%)**
BB	15.70	0.93	8.32 ± 7.38^{d}	76.67	BB	4.91	8.98	6.95 ± 2.04^{b}	66.67
BF	36.54	21.86	29.20 ± 7.34^{abcd}	67.07	BF	11.69	20.28	15.99 ± 4.30^{ab}	66.67
BR	52.03	21.78	36.90 ± 15.10^{ab}	66.67	BR	11.63	1.56	6.59 ± 5.03^{b}	40.00
BW	19.05	10.60	14.82 ± 4.23^{bcd}	60.00	BW	49.30	23.91	36.60 ± 12.70^{a}	53.33
FB	1.09	31.16	16.10 ± 15.0^{bcd}	60.00	FB	13.82	42.77	28.30 ± 14.50^{ab}	60.00
FF	9.35	13.86	11.60 ± 2.26^{d}	80.00	FF	17.89	13.65	15.77 ± 2.12^{ab}	46.67
FR	47.54	40.68	44.11 ± 3.43^{a}	53.33	FR	11.48	15.45	13.46 ± 1.99^{ab}	53.33
FW	24.28	0.39	12.30 ± 11.90^{cd}	73.33	FW	*	*	*	*
RB	9.48	31.09	20.30 ± 10.80^{bcd}	66.67	RB	17.07	48.95	33.00 ± 15.90^{ab}	56.67
RF	15.64	21.18	18.41 ± 2.77^{bcd}	73.33	RF	14.91	22.37	18.64 ± 3.73^{ab}	83.33
RR	7.67	19.02	13.34 ± 5.68^{cd}	63.33	RR	30.42	10.30	20.40 ± 10.10^{ab}	70.00
RW	13.15	23.81	18.48 ± 5.33^{bcd}	86.67	RW	23.15	16.14	19.64 ± 3.51^{ab}	66.67
WB	29.75	40.89	35.32 ± 5.57^{abc}	76.67	WB	10.87	32.37	21.60 ± 10.70^{ab}	63.33
WF	26.58	24.56	25.57 ± 1.01^{abcd}	66.67	WF	22.30	2.38	12.34 ± 9.96^{ab}	63.33
WR	21.74	22.09	21.92 ± 0.18^{abcd}	66.67	WR	26.64	4.47	15.60 ± 11.10^{ab}	70.00
WW	22.57	31.03	26.80 ± 4.23^{abcd}	73.33	WW	*	*	*	*

Means that do not share a letter in the superscripts are significantly different ($P < 0.05$), SR = survival rate, * Data not available due to mortalities caused by rainstorm activities

Mean breeding values and survival rates of offspring of female [two] × male [one] select and control lines cultured in freshwater for 120 days

The mean breeding values of the select line of offspring of female [two] × male [one] ranged between 9.25 ± 4.14 g (RR) and 35.22 ± 5.42 g (FF) (Table 36). The breeding value of FF was comparable to BF but was significantly higher ($P < 0.05$) than BB, BW, FR, FW, RF, RR and WF. Amongst the sexes, males from BF exhibited the highest breeding value of 43.21 g, while RR attained the lowest BV of 5.10 g. For the females, the highest BV of 29.8 g was attained by the purebreed FF, whereas the lowest value of 1.57 g was exhibited by WF. The percentage survival of the hybrids during the trial period range from 30.00 % to 90.00 % for BW and WB respectively.

In the control line, the lowest breeding value of 7.05 ± 2.50 g of offspring of female [two] × male [one] in freshwater was exhibited by the hybrid RW, whereas the highest BV of 30.80 ± 10.60 g was attained by the purebreed WW (Table 36). The breeding value of WW was significantly higher than BF, RW and WB but was not significantly different from the rest of the hybrids under study. For the sexes, the highest BV (41.4 g) among the males was attained by WW, whereas the lowest value (4.55 g) was attained by RW. On the other hand, females from FW had the highest breeding value of 32.32 g, while FF attained the lowest value of 3.27 g among the females.

Due to rainstorm activities during the trial period, some of the experimental fish were lost through mortalities and escapes. For instance, the entire BB, BW and WF stocks were completely lost. Hence, these hybrids were excluded from the analysis.

Table 36: ***Mean breeding values and survival rates of offspring of female [two] × male [one] select and control lines cultured in freshwater for 120 days***

Select line BV (g)					Control line BV (g)				
Hybrids	♀	♂	**Mean ± SE**	**SR (%)**	**Hybrids**	♀	♂	**Mean ± SE**	**SR (%)**
BB	8.97	24.18	16.57 ± 7.61^{bc}	66.67	BB	*	*	*	*
BF	23.83	43.21	33.52 ± 9.69^{a}	73.33	BF	17.25	1.02	9.14 ± 8.11^{b}	46.67
BR	15.70	27.72	21.71 ± 6.01^{abc}	56.67	BR	17.75	16.76	17.25 ± 0.49^{ab}	46.67
BW	23.04	11.61	17.33 ± 5.71^{bc}	30.00	BW	*	*	*	*
FB	25.58	29.85	27.71 ± 2.14^{ab}	63.33	FB	16.84	19.63	18.24 ± 1.40^{ab}	63.33
FF	29.80	40.64	35.22 ± 5.42^{a}	60.00	FF	3.27	22.11	12.69 ± 9.42^{ab}	70.00
FR	23.13	12.87	18.00 ± 5.13^{bc}	56.67	FR	18.32	11.10	14.71 ± 3.61^{ab}	60.00
FW	18.48	14.07	16.27 ± 2.21^{bc}	63.33	FW	32.32	8.92	20.60 ± 11.70^{ab}	50.00
RB	14.80	25.94	20.37 ± 5.57^{abc}	56.67	RB	21.10	21.72	21.41 ± 0.31^{ab}	70.00
RF	16.45	18.63	17.54 ± 1.09^{bc}	66.67	RF	21.26	10.46	15.86 ± 5.40^{ab}	60.00
RR	13.39	5.10	9.25 ± 4.14^{c}	70.00	RR	13.77	29.56	21.66 ± 7.89^{ab}	63.33
RW	23.05	29.64	26.35 ± 3.29^{ab}	66.67	RW	9.55	4.55	7.05 ± 2.50^{b}	86.67
WB	25.86	20.15	23.00 ± 2.86^{abc}	90.00	WB	5.84	15.07	10.45 ± 4.61^{b}	70.00
WF	1.71	17.07	9.39 ± 7.68^{c}	40.00	WF	*	*	*	*
WR	18.86	25.81	22.34 ± 3.47^{abc}	33.33	WR	31.42	22.29	26.85 ± 4.57^{ab}	73.33
WW	27.22	27.14	27.18 ± 0.04^{ab}	80.00	WW	20.19	41.40	30.80 ± 10.60^{a}	46.67

Means that do not share a letter in the superscripts are significantly different ($P < 0.05$), SR = survival rate, * Data not available due to mortalities caused by rainstorm activities

Mean breeding values and survival rates of offspring of female [one] × male [one] select and control lines cultured in brackish water for 120 days

Table 37 shows the breeding values and survival rates of offspring of female [one] × male [one] control and select lines cultured in brackish water for 120 days. The mean breeding values of the select line ranged from 6.78 ± 2.46 g to 27.27 ± 9.57 g. The lowest BV was attained by WF, whereas WB attained the highest. The breeding value of WB was comparable to all the hybrids under study except WF, which attained a significantly lower breeding value. At the level of the sexes, males from RB had the highest BV of 18.54 g, whereas the lowest value of 4.32 g was recorded for WF. Amongst the females, WB exhibited the highest breeding value of 36.84 g, whereas WF attained the lowest BV of 9.24 g. The percentage survival for the hybrids in the select line ranged from 33.33 % for FB to 76.67 % for RW (Table 37). The entire stocks of FF, FR and FW were lost to rainstorm actions and were therefore, excluded from the analysis.

In the control, BB exhibited the lowest mean breeding value of 1.79 ± 1.57 g, whereas RR attained the highest BV of 19.70 ± 0.49 g. The BV of RR was significantly higher than BB and WB, but was not significantly different from the rest of the hybrids. Among the sexes, males from FB had the highest breeding value of 22.6 g, whilst BB attained the lowest BV of 3.36 g. For the females, the highest BV of 26.05 g was exhibited by the hybrid BF, whereas the lowest of 0.22 g was attained by BB. The percentage survival of the hybrids in the control line of female [one] × male [one] ranged between 43.33 % and 90.00 %. The lowest survival was attained by BF, RB and WW, whereas the highest was attained by WR (Table 37).

Table 37: ***Mean breeding values and survival rates of offspring of female [one] × male [one] select and control lines cultured in brackish water for 120 days***

	Select line BV (g)					Control line BV (g)			
Hybrids	♀	♂	**Mean ± SE**	**SR (%)**	**Hybrids**	♀	♂	**Mean ± SE**	**SR (%)**
BB	15.04	15.02	15.03 ± 0.01^{ab}	66.67	BB	3.36	0.22	1.79 ± 1.57^{c}	63.33
BF	10.04	4.39	7.21 ± 2.82^{ab}	40.00	BF	26.05	8.31	17.18 ± 8.87^{a}	43.33
BR	13.47	10.83	12.15 ± 1.32^{ab}	70.00	BR	*	*	*	*
BW	13.36	11.85	12.61 ± 0.76^{ab}	73.33	BW	*	*	*	*
FB	27.92	4.72	16.30 ± 11.60^{ab}	33.33	FB	13.58	22.60	18.09 ± 4.51^{a}	70.00
FF	*	*	*	*	FF	*	*	*	*
FR	*	*	*	*	FR	*	*	*	*
FW	*	*	*	*	FW	*	*	*	*
RB	16.05	18.54	17.30 ± 1.25^{ab}	36.67	RB	9.97	0.91	5.44 ± 4.53^{bc}	43.33
RF	21.29	4.71	13.00 ± 8.29^{ab}	40.00	RF	14.75	11.28	13.02 ± 1.73^{ab}	56.67
RR	19.13	9.93	14.53 ± 4.60^{ab}	43.33	RR	19.22	20.19	19.70 ± 0.49^{a}	50.00
RW	10.74	8.75	9.74 ± 0.99^{ab}	76.67	RW	10.47	8.21	9.34 ± 1.13^{abc}	53.33
WB	36.84	17.70	27.27 ± 9.57^{a}	53.33	WB	15.39	16.82	16.11 ± 0.72^{ab}	53.33
WF	9.24	4.32	6.78 ± 2.46^{b}	56.67	WF	13.97	20.92	17.45 ± 3.48^{a}	53.33
WR	36.28	6.97	21.60 ± 14.70^{ab}	73.33	WR	13.33	13.63	13.48 ± 0.15^{ab}	90.00
WW	14.68	6.99	10.84 ± 3.85^{ab}	50.00	WW	8.06	9.99	9.02 ± 0.97^{abc}	43.33

Means that do not share a letter in the superscripts are significantly different ($P < 0.05$), SR = survival rate, * Data not available due to mortalities caused by rainstorm activities

Mean breeding values and survival rates of offspring of female [two] × male [one] select and control lines cultured in brackish water for 120 days

Data on breeding values and survival rates of offspring of female [two] × male [one] select and control lines cultured in brackish water for 120 days are shown in Table 38. The mean breeding values of the select line ranged from 3.11 ± 1.40 g (WW) to 22.78 ± 1.04 g (FB). The breeding value of FB was significantly higher ($P < 0.05$) than BB, RR, WB, WF and WW, but not significantly different from the rest of hybrids under study. Analysis of the sexes shows that, the highest breeding value of 26.7 g among the males was attained by WR, whereas WB had the lowest BV. The females from FB exhibited the highest BV of 23.82 g among its counterparts, whilst the lowest value (1.56 g) was recorded for WR. The hybrid WR had the lowest percentage survival of 26.67 %, whereas the hybrids BR, BF and RB had the highest survival of 60.00 % (Table 38).

In the control line, the mean breeding values were between 6.41 ± 2.80 g and 17.37 ± 0.24 g. The lowest BV was exhibited by RW, while the highest BV was attained by BF. There was no significant difference ($P > 0.05$) among the means when the control line was subjected to analysis of variance test as shown in Table 39. Amongst the males, BF had the highest breeding value of 17.6 g, while RR attained the lowest BV of 1.83 g. For the females, the highest breeding value of 22.81 g was attained by WR, whereas the lowest value of 3.62 g was recorded for RW. The percentage survival of the hybrids in the control line ranged from 23.33 % (WW) to 53.33 % (BF) (Table 38).

Table 38: ***Mean breeding values and survival rates of offspring of female [two] × male [one] select and control lines cultured in brackish water for 120 days***

Select line BV (g)					Control line BV (g)				
Hybrids	♀	♂	**Mean ± SE**	**SR (%)**	**Hybrids**	♀	♂	**Mean ± SE**	**SR (%)**
BB	2.14	4.65	3.39 ± 1.26^{b}	50.00	BB	*	*	*	*
BF	10.99	11.88	11.43 ± 0.44^{ab}	60.00	BF	17.13	17.60	17.37± 0.24	53.33
BR	9.58	12.74	11.16 ± 1.58^{ab}	60.00	BR	*	*	*	*
BW	*	*	*	*	BW	17.12	5.46	11.29 ± 5.83	26.67
FB	23.82	21.73	22.78 ± 1.04^{a}	53.33	FB	5.92	15.23	10.57 ± 4.65	33.33
FF	*	*	*	*	FF	17.09	16.23	16.66 ± 0.43	40.00
FR	*	*	*	*	FR	10.16	6.10	8.13 ± 2.03	26.67
FW	11.90	10.83	11.36 ± 0.53^{ab}	36.67	FW	*	*	*	*
RB	8.70	10.20	9.45 ± 0.75^{ab}	60.00	RB	14.79	8.33	11.56 ± 3.23	33.33
RF	*	*	*	*	RF	18.16	10.93	14.54 ± 3.61	33.33
RR	9.07	7.25	8.16 ± 0.9^{b}	33.33	RR	14.40	1.83	8.12 ± 6.28	50.00
RW	5.79	17.88	11.83 ± 6.04^{ab}	43.33	RW	3.62	9.21	6.41 ± 2.80	30.00
WB	12.34	0.49	6.41 ± 5.93^{b}	43.33	WB	*	*	*	*
WF	5.54	11.09	8.32 ± 2.78^{b}	50.00	WF	*	*	*	*
WR	1.56	26.70	14.10 ± 12.60^{ab}	26.67	WR	22.81	7.95	15.38 ± 7.43	33.33
WW	4.51	1.70	3.11 ± 1.40^{b}	33.33	WW	7.64	6.80	7.22 ± 0.42	23.33

Means that do not share a letter in the superscripts are significantly different ($P < 0.05$), SR = survival rate, * Data not available due to mortalities caused by rainstorm activities

Table 39: ***ANOVA test on mean breeding values of offspring of female*** *[two]* **×** ***male*** *[one]* ***control line cultured in brackish water***

Source	DF	SS	MS	F	P
Factor	14	403.5	28.8	0.85	0.618
Error	11	372.4	33.9		
Total	25	775.8			

Pooled breeding values across select lines in fresh and brackish water

Table 40 shows the ranked mean breeding values of the hybrids pooled across the select lines of full and half-sibs in fresh and brackish water. In freshwater, the hybrid BF exhibited the highest breeding value of 31.36 ± 2.16 g, whereas the lowest of 11.30 ± 2.04 g was attained by purebreed RR.

In brackish water, the hybrid FB attained the highest breeding value of 19.54 ± 3.22 g, whilst the lowest BV (6.98 ± 3.86 g) was exhibited by the purebreed, WW. On the other hand, the hybrid WB had the best breeding value of 23.00 ± 6.06 g when the performances were pooled across the fresh and brackish water. The worst pooled breeding value (10.83 ± 3.06 g) performance across the two environments was exhibited by the Brimsu strain (BB).

Table 40: ***Ranked mean breeding value estimates for hybrids of S. melanotheron pooled across select lines of full- and half-sibs in fresh and brackish water***

Freshwater		Brackish water		Pooled Fresh & Brackish water	
Hybrids	**Mean ± SE (g)**	**Hybrids**	**Mean ± SE (g)**	**Hybrids**	**Mean ± SE (g)**
BF	31.36 ± 2.16	FB	19.54 ± 3.22	WB	23.00 ± 6.06
BR	29.31 ± 7.59	WR	17.85 ± 3.75	FB	20.72 ± 2.80
WB	29.16 ± 6.16	WB	16.84 ± 10.40	BR	20.48 ± 5.97
WW	26.99 ± 0.19	RB	13.38 ± 3.93	BF	20.34 ± 6.48
RW	22.42 ± 3.94	BR	11.66 ± 0.50	WR	19.99 ± 1.97
WR	22.13 ± 0.21	RR	11.35 ± 3.18	WW	16.98 ± 5.99
FB	21.91 ± 5.80	RW	10.79 ± 1.04	RB	16.86 ± 2.57
RB	20.34 ± 0.04	BW	10.42 ± 2.19	RW	16.60 ± 3.75
RF	17.98 ± 0.44	RF	9.33 ± 3.67	RF	13.65 ± 2.92
WF	17.48 ± 8.09	BF	9.32 ± 2.11	BW	13.25 ± 1.93
BW	16.08 ± 1.25	BB	9.21 ± 5.82	WF	12.52 ± 4.38
BB	12.45 ± 4.13	WF	7.55 ± 0.77	RR	11.32 ± 1.55
RR	11.30 ± 2.04	WW	6.98 ± 3.86	BB	10.83 ± 3.06

Experiment 3: Assessment of fecundity in hybrids of *S. melanotheron*

Body weight and total body length at harvest of *S. melanotheron* hybrids cultured in freshwater for 12 months

Figure 24 shows the mean body weight (a) and total body length (b) of hybrids of *S. melanotheron* cultured in freshwater for 12 months. The hybrid RB had the highest mean weight of 110.33 ± 7.15 g, whereas the lowest weight (49.49 ± 2.50 g) at harvest was attained by FB (Fig. 24a). The mean weight of RB was significantly higher ($P < 0.05$) than all the other hybrids under study.

On the other hand, the total body length of hybrids of *S. melanotheron* ranged from 14.60 ± 0.24 cm for FB to 19.26 ± 0.42 cm for RB (Fig. 24b). The mean body length of RB was not significantly different from BW, FW, RW and WW, but was significantly ($P < 0.05$) higher than all the other hybrids. Data on BB, RF and WF were not available due to mortalities caused by rainstorm activities.

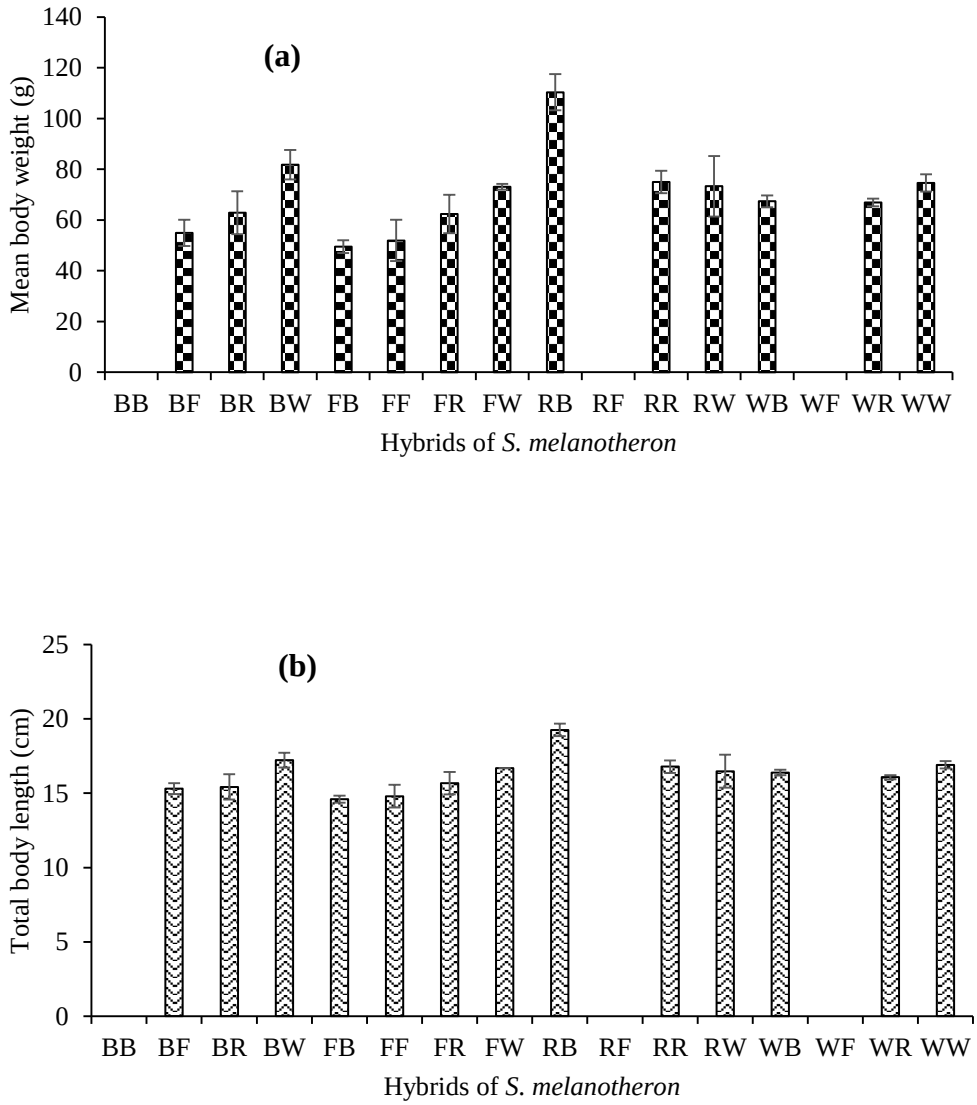


***Figure 24*: Mean body weight (a) and total body length (b) of hybrids of *S. melanotheron* hybrids cultured for 12 months**

Absolute fecundity of *S. melanotheron* hybrids cultured for 12 months

Due to mortalities and escape of some of the hybrids as a result of rainstorm activities, BB, RF and WF were not available for the fecundity evaluations. In some stocks (BF, BR, FF, FR and FW), the females with ripe ovaries were not up to ten (10). The hybrid RB exhibited highest absolute fecundity of 408.31 ± 31.5, whereas the lowest of 257.6 ± 9.99 was attained by the hybrid WR (Table 41). The absolute fecundity of RB was significantly higher than FB, FF, FR and WR, but not significantly different from the rest of the hybrids under investigations. Among the purebreeds, WW exhibited the highest absolute fecundity of 356.3 ± 15.6, whereas FF had the lowest of 314.9 ± 28.6.

Table 41: ***Absolute fecundity of S. melanotheron hybrids cultured for 12 months***

	Replicates of absolute fecundity										
Hybrids	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**Mean ± SE**
BB	*	*	*	*	*	*	*	*	*	*	*
BF	328	330	354	374	369	375	265	*	*	*	342.1 ± 14.8^{abcd}
BR	478	403	90	457	315	349	236	245	310	*	320.3 ± 40.3^{abcd}
BW	345	448	420	441	363	226	320	391	398	311	366.3 ± 21.6^{ab}
FB	315	240	263	185	515	265	285	236	235	228	276.7 ± 28.7^{cd}
FF	448	332	305	453	323	245	245	266	217	*	314.9 ± 28.6^{bcd}
FR	464	48	383	425	340	43	207	524	*	*	294.0 ± 71.2^{bcd}
FW	308	350	*	*	*	*	*	*	*	*	329.0 ± 21^{abcd}
RB	361	433	308	369	470	375	356	527	285	603	408.31 ± 31.5^{a}
RF	*	*	*	*	*	*	*	*	*	*	*
RR	364	287	334	365	375	418	390	311	303	316	346.3 ± 13.4^{abcd}
RW	593	206	308	231	505	*	*	*	*	*	368.6 ± 76.8^{abc}
WB	416	351	327	291	378	256	371	63	553	295	330.1 ± 39.6^{abcd}
WF	*	*	*	*	*	*	*	*	*	*	*
WR	241	191	302	230	255	272	271	288	254	272	257.6 ± 9.99^{d}
WW	383	405	398	266	391	387	377	302	297	357	356.3 ± 15.6^{abc}

Means that do not share a letter in the superscripts are significantly different ($P < 0.05$), * non-availability of data due to mortalities caused by rainstorm actions

Relative fecundity of hybrids of *S. melanotheron* cultured for 12 months

The computed relative fecundity ranged from 3.96 ± 0.24 g^{-1} to 7.82 ± 1.39 g^{-1}. The highest relative fecundity was exhibited by BF, while RB attained the lowest (Fig. 25). The mean relative fecundity of BF was not significantly different from FB, FF and FW but significantly higher ($P < 0.05$) than all the other hybrids. Among the purebreeds, FF attained the highest relative fecundity of 7.70 ± 0.91 g^{-1} as compared to the rest of the purebreeds under study.

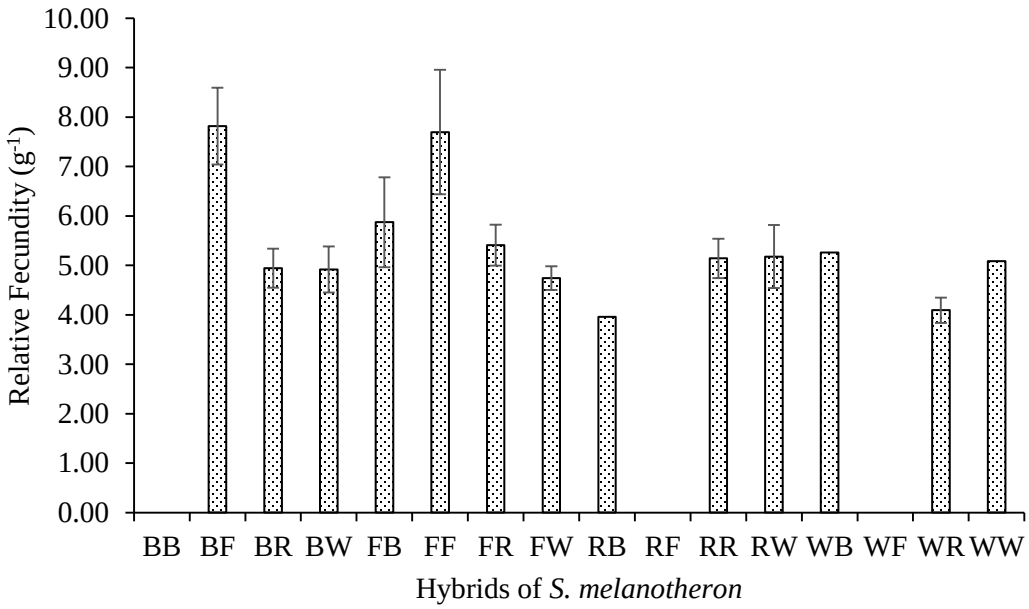


***Figure 25*: Relative fecundity of *S. melanotheron* hybrids cultured in freshwater for 12 months. Vertical bars represent standard errors**

Experiment 4: Assessment of 17α-methyltestosterone on *S. melanotheron*

Percentage sex conversion of treated *S. melanotheron* after 60-day culture

The results of percentage sex conversion determined by gonadal squash method are shown in Table 42. The fish fed 90 mg/kg 17α-MT feed exhibited the highest male percentage conversion of 80.00 ± 3.85 %, whereas the lowest male conversion of 48.67 ± 6.41 % was recorded for those fed 0 mg/kg (control). The percentage male conversion of the fish fed with 90 mg/kg feed was significantly higher than those fed 0, 30 and 120 mg/kg but not significantly different from the fish fed 60 mg/kg.

Table 42: ***Percentage sex conversion of treated S. melanotheron after 60-day culture***

Gonadal Squash Method

Conc. of 17α-MT in feeds (mg/kg)	Replicates (% male) 1	2	3	Mean ± SE (%)
0	36.00	53.33	56.67	48.67 ± 6.41^{b}
30	57.14	56.67	*	56.91 ± 0.24^{b}
60	73.33	60.00	*	66.66 ± 6.66^{ab}
90	73.33	86.67	80.00	80.00 ± 3.85^{a}
120	43.33	63.33	60.00	55.55 ± 6.19^{b}

Means that do not share a letter in the superscripts are significantly different.
*Non-availability of data due to mortalities caused by rainstorm actions

Percentage sex conversion of treated *S. melanotheron* after 180-day culture

Table 43 gives the percentage male conversion of fish treated with various concentrations of 17α-MT for 28 days and cultured further for 180 days to enable the use of the genital papillae to determine the sex. Using the hand sexing method, the highest sex conversion ratio of 84.02 ± 0.97 % was attained by the fish fed 90 mg/kg 17α-MT feed, whereas the lowest of 53.33 ± 3.33 % was recorded for those fed 0 mg/kg. The male conversion rate of fish fed 90 mg/kg was significantly higher than those fed the control feed.

Table 43: ***Percentage sex conversion of treated S. melanotheron after 180-day culture***

Hand Sexing Method

Conc. of 17α-MT in feeds (mg/kg)	Replicates (% males) 1	2	3	Mean ± SE (%)
0	50.0	60.0	50.0	53.33 ± 3.33^{b}
30	61.1	52.0	76.9	63.34 ± 7.28^{ab}
60	82.4	72.0	60.0	71.45 ± 6.46^{ab}
90	84.0	82.4	85.7	84.02 ± 0.97^{a}
120	87.5	28.0	*	57.8 ± 29.70^{ab}

Means that do not share a letter in the superscripts are significantly different
*Non-availability of data due to mortalities caused by rainstorm actions

Mean body weight of *S. melanotheron* fry after 28-day treatment with 17α-methyltestosterone

The mean body weight of the treated fry ranged from 0.41 ± 0.09 g to 0.62 ± 0.03 g as shown in Figure 26. The highest mean body weight was attained by the fry fed 30 mg/kg feed, whereas those fed 120 mg/kg exhibited the lowest mean

body weight. The mean body weight of fish fed 30 mg/kg 17α-MT feed was significantly higher than those fed 60, 90 and 120 mg/kg feed.

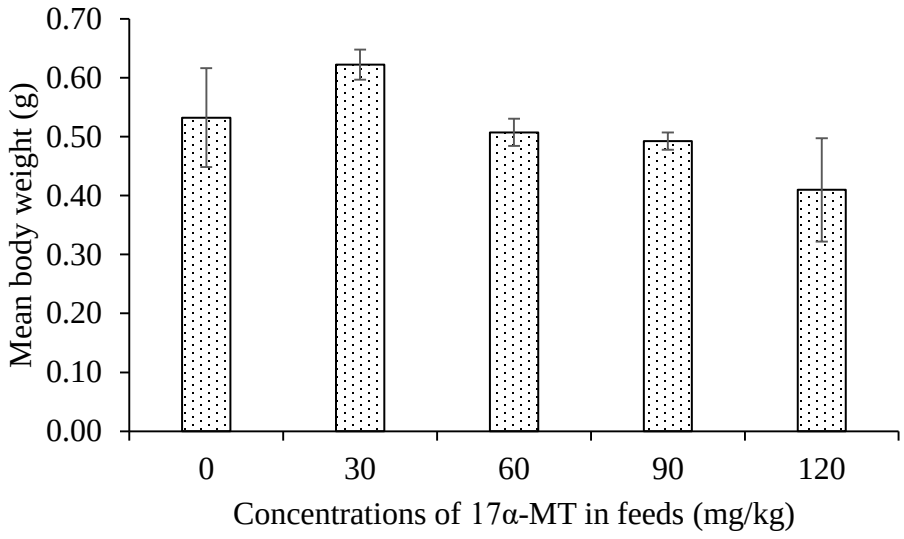


***Figure 26*: Mean body weight of *S. melanotheron* fry after 28-day treatment with different concentrations of 17α-MT in freshwater. Vertical bars represent standard errors**

Mean body weight of treated *S. melanotheron* after 60-day culture

Figure 27 shows the mean body weights of treated *S. melanotheron* cultured for 60 days. The highest mean body weight of 12.85 ± 1.64 g was attained by the fish fed 30 mg/kg 17α-MT feed; whereas those fed 120 mg/kg feed had the lowest mean body weight of 9.70 ± 0.37 g. The mean body weight of the fish fed 30 mg/kg was comparable to those fed 60 and 90 mg/kg 17α-MT feed, but was significantly higher than those fed 0 and 120 mg/kg feed.

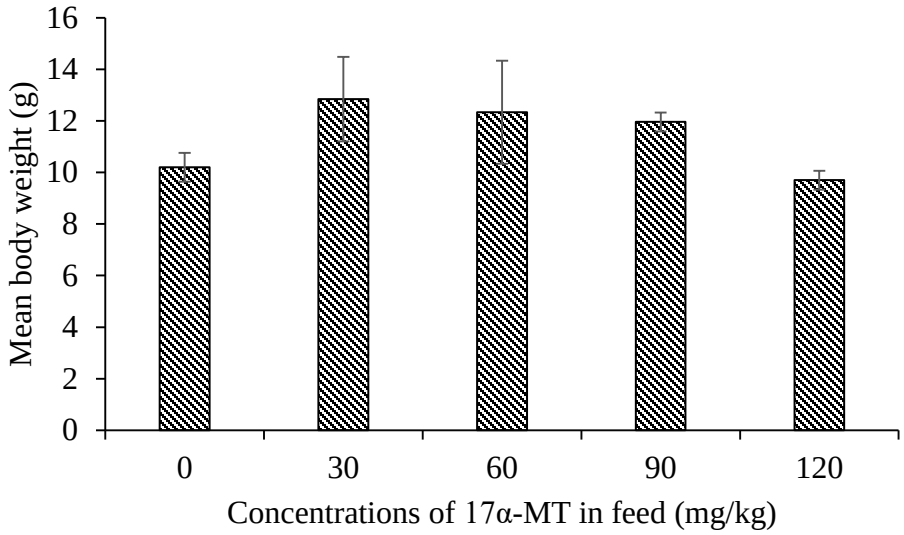


***Figure 27*: Mean body weight of treated *S. melanotheron* fry cultured in freshwater for 60 days. Vertical bars represent standard errors**

Absolute growth rates of treated *S. melanotheron* cultured for 60 days

Table 44 shows the absolute growth rate (AGR) performance of treated *S. melanotheron* cultured in freshwater for sixty days. The fish fed 30 and 60 mg/kg 17α-MT feed had the highest absolute growth rate of 0.20 ± 0.03 g day^{-1}, whereas the lowest AGR of 0.15 ± 0.01 g day^{-1} was attained by those fish fed 120 mg/kg 17α-MT feed. There was no significant difference among the mean AGR when the entire treatment was subjected to analysis of variance test.

Table 44: ***Absolute growth rate of treated S. melanotheron cultured in freshwater for 60 days***

Conc. of 17α-MT in feeds (mg/kg)	Replicates (g day^{-1}) 1	2	3	Mean ± SE (g day^{-1})
0	0.17	0.14	0.17	0.16 ± 0.01
30	0.18	0.17	0.26	0.20 ± 0.03
60	0.26	0.18	0.15	0.20 ± 0.03
90	0.20	0.18	0.19	0.19 ± 0.01
120	0.14	0.16	0.16	0.15 ± 0.01

Specific growth rates of treated *S. melanotheron* cultured for 60 days

The specific growth rate of fish fed 120 mg/kg 17α-MT feed exhibited the highest performance of 5.34 ± 0.29 % day^{-1}, while the least SGR (4.96 ± 0.31 % day^{-1}) was attained by the fish fed the control feed (Table 45). However, the mean SGRs of all the treatments indicated no significant difference when subjected to ANOVA test.

Table 45: ***Specific growth rates of treated S. melanotheron cultured in freshwater for 60 days***

Conc. of 17α-MT in feeds (mg/kg)	Replicates (% day^{-1}) 1	2	3	Mean ± SE (% day^{-1})
0	4.80	4.52	5.57	4.96 ± 0.31
30	4.82	4.90	5.35	5.02 ± 0.17
60	5.63	5.24	4.97	5.28 ± 0.19
90	5.30	5.23	5.42	5.32 ± 0.06
120	5.54	4.78	5.71	5.34 ± 0.29

Mean body weight of treated *S. melanotheron* cultured for 180 days

Figure 28 shows the final mean weight of fish treated with different concentrations of 17α-MT feed and cultured in freshwater for 180 days. The fish fed 0 mg/kg recorded the lowest mean body weight of 70.45 ± 4.67 g, whereas those fed 90 mg/kg exhibited the highest mean body weight of 103.99 ± 7.92 g. The mean body weight of fish fed 90 mg/kg was significantly higher than those fed 0 mg/kg and 120 mg/kg.

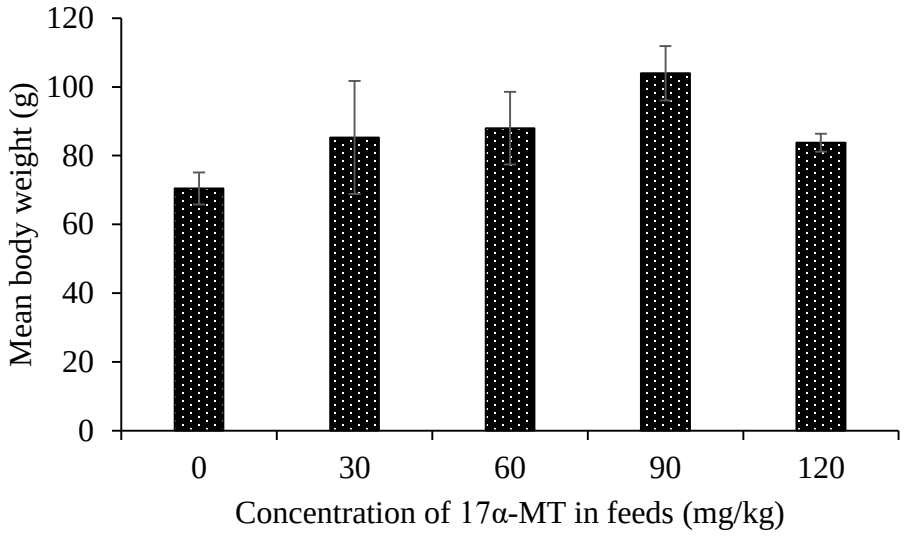


Figure 28: **Mean body weight of treated *S. melanotheron* cultured in freshwater for 180 days. Vertical bars represent standard errors**

Absolute growth rates of treated *S. melanotheron* cultured for 180 days

Table 46 shows the absolute growth rates of *S. melanotheron* treated with different concentrations of 17α-MT and cultured in freshwater for 180 days. The fish fed 90 mg/kg 17α-MT exhibited the highest absolute growth rate of 0.58 ± 0.04 g day^{-1}, while those fed control feed attained the lowest of 0.39 ± 0.03 g day^{-1}. The

mean AGR of fish fed 90 mg/kg was significantly higher ($P < 0.05$) compared to those fed 0 mg/kg.

Table 46: ***Absolute growth rate of treated S. melanotheron cultured in freshwater for 180 days***

Conc. of 17α-MT in feeds (mg/kg)	Replicates (g day^{-1}) 1	2	3	Mean ± SE (g day^{-1})
0	0.35	0.44	0.37	0.39 ± 0.03^{b}
30	0.38	0.38	0.65	0.47 ± 0.09^{ab}
60	0.60	0.44	0.41	0.49 ± 0.06^{ab}
90	0.65	0.58	0.50	0.58 ± 0.04^{a}
120	0.49	0.44	0.46	0.46 ± 0.02^{ab}

Means that do not share a letter in the superscripts are significantly different ($P < 0.05$)

Specific growth rate of treated *S. melanotheron* cultured for 180 days

The specific growth rate of the various treatments in freshwater ranged from 2.72 ± 0.09 % day^{-1} for fish fed 30 mg/kg to 2.98 ± 0.13 % day^{-1} for the fish fed 120 mg/kg 17α-MT (Table 47). There was no significant difference among the entire mean specific growth rates when subjected to ANOVA test.

Table 47: ***Specific growth rate of treated S. melanotheron cultured in freshwater for 180 days***

Conc. of 17α-MT in feeds (mg/kg)	Replicates (% day^{-1}) 1	2	3	Mean ± SE (% day^{-1})
0	2.58	2.71	2.90	2.73 ± 0.09
30	2.59	2.66	2.89	2.72 ± 0.09
60	2.94	2.83	2.81	2.86 ± 0.04
90	3.01	2.98	2.92	2.97 ± 0.03
120	3.11	2.73	3.09	2.98 ± 0.13

Feed conversion ratio of treated *S. melanotheron* cultured for 180 days

Fish fed 30 mg/kg had the highest feed conversion ratio (FCR) of 1.67 ± 0.12 after 180 days of culture in freshwater, whereas those fed 120 mg/kg exhibited the lowest FCR of 1.06 ± 0.21 (Fig. 29). The FCR of fish fed 120 mg/kg 17α-MT feed was not significantly different from those fed 90 mg/kg, but was significantly ($P < 0.05$) lower (better) than those fed 0 mg/kg, 30 mg/kg and 60 mg/kg.

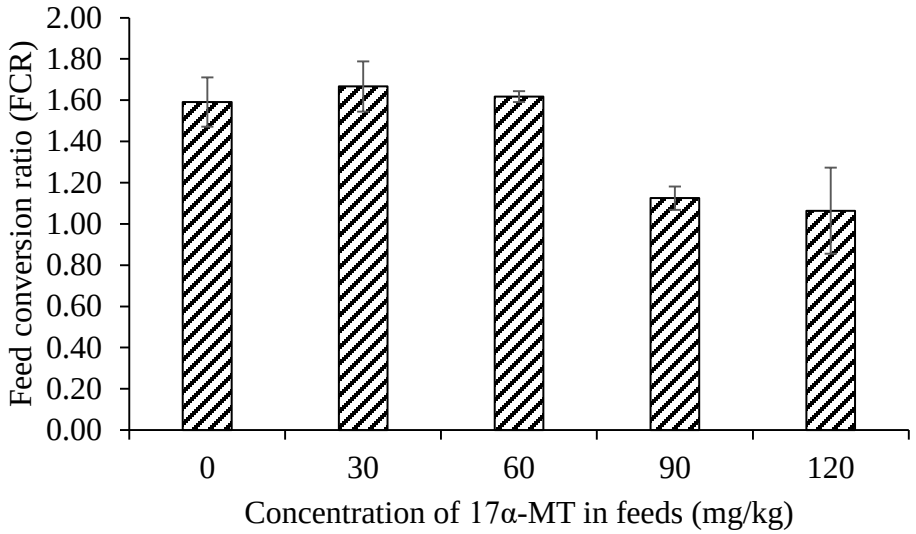


***Figure 29*: Feed conversion ratio of *S. melanotheron* treated with 17α-MT feeds and cultured in freshwater for 180 days. Vertical bars represent standard errors**

Condition factor of treated *S. melanotheron* cultured for 180 days

The condition factors (K) calculated for fish treated with the experimental feeds are recorded in Table 48. The fish treated with the control feed had the highest condition factor of 3.46 ± 0.04, while the lowest condition factor of 3.14 ± 0.05 was

attained by those fed 60 mg/kg. The K of fish fed 60 mg/kg 17α-MT feed was significantly lower ($P < 0.05$) compared to all the other experimental fish.

Table 48: ***Mean condition factor of treated S. melanotheron cultured in freshwater for 180 days***

Conc. of 17α-MT in feeds (mg/kg)	Replicates of K 1	2	3	Mean ± SE
0	3.40	3.45	3.54	3.46 ± 0.04^{a}
30	3.29	3.36	3.32	3.32 ± 0.02^{b}
60	3.04	3.20	3.19	3.14 ± 0.05^{c}
90	3.43	3.38	3.33	3.38 ± 0.03^{ab}
120	3.28	3.36	*	3.32 ± 0.04^{b}

Means that do not share a letter in the superscripts are significantly different ($P < 0.05$), * Data not available due to mortalities after rainstorm actions

CHAPTER FIVE

DISCUSSION

This chapter discusses the results from the five experiments conducted on the crossbreeding of four strains of black-chinned tilapia (*Sarotherodon melanotheron*) in fresh and brackish water. The main aim of these experiments was to develop a hybrid that has superior growth, reproduction and survival qualities compare to the existing strains at the four sources of the fish.

Fry production

Differences in sizes of the parent stocks did not prevent the mating and spawning of the fish during the crossbreeding activities. This could be because the male black-chinned tilapia is the mouth brooder, and the female is the one that initiates the mating (Trewavas, 1983). The females sometimes brood the eggs (Eyeson, 1992) where the male was unable to incubate the eggs for one reason or another. For example, when the male unduly delays in picking up the fertilized eggs due to weak pair bond between the male and the female or when the fertilized eggs were more than what the small buccal cavity of the male could contain. It was observed that the size of the buccal cavity of the brooder determines how many eggs it could incubate at a time. The smaller the buccal cavity, the lesser the number of eggs that could be incubated. The largest number of fry were produced by the purebreed of Brimsu (BB), which happened to be the biggest in terms of size. Similarly, the lowest number of fry were produced by the purebreed of Fosu (FF),

which also happened to be the smallest in terms of size. Incidentally, the crosses between these large size and small size parents produced quantities of fry that were intermediate between those produced by either of their parental stocks. The mating between Fosu female and Brimsu male (FB) produced a mean of 762 ± 439 fry, which was slightly above half of that produced by purebreed, BB (1055 ± 524). For the reciprocal cross BF, a mean of 214 ± 62.6 fry were produced, which was also about twice that produced by the purebreed, FF (142.3 ± 82.8). In the hybrid FB, the male is the B (Brimsu) which is bigger in size so could incubate as much as 762 eggs as compared to BF, where the male is F (Fosu) with its smaller size could incubate only 214 eggs. It can be deduced from this that the size of the buccal cavity of the fish is directly related to the quantity of eggs incubated.

The number of fry produced in both the purebreeds and hybrids in this study ranged from 142 – 1055, which was higher compared to those (20 – 700) reported for *S. melanotheron* by Eyeson (1979) and Trewavas (1983). The higher number of fry produced under the current study could be due to genetic superiority of the broodstocks used in the current breeding experiment.

Generally, the initial mean body weights of the three weeks old hybrid fry showed slight differences. The initial mean body weights (0.11 – 0.57 g) recorded under this study were lower compared to that reported by Kuton, Ayoola and Akapo (2012), who recorded initial mean body weights ranging from 0.56 – 0.90 g for three weeks old all-male *S. melanotheron* fry. The differences could be because the current study used mixed sexes whereas Kuton et al. (2012) used all-males. It is known that male tilapia grow faster than the female (Toguyeni et al., 1997;

Omitogun, 2005). Therefore, in a population where males are dominating, it is expected that their mean body weights would be higher than a mixed sex population.

Growth performance of *S. melanotheron* hybrids from diallel crosses

The results from the diallel crosses indicated that about 50 % (RF, FW, BW, RB, BR and WB) of the twelve hybrids under study exhibited final mean body weights higher compared to either of their parental stocks. These results were in agreement with crossbreeding reports on other tilapia species. El-Zaeem and Salam (2013) observed that the offspring of *O niloticus* × *O. aureus* had final mean body weights, which were higher compared to those of their pure lines. However, the magnitude of the final body weight of the current study was lower compared to that reported by El-Zaeem and Salam (2013). The difference may be due to the difference in species and the type of hybridization used. The current study was on *S. melanotheron* in an intraspecific hybridization, whereas El-Zaeem and Salam (2013) worked on *O. niloticus* and *O. aureus* in an interspecific hybridization. The better growth exhibited by the hybrids compared to their parental lines in the current study could be an indication that the hybrids are genetically superior in terms of weight compared to their parental lines. This implies that fish yield could be increased substantially when these hybrids are used in aquaculture.

On the other hand, the three hybrids (RW, WR and FR), which had final mean body weights lower than either of their parents may not support the method of crossbreeding as a means of increasing fish yield. This could suggest that there

is a genetic distance (Reif, Hahn & Melchinger, 2012) between the Baifikrom and Weija strains. This implies that the two populations may not have genes that complement each other, and may suggest that it is better to use selective breeding methods to improve fish yields in these strains rather than using crossbreeding procedures.

It was observed that the reciprocal crosses of FB yielded final mean weights lower compared to that of FF but higher than the BB. This could mean that the Fosu strain contains a superior gene for growth as compared to that of the Brimsu strain, although the low survival of FF during the trial could have given it an environmental (more space) advantage over BB.

Generally, the final mean body weights of 12.62 ± 1.16 g to 21.52 ± 1.40 g recorded for the hybrids after 90 days of culture in the current study were higher compared to that of Kuton, Ayoola and Akapo (2012), who reported final mean body weight of 12.12 g to 12.45 g for all-male black-chinned tilapia cultured for 90 days. The difference in the final mean body weights could be due to addictive genetic effect of the crossbreeding procedure in the current study. This could mean that the crossbreeding procedure may be a better way of enhancing fish yield as compared to the all-male tilapia production method. However, the poor performance of the all-male black-chinned tilapia in the case of Kuton et al. (2012) as compared to the current study could be due to the differences in the culture systems. Kuton et al. used 1 × 1 × 1.2 m^3 plastic containers to culture their fish whereas the current study used 3 × 2 × 1 m^3 hapas mounted in 1,500 m^2 earthen fish

pond to culture the fish samples. In other words, the current experimental fish had both genetic and environmental advantage over that of Kuton et al. (2012).

The absolute growth rates observed under the current study were lower compared to those reported by Legendre, Hem and Cisse (1989), who observed AGR of 0.32 g day^{-1} for males and 0.43 g day^{-1} for females in a mixed sex culture of black-chinned tilapia in brackish water. The differences in AGR could be due to the age differences of the experimental fish used in the two experiments. The initial mean body weight of the fish used under the current study was 0.11 g to 0.57 g whereas those used by Legendre et al. (1989) was 15 g to 20 g. Therefore, since the growth stage of the fish used by Legendre et al were in the accelerated phase of growth, it is expected to have higher or better AGR as compared to the current study where the fish were in their lag phase of growth.

The SGRs (3.64 % day^{-1} – 5.37 % day^{-1}) recorded in the current study were lower than those reported by Kuton et al. (2012): they reported SGR of 15.83 % day^{-1} to 18.78 % day^{-1}. These high differences could be due to arithmetic error in the calculation of SGR by Kuton et al. (2012). For illustration, they reported an initial body weight (W_i) of 0.56 g and a final body weight (W_f) of 11.56 g for the 90-day rearing period. If these figures were subjected to SGR formula stated in their paper as: $SGR = \frac{\mathrm{Log}_e W_f - \mathrm{Log}_e W_i}{time\ (in\ days)} \times 100$, where W_f = final body weight, W_i = initial body weight, then this should have given them SGR of 3.37 % day^{-1} and not 18.78 % day^{-1} as they reported. So if the values were computed properly, it will indicate that the SGRs recorded under the current study were higher compared to that reported by Kuton et al. (2012). This would further support the initial assertion that

crossbreeding is a better method of improving fish yield as compared to all-male tilapia culture method. In crossbreeding, additional beneficial traits are usually added onto the offspring, giving them advantage over even their own parents. In the case of all-male procedure, the males are made to conserve energy for growth instead of reproduction. This may not give them any genetic advantage over their crossbred counterparts.

The FCR recorded under the current study were better (1.54 ± 0.20 to 2.78 ± 0.42) than those reported by Kuton et al. (2012). They reported FCR ranging from 14.85 to 15.35 for all-male black-chinned tilapia cultured for 90 days. The comparatively higher FCR reported by Kuton et al. (2012) may be due to poor quality of the fish stocks used in their experiment as compared to those used under the current study. In addition, the poor quality of feed in such experiments could also result in high FCR. Similarly, the FCR of 6.1 to 9.1 reported by Legendre et al. (1989) for black-chinned tilapia cultured in brackish water were higher compared to those recorded under the current study. The difference in FCR records could be due to the genetic make-up of the fish samples used by Legendre et al. (1989) as compared to the hybrids used in the current study. The environmental differences; brackish water versus freshwater could also influence the FCR.

Generally, the survival rates of all the experimental fish were very high (90.0 ± 4.99 % to 99.96 ± 0.04 %) compared to rates reported in aquaculture practice. Therefore, the present data demonstrated the feasibility of using these hybrids in fish farming. Gilles, Amon-Kothias and Agnese (1998) reported that, the survival rate of purebreed *S. melanotheron* from Cote d'Ivoire cultured in brackish

water was 87.33 ± 2.03 %. The current data indicated that the crossbreeding of the strains enhanced the survival rates of the hybrids. The high survival rates recorded in the current study were indicative of the potential of the hybrids for aquaculture in Ghana.

Largely, the experimental fish samples exhibited high condition factors ranging from 2.14 ± 0.09 to 3.10 ± 0.03 (Table 7). The hybrids exhibited significantly higher condition factors as compared to the purebreeds. These data demonstrated that the hybrids were comparatively fatter, healthier and could support aquaculture development. According to Abban, Amevenku, Atsakpo and Dankwa (1996), food availability and feeding influence the condition of fish. Therefore, good feeding habit of fish could lead to accumulation of food reserves, which may result in increased condition factor. According to Essa and Haroun (1998), high values of condition factor indicate the suitability of the environment to the fish. Apenuvor (2014) worked on all-male *S. melanotheron* and reported condition factors ranging from 1.86 ± 0.13 to 2.15 ± 2.29. It is worth noting that the upper limit (2.15 ± 2.29) of Apenuvor (2014) is in agreement with the lowest FCR (2.14 ± 0.09) recorded for the current study, which also happened to be the record for FF, the same strain (Fosu) used by Apenuvor (2014). This observation may be a confirmation of an earlier suggestion that crossbreeding is a better method of improving fish yield.

Heterosis in hybrids of *S. melanotheron* cultured in freshwater for 90 days

Heterosis or hybrid vigour denotes a phenomenon in which the first generation (F_1) offspring of different species or populations exhibit greater trait performance (length, weight, fecundity, fertility or development) compared to the better of the two parents or the mean of the two parents (Guy, Jerry & Rowland, 2009; Nielsen et al., 2010; Granier, Audet & Bernatchez, 2011).

The magnitude of heterosis observed in the hybrids of the current study were variable depending on the trait used at stake in the calculation. The results of the current study agree with the findings of earlier researchers who observed manifestations of heterosis for different growth traits and cross types (Chinook salmon (*Oncorhynchus tshawytscha*) Bryden, Heath & Heath, 2004; Lake trout (*Salvelinus namaycush*) and Brook trout (*Salvelinus fontinalis*) Gunther, Moccia, & Bureau, 2005).

The data indicate that seven (7) of the hybrids exhibited positive heterosis ranging from 0.17 ± 1.95 % to 22.17 ± 5.52 %. This means that the hybrids produced can perform better, in terms of growth, compared to the existing pure strains of the four populations. These hybrids (RF, RB, BW, FW, BR, WB and WR) can therefore, be chosen as baseline population for selective breeding to consolidate the gains made for the aquaculture industry. The positive heterosis values for body weight, total length, weight gain, absolute growth rate and specific growth rate observed in these hybrids in the current study indicate that there were positive interactions between the parental genes found at different loci in the intra-generic hybrid genome (Sheridan, 1981). These hybrids could be of high potential

commercial value for fish farming in Ghana. Bentsen et al. (1998) conducted a complete diallel cross of eight strains of *Oreochromis niloticus* and cultured them for 90 days. They observed that seven out of twenty-two crosses exhibited significant heterosis, with a maximum value of 14 %. In another experiment, Maluwa and Gjerde (2006) performed a complete diallel cross of four strains of *Oreochromis shiranus*, cultured them for 180 days and reported a maximum heterosis of 17 % with respect to body weight.

When heterosis was estimated using the survival rate (SR), only three of the hybrids exhibited positive heterosis (BW, FB and WF). This could be an indication that the black-chinned tilapia is naturally hardy and can survive in adverse environments. In general, survival of all the purebreeds was very high, with a minimum of 90 % (Fig. 13) and this may account for why most of the hybrids exhibited negative heterosis with respect to survival rate. Essa and Haroun (1998), worked on *Oreochromis niloticus* hybrids and reported heterosis on survival from 8.71 % to 32.89 % for the species cultured for ninety days. This could mean that the purebreeds of the current study were hardier compared to the purebreeds used by Essa and Haroun (1998). This was deduced from the actual minimum survival rate of 74.4 % reported by Essa and Haroun (1998) for the purebreed as against a minimum of 90.0 % recorded for the purebreed in the current study. With these high survival rates for the hybrids of *S. melanotheron*, it is obvious they could be good candidates for fish farming.

The overall average heterosis estimated indicated positive heterosis for seven (7) hybrids (RF, RB, BW, FW, BR, WB and WR), and this was in line with

Essa and Haroun (1998) who reported positive heterosis values of growth parameters and survival rate for hybrid fingerlings of Nile tilapia (*O. niloticus*), Blue tilapia (*O. aureus*) and Red tilapia (*Oreochromis* sp.). The current results had demonstrated the feasibility of developing a synthetic base population with superior growth performance as compared to the current parent stocks. According to Pingali (1997), the application of heterosis could dramatically increase fish yield levels and reduce production cost through efficient use of inputs. The negative heterosis recorded for FB, RW, FR, BF and WF in all or most of the productive traits in this study suggests that these crosses might not support growth performance improvement through crossbreeding. The negative heterosis indicates that there was no dominant effect but additive inheritance. In the case of the hybrid WR, the overall performance was near zero percent and therefore, their pure parental lines (more especially WW) would be good candidates for selective breeding rather than crossbreeding method of enhancing fish yield.

Response to selection in different culture environments

The efficiency of using within-family selection to improve growth of *Sarotherodon melanotheron* in brackish water and freshwater was investigated in this study. The results of the growth performance assessments in both environments revealed the effectiveness of within-family selection in enhancing the growth of Black-chinned tilapia. The select lines exhibited better growth performance compared to control lines. This improvement in growth was observed in both brackish water (tanks) and freshwater (cages) as reflected in body weight gain in

both environments. The current results agree with that of Bolivar and Newkirk (2000), who studied response to selection for body weight of Nile Tilapia (*Oreochromis niloticus*) in different culture environments, and reported that there was a consistent better growth performance in the selected groups compared to the control populations in both tanks, hapas and ponds. Bolivar, Bartolome and Newkirk (1994) observed that after eight generations the selected *O. niloticus* lines that were developed, using the within-family selection procedure were 8 % to 37 % heavier than the control lines. The current results showed that it is possible to improve the growth of local strains of black-chinned tilapia by using the within-family selection method.

The growth performance of the hybrids in the current study were consistently and significantly higher in freshwater than in brackish water, except the hybrid FB which performed better in brackish water than in freshwater, and FR where the mean body weights at harvest were not significantly different in the two environments. The better performance of the hybrids in freshwater than in brackish water can be explained by the more favourable conditions (physico-chemical parameters) for growth in the freshwater as opposed to the brackish water. The current study, therefore, indicates that although the black-chinned tilapia is a euryhaline fish, their growth performance was significantly enhanced in freshwater than in brackish water. The trend of performance in the current study is in agreement with Pongthana, Nguyen and Ponzoni (2010), who reported that across pure strains and strain combinations of red tilapia (Malaysia, Thailand, Stirling and Taiwan) performance of the fish was significantly lower in saline than in freshwater

environments. It is also consistent with Liao and Chang (1983), who conducted a study on the feasibility of red tilapia culture in saline water and reported that all-male Taiwanese red tilapia exhibited faster growth in freshwater than in saltwater.

Maternal and common environmental effect on growth performance of hybrids of *S. melanotheron*

The pooled maternal and common environment effect (C^2) estimates across freshwater and brackish water were 0.18 ± 0.07 and 0.25 ± 0.06 respectively. This means that about 18 % (freshwater) and 25 % (brackish water) of the weight expressed in the hybrids were as a result of non-heritable factors such as maternal care or improved environment. The results of the current study are in agreement with other estimates reported for other tilapia species. Ponzoni, Hamzah, Tan and Kamaruzzaman (2005) evaluated genetic parameters and response to selection for live weight in the GIFT strain of *Oreochromis niloticus* in freshwater and reported maternal and common environment effect of 0.15. In other studies, Hamzah et al. (2014) reported high maternal and common environment effect of 0.41 when they assessed the performance of the genetically improved farmed tilapia (GIFT) strain over ten generations of selection in Malaysia. The high estimate of C^2 in the GIFT as compared to the current results could be attributed to the keeping of the full-sibs in their respective nursery hapas, for two months, before tagging. Maluwa, Gjerde and Ponzoni (2006), who worked on *Oreochromis shiranus* in three different test environments, reported C^2 of 0.21. On the other hand, Rutten, Komen and

Bovenhuis (2005) recorded low maternal and common environmental effect of 0.09 when they cultured *O. niloticus*. The lower C^2 as compared to the current results could be due to late harvesting of the fish at 609 g. According to Nguyen et al. (2010) maternal and common environmental effects diminish with a longer grow out period and larger weight at harvest. This moderate maternal and common environmental effect exhibited by the current hybrids indicates the existence of great potential for aquaculture in both fresh and brackish water.

Genetic improvement of *S. melanotheron* in different environments

The current study gave varying genetic improvements in the different environments. In freshwater, the pooled genetic improvement recorded for the various hybrids ranged from 5.59 ± 2.35 % to 42.14 ± 6.27 %, whereas in the brackish water it ranged from 20.62 ± 1.48 % to 82.9 ± 26.9 %. This means that the select lines in brackish water were growing far better compared to their control line counterparts, hence the wider range and higher genetic improvement recorded for the hybrids in brackish water as compared to those in freshwater. Generally, the genetic improvements in both fresh and brackish water were large enough to suggest that genetic change was being achieved and in the proposed direction. The current genetic improvement results are in line with the findings of Eknath et al. (1998), who worked on eight strains of *O. niloticus* from Egypt, Ghana, Senegal, Kenya and 4 other commercial strains from Philippines for the production of the genetically improved farmed tilapia (GIFT) strain. They observed genetic

improvement of 26 % after the first generation of selection in the synthetic GIFT strain. They further reported 12 % to 17 % genetic gain per generation across five generations of selection for growth performance. This agrees with the assertion that the genetic gain dwindles as the number of generations of selection increases, and this could partly explain the high genetic gains recorded in the hybrids of the current study. This is because they form the first generation base population where the genetic variations of the different strains have been assembled. The strains of black-chinned tilapia used in the current study were wild undomesticated strains, which probably had not suffered any genetic bottleneck or any form of inbreeding. Gall and Bakar (2002) estimated genetic gain of 13.3 % for *O. niloticus* after 98 days of culture, which agrees with the assertion that wild undomesticated population exhibits higher genetic improvement or response to selection compared to domesticated strains.

When the genetic improvement for the current study were pooled across brackish and freshwater environments, the values ranged from 10.06 ± 3.56 % to 76.56 ± 52.80 %, which may represent the genetic gain per generation of selection in *S. melanotheron* in general. These values were much higher compared to those reported for other fish species. This means there is much variability in the undomesticated strains used in the current study. Ponzoni et al (2011) reported genetic gains of 10 – 15 % per generation for more than six generations in *O. niloticus* (GIFT strain). Gjedrem (2000) reported genetic improvement of 10 – 20 % for cold-water fish species.

Heritability in hybrids of *S. melanotheron*

In freshwater, the high body weight heritability estimates (0.30 ± 0.09 to 0.63 ± 0.21) exhibited in the current study means that the hybrids have the ability to transfer between 30 to 63 percent of their characteristics to their offspring. These results are in line with published results for other tilapia species. Kronert, Horstgen-Schwark and Langholz (1989) investigated the selection prospects for changing sexual maturity to a later stage in *Oreochromis niloticus* under controlled laboratory conditions. They estimated genetic parameters, based on the performance of 91 full-sib families and reported high heritability of 0.65. Oldorf et al. (1989) reported heritability of 0.51 for Lake Manzala strain of *O. niloticus* after testing it under laboratory and commercial farming conditions in Kenya. Similarly, Bolivar and Newkirk (2002) using a single-trait animal model also reported a high heritability of 0.56 in *O. niloticus* selected for growth rate over twelve generations.

However, low to moderate heritability estimates have also been reported. Hamzah et al. (2014) evaluated the growth performance of the genetically improved farmed tilapia (GIFT) strain of *Oreochromis niloticus*, over ten generations of selection in Malaysia. They reported moderate heritability for body weight of 0.24 ± 0.031, indicating that there was still abundant genetic variation for further genetic improvement. Gall and Bakar (2002) observed heritability of 0.20 ± 0.04 in body weight of *O. niloticus* after 98 days of culture. In other aquaculture species, Vandeputte et al. (2004) estimated heritability of 0.33 for the common carp (*Cyprinus carpio* L.), whereas Ma, Saillant, Gatlin and Gold (2008) reported heritability of 0.22 ± 0.06 for body weight in Red Drum (*Sciaenops ocellatus*).

The high heritability estimates in this study suggest that selection for fast growth in black-chinned tilapia would be successful in both brackish and freshwater environments. The high heritability recorded is an indication that there is still abundant genetic variation and scope for further genetic improvement in growth rate (Ponzoni et al., 2008) of black-chinned tilapia.

When heritability estimates across the sexes of half and full-sibs in brackish water were pooled, the females exhibited nominally higher h^2 of 0.47 ± 0.03 compared with the 0.45 ± 0.05 recorded for the males. The slightly higher heritability in the females may be attributed to tank effects, maternal effects, as well as non-additive genetic variations. These observations are in line with the findings of Refstie (1980) who worked on Rainbow trout and reported low heritability of 0.06 for males and a higher value of 1.04 for the females. Eknath et al. (1998) estimated heritability of 0.23 and 0.53 for males and females respectively in the evaluation of genetic parameters in the GIFT strain of *O. niloticus*.

It is worth noting that some of the hybrids exhibited high heritability (0.50 – 0.89) in both sexes, whereas others exhibited high h^2 in only one sex during the trial period. In freshwater, the fish that exhibited high heritability in both sexes when the select, control, half and full-sib lines were pooled, included BF, FB, FF, FR, RR, RW and WB. The hybrids, BR, BW, FW and WR exhibited high h^2 in their females only, whereas WW exhibited high heritability in the males only. These suggest that it is possible to further enhance growth performance and yield by inter-crossing these hybrids with high heritability values. Further investigations could,

therefore be conducted on the following crosses to develop a composite hybrid: female FR (0.89) × male FB (0.84); female BR (0.82) × male FB (0.84); a backcross of female FW (0.84) × male FF (0.89); female WR (0.81) × male FR (0.89), where values in the brackets are estimated h^2 in freshwater.

In brackish water, almost all the fish exhibited high heritability in both sexes, except BB and FF, which recorded high h^2 only in the males, and BW, which exhibited high heritability in only the females. The following inter-crosses could also be evaluated further in brackish water: female WR (0.98) × male FB (0.97); female BF (0.95) × male WR (0.92); female BW (0.97) × male BF (0.94) and female BF (0.94) × male FF (0.93), where values in brackets represent estimated heritability in brackish water.

With these high heritability estimates, a substantial portion of the selection differential (Van Tassell & Van Vleck, 1991) would be expected to be gained in the next generation offspring of the selected parents. The high heritability for body weight across black-chinned tilapia hybrids under the current study justifies its selection as one of the criteria and the most important breeding objective (Hamzah et al., 2014) for this breeding experiment. According to Nguyen, Khaw, Ponzoni, Hamzah and Kamaruzzaman (2007), harvest body weight is the most efficient criterion to improve overall performance of fish as compared to other traits such as length, depth and width. Currently, in Ghana most fish, especially farmed fish, are priced or sold based on the body weight of the fish.

Breeding values of hybrids of *S. melanotheron* cultured in different environments

When breeding value performance of the various crossbreeds in the select lines were pooled across fresh and brackish water environments, the mean ranged from 10.83 ± 3.06 g (BB) to 23.00 ± 6.06 g (WB) (Table 38). These results were in agreement with the findings of Bentsen et al. (2017), who evaluated the genetic improvement of farmed tilapia (GIFT), *Oreochromis niloticus* and reported breeding values ranging from 6.0 g to 29.4 g.

It is worth noting that largely, the range of breeding value estimates were slightly wider for the hybrids in freshwater than those in brackish water. This may be due to better environmental conditions in freshwater as compared to brackish water. The top three best performing hybrids in the different environments demonstrated that some hybrids performed better in one environment than the other, and no one particular hybrid did equally well in both environments. The hybrid, BF was the best in freshwater but did not do equally well in brackish water. Similarly, the hybrid, FB exhibited the highest performance in brackish water but did not do likewise in freshwater. On the other hand, the pooled breeding values across brackish and freshwater lines indicated that the hybrid, WB was the best candidate for the two environments. The results of the present study pointed out that there may be a need to develop separate synthetic base population for the different environments. It is worth stating that due to poor survival and loss of identification tags, some of the hybrids (FF, FR and FW) were excluded from the final breeding value analysis. The good performance of WB coupled with its high

survival rates make it a good candidate to be considered as base population for selective breeding program in both fresh and brackish water environments.

In freshwater, the breeding values estimated for the various sexes in the select lines ranged from 1.09 g to 52.03 g for females, and 0.39 g to 40.89 g for the males. In brackish water, the breeding values ranged from 1.56 g to 36.84 g for the females and 0.49 g to 26.70 g for the males. The range of breeding values in current study was wider compared to other breeding values reported for other fish species. Kohinoor, Rahman, Islam and Hussain (2016), reported breeding values ranging from 4.17 g to 9.70 g for males and 4.24 g to 9.36 g for females when they evaluated growth performance of the genetically improved farmed tilapia (GIFT) strain after six generations of genetic selection for body weight in Bangladesh. The current study had wider range of breeding values because it involves sixteen (16) different hybrids with full-sib, half-sib, select and control lines as compared to Kohinoor et al. (2016) who used only one strain (GIFT) with different families for selective breeding evaluation. Attipoe (2006) reported a breeding value of 1.445 g for the base population when he conducted a diallel cross on *O. niloticus* strains from Nawuni, Yeji, Kpando and Nsawam to evaluate the breeding and selection for faster growth. This low breeding value, as compared to other breeding values reported for *O. niloticus*, still falls within the wide range of estimated breeding values for the current study. Taking into consideration the performance of some of the hybrids at the sex level, few crosses could be conducted further to produce a composite base population for selective breeding for increased body weight at harvest. These may include ♀BW (59.3) × ♂FB (42.77), ♀BW (59.3) × ♂RB (48.95), ♀BR (52.03) ×

♂FR (40.65), ♀BR (52.03) × ♂WB (40.89), ♀BF (36.54) × ♂FR (40.65), where values in brackets represent breeding values in grams. These crosses could be tested in both brackish and freshwater environments.

Fecundity of hybrids of *S. melanotheron*

The mean absolute fecundity estimated for the hybrids under study indicated higher total fecundity for bigger-sized fish. In general, the mean absolute fecundity of the hybrids in the current study increased with increase in maternal weight and length, and this was in agreement with earlier studies conducted on other species of tilapia. Jaspe and Caipang (2011) reported higher absolute fecundity in bigger-sized fish compared to smaller-sized fish, when they evaluated the absolute fecundity, relative fecundity and gonadosomatic index in the F_1 females of different saline-tolerant strains of tilapia, that is *Oreochromis mossambicus*, a tilapia hybrid (*O. spilorus* × *O. niloticus* (GIFT) × *O. aureus*) and their crosses. Similar results were observed in *Sarotherodon aureus* (Payne & Collinson, 1983), *Tilapia zilli* (Coward & Bromage, 1999) and *Oreochromis aureus* (Desprez, Bosc, Baroiller & Mélard, 2008).

In the current study, significant differences were observed in both absolute fecundity and relative fecundity estimates among the different hybrids. This means that the crossbreeding procedure may be a favourable method of enhancing fecundity in *S. melanotheron*. According to Jaspe and Caipang (2011), differences in the reproductive potential can be found between species and among different

strains of the same species. These differences in reproduction parameters among tilapia species and strains may be due to genetic, biological or environmental differences.

In terms of favourable reproductive traits, the hybrids BF, FB, RW and WB can be considered as base population for selective breeding to enhance reproduction. Among the purebreeds, FF (Fosu) could be chosen to form the base population for selective breeding to improve on reproductive parameters of that strain.

Effective concentration of 17α-methyltestosterone on sex-reversal of *S. melanotheron*

In both 90-day and 180-day experiments, no treatment gave 100 % male population of *S. melanotheron* in the current study. In both 90 and 180-day experiments, maximum male population (80.0 % and 84.13 % male) of *S. melanotheron* was obtained at the dose of 90 mg/kg 17α-MT feed, whereas the minimum male proportion was recorded for the dose of 120 mg/kg 17α-MT feed. These results are in line with reports on other tilapia species. Okoko (1996) worked on *O. niloticus*, and observed higher male populations at lower doses of 17α-MT than at higher doses. Okoko (1996) reported 99.3, 97 and 71.9 % males at doses of 30, 60 and 120 mg/kg respectively. The low male to female ratio recorded for 120 mg/kg was in line with the observation of Goudie, Redner, Simco and Davis (1983), who reported that excessive doses of hormone led to sterility or paradoxical

feminization in channel catfish, due to aromatization of androgens to estrogens. According to El-Greisy and El-Gamal (2012), 17α-methyltestosterone functions by suppressing the process of oogenesis, and this inhibitory effect on the development of oocytes is dependent on the dose of methyltestosterone administered to the fish. They reported that the highest sex reversal occurred at 60 mg/kg feed when *O. niloticus* was fed 40, 60 and 80 mg/kg of 17α-MT for 21 days post-hatching to produce all male tilapia population. The current study recorded highest sex reversal of 80 – 84 % male at 90 mg/kg 17α-MT fed for 28 days.

In a related study, Apenuvor (2014) studied the effect of different levels (30, 60 and 120 mg/kg) of 17α-MT on *S. melanotheron* but did not consider 90 mg/kg. The current results were not in agreement with that of Apenuvor (2014), who reported that *S. melanotheron* fed 120 mg/kg 17α-MT exhibited the highest sex reversal of 92.7 % male as compared to 89 % male for those fed 60 mg/kg 17α-MT. The deviation could be due to the differences in the strains (Fosu lagoon, brackish water versus Weija reservoir, freshwater) used. Nevertheless, the purity of the *S. melanotheron* fry that were used by Apenuvor (2014) leaves much to be desired as he tested the hormone on post-fry. Apenuvor collected the swim-up fry for conversion directly from the Fosu lagoon with the aid of a scoop net and stocked them in hapas mounted in concrete fish ponds. He started feeding the fry after five days acclimatization process. This method of acquiring fry for such experiment cannot guarantee that fry (with unknown parents) harvested were made up of only *S. melanotheron* species, even if it was established that only *S. melanotheron* existed in the lagoon. Again, feeding the fry with different doses of 17α-MT in

hapas mounted in the same fish pond cannot also ensure that the right doses were available for the targeted samples due to leaching of the 17α-MT feed from one hapa to the other. Even delaying the feeding of the swim-up fry with the hormone feed for five days during the acclimatization period could also affect the efficacy of the hormone on the sex reversal since the fry could have grown past the recommended length of 9 – 11 mm (Mair & Little, 1991), when the sex of tilapia is not yet developed. Again, the researcher did not indicate how the sex of the treated fish were determined. Therefore, the results from Apenuvor (2014) left many questions unanswered. According to Mair and Little (1991), the rate of masculinization in tilapia can be influenced by the hormone concentration, treatment duration, age and size of fry, availability of natural feed, stocking density and feeding frequency.

In an experiment with *Oreochromis aureus*, McGeachin, Robinson and Neil (1987) reported lowest sex reversal (96 % male) for administering 120 mg/kg 17α-MT as compared to 98 and 99 % male for 90 mg/kg and 60 mg/kg 17α-MT respectively. Although, the species were different from that of Okoko (1996) and the current study (i.e. *O. aureus*, *O. niloticus*, and *S. melanotheron*), the trend and performance of fish fed 120 mg/kg 17α-MT were all in agreement, recorded lower percentage male conversion, indicating a possible paradoxical feminization.

Hormone-treated tilapia usually exhibits higher growth performance compared to the control group. According to Jo, Smitherman and Tave (1995), the higher growth performance in the treated tilapia compared to the control group can

be attributed to anabolic effect of 17α-MT. However, the results of the current study showed that certain doses of 17α-MT exhibited higher anabolic effect on the fry of *S. melanotheron* compared to other doses. The 90 mg/kg 17α-MT feed, which recorded the highest male population also exhibited the highest mean final weight of 103.99 ± 7.92 g, whereas those fed 120 mg/kg 17α-MT attained the lowest weight of 83.78 ± 2.62 g. In another experiment, although 90 mg/kg attained the highest male sex reversal, it did not give the highest mean final weight, there were no significant differences among the mean weights of the treated fish. It could also mean that the anabolic effect of the hormone has not fully taken effect at that early age of the fish. The increased growth performances of the 17α-MT treated tilapia compared to the control group in current study are in agreement with published reports on other tilapia species (Dan & Little, 2000; Little, Bhujel & Pham, 2003; Mair, Abucay, Beardmore & Skibinski, 1995). It could be seen that in all these reports, the optimum dose usually exhibits highest growth performance as well as the highest percentage masculinization. These may suggest that the higher body weights recorded for the optimum dosage groups could be because of the high percentage of males present as compared to the control and other doses, which might contain more females with lower weights because of reallocation of metabolic energy towards reproduction. In males, the metabolic energy is directed towards growth, and they may benefit from anabolism-enhancing androgens (Angienda, Aketch & Waindi, 2010; Tran-Duy, Schrama, van Dam & Verreth, 2008). Ahmad, Shalaby, Khattab and Abdel-Tawwab (2002) studied different doses of 17α-methyltestosterone as a growth promoter in Nile tilapia, *Oreochromis*

niloticus, and reported that the 5 mg/kg was the optimum and effective dose in promoting significant weight gain and specific growth rate when doses of 0.5, 1.0, 2.5, 5, 10, 20 and 40 mg/kg MT were administered to the fish for 90 days. This could explain why the highest dose of 120 mg/kg 17α-MT could not attained the highest final weight, because only a small quantity of 17α-MT was required to promote growth in fish.

The percentage male conversion of 84 % was lower compared to findings in other tilapia species. Vera-Cruz and Mair (1994) obtained 98 % and 99 % males with 40 mg 17α-MT/kg and 60 mg 17α-MT/kg diets respectively for *O. niloticus* fed at 20 % body weight for 25 days. Smith and Phelps (2001) also worked on *O. niloticus* and reported 99 – 100 % male when given 17α-MT at 60 mg/kg of feed. The results of this study exhibited a lower male proportion for optimum dose rate of androgen of 90 mg 17α-MT/kg of feed. The lower conversion rate in the current study may be due to difference in species, poor handling of the hormone by the Chemical sellers and the shelf life of the steroid. According to Phelps and Popma (2000), androgens break down when exposed to sunlight or high temperatures. Varadaraj, Kumari and Pandian (1994) compared storage conditions of 17α-MT stock solutions and treated feed and the impact on efficacy of sex reversal of *O. mossambicus*. They report that when either the stock solution or feed of 17α-MT was exposed to light the efficacy of the treatment was significantly reduced.

CHAPTER SIX

SUMMARY, CONCLUSIONS AND RECOMMENDATIONS

The current study was on crossbreeding of four populations of black-chinned tilapia (*Sarotherodon melanotheron*) from Brimsu, Baifikrom and Weija reservoirs and Fosu lagoon with the main objective of developing an improved hybrid of black-chinned tilapia as base population for selection and breeding of the species for fast growth, reproduction and survival performances in both brackish and freshwater environments. The study assessed indications of magnitude of heterosis in the hybrids produced through complete diallel crosses of the four populations of *S. melanotheron*. The growth performance, feed conversion ratio, response to selection, heritability and breeding values of the hybrids. The fecundity of the various hybrids produced through the diallel crosses were also assessed. The study further determined the effective concentration of 17α-methyltestosterone for production of all-male individuals in *S. melanotheron*.

Growth performance and feed conversion ratio of hybrids of *S. melanotheron*

The current results indicated that five hybrids possessed superior production traits compared to their parent stocks. In terms of weight, the hybrids RF, FW, BW, RB and BR were significantly ($P < 0.05$) heavier compared to their parental lines. Percentage survival was generally high among all the fish ranging from 90.0 % to 99.96 %, and the well-being (condition factor) of the hybrids during the culture period was generally 'good'. The crossbreeding of strains of black-chinned tilapia

yielded high fry production, weight gain, absolute growth rates (AGR), specific growth rate (SGR), enhanced feed conversion (FCR) to fish flesh more efficiently and invariably led to low input usage and high survival rates. This study suggested that culture of the hybrids of black-chinned tilapia could lead to increased productivity and profitability. Generally, the Weija strains exhibited superior growth performance as compared to the strains from Brimsu, Baifikrom and Fosu.

Magnitude of heterosis in the hybrids of *S. melanotheron*

The results of the current study indicated that seven (7) hybrids (BR, BW, FW, RB, RF, WB and WR) out of the twelve exhibited positive heterosis ranging from 0.17 ± 1.95 % to 22.17 ± 5.52 %. The cross between Baifikrom female and Fosu male (RF) exhibited the highest positive average heterosis across all the parameters, whereas WR had the lowest positive average heterosis. On the other hand, WB and BR despite their overall positive performance, the former had 0.00 % heterosis with respect to AGR, whereas the latter exhibited negative heterosis with respect to SGR. The overall performance of the hybrid, WR was near zero percent, suggesting that their pure parental lines may be good candidates (especially WW) for selective breeding rather than crossbreeding method of enhancing fish yield.

Growth performance of hybrids of *S. melanotheron* cultured for 120 days in different environments

Generally, the growth performance of hybrids in freshwater was significantly higher than in brackish water. Nevertheless, the hybrid FB performed better in brackish water compared to freshwater, and the growth performance of FR in freshwater was not significantly different from its performance in brackish water.

Genetic improvement in fresh and brackish water environments

The magnitude of genetic improvement (Selection response**)** per generation of the hybrids was lower in freshwater compared to brackish water, when pooled across select lines of female [one] and female [two]. The highest genetic improvement (42.14 ± 6.27 %) in freshwater was exhibited by BF, whereas in brackish water the highest genetic improvement of 82.9 ± 26.9 % was attained by WB. In general, the genetic gain per generation across all environment ranged from 10.06 ± 3.56 % to 76.56 ± 52.80 %.

Heritability of *S. melanotheron* hybrids in fresh and brackish water

Generally, the magnitude of heritability exhibited by the hybrids in freshwater was not significantly different from those attained by the hybrids in brackish water. The hybrid, FR exhibited the highest heritability of 0.63 ± 0.21 in freshwater, whereas the hybrid FB had the highest heritability of 0.61 ± 0.32 in brackish water. However, the overall mean heritability estimate across all the

hybrids in freshwater was 0.40 ± 0.03, which was the same for the hybrids in brackish water.

Breeding values of hybrids of *S. melanotheron* in fresh and brackish water

The breeding values of the hybrids observed in freshwater were higher compared to those exhibited by the same hybrids in brackish water. None of the hybrids performed equally well in both freshwater and brackish water. The hybrid, BF had the highest breeding value of 31.36 ± 2.16 g in freshwater, whereas in brackish water the highest breeding value of 19.54 ± 3.22 g was attained by FB. However, when the breeding values were pooled across fresh and brackish water environments, the hybrid WB exhibited the highest breeding value of 23.0 ± 6.06 g. This may suggest the necessity to develop separate strains for the different environments.

Absolute and relative fecundity of hybrids of *S. melanotheron*

The absolute fecundity estimates indicated that the crossbreeding process produced three hybrids (RB, RW and BW) with higher fecundity compared to their parental lines. In terms of relative fecundity, the best performing hybrids included BF, FB, FR and WB. Among the purebreeds, FF obtained significantly higher relative fecundity compared to WW and RR.

Effective concentration of 17α-methyltestosterone for production of all-male in *S. melanotheron*

The results demonstrated that 17α-methyltestosterone was effective in producing phenotypic male in *S. melanotheron*. The treatment of *S. melanotheron* with 90 mg/kg of 17α-methyltestosterone for 28 days yielded the highest all-male conversion of 84.13 % male, highest survival of 70 % and highest final mean body weight of 103.99 ± 7.92 g as compared to the other doses of 0, 30, 60 and 120 mg/kg 17α-MT.

CONCLUSIONS

The current study has demonstrated that the differences in body size between female and male *S. melanotheron* strains did not inhibit mating and spawning, provided the individuals involved were sexually mature. However, the quantity of fry produced is directly related to the size of the female and the buccal cavity of the incubating parent.

This study has shown that crossbreeding tends to induce high growth performance of *S. melanotheron* hybrids. The method therefore, favours genetic improvement of *S. melanotheron,* and can be used as a potential tool through which desirable traits could be transmitted to inferior parent stocks.

From the results of the current study, it can be concluded that the aquaculture potential of *S. melanotheron* can be improved by producing intraspecific hybrids

for grow-out purposes. This is because the *S. melanotheron* hybrids exhibited improved feed conversion ratio (FCR) of 1.54 ± 020, increased survival rate of 99.96 ± 0.04 % and high positive heterosis of 22.17 ± 5.52 %.

The study had demonstrated that the hybrids of female Baifikrom × male Fosu (RF), female Baifikrom × male Brimsu (RB) and female Brimsu × male Weija (BW) possess superior productive traits compared to their parent stocks and therefore, could be selected as synthetic base populations for selective breeding.

From the analysis of the results on genetic parameters, it could be concluded that the genetic improvement per generation in *S. melanotheron* hybrids was better in brackish water (82.9 ± 26.9 %) compared to that of freshwater (42.14 ± 6.27 %). Although moderate to high heritability estimates (0.29 ± 0.05 to 0.63 ± 0.21) were observed for the individual hybrids under study, the pooled body weight heritability was the same in both freshwater (0.40 ± 0.03) and brackish water (0.40 ± 0.03) environments, suggesting that body weight heritability trait in *S. melanotheron* may not be influenced by the culture environment.

The results of the current study demonstrate that the culture environment influenced the breeding values of *S. melanotheron* hybrids. The breeding values exhibited by the hybrids were higher in freshwater (31.36 ± 2.16 g) compared to those in brackish water (19.54 ± 3.22 g). This may suggest the need for development of separate synthetic base populations for the two environments. However, when the breeding values were pooled across fresh and brackish water environments, the performance of the hybrid of female Weija × male Brimsu

(WB) was the best. This may imply that WB could be selected, as a synthetic base population, and developed through selective breeding for future use in both fresh and brackish water culture. Nevertheless, it can be concluded that the best performing hybrid in freshwater was BF, whereas in brackish water the hybrid, FB was the best candidate.

The fecundity estimates in the current study indicated that crossbreeding enhanced the absolute fecundity and reproductive parameters of the *S. melanotheron* hybrids. However, further studies should be conducted on the recommended hybrids (BF, FB and WB) to improve on their reproductive traits. It may be concluded that the purebreed from Fosu (FF) may possess superior reproductive traits compared to the other purebreeds from Baifikrom (RR) and Weija (WW), and could therefore, be chosen to form the base population for selective breeding to improve on the reproductive traits of the strain.

The current study had demonstrated that the synthetic androgen, 17α-methyltestosterone was efficient in producing phenotypic males in *S. melanotheron*. The optimum dosage of 17α-methyltestosterone for effective sex reversal in *S. melanotheron* was 90 mg/kg with a feeding duration of 28 days. At a higher dose of 120 mg/kg of 17α-MT, the sex ratio of *S. melanotheron* was not influenced in favour of males.

Based on the results of the current study, it can be concluded that the purebreed from Weija (WW) may possess superior growth performance traits as compared to purebreeds from Brimsu (BB), Baifikrom (RR) and Fosu (FF).

RECOMMENDATIONS

It is recommended from this work that:

1) Information from this study serves as a guide for future studies on relative growth performance of black-chinned tilapia strains and their crosses for establishment of "synthetic" base population for selective breeding programs.
2) The hybrids, RF, RB and BW should be chosen as "synthetic" base populations for selective breeding to consolidate the gains made through the crossbreeding.
3) The hybrid, WB should be chosen as base population for selective breeding to enhance their yield in both fresh and brackish water culture systems.
4) Separate lines should be developed for freshwater and brackish water culture. Thus, the hybrid, BF should be chosen for freshwater culture, whereas FB should be chosen for exclusive brackish water culture.
5) Further research should be conducted on the recommended hybrids (BF, FB, WB, RF, RB and BW) to evaluate their growth performance at higher salinities, e.g. 10, 15 and 20 ppt.
6) These recommended hybrids should be tested in all-male culture systems to assess their yield potentials in commercial culture in both fresh and brackish water cage and pond culture systems.
7) The optimum dose of 90 mg/kg of 17α-MT for sex reversal of *S. melanotheron* should be tested on the recommended best performing hybrids to evaluate its efficacy on these hybrids.

8) For effective sex reversal in *S. melanotheron*, the 17α-MT dosage should not exceed 90 mg/kg because excess dosage would not favour increase in male population.

9) Each of the hybrids suggested for further research should be adopted and named after institutions and researchers so that more attention would be focused on their development for the aquaculture industry in Ghana. These include:

    a. Female Baifikrom × male Fosu (RF) = UCC strain

    b. Female Baifikrom × male Brimsu (RB) = Blay strain

    c. Female Brimsu × male Weija (BW) = Yankson strain

    d. Female Brimsu × male Fosu (BF) = USAID strain

    e. Female Fosu × male Brimsu (FB) = Fisheries Commission strain

    f. Female Weija × male Brimsu (WB) = Ahiah strain

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APPENDIX A

Estimation of heritability

To calculate heritability, first estimate the various components of variance (genetic, phenotypic and environmental) by subjecting the data on harvest body weight to analysis of variance using Microsoft Excel 2013 at a P-value of 0.05. Then use the following formulae to calculate the variances using the mean squared (MS) values in ANOVA table:

$$V_g = \frac{MS_1 - MS_2}{r} \quad \text{_____ (1)}$$

$$V_e = \frac{MS_2}{r} \quad \text{_____ (2)}$$

$$V_p = V_g + V_e \quad \text{_____ (3)}$$

$$h^2 = \frac{V_g}{V_p} \quad \text{_____ (4)} \qquad \text{(Zobel \& Talbert, 1991)}$$

Where V_g = genetic variance, MS_1 = mean square between body weights, MS_2 = mean square within body weights, r = number of replicates, V_e = environmental variance, V_p = phenotypic variance, h^2 = narrow sense heritability. For example, use the values in Table 49 to calculate narrow sense heritability:

$$V_g = \frac{291.7960 - 47.1300}{3} = 81.55532$$

$$V_e = \frac{47.1300}{3} = 15.71003$$

$$V_p = 81.55532 + 15.71003 = 97.26535$$

Heritability, $h^2 = \frac{V_g}{V_p} = \frac{81.55532}{97.26535} = 0.838483$. i.e. $h^2 = 0.84$

Table 49: ***ANOVA on final body weight of FR (Female [one] Male [one] select line) used as an example of how heritability was calculated***

Source of Variation	DF	SS	MS	F	P-value
Between samples	2	583.59208	291.7960	6.19129	0.03477
Within samples	6	282.78046	47.1300		
Total	8	866.37255			

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