

**Relative abundance and molecular characterization of  
stem borers and evaluation of maize genotypes (*Zea mays*  
*L.*) for resistance in Nigeria**

**BY**

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## **DEDICATION**

**This work is dedicated to the glory of Almighty God the Father, the Lord Jesus Christ and Holy Spirit divine.**

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## ABSTRACT

Stem borers constitute a major constraints to cereals production generally and specifically to maize production in Nigeria due to yield and economic losses. Nevertheless there is still dearth of information relative abundance of stem borers, intraspecific differences between stem borers, and genotypes that are resistant to stem borers. The specific objectives of this study therefore were to: (i) determine the relative abundance of stem borers in different locations; (ii) characterize for the purpose of proper identification of the various stem borers collected in Nigeria and (iii) (ii) to evaluate different maize genotypes for their resistance to stem borers.

Sixty maize germplasms including land races and improved varieties were screened at Ibadan, Mokwa, Kontagora, Kabba and Abuja to identify stem borer resistant genotypes. The experimental design used was Randomised Complete Block Design. Agronomic and entomological data were collected and analysed with Statistical analysis system (SAS). Survey of farmers' fields was carried out in Oyo, Ekiti, Osun, Ondo, Kwara and Ogun to determine relative abundance of stem borers in different locations. Collected stem borers were reared to adult stage. DNA was extracted and amplified using primers (CP1, TRs, Tser, 16SAA, 16SBB, LP01 and LP02). This was followed by Sanger dideoxy sequencing. Bioinformatic tools were employed to analyse DNA nucleotide sequences from the resulting electrophoreogram.

The findings of the study were:

- (i) The survey results reveal that stem borer infestation was lowest in Oyo (9.4%) and highest in Ogun (50%). The other states were Ekiti (36%), Osun (38%), Ondo (42%), Kwara (44%), Ogun (50%)
- (ii) The stem borers identified were *Chilo orichalcociliellus* (Strand), *Eldana sacharina* (Walker), *Sesamia calamistis* (Hampson), and Longhorn beetles (subfamily Lamiinae; family Cerambycidae).
- (iii) Based on stem tunneling, deadhearts, stem borer leaf feeding, husk cover, lodging, field weight and yield, none of the evaluated maize varieties was absolutely resistant. Those found tolerant were TZM 1327, TZM 112, Aflatoxin Syn W5, ACR 06 TZL Comp 4C4, PVA Syn 11F2, PVA Syn 9F2, PVA Syn 19F2 and PVA Syn 3F2.
- (iv) Molecular characterization of the identified stem borers were similar to those found in Kenya with average sequence divergence among conspecific individuals averaging 3.3% in the mitochondrial cytochrome c oxidase II (COX II), while two were also similar to those found in Zimbabwe with intraspecific divergence at COX II averaging 3.0%.
- (v) The findings that the evaluated germplasm showed varying degree of tolerance is an indication that many of them can be used in further breeding program aimed at developing complete/ total stem borer resistant varieties. Artificial screening using only one stem borer type in screen house may not produce truly stem borer

resistant maize varieties. The resistant lines produced under this condition may break down in the presence of stemborer complex in the field. The use of resistant varieties is the most promising control measure in reducing yield losses caused by stem borers for resource constrained farmers and may be enhanced by cultural practices.

## CHAPTER ONE

### INTRODUCTION

#### 1.1 Background to the Study

Maize, *Zea mays L.*, is one of the oldest food grains. It belongs to the grass family Poaceae (Gramineae), tribe Maydeae. It is the only cultivated species in this genus. The other species of *Zea*, commonly called teosinte, and the species of *tripsacum*, commonly called gama grass, are important wild relatives of *Zea mays*. These are classified as the “New World” Maydeae because their centre of origin is in the Americas (Paliwal, 2000). Maize is one of the most productive species of food plants. It is a C<sub>4</sub> plant with a high rate of photosynthetic activity. Maize has the highest potential for carbohydrate production per unit area. It was the first major cereal to undergo rapid and widespread technological transformation in its cultivation, as evidenced by the well documented history of hybrid maize in the United States and later in Europe. The success of science-based technology in maize cultivation stimulated a more general agricultural revolution in many parts of the world.

Maize is of great economic significance worldwide as human food, as animal feed and as a source of raw material for a large number of industrial products (Fakorede *et al.*, 2003). The diversity of environments under which maize is grown is unmatched by any other crop. Having originated in the tropics there as an important energy yielding plant, it is now grown within 58°N in Canada and in the Russian Federation and up to 40°S in Chile and Argentina. Most of the maize crop is grown at moderate altitudes, but it is also grown below Sea level in the Capsian plain and up to 3,800m in the Andean mountains. The crop continues to expand to new areas and environments. This is the reason breeders are selecting for various ecological adaptation of mid-altitude, lowland and resistance to different production stresses including stem borer pests.

Maize is classified into distinct types depending on the latitude and the environment in which it is grown. Maize growing in warmer environments between the equator and 30°N and 30°S is often referred to as tropical maize, while those growing in cooler climates beyond 34°N

and 34°S are classified as temperate maize. Subtropical maize types are grown between the 30° and 34° latitudes.

Maize is an increasingly important crop in West and Central Africa, where over 5 million hectares of the crop are grown (Fajemisin, 1992; Fakorede *et al.*, 2003). The crop is grown in all major ecologies of these regions, from the humid tropical forest to the Sudan Savannah and from sea level to over 2000m altitude. Maize has diversified uses, including food, animal feed and industrial use. However, 74% of the output is used for human consumption (Fajemisin, 1992; Olosunde, 2015; Aroga and Ajala, 2005).

Maize production in West and Central Africa is influenced by diverse agro-climate and socioeconomic factors. A larger proportion of maize in these regions is grown in the forest zone. In this agro-ecology, maize production is intense, but yields are limited by numerous diseases, insects as well as weed pests. Similarly, poor rainfall distribution, low solar radiation and high temperature often reduce the yield per ha. (Olosunde, 2015; Badu – Apraku, 2007). Maize is traditionally intercropped with other crops by small scale farmers. Average low yield of about 1 tonne ha<sup>-1</sup> had been reported. Owing to difficulties in grain drying and storage, a great proportion of the crop in the forest is consumed green.

The rainy season in the West Africa forest zone last 6-9 months (March to November); in some regions the rains are interrupted by a short, unpredictable “August break”. The August break divides the rainy season into “first” and “second” seasons popularly called bimodal rainfall pattern. Maize planted at the beginning of the rains is called first – or major season maize and maize planted after the August break is referred to as second – or minor season maize. Maize stem borers are far more prevalent and abundant in the second than in the first season, and the second crop is sometimes a complete loss (Adeyemi *et al.*, 1996; Girling, 1980; Moyal, 2014). Consequently, many farmers in the forest zone of West Africa do not plant second –season maize (Tams and Bowden, 1953; Moyal, 2014), or at best intercrop with other crops to minimize losses.

In the Southern and Northern Guinea Savanna of West Africa, maize production has experienced dramatic increases in the last 15 years, to a great extent due to adoption of improved varieties of maize and the use of fertilizer, especially in Nigeria (Smith *et al.*, 1993). In these areas, maize is often intercropped with other cereals, as well as with legumes, but monocropped maize is becoming more prevalent (Fajemisin, 1992). Yields in savanna

ecological are up to twice that of the forest zone, due to lower pressure from diseases and insects pests, greater solar radiation, lower night temperatures and better rainfall distribution.

In the coastal savanna of Benin and Togo, maize has fewer production constraints than in the forest zone of neighbouring countries, and the crop is normally grown both in first and second seasons. Benin and Togo have the greatest per capita consumption of maize in West Africa, with 96 and 76kg/year, respectively (CIMMYT, 1992).

Globally, maize is grown on 140 million hectares with an annual production of about 600 million tones. Tropical maize is grown in 66 countries and is of major economic significance in 61 of these countries, each having 50,000 hectares or more. The average yield of maize in the tropics is 1.8 tonnes/ha, as against the global average of 4.2 tonnes/ha. The average yield of temperate maize is 7 tonnes/ha (International Maize and Wheat Improvement Centre (CIMMYT), 1994; Oyekunle *et al.*, 2016). Temperate maize, however, is a longer duration crop than most tropical maize. Therefore, the yield of tropical maize when compared, to temperate maize is not so low. Yet productivity of maize in the tropics is much lower than in temperate areas. There are some exceptions where productivity of tropical maize compares well with maize in temperate environments (Fakorede *et al.*, 2003). The maize situation in the tropics is changing rapidly. Superior germplasm with a good harvest index and high productivity is becoming increasingly available for most of the tropical maize environments. The potential of heterosis is beginning to be exploited in maize production on a larger scale in developing countries (Fakorede *et al.*, 2003)

Maize has multifarious uses. It is the only cereal that can be used as food at various stages of plant development. Young maize ear shoots (“baby corn”) are harvested as soon as the plant flowers and used as vegetables. The tender green ears of sweet maize are a delightful delicacy and are consumed in various ways. Green ears of normal field maize are used on a large scale for roasting and boiling and are consumed as food at the dough stage in several countries. The maize plant, which is still green when ears are harvested as baby ears or green ears makes good forage. This is particularly important as the pressure on limited land increases and intensive cropping patterns is practiced in order to produce enough food for increasing population. In Vietnam, cropping intensity is 270 percent and the maize crop, which is often transplanted in the North, with short field occupancy, plays an important role in maintaining this high level of cropping intensity (Paliwal, 2000).



It is projected that in the developing world, the demand for maize as food and feed will grow in the coming decades at a faster rate than for either rice or wheat. Byerlee and Saad (1993) have projected that the rate of increase in demand for maize during the period 1990-2005 will grow at the rate of 14.1% per year in developing countries (as against 2.6% per year globally).

All these suggest that maize is a crop that must be supported in order to feed the increasing world population. Greater increases in food and feed production have to come from coarse cereals including maize, which have a comparative advantage in unfavourable environment. Maize has not yet reached its limit of diffusion in production environments, and the time has come to harvest its high productivity potentials in the tropics.

## **1.2 JUSTIFICATION**

One of the major constraints to maize production in sub-saharan Africa are stem borers. African maize stem borer, *Busseola fusca* (Fuller) is especially serious at altitudes of 500m and above, while African pink stem borer (*Sesamia calamistis* Hamps) is a pest of great economic importance in West African forest zones (Moyal, 2014). The very heavy incidence of *Sesamia calamistis* in the second crop of maize in the West African forest zones can cause yield loss of between 70 and 100%, hence, the need for assessing available genotypes for resistance to stem boring insects. The borer cause dead heart which makes the maize unproductive. The borer can also girdle the stem near a node and tunnel the stem. This results in extensive lodging of the maize plant later in the season. Thus, in some areas farmers do not plant maize during the second season because of devastating borer infestations. This is a great economic loss to the farmers because second crop of maize is very profitable to farmers. This is due to high demand of green maize at this period. Also, the green maize at this time bridge hunger gap between depletion of rainy season harvest and next season harvest. Therefore, the importance of the second season maize in West African forest zones cannot be overemphasized.

Control of lepidopterous borer includes biological, cultural and chemical methods, as well as host plant resistance. Chemical insecticides are used in some countries in Sub-saharan Africa for the control of lepidopterous borer. Many resource-limited African farmers cannot afford insecticides and these chemicals are seldom available. They usually require application equipment and technical knowledge that are beyond the reach of an average farmer.

Insecticides often have a negative impact on the environment, besides its health hazard to humans and non-targeted organisms of the ecosystem. The chemical control of stem and ear borers is more difficult than that of insects which are external plant feeders, since most of their life cycle is spent within plant tissue that could not be reached with contact insecticides.

Host plant resistance, therefore, is an ideal method of pest control, because it requires minimum input and action of the farmer (i.e. purchase of seed of the right variety only). It has no negative environmental effects. Stem borers resistant varieties have been suggested as one of the most promising means of control (Bowden, 1976; Girling, 1980). Moderate levels of resistance could be combined with other methods of control to reduce economic impact of stem borers (Bosque-Perez and Schulthess, 1998). It is conceivable that breeding maize plants for stem borer resistance would progress rapidly if sufficient genetic diversity are present (Alabi, 2006). The probability of identifying sources of resistance to insect attack is closely related to the volume of germplasm of the crop that is evaluated under controlled conditions (Paliwal *et al.*, 2000). Hence the need for artificial screening of the selected crop genotypes for effective host-plant resistance.

Even though there is an extremely large genetic diversity in the *Zea mays* (L) species, the number of genotypes identified as possible sources of resistance to insect species of economic importance is not large (Paliwal *et al.*, 2000). Therefore, the search for resistance genotypes was conducted in a sequential and systematic fashion whereby germplasm evaluation of the elite, high yielding, agronomically desirable and widely adapted germplasm (Paliwal *et al.*, 2000) were considered first. The germplasm evaluation of other desirable unimproved material that appear agronomically promising (Paliwal *et al.*, 2000) were also considered. These include maize landraces or “criollos”, bank collections, germplasm from other programmes and even unadapted cultivars. Similarly, samples of insect pest collected were to be molecularly characterized, identified and documented for effective control research and planned projects on maize.

## **OBJECTIVES OF THE STUDY:**

The general objectives of this study were:

To identify and characterize stem borer populations associated with maize in Nigeria.

The specific objectives are:

1. To survey South West and Kwara state for incidence and occurrence, abundance and diversity of stem borers.
2. Laboratory identification and characterization of stem borers.
3. To determine phylogenetic relationship among these stem borers.
4. To determine resistance of some maize genotypes to stem borer pests.

## CHAPTER TWO

### LITERATURE REVIEW

#### 2.1 BIOLOGY OF MAJOR MAIZE STEM AND EAR BORERS

Lepidopterous borers are among the economically important pests of maize in Africa. Four stem borers and one ear borer cause significant yield loss: the maize stalk borer, *Busseola fusca* Fuller (Noctuidae); the pink stalk borer, *Sesamia calamistis* Hampson (Noctuidae); the African sugar-cane borer, *Eldana saccharina* Walker (Pyralidae); the spotted stalk borer, *Chilo partellus* Swinhoe (Crambidae); and the ear borer *Mussidia nigrivenella* (Ragonot) (Pyralidae). The first three are African in origin and are present in most countries of Sub-Saharan Africa, while *C. partellus* originated in Asia and was accidentally introduced to Eastern Africa approximately 60 years ago (Bowden, 1954; Harris, 1962; Appert, 1970; Breniere, 1971; Boardat *et al.*, 1977; Girling, 1978). *Mussidia nigrivenella* is present in several countries of Sub-Saharan Africa but its area of origin is not clear. In addition to the above five species, other borers of minor importance which have been reported on maize in Africa include *Sesamia botanophaga* Tams and Bowden, *Chilo orichalcociliellus* (Strand), *Chilo agamemnon* Bleszynski, *Chilo diffusilineus* (de Jeannis), and *Coniesta* (=Acigona) *ignefusalis* (Crambidae) (Harris 1962; Endrody-Younga, 1968; Appert, 1970; Bleszynski, 1970; Bonzi, 1982a; Moyal and Tran, 1992). The ear borer *Cryptophlebia leucotreta* (Tortricidae) has also been reported as a minor pest of maize in several West African countries (Libby, 1968; Moyal, 1988; Shanower *et al.*, 1991).

Maize is an exotic crop introduced to Africa from the Americas. The most important pests of maize are however indigenous to Africa and their natural hosts are wild grasses and sedges. The two exceptions are *Chilo partellus* and larger grain borer, *Prostephanus truncatus* (Horn) (Coleoptera: Bostrichidae) which are exotic.

##### 2.1.1. *Sesamia calamistis*

The two most important *Sesamia* species in Africa are *S. calamistis* and *S. botanophaga*. According to Boardat *et al.*, (1977), the former species is present in most countries of Sub-Saharan Africa, Madagascar and the Comoros, while the latter is present in West Africa, Sudan, Uganda and Kenya.

The host range of *S. calamistis* is limited to the family Gramineae (Poaceae) and includes cultivated crops, such as maize, sorghum and millet, as well as wild grasses, such as *Pennisetum purpureum*, *Panicum maximum* and *Setria splendida* (Harris 1962).

Adults of *S. calamistis* emerge in the late evening and mate. Females lay their eggs between the leaf sheaths of the host plant. Longevity of adults is influenced by temperature. In Laboratory tests, females lived 5.7 days at 30<sup>0</sup>C and 10 days at 20<sup>0</sup>C (Shanower *et al.*, 1993a). Boseque-Perez and Dabrowski (1989) found that when larvae were reared on an artificial diet, females laid an average of 320 eggs each in a period of 5 days at temperatures between 24<sup>0</sup>C and 26<sup>0</sup>C. Fecundity tests conducted by Shanower *et al.*, (1993a) revealed that, on the average, a female lays 249 at 25<sup>0</sup>C and 688 eggs at 20<sup>0</sup>C. Under field conditions, eggs hatch in 5-6days and most larvae penetrate the stem shortly after egg hatch.

The development rates of *S. calamistis* have been studied under controlled conditions by Shanower *et al.*, (1993). On an artificial diet, *S. calamistis* required 709 degree – days (DD) to complete its development; eggs needed 122 DD above threshold of 9.7<sup>0</sup>C, larvae 383 DD above 12.2<sup>0</sup>C and pupae 204 DD above 10.2<sup>0</sup>C. The threshold temperatures are well below the mean daily temperatures in the maize growing regions of West Africa, *S. calamistis* is thus theoretically capable of developing throughout the year, provided suitable host plants are available (Shanower *et al.*, 1993a). Development time was also influenced by temperature; immature stages took 162, 94, 50 and 38 days at 15°, 20°, 25° and 30°C, respectively.

Larval feeding might result in the destruction of the growing point, typically referred to as "deadhearts". At later stages, the tunneling and girdling activities of the larvae often result in stalk breakage. In the field, development of the larvae takes 4-6 weeks and most larvae pupate within the stem or cobs.

In contrast to *B. fusca*, *S. calamistis* breeds throughout the year and has no resting stage (Harris, 1962). However, population densities are low during the dry season, when its hosts (mature grasses and maize) are growing in small areas in hydromorphic soils. Bowden (1976) reported that *Sesamia Spp* adults that emerge at the beginning of the cropping season are smaller and less fecund than those emerging later in the year and that the combined effects of smaller numbers of less fecund adults result in lower incidence of *Sesamia spp* in first season maize crops. This report has been confirmed by Shanower *et al.*, (1993b), who studied the

effect of larval diet on the growth and development of *S. calamistis* in the laboratory. Five indigenous African grasses were compared with maize and an artificial diet for their ability to support this insect. While 95% and 30% of the larvae successfully pupated after being reared on artificial diet and maize-stem cuttings, respectively, only 7% pupated on *Sorghum arundinaceum*, 1% or less on *P. maximum*, *P. purpureum* and *Andropogon sp* and none on *Pennisetum polystachion*. Development time was similar on maize and the grasses, but was 50% slower than on the artificial diet. Pupal weights of larvae reared on four of the grasses were much lower than for larvae reared on maize or artificial diet (Shanower *et al*, 1993b). Given that pupal weight is an indicator of fecundity (Setamou *et al*, 1993), it is to be expected that, in areas where *S. calamistis* is forced to feed on wild grasses to bridge the cropping season, larva survivorship and adult fecundity will be greatly reduced. On the other hand, in regions where more than one crop of maize is planted, the potential for pest outbreaks is great because maize provides a suitable host for growth. In West Africa, the population of this borer increases until it peaks around August - September (Harris, 1962; Bosque-Perez and Mareck, 1990). This occurs when second - season maize crops are being grown, and, as a result, *Sesamia spp.* can be a serious problem. *Sesamia* populations often crash around November, in spite of continuous oviposition by adult insects, the mortality factors responsible for this decline are not clearly understood, but natural enemies, especially egg parasitoids and pathogens, are assumed to play a major role (Setamou and Schulthess, 1995).

In addition to host-plant and weather factors, soil nutrients are known to affect bionomics of *S. calamistis*. Detailed studies in this area have been conducted by Setamou *et al.*, (1993). Increasing nitrogen levels in maize plants was found to increase larval survivorship, female fecundity and longevity; nitrogen, however, was not observed to have an effect on development time. Increasing silica applications to maize plants reduced larval survivorship but had no effect on the life parameters (Setamou *et al.*, 1993).

### 2.1.2 *Busseola fusca*

*Busseola fusca* is considered by some authors to be the most important pest of maize in Sub-Saharan Africa (Appert, 1970). However, this is mainly due to its prevalence and abundance on maize in mid altitude regions of East and Southern Africa, as the pest does not appear to be as important as *S. calamistis* or *E. saccharina* on maize in West Africa (Bosque-Perez and Mareck, 1990; Shanower *et al.*, 1991; Gounou *et al*, 1993). *Busseola fusca* is distributed from

approximately 12°N to 30°S (Appert, 1970). This insect was recognized as a major pest of cereals when originally described in 1901 (Tams and Bowden, 1953).

After emergence, larvae crawl over the plants, congregate in the funnel and feed on the rolled leaves. As the leaves grow away from the funnel, a characteristic pattern of holes and "window panes" can be seen (Harris, 1962). Continuous feeding by the larvae might result in deadhearts. After killing a plant, larvae usually migrate to new plants and enter them by boring into the stem near the base. Tunneling of the stem and ears then occurs. Larval development takes between 26 and 33 days.

During the dry season, larvae enter diapause period of arrested development, which usually occurs during adverse environmental conditions - and take up to 6 months to complete their development (Harris, 1962). With the initiation of the rains, the larvae pupate within the stems and 10-12 days later adult moths emerge.

### 2.1.3 *Eldana saccharina*

The stem borer *E. saccharina* was first described in Sierra-Leone and has been known as a pest of graminaceous crops in West Africa for more than a century (Appert 1970). It probably occurs in all suitable areas of Sub-saharan Africa, from approximately latitude 15°N to 30°S (Girling, 1978). In East and Southern Africa, the wild hosts of *E. saccharina* are sedges, *Cyperus* spp, which are preferred over crops, such as sugar cane, as oviposition sites (Atkinson, 1980). On sedges, *E. saccharina* is not a stem borer but feeds in the culms and inflorescence. From several surveys by International Institute of Tropical Agriculture (IITA) Scientists in West Africa this pest was rarely found feeding on wild sedges and grasses. The major hosts of this borer in West Africa are crop plants, such as maize, sugar cane, sorghum and millet. *Eldana saccharina* has been reported as an important pest of maize (Leyenaar and Mareck, 1991) and is often the most prevalent and abundant borer species at the end of the maize -growing season (Bosque-Perez and Mareck, 1990). Infestations of maize plants usually start at anthesis.

The longevity of *E. saccharina* adults is influenced by temperature. In laboratory tests, females lived 6.6 days at 30°C and 15 days at 20°C (Shanower *et al.*, 1993). Females lay eggs on debris on the soil (Atkinson, 1980) or on the hairy margins of maize leaf sheaths

(Cochereau, 1985). Bosque-Perez and Dabrowski (1989) reported that, when larvae are reared on an artificial diet, females lay an average of 380 eggs each during a 5 day period at temperatures between 24 and 26°C. Fecundity tests conducted by Shanower *et al.*, 1993a) using moths from larvae reared on an artificial diet revealed that, on average, a female lay 470 eggs at 20°C and 620 eggs at 30°C. In these studies, fecundity was positively correlated with temperature, but at higher temperatures females survived fewer days (Shanower *et al.*, 1993a), suggesting that temperature is one of the important factors that influences egg population in this insect especially in tropical environment.

Under field conditions, eggs hatch in 5 or 6 days and, after feeding on the leaf sheaths for a few days, larvae enter the stem, where they continue to feed. Larvae may move into the ears and feed on the grain.

Shanower *et al.*, (1993) studied the development rates of *Eldana saccharina* under controlled conditions in the laboratory. On an artificial diet, *E. saccharina* required 709 DD to complete its development; eggs required 125 DD above 9.1°C, larvae 400 DD above 10.6°C and pupae 140 DD above 8.8°C. Like *S. calamistis*, *E. saccharina* is capable of developing throughout the year, provided suitable host plants are available, as these threshold temperatures are below the average mean daily temperatures in the maize growing regions of West Africa (Shanower *et al.*, 1993a). Larval development and growth are also known to be influenced by food source. For example, development was found to be significantly slower when larvae were fed maize stems, compared with artificial diet (Shanower *et al.*, (1993a), indicating that diet determine to a greater extent the growth and development of *Eldana saccharina*, In further experiments by Shanower *et al.*, (1993b), it was found that larval development time was similar on maize and five species of grasses but larval survivorship was <5% on grasses compared with 20% on maize stems and 65% on artificial diet. Temperature also influenced developmental rate, with immature stages lasting 173, 63, 44 and 33 days at 15°, 20°, 25° and 30°C, respectively (Shanower *et al.*, 1993a).

Under field conditions, pupation occurs inside the stem or cobs and the pupa is covered by a cocoon made of site and adult debris. A good external sign of *E. saccharina* attack is the adult exit hole cut by the larvae prior to pupation, after which a large amount of frass hanging from it is observed. Adults emerge 7—14 days after pupation.



Although infestations by this stem borer occur relatively late in the development of the maize plants, damage as a result of their feeding can be severe, with yield losses of up to 20% (Bosque-Perez and Mareck, 1991). Damage done by *E. saccharina* provides access to the stem and cobs for pathogens that can cause rots, and infestations by this borer are associated with high levels of stalk lodging due to tunneling and the effect of stalk rots.

#### **2.1.4 *Mussidia nigrivenella***

The literature on the ear borer *M. nigrivenella* is scanty. The species was first described by Ragonot in 1888, and since then it has been recorded on numerous plant species, including maize, cotton, cocoa, lima beans (*Phaseolus lunatus*), the shea butter tree (*Vitellaria paradoxa* (Syn. *Butrospermum parkii*), *Mucuna pruriens*, *Canavalia* sp and *Stenostylis stenocarpa* (Moyil and Tran, 1991b). This pest infects fruiting structures of mature plants and continues feeding on the stored products (Bordat and Renand, 1987). Infestations of maize start in the field; female moths lay their eggs on the silk and husk leaves. Eggs hatch in 5 — 7 days and young larvae feed within the silk channel for four days before reaching grain. Developing larvae feed on the distal portion of the maize ear and tunnel through the grain, causing extensive damage and often consuming the embryos in a way that it is not superficially visible. Only close inspection reveals the degree of damage. Pupation takes place within the tunnels or on the surface of the grain, and the pupa are surrounded by a silky cocoon. Damage to the grain continues during storage; hence, *M. nigrivenella* can be regarded as both a field and a storage pest, although no reproduction occurs in the store. Preliminary observations indicate that the pest can survive in stored cobs for up to 8 weeks, even at grain moisture contents of 12 - 15% (O. Bolaji, N. A. Bosque-Perez and M. Ivbijaro, unpublished data). This implies that regardless of storage condition this pest can still infest maize in storage, hence the need for effective control measures.

Surveys have been conducted by IITA scientists in farmers' field in Benin, Ghana, Ivory Coast and Nigeria to establish the geographical distribution, host plant ranges and natural enemy complex of this species in West Africa, *Mussidia nigrivenella* has been found in every country and ecological zone from forest to the northern Guinea Savannah but rarely found in the Sudan Savannah. Survey results from Benin indicate that each larva causes, on average, 4% ear damage; five larvae per infested ear are often found (F. Schulthess, unpublished).

Surveys in South-western Nigeria presented this borer to be the most abundant pest of maize at the time of harvest (O. Bolaji, N. A. Bosque-Perez and M. Ivbijaro, unpublished data). In studies conducted at IITA, Ibadan, maize varieties with a short husk-tip extension and loose husk leaves were found to be more severely infested by *M. nigrivenella* than those with a good husk cover; additionally, the abundance of this ear borer was found to increase with delayed harvesting (O. Bolaji and N. A. Bosque-Perez, unpublished data). Breeders may therefore need to select maize genotypes with tight husk and close ear to reduce the effect of this pest.

The life history of *Mussidia nigrivenella* reared on an artificial diet in the laboratory has been studied by Bordat and Renand (1987). On average, at 25°C, the incubation period of the eggs was 4 days, larval stages lasted 18 -21 days and the pupae emerged in 6 - 9 days. Female laid an average of 115 eggs over a period of 5 days, with a maximum of 213 eggs (Bordat and Renand, 1987).

Moyal and Tran (1991a) also studied the life cycle of this borer. When reared on an artificial diet at 22°C, *M. nigrivenella* required between 46 (males) and 49 (females) days to complete its development, on the average, eggs required 6.4, larvae 31.6 and pupae 8.9 days. Although dissection of females revealed over 650 eggs in the ovaries, under the conditions of their experiments the actual number of eggs laid per female (i.e. 69) was low (Moyal and Tran, 1991a). From this report, the virulent nature of this pest depends not on the egg laying potential but damaging effect of few insect pests on the crops.

## **2.2 DISTRIBUTION OF STEM AND EAR BORERS**

During the past few years, surveys conducted by scientists from various national and international institutions in several countries of West and Central Africa to obtain information on the abundance, species composition and relative importance of lepidopterous borers and their natural enemies revealed major findings as summarized below.

In experiments conducted in six locations in Southern Nigeria from August to November of 1985 and 1986, *S. calamistis* was the dominant maize borer up to 8 weeks after planting and *E. saccharina* was the most abundant from 9 weeks after planting onwards (Bosque-Perez and Mareck, 1990). *Mussidia nigrivenella* was found in all the sites, while *B. fusca* was found

in forest/savannah transition zone . Other borer species encountered included *C. ignefusalis* and *Cryptophlebia* sp.

Additional surveys of borers and their natural enemies were carried out by IITA scientists in maize fields in Nigeria during 1991 and 1992 (Bosque-Perez *et al*, 1995, and unpublished data). *Sesamia calamistis* and *E. saccharina* were the most commonly encountered pest of maize; additionally, *B. fusca*, *C. ignefusalis*, *M. nigrivenella* and *C. leucotreta* were detected. Fifty-seven per cent (57%) and 55% of the field sampled had evidence of borer damage in 1991 and 1992, respectively. The percentages of plants with borer damage in individual field varied according to year and ecological zone, with a maximum of 17% for both Southern Guinea and Northern Guinea savanna in 1991 and up to 30% in the Northern and 47% in the Southern Guinea Savannah in 1992. A severe infestation with *C. ignefusalis* was observed in two locations in the Southern Guinea Savanna in 1992. Variations therefore calls for periodic survey of stem borer endemic environments for monitoring of the incidence of these important crop pests.

### **2.3 SAMPLING MAIZE BORERS**

Methods of sampling lepidopterous pest of maize have been developed by Schultess *et al.*, (1991) and Shanower *et al.*, (1991). These methods are enumerative (destructive via dissection) and Presence-absence sampling (non-destructive via visual examination for damage symptoms) methods with a pre-defined precision level. These sampling plans take into consideration the spatial distribution of a species, as defined by Taylor (1961). The species *S. calamistis*, *E. saccharina*, *M. nigrivenella* and *C. leucotreta* all show a highly aggregated distribution in the field. Since two or more lepidopterous species are often found on the same maize plant and almost all plant parts, leaves, stems, tassels and ears - are attacked. Schulthess *et al.*, (1991) therefore recommended sampling whole plants in order to accurately estimate borer populations.

Dissection of maize plants is time consuming and costly. For survey work, it is therefore often more practical to use a non-destructive presence - absence sampling procedure. This procedure requires the establishment of the relationship between the proportion of infested plants and the mean per- plant density for the stem and ear borers described above. For this, 40 plants are taken at random within a field and examined for borer damage (i.e. deadhearts, leaf damage, entrance/exit holes, presence of frass) (Schulthess *et al.*, 1991). Once the

proportion of infested plants in a given field is established, the respective density can be determined. Tables were provided by Shanower *et al.*, (1991) which can be used to obtain the borer mean density per plant, using the per cent plants infested and the species composition. To establish species composition, ten damaged plants should be dissected. Thus approach to researchers seems acceptable.

## **2.4 YIELD REDUCTIONS CAUSED BY MAIZE BORERS**

Reported yield losses due to lepidopterous borers in Africa vary greatly (0 -100%) in various ecological zones, regions and seasons. The severity and nature of stem-borer damages depend upon the borer species, the plant growth stage, the number of larvae feeding on the plant and growth stage. Feeding by borer larvae on maize plants usually results in crop losses as a consequence of death of the growing point (deadhearts), early leaf senescence, reduced translocation, lodging and direct damage to the ears (Appert, 1970; Breniere, 1971; Bosque-Perez and Mareck, 1991). Infestations with stem borers have been found to increase significantly the incidence and severity of stalk rots (Bosque-Perez and Mareck, 1991). Thereby reducing the potential yield of the infested crops.

The degree of stem-borer attack might be assessed by determining the percentage of plants attacked, extent of plant damage - i.e. percentage of the stem tunneled, percentage of the nodes bored, stalk breakage, extent of ear damage - and/or number of larvae or pupae per plant (Walker, 1981). It is also possible to measure the relationship between infestation rate and crop loss by using natural or artificial infestations and by preventing attack, using insecticides or other methods of control (Walker, 1981).

The effect of *E. saccharina* infestations on the field components of maize has been studied by Bosque-Perez and Mareck (1991) through comparisons of infested and insecticide-protected plants. Yield comparison of infested plants versus insecticide-protected ones showed that natural infestations decreased yield by 16, 15 and 28% in the dry, first and second rainy seasons, respectively. For the infested plots, significant negative correlation was found between 100 grain weight and the percentage stem tunneled ( $r = -0.79$ ), suggesting that *E. saccharina* damage to the stem definitely affects grain filling. The regression coefficient obtained from covariance analysis showed that 100-grain weight decreases by  $0.125 \pm 0.0477\text{g}$  for every percentage increase of the stem tunneled.

Using data from surveys conducted in late 1990 in farmers' field in Ghana, Gounou *et al*, (1993) found a positive relationship between the number of *Sesamia sp.* and the extent of the maize stem tunneled and a significant negative relationship between per cent stem tunneling and maize yield. These authors used multiple-regression equations, which include plant parameters (stem diameter, phonological stage) and damage variables (per cent tunneled) to calculate the average ear-weight losses for each ecological zone. The calculated yield-loss figures for late 1990 were 32.5, 14, 19 and 18g plant<sup>-1</sup> for the rain forest, coastal, derived Savanna and Guinea Savanna, respectively, resulting to 27, 15, 18 and 14% yield loss.

In Zaire, the effect of stem-borer attack (mostly *B. fusca*) on maize in farmers' field has been studied by scientists from the IITA/US Agency for International Development (USAID/Zaire Applied Agricultural Research and Outreach Project (Anon, 1990). Multiple-regression analysis was used to identify the relationship between various agronomic practices, observed variables and yield of maize. In maize plots receiving fertilizer treatment, early plantings (i.e. October - November) were estimated to suffer 8 - 9% yield losses (Anon, 1990). Trials conducted by Muyango (1987) in Burundi, using insecticides and exclusion cages, revealed maize-yield reductions of 12 - 15% as a result of *E. saccharina* attacks. In regions between 1500 and 2100m, attacks by *B.fusca* occasionally caused yield reductions of 30 - 50% (Muyango, 1987).

## **2.5 METHODS OF CONTROLLING STEM AND EAR BORERS**

Control of lepidopterous borers includes biological, cultural and chemical methods, as well as host-plant resistance.

### **2.5.1 Biological control**

Natural enemies play an important role in the control of lepidopterous borers in Africa. Approximately 100 genera of parasitoids (Hymenoptera and Diptera) have been recorded attacking cereal stem borers in Africa and its surrounding islands (Polaszek, 1992). In West Africa, several imported natural enemies, such as *Trichogramma spp*, *Sturmiopsis inferens*, *Cotesia flavipes*, *Amyosoma chinense* and *Tetrastichus spp.*, have been released against stem borers in Ghana (Scheibelreiter, 1980), although they failed to establish. In South Africa, introduced parasitoids were first used for the control of *E. saccharina* in 1975 (Carnegie and

Leslie, 1979), but this programme was not successful. Releases of exotic parasites against *S. calamistis*, *B. fusca*, *C. partellus* and *E. saccharina* have also been tried in East Africa, without much success (Ingram, 1983). It appears that the only successful biological control against stem borers in Africa was conducted in Madagascar against *S. calamistis* and *C. partellus* (Appert *et al*, 1969). However, biological control is still considered a viable control option. It is probable that, due to lack of fund, biological control attempts in the past have not been pursued for a sufficiently long period. In most cases, no effort has been made to understand why natural enemy failed to establish. Before releasing a natural enemy, it is important to have a profound knowledge of ecosystem in which it is to be released. This includes crops and wild host plants, which are important over-seasoning sites for pests and natural enemies. In addition, information is needed about the relative importance of a natural enemy in the original habitat and behavioural studies (e.g. Searching behaviour, functional response studies) in the laboratory should be carried out. The results may already give some information about the strengths and weaknesses of a natural enemy and the chance of survival in the new ecosystem. After release, follow up studies are needed to assess establishment, spread and impact on pest populations and crop yield.

Work on cataloguing natural enemies of stem borers has been initiated in several African countries by various institutions, including IITA. Emphasis is being given to parasitoids, although predators, especially ants are known to be important natural enemies of stem borers (Girling 1978). At IITA, the cataloguing is complemented with studies on abundance throughout several cropping seasons, to determine the relative importance of different species in the system. The aim of the work was to compare the natural enemy complexes in various ecological zones and regions of Africa, in order to identify promising candidates to be exchanged between regions. Results of various countries wide surveys and detail on station research carried out by IITA scientists in West Africa have shown that larval and pupal parasitoids of *S. calamistis* and *E. saccharina* are exceedingly rare in this part of the continent (IITA, 1993). Parasitization rates of less than 3% were found at the end of the second planting season, when the damage to maize is already done. Similar surveys and on-station trial done in Benin, Nigeria, Ghana and Ivory Coast suggests that most common natural enemies of *S. calamistis* are the egg parasitoids *I. busseolae* and *I. isis* (Polaszek *et al.*, 1993; Setamou and Schulthess. 1995).

Several pathogens have been detected on *S. calamistis* and *E. saccharina* in Nigeria (Bosque-Perez and Dabrowski, 1989) and some of these appear to be important field-mortality factors (IITA, 1986). Ecological studies, selection for greater virulence and application technology of these pathogens could make them a useful pest management tool provided their ecology is understood and means are developed for their adequate dispersal/perpetuation in the field.

### **2.5.2 Cultural Practices**

In West Central Africa, farmers do not usually make a conscious effort to control borer populations.

Some authors have reported greater infestations of stem borers in late-season maize compared to early plantings i.e. in Benin (Shanower *et al.*, 1991), Burundi (Muyango, 1987), Cameroon (Aroga, 1988), Ghana (Girling, 1986; Gounou *et al.*, 1993), Nigeria (Harris, 1962; Adeyemi *et al.*, 1966; Carter, 1985) and Zaire (Alam, 1990). Thus, in some areas farmers do not plant maize during second season, because of devastating borer infestations. This may influence the population dynamics of the pest, especially during the dry season. For example, crop residue left after harvest or used as building materials are important overwintering site for diapausing species, such as *B. fusca* (Harris, 1962). Burning at the end of the dry season to clear bush fallow may have an effect on both pests and natural enemies.

As reported by Setamou *et al.*, (1995), nitrogen fertilization enhances both borer development and the plants' tolerance to borer attack. Yield losses decreased linearly from 20% with no fertilizer to 11% with 120kg nitrogen ha<sup>-1</sup>

Survey results from Ghana and information from the literatures suggest that mixed cropping does not have an important effect on borer infestations on maize (Gounou *et al.*, 1993). Recently Olaoye and Ajala (personal communication) have found out that Telefaria and cassava intercropped with maize reduces infestation of stem borers in S.E Nigeria. However occurrence and abundance of wild hosts appear to be more important in explaining fluctuations in borer populations than intercropping. In most cases, the role of wild hosts is unknown and misunderstood. They can be sources for new outbreaks of pests and reservoirs of natural enemies (and thus prevent or reduce pest outbreaks), be overwintering sites for both pest and their natural enemies, or even act as trap plants. Consequently, the effect of a sustainable intervention technique, such as biological control or resistant varieties can only be understood in the context of the natural habitat of stem borers and their antagonists.

### 2.5.3 Chemical control

Chemical insecticides are used in some countries of Sub-Saharan Africa for the control of lepidopterous borers. Many of Africa farmers cannot afford these insecticides and required application equipment and knowledge seldom available to farmers in the continent. Additionally, insecticides often are not eco-friendly they constitutes health hazards to humans life. Many of the insecticides originally recommended for borer control in Africa have been banned in the USA and Europe because of their toxicity and the negative effect on non-targeted organsims.

The chemical control of stem and/ or ear borer is more difficult than that of insects which are external plant feeders, as most of their life cycle is spent within plant tissues that cannot be reached with contact insecticides. Thus, foliar applications of insecticides for the control of stem borers have generally been found to be unsatisfactory (Jotwani, 1983). For borers such as *B. fusca* and *C. partellus*, contact insecticides have been recommended for use during early plant stages, when insects are feeding in the plant funnel. Pyrethroids have been found to be effective for the control of *B. fusca* in Burundi (Muyango, 1987).

### 2.5.4 Use of Resistant Varieties

Host plant resistance is an ideal method of pest control, because it requires minimum input and action by the farmer. Purchase of seed of the right variety is only what it required. Maize varieties resistant to stem borers have been suggested as one of the most promising means of control (Bowden, 1976; Girling, 1980) for some years back, IITA has devoted efforts to developing sources of resistance to *S. calamistis* and *E. saccharina* (Bosque-Perez *et al*, 1989). In fact some stem borer resistant open pollinated maize varieties have been jointly developed and released by IITA and IAR&T. These include BR9928DMR and BR9943DMR.

Moderate levels of resistance could be combined with other methods of control to reduce the economic impact of stem borer. Generally integrated pest management may be adopted for effective control of borer in Africa.



## **2.6 THE ROLE OF BIOTECHNOLOGY IN AGRICULTURAL RESEARCH**

The growing human population and the concomitant increase in use of natural resources are generating a series of negative effects on ecosystems, such as pollution, loss of genetic diversity, soil fertility decline, climatic changes, deforestation, and desertification. Agriculture is asked to satisfy two apparently contradictory needs, to become more productive and at the same time more sustainable: that is, to supply the food needed without depleting renewable resources. Breeding crops that produce higher yields of better quality but do not adversely affect the ecosystem can be achieved only through a very broad scientific input.

### **2.6.1 The New Biotechnologies**

Biotechnologies could play a decisive role in agriculture because of their ability to directly modify plants, animals and agricultural processes in response to new needs. What were seen as promising technologies a few years ago have already produced new varieties. The concern about biotechnology is the high risk. Basic research should be reviewed in the light of results that have been obtained from this applied research.

### **2.6.2 Current Role of Biotechnology in Crop Improvement**

Some of the applications of biotechnology that are currently being used in crop production are shown in Table 1 below. The first of these invitro cultures of meristems and buds is now widely used for the micro-propagation of many species for commercial purposes. It is also used for germplasm conservation of vegetatively propagated species and for the exchange of virus free material.

### **Molecular Markers and their Application in Cereals Breeding**

Conventional cereal breeding like other crops is time consuming and very dependent on environmental conditions. Breeding a new variety takes between eight and twelve years, and even then the release of an improved variety cannot be guaranteed. Hence, breeders are

extremely interested in new technologies that could make this procedure more efficient. Molecular marker technology offers such a possibility by adapting a wide range of novel approaches to improving the selection strategies in cereal breeding (Thottapilly et al., 1992).

**Table 1: Biotechnology in Crop Production**

Technology	Application
Meristem and bud culture	Micro-propagation for commercial purposes, genetic conservation and exchange of material
Zygotic embryo culture	Inter-specific crosses
Anther and micro-spore culture	Haploid production
Cell and tissue culture	In vitro selection, somaclonal variation, Somatic embryo genesis, artificial seeds
Chromosome engineering	2 <sup>nd</sup> gametes for inter specific crosses
Protoplast culture	Fusion for somatic hybridization
Genetic engineering	Gene transfer
Molecular markers	Aid to breeding programmes
Monoclonal antibodies	Diagnosis of plant diseases

Source:(Thottapilly, G.L.M, Monti, D.R., Mohan-Raj, A.W., Moore (1992)

## **CHAPTER THREE**

### **MATERIALS AND METHODS**

#### **3.1 FIELD EXPERIMENTS**

##### **EVALUATION OF MAIZE VARIETIES FOR RESISTANCE TO STEM BORERS:**

Field evaluation of maize varieties for resistance to stem borers were conducted as follows:

- (1) Evaluation of 20 maize varieties (white and yellow) at NACGRAB field in 2010.
- (2) Evaluation of 10 yellow maize varieties at NACGRAB field in 2012
- (3) Evaluation of 10 white maize varieties screened at NACGRAB field in 2012.
- (4) Crosses of 10 white parent maize varieties with resistant check BR 9943DMRSR in 2014
- (5) Crosses of 10 yellow parent maize varieties with resistant check BR9928DMRSR in 2014
- (6) Evaluation of 10 yellow maize (parents of crosses) at NACGRAB field in 2014
- (7) Evaluation of 10 white maize (parent of crosses) at NACGRAB field in 2014
- (8) Evaluation of F1 of ten yellow maize varieties crossed and evaluated at NACRAB field in 2014.
- (9) Evaluation of F1 of ten white maize varieties crossed and evaluated at NACGRAB field in 2014
- (10) Evaluation of ten yellow maize parents in 5 locations in 2015.
- (11) Evaluation of ten white maize parents in 5 locations in 2015

##### **3.1.1 Experimental Sites**

The experiments were carried out in different sites. One of the sites included National Centre for Genetic Resources and Biotechnology (NACGRAB) research field at Ibadan (Latitude N7° 23.07'; Long 003°50.412' ; Altitude 183m above sea level). The field has been used over the years to plant various arable crops. The arable crops include cassava, pepper, sorghum, maize, yams etc.

The vegetation prevalent in NACGRAB is that of Forest/Forest transition (derived savanna). The area is however currently experiencing climate change. Though the rainfall is still bimodal, however since 2008, the early rainfall normally comes by April and becomes stable by June, the usual August break comes late August to September while the later rains are torrential and normally last till late November or first week in December.

The second site was at Koloko, off Lalupon, Ibadan (Lat 7°25' 04.7''N; Long 4° 07' 27.85''E). The climate was similar to that of NACGRAB, Ibadan, except that the place was closer to a major river called Asejire.

The other sites used in 2015 include Mokwa (Lat 9° 17' 34.17'' N; Long 5° 3' 16.79''E). The elevation is 153.64m above the sea level. Mokwa is in Southern Guinea Savanna. It is about 31.7km South West to Jebba and 62.2km South East to Patigi, Kwara State. It is 267.9km West of Abuja, FCT. Kontagora was another site (Lat 10° 24' 25.73''N; Long 5° 28' 11.7''E). The elevation is 330.92m above the sea level. Kontagora is 36.5km to West North West (WNW) Ibeto , 80.5km North North West (NNW) to Rijau and 85.3km East South East (ESE) to Tegina. Another site was Lugbe, FCT (Lat 8° 58' 30.263''N; Long 7° 22' 34.702''E). The Elevation is 399m above the sea level. The other sites includes Okebukun (Kabba) (Lat E 006° 11' 05.1''; Long N07° 49' 01.1'' Alt 507m).

### **3.1.2 Experimental Lay-out**

The first experiment was carried out at NACGRAB, Ibadan field in 2010. The twenty maize germplasm which were both white and yellow maize varieties and improved and land races were evaluated for resistance to stem borers in 2010 to identify resistance donors.

The second experiment was also carried out at NACGRAB, Ibadan field in the year 2012. The twenty maize varieties were landraces consisting of 10 yellow maize varieties and 10 white maize varieties .They were obtained from IITA.

The next experiment involved crosses of IITA open pollinated varieties (which previously have been bred for resistance to aflatoxin) with IITA Borer resistant checks. The ten yellow varieties were crossed with yellow borer resistant check BR 9928DMRSR while ten white varieties were crossed with white borer resistant check BR 9943 DMRSR. This was done between March and April 2014 at Koloko, off Lalupon, Ibadan, Oyo State.

The next experiment was carried out in July, 2014. This is with the third set of maize (which was used for the crosses carried out at Koloko, off Lalupon, Ibadan). The third set of twenty improved maize varieties (ten yellow and ten white) were evaluated in 2014 for resistance to stem borers to identify resistance donors in the second season in the field, at NACGRAB, Moor Plantation, Ibadan, South West Nigeria.

The next experiment was carried out in 2015. The third set of twenty improved maize varieties (ten yellow and ten white) together with reference checks were re-evaluated in 2015 for resistance to stem borers to identify resistance donors in the season in five locations, namely: Ibadan (Oyo State), Mokwa (Niger State), Kontagora (Niger State), Lugbe (Abuja, FCT) and Kabba (Kogi State).

All the experiments were under rain fed except the crosses that were carried out at Koloko, Lalupon, Ibadan that were both irrigated and rain fed. The experimental design used was Randomized Complete Block Design (RCBD). The plots were one row per plots. The rows were 5m long each, while intra row spacing was 25cm with the inter row spacing of 75cm. The experiments were replicated three times except in 2012 which were replicated 4 times as a result of more available seeds and space for evaluation.

### **3.1.3 Cultural Practices**

The land was ploughed, harrowed and ridged using tractor where possible and hoes where necessary. The planting was done manually following the farmers' method. No mechanical method was employed in planting. Some of the stands that did not germinate were supplied 8 days after planting.

Fertilizer application was done according to the farmers practice i.e. 120KgN ha<sup>-1</sup>, 30Kg P ha<sup>-1</sup> and 30Kg ha<sup>-1</sup>. The fertilizers used to achieve these doses were both Urea and NPK. No insecticide was used. However, pre-emergence herbicide was used to control the weeds, while post emergence weed control was achieved using hand weeding.

### **3.1.4 Collection of Entomological Data**

(1) Plant counts: The plants were thinned to one or two plants per stand. The plant counts per row was counted at 4 Weeks after emergence (4WAE)

- (2) Stem borer leaf feeding 4 WAE (SBLF4) - The number of leaves with leaf feeding symptoms caused by stem borer was done at 4 weeks after emergence per plot
- (3) Leaf feeding score (LFS4)- Record of leaf feeding score with the help of visual damage rating scale for leaf feeding 4 weeks after emergence per plot .
- (4) Stem borer dead heart WAE (SBDH4) - Record of the number of plants per plot with dead heart was done at 4 weeks after emergence.
- (5) Stem borer leaf feeding 6WAE (SBLF6) – The number of leaves with leaf feeding by stem borer 6 weeks after emergence.
- (6) Leaf feeding score 6 WAE (LFS6)- Leaf feeding score with help of visual damage rating scale for leaf feeding 6 weeks after emergence per plot.
- (7) Stem borer dead heart 6 WAE (SBDH6) - The number of plants per plot with dead heart due to stem borers 6 weeks after emergence.
- (8) Stem borer leaf feeding (SBLF8) - The number of leaves with leaf feeding by stem borer 8 weeks after emergence per plot.
- (9) Leaf feeding score 8WAE (LFS8)- Leaf feeding score with help of visual damage rating scale for leaf feeding 8 weeks after emergence per plot .
- (10) Stem borer dead heart 8 WAE (SBDH8) - The number of plants per plot with dead heart due to stem borers 8 weeks after emergence.
- (11) Root lodging (RL)-Number of plants which were leaning (starting from the root base) more than 45 degree from the upright position.
- (12) Stalk lodging (SL) - Number of plants with stalks broken below the ear or bending more than 45 degree from the upright position.
- (13) Number of holes per stem at harvest (NOH) – 5 or 10 plants were randomly selected per plot at harvest. The number of holes on each stem due to stem borer were counted. Average number of holes per plot were recorded.

(14) Length of tunnels per stem at harvest (LOT) - 5 or 10 plants were randomly selected per plot at harvest. Each plant was dissected and the length of tunnel made by stem borers on each plant were measured in centimetres. The average length of tunnel per plot was used.



Table 2: Visual Damage Rating Scale for Leaf Feeding

Score	No of leaves with feeding systems	Leaf eaten ( $mm^2$ )
1	1 -2	<150
2	1 -2	150 -300
3	2 -3	300 -450
4	2 - 3	450 -600
5	3 -4	600 -750
6	3 -4	750 -900
7	4 -5	900-1,050
8	4 - 5	1,050 - 1,200
9	5 -6	>1 ,200

### 3.1.5 Collection of Agronomic data

(1) Days to 50% flowering or anthesis (DF 50%) - Number of days from planting to the date when 50% of the plants tasseled.

(2) Days to 50% silking (Days to 50% SILK) - Number of days from planting to the date when 50% of the plants have silk emerged.

(3) Anthesis-Silking interval (ANSI) - The difference between number of days to anthesis and silking.

(4) Plant height at harvest (PHT)- 5 or 10 plants randomly selected from each plot were taken and measured in Centimetres (cm) the distance from the base of the plant to the base of tassel(the place where tassel begins branching).

(5) Ear height (EHT) - Measured in Centimeters (cm) as the distance from the base of the plant to the node which bears the top ear.

(6) Plant aspect (PASPECT) – These was taken before harvest, after flowering (at brown silk stage) when plants are still green and ears fully developed. Scores of 1-5(1-excellent; 5-very poor). This is general score for “look” or “appeal” of the whole row of plants per plot. In short how good looking the plants were.

(7)Root lodging (RL)-Actual number of plants which were leaning (starting from the root base) more than 45 degree from the upright position were taken as root lodging

(8) Stalk lodging (SL) - Number of plants with stalks broken below the ear or bending more than 45 degree from the upright position were taken as stalk lodging

(9) Husk cover (HC) - The ears husk cover were rated on a scale of 1-5 (1-very tight and complete husk cover and 5- very loose husk cover and exposed ear tips).

(10) Number of plants harvested (PHARV) - The actual number of plants per plot were counted. The count were done just before or at harvest.

(11)Number of ears harvested (EHARV) - Actual number of ears harvested and weighed per plot.

(12) Ear aspect (EASPECT) - Scores of 1-5 (1-Excellent; 5-very poor). The score was given when the pile of harvested ears of each plot was spread out and general look of the ears was taken into account. (Ear size, uniform size, uniformity of color and texture, grain fill, disease and insect damage were all considered for this score).

(13) Ear rot (EROT) - Based on proportion of ears showing rot using scale (1-5) (1 little or no rot; 5-most of the ears were badly rotten).

(14) Field weight (FWT) - The weight (in Kilograms (kg)) of all the ears recorded per plot were taken.

(15) Moisture content (MC) - Grain moisture taken by moisture tester at harvest in percentage.

(16) Grain weight- The weight of grains in kg/plot.

### **3.1.6 Data Analysis**

Analysis of variance (ANOVA) was performed on plot means of individual characters evaluated for both agronomic data and entomological data by using General Linear Model procedure (GLM) of the Statistical Analysis System (SAS) package (SAS Institute Inc. 1990). Significantly different means were separated using Duncan's multiple range test at 1% and 5% probability levels.

## **3.2 SURVEY OF STEM BORERS IN SELECTED LOCATIONS**

Five states in South West Nigeria ( Oyo, Ogun, Ondo, Osun and Ekiti States together with Kwara State) in tropical humid ecology were surveyed for occurrence and abundance of stem borers infestation. The survey was done on maize farms in all these states from July 16, 2014 to 1<sup>st</sup> August, 2014. The Geographical Positioning System (GPS) of all the farms surveyed were captured. On each farm, five (5) rectangles of 50m by 100 m were surveyed. In each of the five (5) rectangles taken across each farm, about one hundred plants were counted and the number affected by stem borers were counted and expressed as percentage. Average percentage of the five rectangles were taken to represent the infestation per farm. Five farms were surveyed per state.

### **3.2.1 Sampling of immature stages**

During the survey, the immature stages (larvae and pupae) samples of the stem borers were collected. This was to identify the type and the damage of the stem borers. The maize that was affected by stem borers were characterized by dry chaffy tassels and sometimes broken chaffy tassels. The stems are usually broken as well. They were susceptible to root and stalk lodging. Such stems were dissected and the immature stem borer samples were collected from them. This was done for all locations and borer samples were labelled.

### **3.2.2 Rearing of immature stages in the laboratory**

The samples collected from each location were divided into two. One half was subjected to molecular analysis. The other half was reared at National Centre for Genetic Resources and Biotechnology (NACGRAB) at room temperature between 25°C to 33° C until they become

adult. The immature larva stages were fed with fresh maize leaves and stems till they turn to pupae. From pupae they turned to adults.

### **3.2.3 Data Analysis**

Analysis of variance was also performed on the results collected from survey of stem borers carried out in some selected locations by using SPSS software. The data for stem borer complex and cropping system were coded to enable analysis. The number of types of stem borer found in each location determine the number inputted in that location under stem borer complex. For cropping system, sole cropping was denoted 1 while intercropping was denoted 2 but sole cropping with few stands of other crops was denoted 1.5.

## **3.3 MOLECULAR CHARACTERIZATION OF STEM BORERS**

### **3.3.1 Collection and preservation of stem borers**

The samples of immature stages of stem borers collected were preserved in 96% Ethanol. This was to keep the samples intact until they were taken to IITA Bioscience Centre for further analysis.

### **3.3.2 DNA EXTRACTION OF THE STEM BORER SAMPLE**

The larvae collected from the field were preserved in Ethanol before taken to IITA Bioscience Centre. The DNA of the larvae were extracted using QIAGEN KIT. The Kit used was QIAamp DNA Mini Kit (250) Catalogue no 51306.

Buffer AE was placed in 70°C water bath. 180µl of ATL was added to the isolate. 20µl of proteinase K was added to remove protein contaminants. The samples were incubated at 56°C until completely lysed. The samples were vortexed occasionally or heat block were shaken at 500rpm. Lysis was completed in 2 to 3 hours. The tubes were centrifuged to collect the condensation. 200 µl of buffer AL was added and the samples were vortexed for 15 seconds. The samples were then incubated for 70°C for 10 minutes. The tubes were centrifuged again to collect the condensation. 230µl of ethanol (96-100%) was added and the samples were mixed by vortexing for 30 seconds. The samples were then applied to QIAamp spin column. The samples were centrifuged at 6000g for 1 minute. 500µl of buffer AW1 was then added. The mixtures were then spinned for 1 minute and the spin was placed in a collection tube. The filtrate was then discarded. After this, 500µl of buffer AW2 was added. The mixtures were spinned again at 6000g for 1 minute and the spin was placed in a collection tube. The filtrate was discarded. The samples were centrifuged again at full speed for 3 minutes. The column was placed in labeled 1.5ml tube. 200µl of the preheated (70°C) Buffer AE was

added and the mixtures in the tube were incubated at 70°C for 5 minutes and centrifuged at 6000g for 1 minute. The filtrate solutions were placed back into the spin column. 200 µl of Buffer AE was added again, and the reaction mixture tube was incubated at 70°C for 5 minutes and then centrifuged at 6000g for 1 minute. The spin column was then discarded. The pure DNA was then collected.

### **3.3.3 AGAROSE GEL PREPARATION FOR QUANTIFICATION OF DNA**

Agarose can come in either powder or tablet. The one used for this experiment was tablets. Each tablet was 0.5g. Two tablets were placed into 500ml cylinder and 100ml of 0.5×TBE buffer into it. This gave 1% (W/V) agarose gel. That is 1g of Agarose was dissolved in 100ml of 0.5 × TBE buffer. This is for DNA quantification. The cylinder was incubated by placing it in microwave for 3 minutes. The cylinder was cooled by placing it under running water. In the fumehood 5µl of ethidium bromide was added. The melted agarose gel was poured into a casting tray in which a plastic comb has been inserted. As it cools, the agarose solidifies to a gelatinous substance consisting of a dense network of cross linked molecules. The solidified gel slab was immersed in a chamber filled with buffer solution containing ions needed to conduct electricity. Removal of comb leaves behind a series of wells into which the DNA samples were loaded using pipette for electrophoresis. Before loading, the DNA was mixed with loading dye. The dye molecules do not interact with the DNA, but since they are negatively charged, they migrate independently towards the positive electrode. Bromophenol blue, commonly used dye migrates at a rate equivalent to a DNA fragment of approximately 300bp in a 1% gel. Thus the movement of the visible dye allows one to monitor the migration of the unseen DNA fragments. The current was applied through electrodes at both ends of the electrophoresis chamber. The negatively charged DNA fragments move from wells into the gel, through the pores in the matrix towards the positive pole. The range of electrical power supply is 50 to 150 V and 20 to 75mA. The current can be adjusted as desired, noting the lower the current the longer it takes to complete the process of electrophoresis ranging from 30 minutes to several hours.

### **3.3.4 PRIMERS USED FOR THE POLYMERASE CHAIN REACTION**

Literatures were searched for the primers used by authors that have worked on cereals stem borers. The following four pairs of forward and reverse primers were discovered and used in the present work:

CP1 (5'-GATGATGAAATTTTGGATC-3') [modified from Harry *et al.* (1998); Moyal and Le Ru (2013); Mehdi et al (2015); Ongamo (2008)]

TRs (5'-TCTATCTTATGTTTTCAAAAG-3') (Simon *et al.* 1994); Moyal and Le Ru (2013); Mehdi et al (2015)

CP1 (5'-GATGATGAAATTTTGGATC-3') (modified from Harry et al., 1998); Moyal and Le Ru (2013); Mehdi et al (2015); Ongamo et al (2008)

Tser (5'-TATTTCTTTATTATGTTTTCAAAAC-3') (Simon et al., 1994); Ongamo et al (2008)

16SAA (5'-ATGCTWCCTTTGCACRGTCAGATACYGCGGC-3') (Chai and Du (2012)

16SBB (5'-CTTATCGAYAAAAAAGWTTGCGACCTCRATGTTG-3'), (Chai and Du (2012)

LP01 (5'-TGATTAGCTCCACAAATTTCTGAACATTGACC-3'), (Chai and Du (2012)

LP02 (5'-WACACCAGTTCATATTTDAACCAGAATGATATT-3') (Chai and Du (2012)

### 3.3.5 POLYMERASE CHAIN REACTION (PCR)

Table 3: **PCR Cocktail mix**

The DNA was subjected to the following cocktail mix and condition for the PCR

#### Reaction 1

10× PCR buffer	1.0
25mM MgCl <sub>2</sub>	1.0
5pMol forward primer	0.5
5pMol reverse primer	0.5
DMSO	1.0
2.5Mm DNTPs	0.8
Taq 5u/ul	0.1
10ng/μl DNA	2.0
Sterile H <sub>2</sub> O	3.1
	10μL

A reaction mixture of 10μl was constituted as stated above. The samples were first optimized because more than one pair of primers were used. Amplification of the DNA extract was done using PCR in a programmable thermo cycler or PCR machine. The reaction mixture was subjected to touch down PCR reaction because more than one pair of primers was used. Taq polymerase was first activated by heating the reaction mixture at 94°C for 5 minutes. This was followed by initial denaturation at 94°C for 30 seconds, annealing at 65°C for 30 seconds and extension at 72°C for 30 seconds. This was followed by nine cycles of another denaturation at 94°C for 30 seconds, annealing at 65°C for 30 seconds and extension at 72°C



for 30 seconds. This was followed by another denaturation at 93°C for 15 seconds, annealing at 55°C for 20 seconds and extension at 72°C for 30 seconds followed by thirty four cycles of another denaturation at 93°C for 15 seconds and annealing at 55°C for 20 seconds and extension at 72°C for 30 seconds. This was followed by final extension at 72°C for 5minutes before it was left at 10°C until PCR products was needed.

**TABLE 4**

<b>PCR CONDITION</b>	<b>TIME</b>
1. 94°C	5min
2. 94°C	30sec
3. 65°C	30sec
4. 72°C	30sec
5. Go to 2 nine times	
6. 93°C	15sec
7. 55°C	20sec
8. 72°C	30sec
9. Go to 6 thirty four times	
10. 72°C	5min
11. 10°C	∞

### **3.3.6 PCR AGAROSE GEL ELECTROPHORESIS**

1.5% agarose gel was prepared for PCR agarose gel electrophoresis. 2µl amplicons of the PCR reaction was then loaded on 1.5% agarose gel. The loading dye was added to the amplicons. The ladder used was 1Kbplus from Thermo Scientific. This helps to quantify the loaded DNA by comparison with visible bands. The gel was later placed in Gel Documentation System (GDS) and UV light was put on to measure the gel.

### 3.3.7 PCR PRODUCT PURIFICATION

2 vol (20µl) of absolute ethanol was added to the PCR product (amplicons). The reaction mixtures were then incubated at room temperature for 15 minutes. The result was spun down at 10000rpm for 15 minutes. The supernatant was then decanted. This was followed by another round of spinning at 10000 rpm for 15 minutes. Another 2 volumes (40µl) of 70% ethanol was added. The supernatant was again decanted. The samples were air dried. Then 10 µl of ultra-pure water was then added. The amplicons were checked on 1.5 agarose gel.

**Table 5: SEQUENCING REACTION**

Big dye® terminator v1.1/3.1 sequencing buffer (5 ×) is supplied at 5× concentration.

Reagent	Concentration	Volume
Reaction premix	2.5×	1µl
Big dye sequencing buffer	5×	1µl
Primer 5pmol	-	1.0 µl
PCR(Template quantity 10-40ng)	-	2.5µl
Water	-	4.5µl
Final volume	1×	10µl

The tubes (1.8 ml or 2 ml) were placed in thermal cycler and set to the correct volume (10µl). Initial denaturation was 96°C for 1 minute followed by 25 cycles of denaturation at 96°C for 10 seconds, annealing at 50°C for 5 seconds and final extension at 60°C for 4 minutes. The reaction was then left at 4°C until ready for purification.

### **3.3.8 PURIFICATION OF SEQUENCING PRODUCT PRIOR TO ELECTROPHORESIS (PRECIPITATION OF 10 $\mu$ L SEQUENCING REACTION) (EDTA/ETHANOL PRECIPITATION)**

Frozen Hi.Di formamide was thawed on ice. After thawing, the tube was mixed on vortex for 5 seconds and centrifuged briefly. 96 well reaction plates with full plate cover were removed from PCR machine and centrifuged briefly for 1 minute without cooling. 2.5 $\mu$ l of 125mM EDTA was added to each well. 30 $\mu$ l of absolute ethanol was also added to each well. The plates were covered and vortexed for 5 seconds. The extension products were incubated at room temperature. The common errors such as precipitation time less than 15 minutes which will usually result in loss of very short extension product or precipitation longer than 24 hours which normally increase the precipitation of unincorporated dye terminator were completely avoided. The plates were centrifuged at 3000rpm for 45 minutes at 4°C. The plate covers were removed and the plates were inverted on paper towel. The plates were centrifuged at 900rpm for 1 minute without cooling. This was to remove supernatants completely to ensure that no unincorporated dye terminators remain in the sequencing product. 100 $\mu$ l of 70% ethanol was added and the plates covered. The samples were centrifuged at 3000rpm at 15 minutes for 4°C. The samples were then decanted by inverting on paper towel and centrifuged for 1 minute to remove supernatant. The plates were heated to 90°C for 1 minute in a heating block. The plates were then removed from the heating block. 10 $\mu$ l Hi.Di of formamide was added to each sample. The samples were covered and vortexed for 5 seconds. The samples were then centrifuged for 1000rpm for 1 minute without cooling. The plate was heated to 95°C for 2 minutes to denature the sample for at least 5 minutes. The plates were stored at this point for up to 1 week. Samples at this level were ready to run perfectly.

### **3.3.9 ANALYSING THE SEQUENCES USING GENETIC ANALYZER (ABI 3130 $\times$ L)**

The ABI 3130 $\times$ L Genetic analyzer makes use of capillary electrophoresis. Like gel electrophoresis DNA samples are separated according to size. Samples are run through capillaries in a special liquid matrix called polymer. Much less manual handling is required. Less reagents is required (lower volumes) .Thousands of samples can be run in a day. Different primers can be labeled with different dyes and may run in the same capillary. During sample preparation, the DNA fragments in a sample are chemically labeled with

fluorescent dyes. The dyes facilitate the detection and identification of the DNA. Typically each DNA molecule is labeled with one dye molecule but up to five dyes can be used to label the DNA sample. The operator creates a plate record and specifies the sample type and run module. The operator places the plates on the instrument and starts the run. The auto sampler automatically moves the sample plate into position to be sampled by the 16 capillaries. Molecules are electrokinetically injected into thin, fused silica capillaries that have been filled with polymer. Electrophoresis of all samples begins at the same time when a voltage is applied across all capillaries. The DNA fragments migrate towards the other end of the capillaries with the shorter fragments moving faster than longer fragments. As the fragments enter detection cell, they move through the path of a laser beam. The laser light causes the dye on the fragments to fluoresce. The fluorescence is then captured by the charge-coupled device (CCD) camera. The CCD camera converts the fluorescence information into electronic information, which is then transferred to the computer workstation for processing by the 3130xL data collection software.

#### **3.4.0 VIEWING AND ANALYSIS OF SEQUENCING DATA**

The sequences data was viewed and analyzed with Mega 7.0.14 and Geneious 9.1.3. The sequences were compared with sequences in NCBI. This was done by BLAST.

## CHAPTER FOUR

### 4.0 RESULTS

#### 4.1 Field Experiments

##### 4.1.1 EVALUATED MAIZE VARIETIES

The varieties were not significantly different for stem borer leaf feeding 6 WAP, leaf feeding score 6 weeks after planting, stem borer dead heart 6 weeks after planting, stem borer leaf feeding 8 weeks after planting, leaf feeding score 8 weeks after planting and stem borer dead heart 8 weeks after planting ( $P < 0.05$ ). Varieties were not significantly different for root lodging and stalk lodging ( $P > 0.05$ ) (Tables 6A & 6B).

Varieties were significantly different for plant count at germination, plant count at harvest and yield ( $P < 0.01$ ). The varieties were highly significantly different for days to 50% flowering, husk cover and days to 50% silking ( $P < 0.01$ ) and also significantly different for plant aspect ( $P < 0.05$ ). Varieties were highly significantly different for number of cobs ( $P < 0.01$ ), but not significantly different for anthesis-silking interval and 1000 grain weight. The blocks were significantly different for Anthesis-silking interval which shows fertility gradient (Tables 6C and 6D)

##### 4.1.2 TWENTY MAIZE VARIETIES EVALUATED FOR STEM BORER RESISTANCE ON NACGRAB FIELD IN 2010

Table 6 A: Stemborer damage attributes of twenty maize varieties evaluated in 2010.

SV	DF	SBLF4	LFS4	SBDH4	SBLF6	LF6	SBDH6	SBLF8	LFS8
Block	2	2.65	1.39	0.12	9.21	1.47*	191	5.54	0.49
Var	19	21.60	2.20	2.85	5.36	0.63	1.72	2.49	0.70
Error	38	12.58	1.66	3.38	4.80	0.46	1.21	2.56	0.45
Total	59	15.30	1.82	2.98	5.01	0.54	1.34	2.57	0.53
R <sup>2</sup>		0.48	0.42	0.31	0.42	0.49	0.46	0.39	0.47
CV		89.42	58.45	183.90	98.53	52.17	255.05	116.05	53.55
Means		3.97	2.21	1.00	2.22	1.29	0.43	1.38	1.26

Table 6B: Stemborer damage attributes of twenty maize varieties evaluated in 2010

SV	DF	SBDH8	RLOD	SLOD
Block	2	0.37	64.23**	8.21*
Var	19	0.22	18.73	3.20
Error	38	0.24	12.17	2.15
Total	59	0.23	15.40	2.59
R <sup>2</sup>		0.37	0.52	0.49
CV		314.92	154.47	88.69
Means		0.16	2.26	1.66

\*, \*\* Significant at probability level of 0.05 and 0.01 respectively;

SBLF4= Stem borer leaf feeding 4 weeks after planting; LFS4= Leaf feeding Score 4 weeks after planting; SBDH4= stem borer dead heart 4 weeks after planting; SBLF6= Stem borer leaf feeding at 6 weeks after planting; LF6= Leaf feeding score 6 weeks after planting; SBDH6 =stem borer dead heart 6 weeks after planting; LFS8= leaf feeding score 8 weeks after planting; SBDH8= stem borer dead heart 8 weeks after planting. RLODG= Root lodging; SLODG= Stalk lodging;

Table 6C: Agronomic attributes of 20 varieties of Maize evaluated in 2010

SV	DF	PC	D50FL	D50SLK	ANSI	PLASP	HSC
BLOCK	2	30.0	446.54**	573.24**	28.46	0.02	0.00
GENOTYPE	19	450.61**	210.25**	168.80**	10.18	1.48**	0.50**
ERROR	38	91.70	4.93	18.94	6.16	0.68	0.00
TOTAL	59	205.19	98.06	100.78	8.01	0.90	0.02
R <sup>2</sup>		0.71	0.97	0.89	0.54	0.54	1.0
CV		49.53	3.78	7.50	77.76	24.73	0
MEANS		19.22	58.79	58.04	0.19	3.34	0.98

Table 6D: Agronomic attributes of 20 varieties of Maize evaluated in 2010

SV	DF	PCHARV	COBS	1000GRWT	YLD
BLOCK	2	6.82	28.61	117.57	0.46
VAR	19	78.72**	114.70**	386.19	1.49**
ERROR	38	20.36	34.01	289.72	0.35
TOTAL	59	37.41	54.85	271.79	0.72
R <sup>2</sup>		0.65	0.71	0.49	0.69
CV		67.52	88.63	68.40	0.03
MEANS		6.68	6.53	24.89	2133.92

\*, \*\* Significant at probability level of 0.05 and 0.01 respectively;

PC= Plant count at germination; D50FL= days to 50% flowering; D50\_SLK=Days to 50% silking ANSI= Anthesis –silk interval; PLASP= Plant aspect; HSC=Husk cover; PCHARV= Plant count at harvest; COBS= Number of cobs; 1000 GRWT= 1000 grain weight; YLD= Yield per hectare;

Table 7: Means of different agronomic and entomological data observed separated by Duncan Multiple Range Test of Twenty maize varieties evaluated at Ibadan in 2010

Variety	PC	PCHARV	YLD g/ha	SBLF4	LFS4	SBDH4
TZE COMP3C2DT	41.33a	14.33abc	2139.63a	8.00ab	2.67ab	1.00a
SAMMA3 15	39.67ab	15.00ab	2138.10ab	5.67abc	3.00a	1.33a
DTSR-WC1	32.67abc	16.33a	2137.02ab	6.67abc	3.33a	1.33a
99TZEE-YSTR	31.33abc	5.33efd	2133.37d	6.67abc	3.33a	2.00a
DTSRW-CO	28.33abcd	9.33abcde	2134.26bcd	5.00abc	2.33ab	0.00a
TZM 96	28.00abcd	11.00abcd	2134.78abc	3.33abc	1.67ab	1.00a
2000SYN-EE WSTR	26.33abcde	8.00abcdef	2133.52d	6.00abc	3.33a	2.67a
ACR94TZEECOMP5-W	23.67abcde	7.67abcdef	2133.52d	5.00abc	2.00ab	1.67a
TZM 100	22.67bcde	1.00ef	2133.40d	6.33abc	2.67ab	0.67a
AMATZBR-W	20.00bcde	8.33abcdef	2133.73cd	2.33abc	2.00ab	1.00a
SAMMAZ 17	18.33cdefg	5.67cdef	2133.40d	9.33a	3.67a	3.00a
TZM 108	17.00cdefge	8.67abcdef	2134.32bcd	1.67bc	1.33ab	0.33a
LNTP-Y-LNC5	12.33defg	7.33bcde	2134.14bcd	1.67bc	1.67ab	0.33a
TZM 104	10.67defge	1.00ef	2133.33d	1.33bc	1.33ab	0.00a
TZM 106	10.33defg	1.00ef	2133.33d	1.00bc	1.33ab	0.00a
SAMMAZ 18	9.00efg	8.00abcdef	2133.62d	4.00abc	2.67ab	2.67a
LNPTP x LNP-WC3	8.00efg	3.00def	2133.80cd	1.33bc	1.67ab	0.33a
SAMMAZ 19	3.00fg	1.33ef	2133.38d	1.00bc	1.33ab	0.00a
BR 9943 DMRSR	1.67fg	1.33ef	2133.47d	0.33c	1.33ab	0.00a
TZBR-ELD3-C5	0.00g	0.00f	0	0.00c	0.00b	0.00a
Means	19.22	6.68	2133.92	3.97	2.21	1.0
SE(0.05)	1.85	0.79	0.11	0.51	0.18	0.23

Means with the same letter(s) in each column were not significantly different at  $p < 0.05$ . PC=Plant count at germination; PCHARV=Plant count at harvest; YLD=Yield; SBLF4= Stem borer leaf feeding 4 weeks after planting; LFS4= Leaf feeding Score 4 weeks after planting; SBDH4= stem borer dead heart 4 weeks after planting;



TZE COMP3C2DT had the highest plant count at germination with mean of 41.33 plants while the resistant check BR 9943DMRSR had mean of 1.67 plants. DTSR-WC1 has highest mean of 16.33 plants count at harvest while BR 9943 DMRSR and SAMMAZ 19 mean of 1.33 plant and TZM 104 and TZM 106 both has mean of 1.0 plant at harvest. TZE COMP 3C2DT had mean yield of 2135.63Kg per hectare. SAMMAZ 17 has the highest mean of 9.33 stem borer leaf feeding at 4 weeks after planting while resistant check has mean of 0.33 stem borer leaf feeding at 4WAP. SAMMAZ 17 had the highest leaf feeding score of 3.67 at 4 WAP while the resistant check BR 9943 DMRSR and SAMMAZ 19 and TZM 104 and TZM 106 and TZM 108 had the mean of 1.33 leaf feeding score at 4 weeks after planting. There was no significant difference in stem borer dead heart 4 weeks after planting, though Sammaz 17 had the highest stem borer dead heart with 3.00 while resistant Check had no stem borer dead heart 4 weeks after planting (Table 7)

DTSRW-CO has highest stem borer leaf feeding 6 weeks after planting with mean of 4.67 while resistant check and TZM 106 have mean of 0.33 stem borer leaf feeding. 99TZE-YSTR has the highest leaf feeding score with mean of 2.67, while resistant check BR 9943 DMRSR , Sammaz 19 , LNPTPXNLP-WC3, TZM 104, TZM 106, TZM 108, AMATTZBR-W, TZM 100, ACR 94 TZEE COMP5-W and DTSR-WC1 has mean of leaf feeding score 1.00 making them better varieties for stem borer resistance . 99TZEE-YSTR has the highest mean stem borer dead heart 6 weeks after planting of 3.00 while resistant check BR 9943 DMRSR, DTSRW-C0, TZM 96, 2000-SYN EE WSTR, ACR94 TZEE COMP5-W, TZM 100, TZM 108, LNTP-Y-LNC5, TZM 104, SAMMAZ 18, LNP TPX LNP-WC3 and TZBR-ELD3-C5 have no stem borer dead heart 6 weeks after planting. There was no significant difference in stem borer leaf feeding 8 weeks after planting but DTSRW-C0 has the highest leaf feeding with mean of 3.00. Both DTSR-WC1 and DTSRW-CO have the highest mean leaf feeding score 8 weeks after planting with mean of 2.33 while the resistant check have mean of 1.00. There was no significant difference in stem borer dead heart 8 weeks after planting though SAMMAZ 15 has the highest number with mean of 1.00 plant while resistant check has no stem borer dead heart. The resistant check has the highest number of days to flowering with mean of 69.33 days. TZM has the best plant aspect with mean of 2.33 while the resistant check BR 9943 DMRSR , ACR 94 TZEE COMP 5- W and 99 TZEE- YSTR have means of 4.00 while Sammaz 19 and TZM 104 both have poor plant aspect with mean rating of 4.33 (Table 8 ).

Variety 99TZEE-YSTR has the highest root lodging incidence with mean of 10.33 while the resistant check BR 9943 DMRSR has no root lodging. Also LNTP-Y-LNC5 and LNP TPX LNP-WC3 both have mean root lodging of 0.33. TZM 100 and TZE COMP3 C2DT both have the highest stalk lodging with mean of 3.33 plants. All the varieties have excellent husk cover. Resistant check BR 9943 DMRSR has highest days to 50% silking with mean of 68 days while DTSRW-CO has the lowest days to silking with mean of 52.67 days. TZM 104 and LNTP TP x LNP-WC3 have the highest anthesis-silking interval with mean of 6.0 days, while resistant check BR 9943 DMRSR has mean of 5.0 days of anthesis-silk interval. TZE COMP 3 C2DT has the highest number of cobs with mean of 22.0 cobs, while resistant check BR 9943 DMRSR, SAMMAZ 17 and SAMMAZ 19 has mean of 1.0 cob (Table 9)

2000 SYN- EE WSTR has the highest one thousand grain weight with mean weight of 47.33g, while the resistant check has mean weight of 17.0g. TZE COMP3 C2DT has mean field weight per plot of 2.30 kg while resistant check BR 9943DMRSR, SAMMAZ 19, SAMMAZ 17, TZM 104 and TZM 106 has 0.20 kg, 0.08 kg, 0 and 0 kg field weight per plot respectively. TZE COMP3 C2 DT has the highest grain weight per plot of 1.93 kg while resistant check BR9943 DMRSR, as well as and also TZM 104 and TZM 106 have mean grain weight per plot of 0.14 kg and 0 kg and 0 kg respectively (Table 10).

Table 8: Means of different agronomic and entomological parameter observed and ranked by Duncan Multiple Range Test of Twenty maize varieties evaluated at Ibadan in 2010.

Variety	SBLF6	LF6	SBDH6	SBLF8	LFS8	SBDH8	D50-FL	PLASP
TZE COMP3C2DT	3.33ab	1.67ab	0.67b	1.33a	1.00abc	0.00a	54.33ef	3.00ab
SAMMA3 15	4.33ab	2.00ab	1.00ab	2.33a	2.00ab	1.00a	60.33cd	3.00ab
DTSR-WC1	2.67ab	1.00bc	1.67ab	3.00a	2.33a	0.00a	58.33de	3.00ab
99TZEE-YSTR	3.33ab	2.67a	3.00a	1.33a	1.00abc	0.00a	53.33f	4.00ab
DTSRW-CO	4.67a	1.67ab	0.00b	3.00a	2.33a	0.67a	58.33de	3.00ab
TZM 96	2.67ab	1.33abc	0.00b	1.00a	1.00abc	0.33a	63.67bc	2.33b
2000SYN-EE WSTR	2.67ab	1.67ab	0.00b	2.00a	1.33abc	0.33a	54.67ef	3.33ab
ACR94TZEECOMP5-W	3.00ab	1.00bc	0.00b	2.00a	1.00bc	0.00a	57.33def	4.00ab
TZM 100	1.67ab	1.00bc	0.00b	0.33a	0.67bc	0.33a	54.67ef	3.33ab
AMATTZBR-W	1.33ab	1.00bc	0.33b	2.00a	1.67ab	0.00a	64.33bc	3.67ab
SAMMAZ 17	4.33ab	1.33bc	1.00ab	2.00a	1.33abc	0.00a	63.33bc	3.00ab
TZM 108	1.33ab	1.00bc	0.00b	0.67a	1.00abc	0.00a	61.67cd	2.67ab
LNTP-Y-LNC5	1.33ab	1.33abc	0.00b	0.67a	1.00abc	0.00a	63.00bc	3.33ab
TZM 104	1.33ab	1.00bc	0.00b	0.67a	1.00abc	0.33a	56.00ef	4.33a
TZM 106	0.33ab	1.00bc	0.33b	1.33a	1.33abc	0.00a	53.33f	3.67ab
SAMMAZ 18	2.67ab	1.33abc	0.00b	2.33a	1.33abc	0.00a	61.33cd	3.67ab
LNPTPXLNP-WC3	1.00ab	1.00bc	0.00b	0.33a	1.00abc	0.00a	63.00bc	3.00ab
SAMMAZ 19	0.67ab	1.00bc	0.33b	0.33a	1.00abc	0.00a	66.33ab	4.33a
BR 9943 DMRSR	0.33ab	1.00bc	0.00b	0.00a	1.00abc	0.00a	69.33a	4.00ab
TZBR-ELD3-C5	0.00b	0.00c	0.00b	0.00a	0.00c	0.00a	0.00g	0.00c
Means	2.22	1.29	0.43	1.38	1.26	0.16	58.79	3.34
S.E(0.05)	0.30	0.10	0.16	0.21	0.10	0.06	1.32	0.13

Means with the same letter(s) in each column are not significantly different at  $p>0.05$ .

SBLF6=Stem borer leaf feeding 6 weeks after planting; LF6= Leaf feeding Score 6 weeks after planting;SBDH6= Stem borer dead heart 6 weeks after planting;SBLF8= Stem borer leaf feeding 8 weeks after planting;LFS8= Leaf feeding score 8 weeks after planting; SBDH8= stem borer dead heart 8 weeks after planting; D50FL= Days to 50% flowering; PLASP= plant aspect.

Table 9: Means of different agronomic and entomological parameter observed and ranked by Duncan Multiple Range Test of Twenty maize varieties evaluated at Ibadan in 2010.

Variety	RLODG	SLODG	HUSKCOVER	D50-SLK	ANSI	COBS
TZE COMP3C2DT	3.33b	3.33a	1.00a	55.33bc	1.00abc	22.00a
SAMMAZ 15	1.67b	3.00ab	1.00a	61.00abc	2.67abc	19.67ab
DTSR-WC1	1.00b	2.33ab	1.00a	55.33bc	3.67abc	14.33abc
99TZEE-YSTR	10.33a	2.33ab	1.00a	57.00bc	3.67abc	0.50d
DTSRW-CO	2.00b	1.33ab	1.00a	52.67c	5.67ab	7.33bcd
TZM 96	2.67b	2.33ab	1.00a	63.33ab	1.00abc	11.00abcd
2000SYN-EE WSTR	2.33b	3.00ab	1.00a	55.33bc	0.67bc	4.67cd
ACR94TZEECOMP5-W	1.00b	1.33ab	1.00a	58.67bc	4.00abc	3.00cd
TZM 100	6.33ab	3.33a	1.00a	55.33bc	0.67bc	1.33d
AMATTZBR-W	2.33b	1.00ab	1.00a	63.67ab	0.67bc	9.500bcd
SAMMAZ 17	1.33b	2.00ab	1.00	60.33abc	3.67abc	1.00d
TZM 108	4.67ab	1.00ab	1.00a	58.67bc	3.00abc	8.00cdb
LNTP-Y-LNC5	0.33b	1.00ab	1.00a	62.00ab	3.00abc	8.00cdb
TZM 104	1.33b	1.33ab	1.00a	62.00ab	6.00a	0.00d
TZM 106	1.00b	1.67ab	1.00a	55.33bc	2.00abc	0.00d
SAMMAZ 18	1.00b	0.67ab	1.00a	60.33abc	4.33abc	2.50cd
LNPTPXLNP-WC3	0.33b	0.33ab	1.00a	60.33abc	6.00a	3.33cd
SSAMMAZ 19	0.67b	0.67ab	1.00a	60.67abc	5.67ab	1.00d
BR 9943 DMRSR	0.00b	0.00b	1.00a	68.00a	5.00abc	1.00d
TZBR-ELD3-C5	0.00b	0.00b	0.00b	0.00d	0.00c	0.00d
Means	2.26	1.66	0.98	58.04	3.19	6.58
SE (0.05)	0.52	0.22	0.02	1.36	0.38	1.15

Means with common letter in each column were not significantly different at  $p>0.05$ .

RLODG= Root lodging; SLODG= Stem lodging; HUSKCOVER= Husk cover; d50SLK= Days to 50% silking; ANSI= Anthesis –Silking interval; COBS=Number of cobs at harvest.

Table 10: Means of different agronomic data observed and separated by Duncan Multiple Range Test of Twenty maize varieties evaluated at Ibadan in 2010.

Variety	1000-GRWT(g)	FWT(kg)	GRWT(kg)
TZE COMP3C2DT	31.33ab	2.30a	1.93a
SAMMAZ 15	34.67ab	1.77ab	1.19abc
DTSR-WC1	33.00ab	1.68ab	1.33ab
99TZEE-YSTR	18.50ab	0.10b	0.09bc
DTSRW-CO	17.7ab	0.93ab	0.56bc
TZM 96	35.67ab	1.45ab	1.10abc
2000SYN-EE WSTR	47.33a	0.19b	0.13bc
ACR94TZEECOMP5-W	26.50ab	0.28b	0.23bc
TZM 100	14.67a	0.10b	0.06bc
AMATTZBR-W	22.50ab	0.60ab	0.61bc
SAMMAZ 17	20.00ab	0.07b	0.31b
TZM 108	29.00ab	0.98ab	0.76abc
LNTP-Y-LNC5	40.50a	1.21ab	0.99abc
TZM 104	0.00a+	0.00b+	0.00c+
TZM 106	0.00a+	0.00b+	0.00c+
SAMMAZ 18	26.00ab	0.85ab	0.65bc
LNPTPXLNP-WC3	23.00ab	0.47b	0.40bc
SAMMAZ 19	31.50ab	0.08b	0.05bc
BR 9943 DMRSR	17.00ab	0.20b	0.14bc
TZBR-ELD3-C5	0.00a	0.00b	0.00c
Means	24.89	0.79	0.59
SE(0.05)	2.56	0.13	0.10

+ The genotypes have inbreeding depression

Means with the same letter(s) in each column are not significantly different at  $p>0.05$ .

1000GRWT= One thousand grain weight in grammes; FWTKG= Field weight per plot in Kg; Grain weight in Kg.

#### 4.1.3 EVALUATION OF TEN YELLOW MAIZE VARIETIES AT IBADAN IN 2012

**Table 11:** Mean squares, Coefficient of determination ( $R^2$ ), Coefficient of variation (CV) for stem borer damage traits measured on 10 yellow maize varieties in 2012

SV	DF	SBLF7	LFS7	SBDH7	SBLF8	SBDH8	NOHPP	LOT(CM)	RLOD	SLOD
Rep	3	38.57	0.30	0.00	2995.28**	0.00	4.87	47.71	0.13	3.33
Var	9	53.34	0.46	0.00	2010.49*	0.00	43.21	62.07*	0.27	8.94**
Error	27	19.38	0.15	0.00	695.62	0.00	14.18	29.12	0.21	1.11
Total	39	28.69	0.23	0.00	1327.46	0.00	20.16	38.16	0.21	3.09
$R^2$		0.53	0.55	0.00	0.64	0.00	0.513	0.47	0.33	0.75
CV		77.91	33.89		36.09		117.68	71.88	151.81	81.08
Mean		5.65	1.15	0.00	73.08	0.00	3.20	7.51	0.30	1.30
S.E	(0.05)	0.85	0.08	0.00	5.76	0.00	0.71	0.98	0.07	0.28

\*\* Significant at probability level of 0.05 and 0.01 respectively.

SBLF7= Stem borer leaf feeding 7 weeks after planting; LF7=Leaf feeding score 7weeks after planting; SBDH7= Stem borer dead heart 7 weeks after planting; SBLF8= Stem borer leaf feeding 8 weeks after planting; SBDH8= Stem borer dead heart 8 weeks after planting; NOHPP= Number of holes per plant; CM\_ LOT= Length of tunnels in cm; RLOD= Root lodging; SLOD= Stalk lodging;

Table 12: Mean squares, Coefficient of determination ( $R^2$ ), Coefficient of variation (CV) for agronomic traits measured on 10 yellow maize varieties in 2012

	DF	PC	DTO50FL	D50SLK	STG	PHT_CM	EHT_CM	HUSKCOV	PLASP	EASP	PCHARV	COBS	FWT_KG	GRWT_KG	YLD_KG
SV															
REP	3	3.37	0.60	15.57**	1.17**	2695.18**	357.73	0.03	0.00	0.13	78.91**	10.23	0.09	0.08**	2.52
VAR	9	54.54*	44.93**	0.83	0.83	1144.89	1021.83**	0.03	3.11	3.38**	4.89	143.47**	0.60**	0.14**	1.90**
ERROR	27	7.59	7.71	1.28	0.09	584.05	207.06	0.03	0.30	0.28	9.97	17.26	0.06	0.02	6.33
TOTAL	39	18.10	15.75	4.54	0.39	1114.37	406.67	0.03	0.92	0.98	25.49	45.85	0.19	0.05	8.97
$R^2$		0.71	0.66	0.81	0.84	0.64	0.65	0.31	0.78	0.80	0.73	0.74	0.77	0.77	0.15
CV		18.30	5.57	1.99	17.39	12.85	19.17	16.22	18.14	16.58	27.99	43.62	56.92	58.76	3.70
MEAN		15.05	49.80	56.65	1.75	188.08	75.06	0.98	3.00	3.20	11.28	9.53	0.43	0.22	2.15
S.E	(0.05)	0.67	0.63	0.34	0.10	5.28	3.19	0.03	0.15	0.16	0.80	1.07	0.07	0.04	14.97

\*\* Significant at probability level of 0.05 and 0.01 respectively.

DTO50\_FL= Days to 50% flowering; D50\_SL= Days to 50% silking; STG= Stay green; PHT\_CM= Plant height at harvest;

EHT\_CM=Ear height; Huskcov= Husk cover; PLASP= Plant aspect; EASP= ears aspect; PCHARV=Plant count at harvest; FWT\_KG=Field weight in Kg; GRWT\_Kg=Grain weight in kg; YLD=Yield/hectare.

Data were collected on 10 yellow maize varieties and 10 white varieties planted on the Field in NACGRAB in 2012 during the raining season.

Analysis of variance shows that Varieties were highly significantly different for parameters taken such as leaf feeding score 7 weeks after planting(7WAP)( $p>0.01$ ) and Varieties were significantly different for Stem borer leaf feeding 7 weeks after planting(7WAP) and stem borer leaf feeding 8 weeks after planting(8WAP) ( $p>0.05$ ) . However, varieties were not significantly different for stem borer dead heart 7 weeks after planting, Leaf feeding score 8 weeks after planting, stem borer dead heart 8 weeks after planting. Varieties were highly significant for stalk lodging, number of holes per plant ( $p>0.01$ ) but were significantly different for length of tunnels ( $p>0.05$ ) .However the varieties were not significantly different for root lodging. (Table 11).

Analysis of variance shows that Varieties were highly significantly different for parameters taken such as Plant count at germination, days to 50% flowering and stay green, ear height, number of cobs(ears), plant aspect, ear aspect, field weight, grain yield per plot and yield per hectare ( $p>0.01$ ) . However the varieties were not significantly different for husk cover, days to 50% silking, plant count at harvest and plant height at harvest. (Table 12)



Table 13: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of ten yellow maize varieties evaluated at Ibadan in 2012.

VAR	PCG	LFS7	SBDH7	SBLF7	DTO50FL	LFS8	SBDH8	SBLF8	D50SL	STG	PCH
TZM 13270	18.75a	1.00b	0.00a	7.50abc	49.50bc	2.00a	0.00a	112.00a	55.00c	2.00b	10.25bcd
TZM 223	18.75a	1.00b	0.00a	2.00c	49.25bc	2.00a	0.00a	79.50abcd	56.00bc	2.00b	15.25ab
TZM 132	18.00a	2.00a	0.00a	11.00ab	52.75ab	2.00a	0.00a	59.50bcde	56.75bc	1.50c	14.75ab
TZM 138	16.75ab	1.50ab	0.00a	7.50abc	52.50ab	2.00a	0.00a	79.25abcd	55.50bc	2.00b	16.00a
TZM 226	16.25ab	1.00b	0.00a	3.00c	47.00c	2.00a	0.00a	89.50abc	57.00b	1.50c	14.25abc
TZM 137	15.75ab	1.00b	0.00a	4.00bc	45.25c	2.00a	0.00a	97.00ab	55.75bc	2.50a	9.50cd
TZM 99	15.25abc	1.00b	0.00a	2.50c	49.50bc	2.00a	0.00a	92.75abc	56.25bc	1.00d	14.50ab
TZM 2230	12.50bc	1.00b	0.00a	12.00a	56.50a	2.00a	0.00a	46.25de	62.00a	1.50c	9.25d
TZM 20112	11.25c	1.00b	0.00a	2.50c	49.25bc	2.00a	0.00a	53.00cde	55.75bc	1.00d	6.50de
TZM 1327	7.25d	1.00b	0.00a	4.5bc	46.50c	2.00a	0.00a	22.00e	56.50bc	2.50a	2.50e
Mean	15.05	1.15	0.00	5.67	49.80	2.00	0.00	73.08	56.65	1.75	11.28
S.E	0.67	0.08	0.00	0.85	0.63	0.00	0.00	5.76	0.34	0.10	0.80

Means with the same letter(s) in each column are not significantly different at  $p>0.05$ . PCG= Plant count at germination; LFS7= Leaf feeding score at 7 weeks after planting; SBDH7= Stem borer dead heart 7 weeks after planting; DTOFL= Days to 50% flowering; LFS8= Leaf feeding score 8 weeks after planting; SBDH8= Stem borer dead heart 8 weeks after planting; SBLF8= Stem borer leaf feeding 8 weeks after planting; D50SL= Days to 50% silking; STG= Stay green; PCH= Plant count at harvest

TZM 13270 and TZM 223 had the highest plant count at germination with mean of 18.75; TZM 132 has the highest leaf feeding score 7 weeks after planting with mean of 2.0 while there was no significant difference in the rest varieties; No stem borer dead heart 7 weeks after planting; TZM 2230 has the highest stem borer leaf feeding with mean of 12.00a while TZM 223 had the lowest stem borer leaf feeding with 2.0. TZM 2230 had the highest number of days to flowering with mean of 56.5 days while TZM 137 had the lowest number of days to flowering with 45.5 days. There was no significant difference in the leaf feeding score 8 weeks after planting. Also there was no stem borer dead heart 8 weeks after planting. TZM 13270 had the highest stem borer leaf feeding after planting with mean of 112.0 while TZM 1327 had the lowest stem borer leaf feeding with 22.00. TZM 2230 has the highest number of days to silking with 62 days while TZM 13270 had the lowest number of days to silking with 55.0 days. TZM 99 and TZM 20112 have the best stay green rating with mean of 1.0 while TZM 1327 had the worst rating with a mean of 2.5. TZM 138 had the highest number of plant count at harvest with 16.0 while TZM 1327 had the least number of plant counts at harvest with mean of 2.5 plants (Table 13).

TZM 226 had the mean plant height of 228.63 to rank highest while TZM 132 had the lowest plant height with mean of 150.05. TZM 226 also had the highest ear height with mean of 96.83 while TZM 1327 had the lowest ear height with 39.68. There was no significant difference in root lodging. TZM 13270 has the highest stalk lodging with mean of 5.0 plants while TZM 226 and TZM 2230 had no root lodging. There is no significant difference in the huskcover of all the varieties. TZM 223 had the highest mean number of ears with 18.25 while TZM 1327 had the least mean number of 0.5 ears. TZM 1327 had the worst plant aspect with mean of 5.0 and TZM 99 had the best plant with mean of 1.50. TZM 99 has the best ear aspect with 1.50 while TZM 1327 had the worst ear aspect with mean rating of 5.0. TZM 223 had the highest number of holes per plant with mean of 8.25 while TZM 99 and TZM 2230 had the lowest number of holes per plant. TZM 20112 had the mean highest length of

tunnel with 13.24 cm while TZM 99 had the least length of tunnel with 1.775 cm. TZM 99 had the highest field weight with 1.25 kg while TZM 1327 had the least field weight with mean of 0.0025kg. TZM 99 had the highest mean grain weight with 0.575kg while TZM 1327 had the least grain weight with mean of 0.0 kg. TZM 99 had the highest yield per hectare with 2134.58kg while TZM 1327 had the lowest yield per hectare of 2133.33kg (Table 14)

Varieties were highly significantly different for parameters such as plant count at germination, stem borer leaf feeding 8 weeks after planting and root lodging ( $p < 0.01$ ). However, the varieties were not significantly different for stem borer leaf feeding 7 weeks after planting, leaf feeding score 7 weeks after planting, stem borer dead heart 7 weeks after planting, leaf feeding score 8 weeks after planting, stem borer dead heart 8 weeks after planting, plant count at harvest and stalk lodging, number of holes per plant and length of tunnels (Table 15).

Varieties were highly significantly different for parameters such as days to 50% flowering, days to 50% silking, and stay green ( $p > 0.01$ ), and also significantly different for moisture content and yield per hectare ( $p < 0.05$ ). Varieties were not significantly different for plant height at harvest, husk cover, number of ears, plant aspect, ear aspect, and field weight per plot and grain yield per plot showing uniformity among the varieties for these traits (Table 16).

Table14: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of ten yellow maize varieties evaluated at Ibadan in 2012.

VAR	CMPHT	CMEHT	RLOD	SLOD	HUSKC OVER	NOERS	PLASP	EASP	NOHPP	CMLOT	KGFWT	KGSY	YLD
TZM 13270	176.88bc	71.00bc	0.50a	5.00a	1.00a	4.50efg	3.00bc	3.00cd	4.00ab	8.345abc	0.0688ef	0.03e	2133.40c
TZM 223	211.88ab	91.00ab	0.50a	0.50bc	1.00a	18.25a	3.00bc	4.00b	8.25a	8.733abc	0.60bc	0.325b	2133.93b
TZM 132	150.05c	71.73bc	0.50a	1.00bc	1.00a	11.00bcde	3.00bc	3.00cd	0.50b	7.00abc	0.50bcd	0.30b	2133.83b
TZM 138	177.28bc	71.95bc	0.00a	0.50bc	1.00a	14.50abc	3.00bc	3.50bc	0.50b	4.682abc	0.4500bcde	0.20bc	2133.78b
TZM 226	228.63a	96.83a	0.50a	0.00c	1.00a	17.00ab	2.50c	3.00cd	7.50a	4.600abc	0.8000b	0.35b	2134.13a
TZM 137	182.70bc	75.80abc	0.50a	2.00b	1.00a	8.00cdef	3.00bc	3.00cd	4.75ab	3.183bc	0.4000cde	0.288b	2133.73b
TZM 99	210.48ab	78.68abc	0.00a	2.00b	1.00a	12.00abcd	1.50d	1.50e	0.00b	1.775c	1.2500a	0.575a	2134.58a
TZM 2230	203.98ab	88.43abc	0.00a	0.00c	0.75a	6.00defg	2.50c	2.50d	0.00b	11.425ab	0.1528def	0.051c	2133.48c
TZM20112	188.05bc	65.63c	0.00a	1.50bc	1.00a	3.50fg	3.50b	3.50cd	6.00ab	13.24a	0.1025ef	0.037c	2133.43c
TZM 1327	150.90c	39.68d	0.50a	0.50bc	1.00a	0.50g	5.00a	5.00a	0.50b	12.09ab	0.0025f	0.00c	0.00d
Mean	188.08	75.06	0.30	1.30	0.98	9.53	3.00	3.20	3.20	7.51	0.43	0.22	2150.46
S.E(0.05)	5.28	3.19	0.07	0.28	0.03	1.07	0.15	0.16	0.71	0.98	0.07	0.04	14.97

Means with the same letter(s) in each column are not significantly different at  $p < 0.05$ . CMPHT=Plant height in cm; CMEHT= Ear height in cm; RLOD= Root lodging; SLOD= Stalk lodging; HUSKCOVER= Husk cover; NOERS= Number of ears; PLASP= Plant aspect; EASP= Ear aspect; NOHPP= Number of ears per plant; CMLOT= Length of tunnels; KGFWT= Field weight in Kg; KGSY= Grain weight per plot; YLD= Yield per hectare.

#### 4.1.4 TEN WHITE MAIZE VARIETIES EVALUATED IN 2012

Table 15: Mean squares, Coefficient of Determination, Coefficient of Variation for stem borer traits measured on 10 white maize varieties in Ibadan in 2012

SV	DF	PC	SBLF7	LFS7	SBDH7	SBLF8	LFS8	SBDH8	NOHPP	LOT	RLOD	SLOD
REP	3	17.90	4.27	0.16	0.23	1142.03	0.03	0.63	9.00	24.17	0.10	0.23
GENOTYPE	9	47.77**	10.62	0.24	0.23	1977.71**	0.03	0.63	3.67	9.67	0.89**	8.23
ERROR	27	15.34	7.56	0.29	0.23	675.20	0.03	0.63	4.52	10.17	0.19	6.15
TOTAL	39	23.02	8.55	0.27	0.23	1011.69	0.03	0.63	4.67	11.13	0.35	6.17
R <sup>2</sup>		0.54	0.39	0.25	0.31	0.54	0.31	0.31	0.33	0.37	0.61	0.31
CV		34.21	70.51	47.70	632.46	38.18	15.43	632.46	106.28	121.19	175.5	84.79
MEAN		11.45	3.90	1.13	0.08	64.93	1.03	0.13	2.00	2.63	0.25	2.93
S.E	(0.05)	0.76	0.46	0.08	0.08	0.763	0.03	0.13	0.34	0.53	0.09	0.39

\*, \*\* Significant at probability level of 0.05 and 0.01 respectively; ns=not significant;

PC= Plant count at germination; SBLF7=Stem borer leaf feeding 7 weeks after planting; LFS7= Leaf feeding score 7 weeks after planting; SBDH7= stem borer dead heart 7 weeks after planting; LFS7= Leaf feeding score 7weeks after planting; SBDH8= Stem borer dead heart 8 weeks after planting; SBLF8= Stem borer leaf feeding 8 weeks after planting; NOHPP= Number of holes per plant; LOT\_CM=Length of tunnel in Cm; RLOD=Root lodging; SLOD=Stalk lodging;

Table 16: Mean squares, Coefficient of Determination, Coefficient of Variation for agronomic traits measured on 10 white maize varieties in Ibadan in 2012

SV	DF	PC	D50_FL	D50_SL	STG	PHT_CM	HUSKCOV	PLASP	EASP	PCHARV	COBS	FWT	GRWT	MCP	YLD
Rep	3	17.90	32.29*	58.43**	0.83	1330.75	0.20	0.76	0.20	23.30	73.09**	0.22	0.05	21.54	13465.23
Gentype	9	47.77**	92.19**	42.78**	1.97**	405.02	0.21	0.79	1.07	21.01	23.19	0.13	0.04	32.07*	20140.39*
Error	27	15.34	9.31	12.91	0.62	387.40	0.14	0.59	0.57	10.63	16.70	0.08	0.02	14.21	8938.12
Total	39	23.02	30.20	23.30	0.95	464.03	0.16	0.65	0.66	13.93	22.54	0.10	0.03	18.90	11871.50
R <sup>2</sup>		0.54	0.79	0.62	0.55	0.42	0.39	0.37	0.40	0.47	0.49	0.46	0.46	0.48	0.48
CV		34.21	5.51	5.53	39.92	10.28	31.67	47.34	39.75	39.05	58.60	72.25	75.60	22.43	4.53
Mean		11.45	59.28	64.93	2.03	191.52	1.20	1.63	1.90	8.35	6.98	0.39	0.19	16.81	2088.30
S.E	(0.05)	0.76	0.87	0.79	0.15	3.14	0.06	0.13	0.13	0.59	0.75	0.05	0.03	0.68	17.23

\*, \*\* Significant at probability level of 0.05 and 0.01 respectively; ns= not significant;

PC=Plant count at germination; D50\_FL= Days to 50% flowering; D\_SL= Days to 50% silking; STG=Stay green; PHT\_CM= Plant height at harvest in Cm; HUSKCOV=Husk cover; PLASP= Plant aspect; EASP= Ear aspect; PCHARV= Plant count at harvest; COBS=Number of cobs or ears; FWT= Field weight in Kg; GRWT= Grain yield in Kg; MCP=Moisture content of grain at harvest; YLD= yield per hectare in Kg.

Table 17: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of ten white maize varieties evaluated at Ibadan in 2012.

Variety	PC	LFS7	SBDH7	SBLF7	DTO50FI	LFS8	SBDH8	SBLF8	SILK	STG	PCH
TZM 1302	16.00a	1.00a	0.00a	5.50a	55.25cd	1.00a	0.00a	96.50a	62.00b	2.75ab	10.75a
TZM 150	14.50ab	1.75a	0.75a	4.50a	62.00b	1.25a	0.00a	70.75ab	66.25b	2.00abc	9.75a
TZM 1277	14.25ab	1.00a	0.00a	3.75a	54.25d	1.00a	0.00a	85.75ab	61.75b	3.00a	10.00a
TZM 156	12.50ab	1.25a	0.00a	6.00a	60.25bc	1.00a	1.25a	57.75abc	64.75b	2.50ab	8.00ab
TZM 1291	12.25ab	1.00a	0.00a	2.50a	59.50bc	1.00a	0.00a	71.50ab	66.25b	1.00c	11.25a
TZM 217	12.00ab	1.25a	0.00a	4.50a	57.00cd	1.00a	0.00a	67.50ab	62.25b	2.50ab	8.50ab
TZM 224	11.75ab	1.00a	0.00a	5.75a	57.75bcd	1.00a	0.00a	97.50a	64.00b	2.25abc	7.25ab
TZM 219	8.75bc	1.000a	0.00a	1.75a	58.75bcd	1.00a	0.00a	57.75abc	65.00b	1.75abc	8.00ab
TZM 227	8.00bc	1.00a	0.00a	1.50a	56.75cd	1.00a	0.00a	51.75bc	64.00b	1.50bc	6.50ab
TZM 112	4.50c	1.00a	0.00a	3.25a	71.25a	1.00a	0.00a	23.75c	73.00a	1.00c	3.50b
Mean	11.45	1.13	0.08	3.90	59.28	1.03	0.13	64.93	2.03	8.35	191.52
S.E(0.05)	0.76	0.08	0.08	0.46	0.87	0.03	0.13	0.763	0.15	0.59	3.41

Means with the same letter in each column are not significantly different at  $p < 0.05$ .

PCG=Plant count at germination; LFS7= Leaf feeding Score at 7 weeks after planting; SBDH7= stem borer dead heart 7 weeks after planting; SBLF7= Stem borer leaf feeding 7 weeks after planting; DTO50FL= Days to 50% flowering; LFS8= Leaf feeding Score 8 weeks after planting; SBDH8= Stem borer dead heart 8 weeks after planting; SBLF8= Stem borer leaf feeding 8 weeks after planting. SILK=Days to 50% silking; STG= Stay green; PCH= Plant count at harvest;

Table 18: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of ten white maize varieties evaluated at Ibadan in 2012.

Varieties	CMPTH	CMEHT	RLOD	SLOD	HUSKCOVER	NOERS	PLASP	EASP	NOHPP	CMLOT
TZM 1302	184.25a	81.95a	0.25b	4.00ab	1.00a	8.00ab	1.50ab	1.75ab	1.50a	1.65a
TZM 150	202.20a	87.85a	0.00b	3.25ab	1.25a	7.50ab	1.50ab	1.75ab	3.50a	2.815a
TZM 1277	177.45a	78.65a	0.00b	3.25ab	1.50a	6.00ab	1.50ab	2.25ab	3.25a	5.325a
TZM 156	182.85a	75.40a	0.25b	4.00ab	1.50a	4.75b	2.75a	3.00a	2.00a	1.975a
TZM 1291	194.70a	81.03a	0.00b	1.75ab	1.00a	12.50a	1.00b	1.00b	1.25a	1.925a
TZM 217	202.15a	94.65a	0.50b	2.75ab	1.00a	7.00ab	1.50ab	1.50b	2.25a	3.125a
TZM 224	194.70a	87.30a	0.00b	5.50a	1.25a	7.50ab	1.75ab	2.00ab	2.50a	3.725a
TZM 219	182.00a	72.95a	0.00b	2.75ab	1.50a	7.75ab	1.50ab	2.00ab	1.00a	0.85a
TZM 227	188.00a	94.75a	1.50a	1.50ab	1.00a	4.50b	1.75ab	2.00ab	2.25a	4.450a
TZM 112	206.85a	79.30a	0.00b	0.50b	1.00a	4.25b	1.50ab	1.75ab	0.50a	0.475a
Mean	191.52	83.38	0.25	2.93	1.20	6.98	1.63	1.90	2.00	2.63
S.E(0.05)	3.41	3.15	0.09	0.39	0.06	0.75	0.13	0.13	0.34	0.53

Means with the same letter(s) in each column were not significantly different at  $p < 0.05$ .

CMPTH= Plant height in cm; CMEHT= Ear height in cm; RLOD= Root lodging; SLOD= Stalk lodging; HUSKCOVER= Husk cover;

NOERS= Number of ears or cobs; PLASP= Plant aspect; PLASP= Plant aspect; EASP= Ear aspect; NOHPP= Number of holes per plant; CMLOT= Length of tunnel in cm.



Table 19: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of ten white maize varieties evaluated at Ibadan in 2012.

	FWT(KG)	GRWT(KG)	MCP	YLD(Kg/ha)
TZM 1302	0.3875ab	0.1888b	16.35a	2099.84b
TZM 150	0.50ab	0.2875ab	18.650a	2042.23b
TZM 1277	0.2325b	0.09b	16.625a	2092.78b
TZM 156	0.2875b	0.1263b	16.150a	2104.76b
TZM 1291	0.8250a	0.425a	19.125a	2030.63b
TZM 217	0.3750ab	0.20ab	17.85a	2062.18b
TZM 224	0.3625b	0.20ab	17.50a	2070.95b
TZM 219	0.45ab	0.1625b	18.40a	2048.45b
TZM 227	0.2875b	0.1625b	18.175a	2053.93b
TZM 112	0.20b	0.10b	19.275a	2277.22a
Mean	0.39	0.19	16.81	2088.30
S.E	0.05	0.03	0.68	17.23

Means with the same letter(s) in each column are not significantly different at  $p < 0.05$ .

FWT (KG) = Field weight in Kg; GRWT=Grain Yield; MCP=Moisture content; YLD =Yield per hectare.

TZM 1302 had the highest plant count at germination of 16.00 plants while TZM 112 had the lowest plant count of 4.5. There is no significant difference in leaf feeding score 7 weeks after planting. Also, there was no significant difference in stem borer dead heart 7 weeks after planting. Likewise there was no significant difference in stem borer leaf feeding 7 weeks after planting. TZM 112 had the highest number of days to flowering with mean of 71.25 days while TZM 1277 had the least number of days to flowering of 54.25 days. There was no significant difference in leaf feeding score 8 weeks after planting. Also, there was no significant difference in the stem borer dead heart 8 weeks after planting. TZM 224 has the highest stem borer leaf feeding with 97.50 while TZM 112 has the least stem borer leaf feeding with 23.75. TZM 112 had the highest number of days to silking with 73 days while TZM 1277 had the least number of days with mean of 61.75 days. TZM 1277 had the worst

stay green property with mean of 3.0 while TZM 112 and TZM 1291 had the best stay green attribute with mean of 1.00. TZM 1291 had the highest number of plant count at harvest with mean of 11.25 while TZM 1277 had lowest plant count at harvest with mean of 3.50. (Table 17)

There was no significant difference in the plant height although TZM 112 has the highest plant height with 206.85cm while TZM 1277 has the lowest plant height with 177.45cm. Also there is no significant difference in the ear height but TZM 227 has the highest ear height with 94.75cm while TZM 219 had the lowest ear height with 72.95cm. There was no significant difference in the root lodging, stalk lodging and husk cover although TZM 224 has the highest with mean of 5.50 while TZM 112 has the lowest stalk lodging with mean of 0.5. TZM 1291 has the highest number of ears with mean of 12.50 ears while TZM 112 had the lowest number of ears with mean of 4.25 ears. TZM 1291 had the best plant aspect with mean of 1.0 while TZM 156 had the worst plant aspect with mean of 2.75. TZM 1291 had the best ear aspect with mean of 1.0 while TZM 156 had the worst ear aspect with mean of 3.0. There was no significant difference in the number of holes per plant and length of tunnels among the varieties, however TZM 150 had the highest number of holes per plant with mean of 3.5 holes while TZM 112 had the lowest number of holes per plant with mean of 0.5. Likewise TZM 1227 had the highest length of tunnel with mean of 5.325 cm while TZM 112 had the lowest length of tunnel with mean of 0.475cm. (Table 18)

TZM 1291 had the highest field weight with 0.8250 Kg while TZM 112 had the least field weight with 0.20 Kg. TZM 1291 had the highest grain weight with 0.425 Kg while TZM 1277 had the least grain weight with 0.09 Kg, although there was no much difference in the grain weight among all the varieties. There was no significant difference in moisture content at harvest. (Table 19)

Varieties were also significantly different for stalk lodging ( $p > 0.05$ ). However the varieties were not significantly different for parameters such as stem borer leaf feeding 4 weeks after planting, stem borer leaf feeding 6 weeks after planting, for number of holes, and length of tunnels (Table 20).

Varieties were significant for parameters such as plant count at germination, days to 50% flowering, number of cobs ( $p > 0.05$ ) and highly significantly different for days to 50% silking,

and plant height ( $p>0.01$ ). Varieties were not significantly different for anthesis-silking interval, stay green and yield per hectare (Table 21).

#### 4.1.5 TEN WHITE MAIZE VARIETIES (PARENTS) EVALUATED IN 2014

Table 20: Mean squares, Coefficient of Determination, Coefficient of Variation for stem borer traits measured on 10 white maize varieties (parents of crosses) evaluated at Ibadan in 2014.

SV	DF	PC	SBLF4	SBLF6	NOHPP	LOT_CM	RLOD	SLOD
REP	2	0.64	0.21	0.21	13.13	2747.09*	3.76	46.91**
GENOTYPE	10	8.07*	1.13	0.63	18.43	300.54	6.81	22.62**
ERROR	20	3.14	2.08	1.01	10.28	611.26	17.32	7.61
TOTAL	32	4.52	1.67	0.84	13.00	1727.07	13.18	14.76
R <sup>2</sup>		0.57	0.22	0.25	0.51	0.41	0.18	0.68
CV (%)		11.74	61.79	59.28	32.90	31.04	116.40	42.14
MEAN		15.09	2.33	11.70	9.75	79.64	3.58	6.55

\*, \*\*=Significant at 0.05 and 0.01 respectively.

PC=plant count at germination; SBLF4=Stem borer leaf feeding 4 weeks after planting; SBLF6= Stem borer leaf feeding 6 weeks after planting; NOHPP=Number of holes per plant; LOT= Length of tunnels; RLOD= Root lodging; SLOD= stalk lodging;

**Table 21: Mean squares, Coefficient of Determination, Coefficient of Variation for agronomic traits measured on 10 white maize varieties (parents of crosses) evaluated at Ibadan in 2014.**

SV	DF	PC	D50_FL	D50_SLK	ANSI	STG	PHT_CM	EHT_CM	HUSKCOV	PLASP	COBS	YLD
Rep	2	0.64	34.74**	15.03	11.30	3.36	147.12*	654.95*	0.39	0.03	60.48**	253.60
Var	10	8.07*	11.28*	23.08**	2.22	3.48	1253.60**	165.37	0.19	0.34	15.69*	708.19
Error	20	3.14	4.26	5.53	1.77	1.63	370.52	115.83	0.36	0.63	5.92	1076.30
Total	32	4.52	8.36	11.61	2.51	2.32	715.59	165.00	0.31	0.50	12.38	909.85
R <sup>2</sup>		0.57	0.68	0.70	0.56	0.56	0.68	0.56	0.27	0.22	0.70	0.26
CV (%)		11.74	3.81	4.45	71.97	36.96	8.68	11.15	30.96	30.82	47.22	1.60
Mean		15.09	54.12	52.89	1.85	3.45	221.77	96.52	1.93	2.58	5.15	2045.75

\*, \*\* Significant at probability level of 0.05 and 0.01 respectively.

PC=plant count at germination; D50FL= Days to 50% flowering; D50\_SLK= days to 50% silking; ANSI= Anthesis silking interval; STGR=Stay green;PHT=Plant height; EHT=Ear height; HUSKCOV=Husk Cover;PLASP=Plant aspect; COBS= Number of cobs; YLD=Yield per hectare.

Table 22: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of ten white maize varieties evaluated at Ibadan in 2014.

Varieties	PC	SBLF4	SBLF6	D50FL	D50SILK	ANSI	EHT	PHT	PLASPE CT	RLOD GING
Obatanpa/TZL Comp3C3	17.00a	2.00a	2.67a	52.33bc	50.67c	1.67ab	102.27ab	222.13abc	3.00a	2.33a
SynLDFO/Obatanpa/TZL Comp3C3*2	16.67a	3.67a	1.67a	51.00c	50.00c	1.00ab	98.00ab	200.53cd	3.00a	4.67a
BR9943 DMRSR	16.00ab	2.00a	1.33a	56.00ab	51.33c	3.00a	88.20ab	231.87abc	2.33a	4.67a
Obatanpa/IWDC2Syn	16.00ab	2.67a	2.00a	54.33abc	51.33c	3.00a	88.20ab	218.40bcd	2.33a	2.33a
TZLcomp4C4	16.00ab	2.33a	1.00a	54.33abc	52.00c	2.33ab	106.73a	241.80ab	2.67a	2.67a
ACR06 TZLComp4C4	15.67ab	1.67a	1.67a	55.00abc	52.67c	3.00a	91.40ab	230.60abc	2.00a	2.00a
Aflatoxin Syn w4	15.00abc	1.33a	2.00a	53.00bc	50.67c	2.33ab	93.07ab	203.47cd	2.67a	3.33a
Aflatoxin Syn w5	14.00abc	2.33a	1.67a	53.00bc	51.33c	1.67ab	81.33b	183.67d	3.00a	7.00a
TZLComp3C3DTC2	14.00abc	2.67a	2.00a	53.00bc	52.00c	1.67ab	102.80a	218.07bcd	2.67a	3.67a
ACR06 TZL Comp3C4	13.00bc	2.33a	1.33a	58.00a	58.00a	0.00b	98.13ab	232.13abc	2.33a	2.33a
TZLComp4c3	12.00c	2.67a	1.33a	55.33ab	56.33ab	1.67ab	97.80ab	256.80a	2.33a	4.33a
Mean	15.0	2.33	1.70	54.12	52.88	1.85	96.52	221.77	2.58	3.58
S.E(0.05)	0.37	0.22	0.16	0.50	0.59	0.28	2.24	4.66	0.12	0.63

Means with common letter in each column were not significantly different at  $p>0.05$ .

PC=Plant count; SBLF4=Stem borer leaf feeding 4WAP; SBLF6= Stem borer leaf feeding 6WAP; D50FL=Days to 50% flowering; D50SLK= Days to 50% silking; ANSI= Anthesis- silk interval; EHT=Ear height; PHT=Plant height; PLASP=Plant aspect; RLOGD=Root lodging.

Obatanpa/TZLComp3C3 had the highest plant count with 17.00 while TZL Comp 4C3 had the lowest plant count with 14 plant stands. There was no significant difference in stem borer leaf feeding 4 weeks after planting (4WAP) and at 6 weeks after planting (6WAP). ACR06TZLComp3 C4 has the highest number of days to flowering with mean of 58 days to flowering while SynLDFO/Obatanpa/TZL Comp3C3\*2 had the lowest number of days to flowering with mean of 51 days. ACR06TZL Comp 3 C4 has the highest number of days to silking with mean of 58 days while SynLDFO/Obatanpa/TZL Comp3C3\*2 has the lowest number of days to silking with mean of 50 days. BR 9943DMRSR, Obatanpa/IWDC2 Syn and ACR06 TZLComp4C4 had the highest Anthesis-Silk interval with mean of 3.00 days while ACR06 TZL Comp3 C4 have lowest number days to Anthesis-Silking interval of 0.00 day. Obatanpa/TZL Comp3C3 has the highest ear height with 102.27 cm while Aflatoxin Syn W5 had the lowest ear height with 81.33cm. TZL Comp 4 C3 has the highest plant height with 256.80 cm while SynLDFO/Obatanpa/TZL Comp3C3\*2 has the lowest plant height with 200.53cm. There was no significant difference between the varieties in terms of Plant aspect and root lodging. (Table 22)

Syn LDFO/Obatanpa/TZL Comp3C3\*2 has the highest stalk lodging incidence with mean of 11.67 plants while TZL Comp4 C3 has the lowest stalk lodging with mean of 2.67 plants. TZL Comp4 C4 has the highest mean number of cobs with mean number of 10.33 cobs while SynLDFO/Obatanpa/TZL Comp 3C3\*2 and BR 9943 DMRSR both have mean number of 2.67 cobs. TZLComp 4C4 have the highest number of holes with 14.0 holes while BR 9943 DMRSR has the lowest number of holes with mean of 5.80 holes. SynLDFO/Obatanpa/TZL Comp 3 C3\*2 has the highest length of tunnel with 96.50 cm while ACR06TZLComp4C4 has the lowest length of tunnel with 67.00 cm, although there is no significant difference in the length of tunnel. (Table 23)

Varieties were highly significantly different for plant count at germination and for stalk lodging attribute ( $p > 0.01$ ). Varieties however were not significantly different for stem borer leaf feeding 4 weeks after planting, stem borer leaf feeding 6 weeks after planting, root lodging and number of holes and length of tunnels (Table 24).

Variety were highly significantly different for days to 50% flowering, days to 50% silking, for both grain weight and stay green ( $p > 0.01$ ). Varieties were also significantly different for number of cobs and field weight ( $p > 0.05$ ). Varieties were however not significantly different for for anthesis-silking interval , plant height, ear height, plant aspect, ear aspect and ear rot. (Table 25).



Table 23: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of ten white maize varieties evaluated at Ibadan in 2014.

Varieties		SLODGING	COBS	NOH	LOT	ST GREEN	EROT	FLDWT	GRWTG	EASP	FLDWT KG	YLD
Obatanpa/TZL Comp3C3		9.33ab	5.676	11.07ab	92.47a	2.33b	3.00ab	366.70ac	162.80c	3.00ab	0.37bc	2481.60bc
SynLDFO/Obatanpa/ Comp3C3*2	TZL	11.67a	2.67b	10.00ab	96.50a	2.00b	3.00ab	144.90c	84.30c	3.33a	0.14c	2276.60c
BR9943 DMRSR		8.33abc	2.67b	5.80b	71.30a	3.00ab	2.00c	337.50c	194.80bc	2.50abc	0.34c	2398.00c
Obatanpa/IWDC2Syn		5.00bcd	6.33ab	11.00b	76.37a	4.00ab	3.33a	816.70ab	408.70ab	2.33bc	0.82ab	2877.20ab
TZLcomp4C4		3.67cd	10.33a	14.00a	75.57a	5.00a	2.33bc	958.30a	521.90a	2.33bc	0.96a	3018.00a
ACRO6 TZLComp4C4		3.67cd	7.33ab	7.20b	67.00a	5.00a	3.67a	408.30bc	154.50c	3.00ab	0.41bc	2497.30bc
Aflatoxin Syn W4		5.67bcd	3.67b	9.87ab	92.47a	2.00b	3.00ab	250.00c	92.40c	3.33a	0.25c	2367.40c
Aflatoxin Syn W5		7.67abcd	4.33b	6.53b	74.63a	3.00ab	2.50bc	550.00abc	256.80bc	2.50abc	0.55c	2588.00bc
TZLComp3C3DTC2		8.00abcd	4.67b	11.60ab	80.77a	4.00ab	3.00ab	375.00bc	164.00c	3.00ab	0.38bc	2515.00bc
ACR06 TZL Comp3C4		3.67cd	3.33b	8.67ab	69.37a	3.67ab	2.33bc	308.30c	138.30c	2.67abc	0.31c	2448.40bc
TZLComp4c3		2.67d	5.67b	11.47ab	79.60a	4.00ab	2.00c	466.70bc	254.10bc	2.00c	0.47bc	2581.66c
Mean		6.55	5.15	9.75	79.64	3.45	2.77	0.45	220.86	2.74	453.54	25533.60
S.E(0.05)		0.67	0.61	0.63	4.43	0.27	0.11	0.06	31.27	0.10	57.64	0.03

Means with common letter in each column were not significantly different at  $p>0.05$ . SLODGING= Stalk lodging; COBS= number of cobs or ears; NOH= Number of holes; LOT= Length of tunnels; ST GREEN= stay green; EROT= Ear rot; FLDWT= Field weight; GRWTG= Grain weight in g; EASP= Ear aspect; FLDWT KG= Field weight in Kg; YLD= Yield in Kg

#### 4.1.6 TEN YELLOW MAIZE VARIETIES (PARENTS) EVALUATED AT IBADAN IN 2014 .

Table 24: Mean squares, Coefficient of Determination, Coefficient of Variation for stem borer traits measured on 10 yellow maize varieties (parents of crosses) evaluated at Ibadan in 2014.

SV	DF	PC	SBLF4	SBLF6	RLOD	SLOD	NOH	LOT(CM)
REP	2	9.48	0.39	0.58	3.27	16.76	23.57	777.61
VAR	10	45.27**	0.62	0.61	6.82	35.28**	13.70	709.02
ERROR	20	4.02	0.86	0.54	7.41	6.96	9.82	353.08
TOTAL	32	17.25	0.56	0.56	6.97	16.42	11.89	490.84
MEAN		13.42	1.48	0.58	3.18	6.88	8.69	68.55
R <sup>2</sup>		0.85	0.29	0.40	0.34	0.74	0.48	0.55
CV (%)		14.93	62.48	127.92	85.53	38.35	36.07	27.41

\*, \*\* Significant at probability level of 0.05 and 0.01 respectively; ns= not significant

PC=Plant count; SBLF4=Stem borer leaf feeding 4 weeks after planting; SBLF6= Stem borer leaf feeding 6 weeks after planting; RLOD= Root lodging; SLOD= Stalk lodging; NOH= Number of holes; LOT= length of tunnels in CM

**Table 25: Mean squares, Coefficient of Determination, Coefficient of Variation for agronomic traits measured on 10 yellow maize varieties (parents of crosses) evaluated at Ibadan in 2014.**

SV	DF	PC	D50FL	D50SLK	ANSI	PHT	EHT	STGREEN	PLASP	EASP	EROT	EHARV	FWT	GRWT
REP	2	9.48	6.30*	14.03*	0.21	324.01	92.11	0.39	1.48	0.94	3.55	5.30	2409.17	6729.71
VAR	10	45.27**	5.65**	13.62**	1.72	636.80	45.7	3.42**	1.33	2.14	1.54	7.74*	27808.09*	9926.48**
ERROR	20	4.02	1.44	3.00	2.21	385.17	36.42	0.99	0.95	1.64	1.91	2.70	9230.89	2349.59
TOTAL	32	17.25	3.06	7.01	1.93	459.98	42.83	1.72	1.10	1.75	1.90	4.44	14609.90	4991.13
MEANS		13.42	53.06	52.52	1.39	211.91	94.38	2.70	2.33	2.42	2.09	2.42	146.24	73.69
R <sup>2</sup>		0.85	0.71	0.73	0.29	0.48	0.47	0.64	0.46	0.42	0.37	0.62	0.61	0.71
CV(%)		14.93	2.26	3.30	106.70	9.26	6.39	36.97	41.81	52.82	66.13	67.82	65.70	65.78

\*, \*\* Significant at probability level of 0.05 and 0.01 respectively.

PC=Plant count at germination; D50FL =Days to 50% flowering; D50SLK= Days to 50% silking; ANSI= Anthesis-Silking interval; PHT= Plant height;EHT= Ear height; FWT= Field weight; STGREEN= Stay green; PLASP= Plant aspect; EASP= Ear aspect;EROT= Ear rot; EHARV= Number of ears or cobs at harvest; FWT= Field weight; GRWT=Grainweight..

Table 26: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of ten Provitamin A Yellow maize varieties evaluated at Ibadan in 2014.

Varieties	PC	SBLF4	SBLF6	D50-FL	D50-SIL	ANSI	EHT
1. PVA Syn 19F2	18.33a	1.67a	0.33a	53.00b	51.00b	2.00a	88.00a
2. ACR 91Suwan -1 SR-C1	17.67a	1.67a	0.33a	52.33b	50.67b	1.67a	98.00a
3. PVA Syn 17F2	16.33ab	1.33a	0.67a	52.33b	51.33b	1.00a	96.00a
4. PVA Syn 6F2	16.00ab	1.33a	0.67a	51.67b	51.00b	0.67a	95.33a
5. PVA Syn 9 F2	15.67ab	1.67a	0.00a	52.00b	52.67b	0.67a	95.33a
6. Aflatoxin Syn 2-y	13.67cb	2.67a	1.33a	52.67b	52.33b	0.33a	99.00a
7. PVA Syn 11 F2	15.67cb	1.00a	0.67a	53.33b	52.33b	1.00a	87.67a
8. BR 99 28 DMRSR	11.67cd	1.33a	1.33a	56.67a	58.33a	3.00a	99.00a
9. PVA Syn 3F2	10.67cd	1.33a	0.33a	53.33b	51.67b	1.67a	94.07a
10. PVA Syn 10 F2	8.67ed	1.33a	0.67a	52.33b	52.67b	1.67a	93.97a
11. PVA Syn 1 F2	6.00e	1.00a	0.00a	54.00b	53.67b	1.67a	91.87a
Mean	13.42	1.48	0.58	53.06	52.52	1.39	94.38
S.E (0.05)	0.72	0.15	0.13	0.30	0.46	0.24	1.14

Means with common letter in each column were not significantly different at  $p>0.05$ .

PC=Plant Count; SBLF4=Stem borer leaf feeding 4 weeks after planting; SBLF6= Stem borer leaf feeding 6 weeks after planting; D50\_FL= Days to 50% flowering; D50\_SIL= days to 50% silking; ANSI=Anthesis silk interval; EHT= ear height;

PVA Syn 19F2 had the highest number of plant count at germination with mean of 18.33 whereas PVA Syn 1F2 had the lowest plant count with mean of 6.00. There was no significant difference in the number of stem borer leaf feeding both at 4 weeks and 6 weeks respectively. Resistant check BR 9928 DMRSR had the highest number of days to flowering with mean of 56.67 days whereas there was no significant difference in the number of days to 50% flowering of other varieties. Likewise BR 9928 DMRSR had the highest number of days to silking with mean of 58.33 days. There was no significant difference in the number of anthesis-silk interval days for all the tested varieties including the check. Also, there was no significant difference in the ear height for all the varieties. (Table 26)

BR 9928 DMRSR had the highest plant height with mean of 249.33 cm while Aflatoxin Syn 2-Y had the highest plant height with mean of 196.60cm. BR 99228 DMRSR had the best plant aspect with mean of 1.00b. There was no significant difference in the root lodging but PVA Syn 17 F2 had the highest root lodging with mean of 5.33 plants while resistant check BR 9928 DMRSR had lowest root lodging with mean of 1.67 plants. PVA Syn 17 F2 had the highest stalk lodging with 11.67 plants while the resistant check BR 9928 DMRSR had the lowest stalk lodging with 1.33 plants. PVA Syn 19 F2 and PVA Syn 9 F2 both had the highest number of cobs while PVA Syn 3 F2 had mean of 0.33 numbers of cobs. PVA Syn 3 F2 and PVA Syn 1 F2 both had the best ear rot rating with mean of 1.00.(Table 27)

PVA Syn 9F2 had the highest field weight with 366.67g while PVA Syn 3 F2 had the lowest field weight of 33.33g yield. Also PVA Syn 9F2 had the highest grain weight per plot of 195.24g while PVA Syn 3 F2 had the lowest grain weight with 5.78g. PVA Syn 6 F2 had the highest number of holes with mean of 12.67 holes while PVA Syn 11 F2 had the lowest number of holes with mean of 6.17 holes. Aflatoxin Syn 2-Y had the highest length of tunnels with mean of 92.87 cm while the resistant check BR 9928 DMRSR had the least length of tunnel with mean of 37.43 cm. PVA 17 F2 had the best stay green with mean rating of 1.33 while resistant check BR 9928 DMRSR had the worst stay green rating with mean of 5.00. Both PVA Syn 3 F2 and PVA Syn 1 F2 had the the best ear aspect with mean of 1.00 although there was no significant difference in the ear aspect of all the remaining varieties. (Table 28).

Varieties were not significantly different for plant count at germination, stem borer leaf feeding 6 weeks after planting, leaf feeding score 6 weeks after planting, stem borer dead

heart 6 weeks after planting, stem borer leaf feeding 8 weeks after planting , leaf feeding score 8 weeks after planting and stem borer dead heart 8 weeks after planting (Table 29).

Varieties were significantly different for days to 50% flowering and number of cobs at harvest ( $p>0.05$ ), Varieties were however highly significantly different for parameters like grain weight and yield per hectare ( $p>0.01$ ). Varieties were not significant for other parameters such as days to 50% silking, anthesis-silking interval, stay green, ear height and plant height, plant aspect, ear aspect, prolificity, and moisture content.(Table 30).

Table 27: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of ten Yellow maize varieties evaluated at Ibadan in 2014.

Varieties	PHT	PLASPECT	RLODGING	SLODGING	COBS/PLOT	EROT
1. PVA Syn 19F2	198.67b	2.33ab	4.00a	8.67abc	4.67a	2.67a
2. ACR 91 Suwan -1 SR-C1	209.00b	2.67ab	2.33a	11.00ab	2.33abc	3.00a
3. PVA Syn 17F2	213.60ab	2.33ab	5.33a	11.67a	3.00abc	2.67a
4. PVA Syn 6F2	202.67b	2.33a	4.33a	9.33abc	4.33ab	2.00a
5. PVA Syn 9 F2	209.67b	2.33ab	2.00a	8.33abc	4.67a	2.67a
6. Aflatoxin Syn 2-Y	196.60b	2.67ab	3.00a	6.00bdc	2.33abc	2.67a
7. PVA Syn 11 F2	207.67b	2.00ab	5.00a	7.67abc	2.33abc	2.00a
8. BR 99 28 DMRSR	249.33a	1.00b	1.67a	1.33d	0.67c	1.33a
9. PVA Syn 3F2	220.53ab	2.67ab	2.33a	5.00cd	0.33c	1.00a
10. PVA Syn 10 F2	203.97b	3.67a	4.33a	5.00cd	0.67c	2.00a
11. PVA Syn 1 F2	219.33ab	1.67b	0.67a	1.67d	1.33bc	1.00a
Mean	211.91	2.33	3.18	6.88	2.42	2.09
S.E(0.05)	3.73	0.18	0.46	0.71	0.37	0.24

Means with common letter in each column were not significantly different at  $p>0.05$ . PHT=Plant height; PLASPECT= Plant Aspect; RLODGING= Root lodging; SLODGING= Stalk Lodging; COBS/PLOT= Number of Cobs per plot.

Table 28: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of ten Yellow maize varieties evaluated at Ibadan in 2014.

Varieties	FIELDWT-G	GRWT-G-	NOH	LOT-CM-	STGREEN	EASP
1. PVA Syn 19F2	196.97ab	129.300ab	8.80ab	62.93abc	3.00bcd	3.00a
2. ACR91SUWAN -1 SR-C1	200.00ab	43.71bc	7.60ab	85.80ab	2.00cd	3.33a
3. PVA Syn 17F2	150.00b	93.01bc	7.37ab	78.27ab	1.33d	3.00a
4. PVA Syn 6F2	181.67b	126.10ab	12.67a	80.20ab	2.33bcd	3.00a
5. PVA Syn 9 F2	366.67a	195.24a	9.87ab	55.13bc	3.33abc	3.00a
6. Aflatoxin Syn 2-Y	175.00b	40.87bc	7.50ab	92.87a	1.67cd	2.33a
7. PVASyn 11 F2	121.84b	81.18bc	6.17b	68.17abc	2.33bcd	3.00a
8. BR 99 28 DMRSR	91.67b	42.74bc	6.50ab	37.43c	5.00a	1.67a
9. PV Syn 3F2	33.33b	5.78	7.10ab	66.31abc	2.33bcd	1.00a
10. PVA Syn 10 F2	49.86b	23.48c	11.10ab	65.43abc	2.33bcd	2.33a
11. PVA Syn 1 F2	41.67b	29.20c	10.87ab	61.50abc	4.00ab	1.00a
Means	146.24	73.69	8.69	68.55	2.70	2.42
S.E(0.05)	21.04	12.30	0.60	3.86	0.23	0.23

Means with common letter in each column were not significantly different at  $p>0.05$ . FIELDWT-G= Field weight per plot in grammes; GRWT-G= Grain weight per plot in grammes; NOH=Number of holes; STGREEN= Stay green; EASP= Ear aspect



#### 4.1.7 F1 OF WHITE MAIZE HYBRID EVALUATED AT IBADAN IN 2014

**Table 29: Mean squares, Coefficient of Determination, Coefficient of Variation of stem borer traits measured on 10 F1 white maize hybrids evaluated at Ibadan in 2014.**

SV	DF	PC	SBLF6	LF6	SBDH6	SBLF8	LF8	SBDH8	RLOD	SLOD
REP	2	38.82*	0.94	0.12	0.03	4.03	0.39	0	4.58	26.39**
VAR	10	7.27	0.96	0.15	0.03	0.59	0.16	0	3.67	2.46
ERROR	20	6.22	1.17	0.09	0.03	1.20	0.19	0	4.51	3.73
TOTAL	32	8.59	1.09	0.11	0.03	1.18	0.19	0	4.25	4.75
MEAN		14.91	1.97	1.12	0.03	1.61	1.15	0	0.34	0.51
R <sup>2</sup>		0.55	0.33	0.5	0.38	0.37	0.38	0	280.30	93.69
CV(%)		16.73	54.98	26.44	574.46	68.13	38.24		0.76	2.06

\*, \*\* Significant at probability level of 0.05 and 0.01 respectively; ns= not significant

PC=Plant count; SBLF6= Stem borer leaf feeding; LF6= Leaf feeding score 6 weeks after planting; SBDH6=Stem borer dead heart 6 weeks after planting; stem borer leaf feeding 8 weeks after planting; LF8= leaf feeding score 8 weeks after planting; SBDH8=Stem borer dead; RLOD= Root lodging; SLOD= Stalk lodging.

Table 30: Mean squares, Coefficient of Determination, Coefficient of Variation for agronomic traits measured on F1 of White maize hybrid evaluated at Ibadan in 2014.

SV	D F	PC	D50FL	D50SLK	ANSI	STG	PHT	EHT	PLASP	EASP	COBS	PROLIFICITY	GRWT	MCT	YLD
REP	2	32.82*	8.12**	21.55**	10.18**	0.48	1119.52**	227.89	5.18**	0.58	45.48	1.18	0.55**	0.42	1.07*
VAR	10	7.27	1.81*	2.52	1.56	0.42	57.43	133.29	0.81	1.48	36.62*	0.66	0.22**	1.13	0.52**
ERROR	20	6.22	0.75	1.81	1.38	0.65	225.76	137.11	0.92	0.78	16.85	1.25	0.10	1.71	0.14
TOTAL	32	8.59	1.55	2.90	1.99	0.57	229.01	141.59	1.15	0.98	25.76	1.06	0.16	1.45	0.32
MEAN		14.91	59.12	61.73	2.64	2.48	247.36	100.15	0.50	2.12	0.57	1.00	1.17	0.26	0.72
R <sup>2</sup>		0.55	0.70	0.65	0.57	0.29	0.38	0.40	30.95	0.51	28.34	0.27	0.63	7.05	21.98
CV(%)		16.73	1.47	2.18	44.59	32.48	6.07	11.69	3.09	41.52	14.48	111.74	26.63	18.56	1.70

\*, \*\* Significant at probability level of 0.05 and 0.01 respectively; ns= not significant SBDH8=Stem borer dead heart at 8 weeks after planting; D50%FL= Days to 50% flowering; D50% SLK= Days to 50% silking;ANSI=anthesis silk interval; EHT= Ear height; PHT= Plant height.PLASP=Plant aspect; RLOD= Root lodging; SLOD= Stem lodging; NOCOBS= Number of cobs; Yield = Yield per hectare; MCT= moisture content at harvest.GRWT= grain weight; NOH=Number of holes; LOT=Length of tunnels; STY-GREEN= Stay green; Prolificity.

Obatanpa/ TZL Comp3 C3 has the highest plant convent at germination with mean of 17.0 plants while the resistant check BR 9943 DMRSR has mean of 11.0 plants. Aflatoxin Syn W4 and ACR06 TZL Comp4 C4 both had the highest stem borer leaf feeding 6 weeks after planting with mean of 2.67, while the resistant check BR 9943 DMRSR and Syn LDFO/Obatanpa/ TZL Comp 3 C3\*2 and ACR06 TZL Comp3 C4 and Obatanpa/IWD C2 Syn had mean stem borer leaf feeding of 1.33plants. Obatanpa/TZL Comp 3 C3 has the highest leaf feeding score of 1.67 while resistant check BR 9943 DMRSR and Syn LDFO/Obatanpa/ TZL Comp 3 C3 \*2 had mean leaf feeding score of 1.33,but the rest varieties have mean leaf feeding score of 1.00. All the varieties had no stem borer dead heart 6 weeks after planting. There was no significant difference in the leaf feeding 8 weeks after planting,but Syn LDFO/Obatanpa/TZL Comp 3 C3\*2 had the highest SBLF8 with mean of 2.0 while Resistant Check BR 9943 DMRSR had mean of 1.33. There was no significant difference in leaf feeding score 8 weeks after planting but Aflatoxin Syn W4 had highest mean leaf feeding score of 1.67 while BR 9943 DMRSR has mean leaf feeding score of 1.00.(Table 31).

Table 31: Means of different entomological data observed and separated by Duncan Multiple Range Test of ten F1 white maize hybrid evaluated at Ibadan in 2014.

Variety *BR 9943DMRSR	PC	SBLF6	LF6	SBDH6	SBLF8	LF8
Obatanpa/TZL Comp3 C3	17.00a	2.00a	1.67a	0.00a	1.33a	1.33a
ACR 06 TZL Comp3 C4	15.68ab	1.33a	1.00b	0.00a	1.67a	1.00a
Aflatoxin Syn W4	15.67ab	2.67a	1.00b`	0.00a	2.67a	1.67a
TZL Comp3 C3 DTC2	15.67ab	2.00a	1.00b	0.33a	1.33a	1.00a
Obatanpa/IWD C2 Syn	15.67ab	1.33a	1.00b	0.00a	1.67a	1.00a
Aflatoxin Syn W5	15.33ab	2.00a	1.00b	0.00a	1.33a	1.00a
TZLComp4 C3	15.33ab	2.33a	1.00b	0.00a	1.67a	1.00a
TZLComp4 C4	14.67ab	2.67a	1.00b	0.00a	1.67a	1.00a
Syn LDFO/Obatanpa/TZL Comp3C3*2	14.33ab	1.33a	1.33ab	0.00a	2.00a	1.33a
ACR06TZL Comp4 C4	13.67ab	2.67a	1.00b	0.00a	1.00a	1.33a
BR 9943 DMRSR	11.00b	1.33a	1.33ab	0.00a	1.33a	1.00a
Mean	14.91	1.97	1.12	0.03	1.61	1.15
S.E(0.05)	0.51	0.18	0.06	0.19	0.08	0.22

Means with common letter in each column were not significantly different at  $p>0.05$ .

PC=Plant count at germination; SBLF6= Stem borer leaf feeding 6 weeks after planting; LF6= Leaf feeding score 6 weeks after planting;SBDH6= Stem borer dead heart 6 weeks after planting; SBLF8= Stem borer leaf feeding 8 weeks after planting; LF8= Leaf feeding score 8 weeks after planting;

There is no significant difference in the plant aspect, but TZL Comp4 C4, TZL Comp 4 C3, and TZL Comp 3 C3 DTC2 have the best plant aspect with mean rating of 2.33. The resistant check BR 9943DMRSR has the mean rating of 3.33 while Aflatoxin Syn W5 has the worst plant aspect with mean 4.00. There is no significant difference in the root lodging among the varieties but ACR06 TZL Comp 4 C4 has the worst root lodging with mean of 3.67 plants

while resistant check BR 9943 DMRSR has mean of 1.67 plants. ACR06 TZL Comp 4 C4 has the worst stalk lodging with mean of 4.0 plants while Aflatoxin Syn W4 and TZL Comp 4 C4 both have the lowest number of stalk lodging with mean of 1.00 while the resistant check BR 9943DMRSR has mean of 1.33 plants. ACR06 TZL Comp 3 C4 and Aflatoxin Syn W5 both have the highest number of cobs at harvest with mean of 17.67 cobs or ears. Syn LDFO/Obatanpa/TZL Comp3 C3\*2 has the lowest number of cobs or ears with mean of 1.33 cobs, while the resistant check has mean number of 5.67 cobs or ears. ACR06 TZL Comp 3 C4 has the highest yield per plot with mean of 2.15 kg per plot while the resistant check BR 9943 DMRSR has the lowest yield with 0.72kg per plot. There was no significant difference in the moisture content at harvest but Aflatoxin Syn W5 has mean of 19.90 while the resistant check BR 9943 DMRSR has mean of 18.70. (Table 32)

Aflatoxin Syn W5 had the highest grain weight per plot with mean weight of 1.38kg while the resistant check BR9943 DMRSR has the lowest grain yield per plot with mean of 0.43 kg per plot. Obatanpa/TZL Comp 3 C3 has the highest number of holes per plant with mean of 2.53 while TZL Comp 4 C3 has the lowest number of holes with mean of 1.13 holes, but the resistant check BR 9943 DMRSR has mean number of holes of 1.73. ACR06 TZL Comp 4 C4 has the highest length of tunnels with mean of 52.11cm, while the resistant check BR 9943 DMRSR had the lowest length of tunnels with mean of 21.64 cm, although differences among all the varieties are not statistically significant. The stay green characteristic is not significant among the varieties but ACR 06 TZL Comp 3 C4 has the best stay green characteristic with mean of 1.67 while resistant check BR 9943 DMRSR and Aflatoxin Syn W5 both have the worst stay green characteristic with mean of 3.00. There is no significant difference in the ear aspect of all the varieties but Obatanpa/IWD C2 Syn has the best ear aspect with mean rating of 1.33, while the resistant check BR 9943 DMRSR and Aflatoxin Syn W5 both have the worst ear aspect with mean of 3.33. There is no significant difference in the prolificity among the varieties. (Table 33)

Table 32: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of ten F1 white maize hybrids evaluated at Ibadan in 2014.

HYBRID	PLASPECT	R LODGIN	S-LODGING	NO-COBS	YIELD-WT	MCT-
Obatanpa/TZL Comp3 C3 X BR	3.00a	0.00a	2.33a	15.00a	1.48ab	18.07a
ACR 06 TZL Comp3 C4 X BR	3.00a	0.33a	4.00a	17.67a	2.15a	18.40a
Aflatoxin Syn W4 X BR	3.33a	0.00a	1.00a	16.33a	1.78ab	17.87a
TZL Comp3 C3 DTC2 X BR	2.33a	0.33a	1.33a	12.33ab	1.77ab	18.17a
Obatanpa/IWD C2 Syn X BR	2.67a	0.00a	2.33a	17.00a	1.88ab	19.03a
Aflatoxin Syn W5 X BR	4.00a	0.33a	1.67a	17.67a	2.03a	19.90a
TZLComp4 C3 X BR	3.33a	1.00a	2.67a	15.33a	1.90ab	18.47a
TZLComp4 C4 X BR	2.33a	0.00a	1.00a	17.33a	2.02a	18.70a
Syn LDF0/Obatanpa/TZL Comp3C3*2 X BR	3.00a	1.00a	2.33a	1.33ab	1.23bc	19.00a
ACR06TZL Comp4 C4 X BR	3.67a	3.67a	2.67a	13.67a	1.73ab	17.80a
BR 9943 DMRSR	3.33a	1.67a	1.33a	5.67b	0.72c	18.73a
<b>Mean</b>	3.09	0.76	2.06	14.48	1.7	18.56
S.E(0.05)	0.19	0.36	0.38	0.88	0.10	0.21

Means with common letter in each column were not significantly different at  $p>0.05$ .

PLASPECT=Plant aspect; RLODGING= Root lodging; S-LODGING=Stalk lodging; NO COBS= Number of cobs; YIELD-WT= Yield weight; MCT= Moisture content; BR= BR 9943 DMRSR

Table 33: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of ten F1 White maize hybrids evaluated at Ibadan in 2014.

HYBRID	GRWT	NOH	LOT	ST-GREN	E-ASPECT	PROLIFCT
Obatanpa/TZL Comp3 C3 X BR	1.02ab	2.53a	43.85a	2.67a	2.00a	0.67a
Obatanp/IWDC2Syn X BR	1.25a	2.33a	46.53a	2.33a	1.33a	1.67a
ACR 06 TZL Comp3 C4 X BR	1.35a	1.73a	32.94a	1.67a	2.33a	1.67a
ACR 06TZL Comp4C4 X BR	1.33a	2.00a	52.11a	2.67a	1.67a	0.67a
Aflatoxin Syn W4 X BR	1.23a	2.13a	42.27a	2.67a	1.67a	0.67a
Aflatoxin Syn W5 X BR	1.38a	1.53a	24.63a	3.00a	3.33a	0.67a
TZL Comp3 C3 DTC2 X BR	1.28a	1.40a	40.08a	2.33a	1.67a	1.00a
TZL Comp4 C3X BR	1.22a	1.13a	38.51a	2.33a	2.67a	1.00a
TZL Comp4 C4 X BR	1.32a	2.47a	49.21a	2.33a	1.67a	0.33a
SynLDFO/Obatanpa/TZL X BR	1.02ab	1.67a	25.23a	2.33a	1.67a	1.00a
BR 9943 DMRSR	0.43b	1.73a	21.64a	3.00a	3.33a	1.67
<b>Mean</b>	1.17	1.88	37.91	2.48	2.12	1.00
<b>S.E (0.05)</b>	0.07	0.15	2.63	0.13	0.17	0.18

Means with common letter in each column were not significantly different at  $p>0.05$ .

GRWT= Grain weight; NOH= Numbr of holes; Lot= Length of tunnels; ST-GREEN= Stay green; E-ASPECT=Ear aspect; PROLIFCT= Prolificity;BR=BR9943DMRSR

Table 34: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of ten F1 white maize hybrids evaluated at Ibadan in 2014.

<b>HYBRID</b>	<b>HUSK-COVER</b>	<b>YLD</b>
Obatanpa/TZL Comp3 C3 X BR	2.00a	2057.85a
Obatanp/IWDC2Syn X BR	1.67a	2033.99a
ACR 06 TZL Comp3 C4 X BR	2.00a	2050.15a
ACR 06TZL Comp4C4 X BR	1.67a	2064.79a
Aflatoxin Syn W4 X BR	2.00a	2063.17a
Aflatoxin Syn W5 X BR	2.00a	2012.39a
TZL Comp3 C3 DTC2 X BR	2.33a	2055.62a
TZL Comp4 C3 X BR	1.67a	2048.23a
TZL Comp4 C4 X BR	2.00a	2042.49a
SynLDFO/Obatanpa/TZL	1.67a	2034.17a
Comp 3 C3*2 X BR		
BR 9943 DMRSR	2.33a	2040.35a
Mean	1.94	2045.75
S.E(0.05)	0.10	5.25

Means with common letter in each column were not significantly different at  $p>0.05$ .

HUSK-COVER= Husk cover; YLD= Yield per hectare in Kg; BR= BR 9943 DMRSR



There was no difference in the husk cover among the varieties but the resistant check BR 9943 DMRSR has the worst husk cover with mean rating of 2.33 while Obatanpa/IWD C2 Syn, ACR06 TZL Comp4 C4, TZL Comp 4 C3 and Syn LD FO/Obatanpa/TZL Comp 3 C3\*2 have the best husk cover rating with mean rating of 1.67. ACR06 TZL Comp 4 C4 has the highest grain yield per hectare with 2064.79 while Aflatoxin Syn W5 has the worst grain yield per hectare of 2012.39 kg but resistant BR 9943 DMRSR check has grain yield of 2040.35 Kg per hectare. (Table 34).

Variety was highly significant for plant count ( $p>0.01$ ) and stem borer leaf feeding 6 weeks after planting ( $p>0.05$ ). However variety was not significantly different for leaf feeding score 6 weeks after planting, stem borer dead heart 6 weeks after planting, stem borer leaf feeding 8 weeks after planting, leaf feeding score 8 weeks after planting, stem borer dead heart 8 weeks after planting, root lodging, stalk lodging and number of holes (Table 35)

Variety was highly significant for plant count, stay green, plant height, ear height and plant aspect ( $p>0.01$ ). But variety was not significant for prolificity and husk cover. (Table 36)

Plant count at germination was not significant. However, PVA Syn 9F2 and PVA Syn 17 F2 both have the highest plant count at germination with mean of 18.00, while PVA Syn 11 F2 had the least plant count at germination with mean of 5.33 but resistant check BR 9928 DMRSR had mean of 10.00 plants. PVA Syn 3 F2 has the highest stem borer leaf feeding 6 weeks after planting with mean of 4.67, but the borer resistant check BR 9928 DMRSR and PVA Syn 11 F2 had mean of 1.0 stem borer leaf feeding 6 weeks after planting. There was no significant difference in the leaf feeding score 6 weeks after planting. PVA Syn 17 F2, PVA Syn 3 F2 and PVA Syn 19 F2 all had the highest leaf feeding score 6 weeks after planting with mean of 1.67, while BR 9928 DMRSR, PVA 9 F2, PVA Syn 1 F2, Aflatoxin Syn 2-Y, PVA Syn 10 F2, ACR 91 SUWAN-1 SR-C1, PVA Syn 11 F2 had mean leaf feeding score of 1.0 six (6) weeks after planting. There was no significant difference in the stem borer dead heart six weeks after planting, but PVA Syn 19 F2 and ACR91 SUWAN-1 SR-C1 both have mean of 0.33 while other varieties are comparable to stem borer resistant check BR 9928 DMRSR with no stem borer dead heart 6 weeks after planting. There is no significant difference in stem borer leaf feeding 8 weeks after planting. All the varieties have mean of 1.0 stem bore leaf feeding 8 weeks after planting. There was also no significant difference in leaf feeding score in all the varieties 8 weeks after planting. All the varieties had mean leaf feeding score of 1.0. Likewise all the varieties have no stem borer dead heart 8

weeks after planting. PVA 17 F2 has the highest ear height with mean of 115.67cm while PVA Syn 11 F2 has lowest ear height of 72.82 cm but the resistant check BR 9928 DMRSR has mean of 75.20 cm. PVA Syn 9 F2 has the highest plant height with mean of 260.60cm while PVA Syn 11F2 has the lowest plant height of 200.96cm but the resistant check BR 9928 DMRSR has the plant height of 243.0 cm. (Table 37).

#### 4.1.8 FI OF YELLOW MAIZE HYBRIDS EVALUATED AT IBADAN IN 2014 FOR STEM BORER TOLERANCE

Table 35: Mean squares, Coefficient of Determination, Coefficient of Variation of stem borer traits measured on ten F1 yellow maize hybrids evaluated at Ibadan in 2014.

SV	DF	PC	SBLF6	LF6	SBDH6	SBLF8	LFS8	SBDH8	RLOD	SLOD	NOH
REP	2	1.91	1.18	0.39	0.03	0.03	0.00	0.00	2.76	22.21	0.32
VARIETY	10	46.69**	4.70*	0.28	0.05	0.03	0.00	0.00	7.25	12.79	0.44
ERROR	20	7.8	1.82	0.40	0.06	0.03	0.00	0.00	4.39	6.38	1.05
TOTAL	32	19.59	2.68	0.36	0.06	0.03	0.00	0.00	5.18	9.37	0.82
MEAN		2.36	1.21	0.06	1.03	1.00	0.00	0.00	0.47	0.57	0.19
R <sup>2</sup>		0.58	0.32	0.32	0.38	0.00	0.00	0.00	150.33	49.91	50.06
CV(%)		57.00	51.78	416.23	16.90	0.00	0.00	0.00	1.39	5.06	2.05

\*, \*\*=Significant at probability level of 0.05 and 0.01 respectively;

PC= Plant Count; SBLF6= Stem borer leaf feeding 6 weeks after planting; LFS6= Leaf feeding score; SBDH6= Stem borer dead heart 6 weeks after planting; Stem borer feeding 8 weeks after planting; LFS8= Leaf feeding score 8weeks after planting;SBH8= Stem borer dead heart 8 weeks after planting; RLOD=root lodging; Stalk lodging; NOH=Number of holes.

**Table 36: Mean squares, Coefficient of Determination, Coefficient of Variation for agronomic traits measured on ten F1 yellow maize hybrid evaluated at Ibadan in 2014.**

SV	DF	PC	STG	PHT	EHT	PLASP	HCOV	PROL
REP	2	1.91	5.12**	2206.32**	123.91	3.27**	1.85*	1.48**
VAR	10	46.69**	2.07**	934.61**	459.84**	1.67**	0.36	4.52
ERROR	20	7.81	0.55	11.37	92.76	0.47	0.48	2.38
TOTAL	32	19.59	1.31	499.57	209.42	1.02	0.53	4.00
MEANS		2.36	0.74	0.86	96.10	0.71	0.43	0.63
R <sup>2</sup>		0.58	21.74	4.24	0.72	23.63	34.19	74.94
CV(%)		57.00	3.42	248.84	10.02	2.90	2.03	2.06

\*,\*\*=Significant at probability level of 0.05 and 0.01 respectively.

PC=Plant count at germination; STG=Stay green; PHT= Plant height; EHT=ear height; PLASPECT= Plant aspect; H-COV= husk cover; PROL=Prolificity

Table 37: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of ten F1 white maize hybrids evaluated at Ibadan in 2014.

Variety	PC	SBLF6	LF6	SBDH6	SBLF8	LFS8	SBDH8	EHT	PHT
PVASyn9F2	18.00a	1.33c	1.00a	0.00a	1.00a	1.00a	0.00a	97.40ab	260.60a
PVASyn17F2	18.00a	1.67c	1.67a	0.00a	1.00a	1.00a	0.00a	115.67a	258.73a
PVASyn1F2	17.67a	2.67abc	1.00a	0.00a	1.00a	1.00a	0.00a	101.00ab	259.07a
Aflatoxin Syn2-Y	17.33a	1.67c	1.00a	0.00a	1.00a	1.00a	0.00a	96.80b	256.73a
PVASyn3F2	16.67a	4.67a	1.67a	0.00a	1.33a	1.00a	0.00a	99.07ab	236.27b
PVASyn19F2	16.33a	2.67abc	1.67a	0.33a	1.00a	1.00a	0.00a	95.20b	257.73a
PVASyn10F2	16.00a	3.00abc	1.00a	0.00a	1.00a	1.00a	0.00a	99.67ab	251.67ab
PVASyn6F2	16.00a	4.33ab	1.33a	0.00a	1.00a	1.00a	0.00a	107.13ab	260.13a
ACR91Suwan-1 SR-C1	15.67a	2.00bc	1.00a	0.33a	1.00a	1.00a	0.00a	97.13ab	252.40ab
BR 9928 DMRSR	10.00b	1.00c	1.00a	0.00a	1.00a	1.00a	0.00a	75.20c	243.00ab
PVASyn11F2	5.33b	1.00c	1.00a	0.00a	1.00a	1.00a	0.00a	72.82c	200.96c
Mean	15.18	2.36	1.21	0.06	1.03	1.00	0.00	96.10	248.84
S.E (0.05)	0.77	0.28	0.10	0.04	0.03	0.00	0.00	2.52	3.8

Means with common letter in each column were not significantly different at  $p>0.05$ . PC= Plant count; SBLF6=Stem borer leaf feeding 6 weeks after planting; LF6= Leaf feeding score 6 weeks after planting; SBDH6= Stem borer dead heart 6 weeks after planting; SBLF8= Stem borer feeding 8 weeks after planting; LFS8= Leaf score 8 weeks after planting; SBDH8 = Stem borer dead heart 8 weeks after planting; EHT= Ear height; PHT= Plant height.

Table 38: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of ten F1 Yellow maize hybrids evaluated at Ibadan in 2014.

Variety	PLASPECT	R-LODGING	S-LODGING	NOH	ST-GREE	PROLIFIC	H-COV
PVA Syn 9F2	2.67bc	0.67b	5.33ab	1.33a	2.67cd	4.00ab	2.00a
PVA Syn 17F2	2.33bc	2.00ab	6.67a	2.27a	3.67abcd	2.67abc	2.33a
PVA Syn 1F2	2.00c	0.33b	6.67a	1.67a	3.33bcd	2.00abc	2.33a
Aflatoxin Syn2-y	3.33bc	5.33a	6.33a	2.27a	3.67abcd	1.33abc	2.33a
PVA Syn 3F2	3.67ab	2.33ab	6.67a	1.93a	4.33ab	1.67abc	2.00a
PVA Syn 19F2	3.67ab	2.33ab	2.67ab	1.73a	2.33d	2.67abc	2.00a
PVA Syn 10F2	3.00bc	0.33bc	5.33ab	2.27a	4.00abc	1.33abc	2.33a
PVA Syn 6F2	2.33bc	0.33bc	5.00ab	2.60a	3.33bcd	1.00bc	2.33a
ACR91 Suwan-1 SR-C1	2.67bc	1.67ab	7.33a	1.93a	3.00bcd	1.00bc	1.67a
BR 9928 DMRSR	2.67bc	0.33b	2.67ab	2.53a	2.33d	4.33a	1.67a
PVA Syn 11F2	4.67a	2.00ab	1.00b	2.00a	5.00a	0.67c	1.33a
Mean	2.91	1.39	5.06	2.05	3.42	2.06	2.03
S.E (0.05)	0.18	0.40	0.53	0.16	0.20	0.35	0.13

Means with common letter in each column were not significantly different at  $p>0.05$ .

PLASPECT= Plant aspect; R-LODGING= Root lodging; SLODGING= Stalk lodging; NOH= Number of holes; ST-GREEN= Stay green; PROLIFIC= Prolificity; H-COV= Husk cover.

PVA Syn 1 F2 has the best plant aspect with mean of 2.0 while PVA Syn11 F2 has the worst plant aspect with mean of 4.67, but resistant check BR 9928 DMRSR has plant aspect rating of 2.67. Aflatoxin Syn 2-Y has the highest root lodging with mean of 5.33 plants while the resistant BR 9928 DMRSR, PVA Syn F2, PVA Syn 6 F2 and PVA Syn 10 F2 have mean root lodging of 0.33 plants. ACR 91 Suwan-1 SR C-1 has highest stalk lodging with mean of 7.33 plants while PVA Syn 11 F2 has lowest number of stalk lodging with mean of 1.0 plant but the resistant check BR 9928 DMRSR has mean stalk lodging of 2.67 plants. There was no significant difference in the number of holes in all the varieties; however, PVA Syn 6 F2 has the highest number of holes with mean of 2.60 holes while PVA Syn 9 F2 has the lowest number of holes with mean of 1.67 holes but resistant check BR 9928 DMRSR has mean of 2.53 holes. BR 9928 DMRSR and PVA Syn 19 F2 have the best stay green characteristics with mean rating of 2.33 while PVA Syn 11 F2 has the worst stay green characteristics with mean rating of 5.0. The difference in husk cover was not significant but PVA Syn 11 F2 has best husk cover mean rating of 1.33 while PVA Syn 17 F2, PVA Syn 1 F2, Aflatoxin Syn 2-Y, PVA Syn 10F2, PVA Syn 6 F2 have the worst husk cover rating of 2.33, but the resistant check BR 9928 DMRSR and ACR 91 Suwan-1 SR-C1 have the best husk cover with mean rating of 1.67. (Table 38)

Varieties were highly significantly different for plant count at germination ( $p < 0.01$ ). Varieties were significantly different for root lodging ( $p > 0.05$ ). Location was highly significantly different for plant count, stem borer leaf feeding 4 weeks after planting, leaf feeding score 4 weeks after planting, stem borer dead heart 4 weeks after planting, stem borer leaf feeding 6 weeks after planting, leaf feeding score 6 weeks after planting, stem borer dead heart 6 weeks after planting, stem borer leaf feeding 8 weeks after planting, leaf feeding score 8 weeks after planting and stalk lodging ( $p < 0.01$ ). Location was also significantly different for root lodging ( $p < 0.05$ ). Varieties were not significantly different for stem borer leaf feeding 4 weeks after planting, leaf feeding score 4 weeks after planting, stem borer dead heart 4 weeks after planting, stem borer leaf feeding 6 weeks after planting, leaf feeding score 6 weeks after planting, stem borer dead heart 6 weeks after planting, stem borer leaf feeding 8 weeks after planting, leaf feeding score 8 weeks after planting and stalk lodging ( $p < 0.05$ ). Location was not significantly different for stem borer dead heart 8 weeks after planting ( $p < 0.05$ ). Varieties by locations interaction (VxL) was significantly different for stem borer dead heart 8 weeks after planting and stalk lodging ( $P < 0.05$ ). Varieties by locations interaction was not

significantly different for plant count, stem borer leaf feeding 4 weeks after planting, leaf feeding score 4 weeks after planting, stem borer dead heart 4 weeks after planting, stem borer leaf feeding 6 weeks after planting, leaf feeding score 6 weeks after planting, stem borer dead heart 6 weeks after planting, stem borer leaf feeding 8 weeks after planting, leaf feeding score 8 weeks after planting and root lodging ( $p < 0.05$ ) (Table 39)

Varieties were highly significant for plant count, ear height, ear aspect, plant count at harvest, ear count or number of cobs at harvest, ear rot and field weight ( $p < 0.01$ ). Varieties were also significant for days to 50% silking, anthesis-silking interval, and plant height ( $P < 0.05$ ). Varieties were not significantly different for days to 50% flowering, plant aspect and husk cover ( $p < 0.05$ ). Locations were however significantly different for plant count, days to 50% flowering, days to 50% silking, anthesis-silking interval, plant height, ear height, ear aspect, plant count at harvest, number of ears or cobs at harvest, husk cover, ear rot and field weight ( $p > 0.01$ ). Varieties by locations interaction were however not significantly different for plant count at germination, days to 50% flowering, days to 50% silking, anthesis-silking interval, plant height, ear height, plant aspect, ear aspect, plant count at harvest, number of ears or cobs at harvest, husk cover, ear rot and field weight ( $p > 0.05$ ) (Table 40)



#### 4.1.9 TEN WHITE MAIZE VARIETIES EVALUATED IN FIVE LOCATIONS IN 2015

Table 39: Mean squares, Coefficient of Determination, Coefficient of Variation of stem borer traits measured on 10 white maize varieties evaluated in 5 locations in 2015.

SV	DF	PC	SBLF4	LFS4	SBDH4	SBLF6	LFS6	SBDH6	SBLF8	LFS8	SBDH8	RLOD	SLOD
REP	2	37.61	124.91*	1.30	0.04	135.38**	3.08	0.19	43.17	2.34	0.27	5.16*	0.74
VAR(V)	10	162.39**	23.88	1.05	0.20	36.55	2.02	0.18	12.09	1.54	0.14	4.28*	1.70
LOC(L)	20	412.44**	2421.82**	160.89**	2.60**	1908.42**	117.24**	1.96**	830.71**	90.62**	0.14	19.80*	20.39**
VxL	40	9.66	23.17	1.08	0.23	32.25	1.55	0.11	15.84	2.43	0.18*	1.99	2.09*
ERROR		13.35	27.52	1.16	0.24	26.58	1.22	0.17	20.30	3.11	0.12	1.76	1.24
TOTAL		32.48	97.03	5.62	0.30	85.22	4.72	0.20	41.39	5.19	0.14	2.49	2.00
MEANS		16.27	4.91	2.36	0.18	5.31	2.76	0.13	5.13	3.46	0.12	2.60	14.85
R <sup>2</sup>		0.73	0.81	0.86	0.50	0.80	0.83	0.45	0.68	0.61	0.44	0.54	0.59
CV (%)		22.46	106.88	45.68	277.91	97.16	39.97	316	87.74	51.05	298.15	116.87	109.47

\*, \*\*= significant at 5% and 1% respectively.

PC= Plant count at germination; SBLF4= Stem borer leaf feeding 4 weeks after planting; LFS4= Leaf feeding score 4 weeks after planting; SBDH4= stem borer dead heart 4 weeks after planting. SBLF6= Stem borer leaf feeding 6 weeks after planting; LFS6= Leaf feeding score 6 weeks after planting; SBDH6=Stem borer dead heart 6 weeks after planting; SBLF8= Stem borer leaf feeding 8 weeks after planting; LFS8= Leaf feeding score 8 weeks after planting; SBDH8= Stem borer dead heart 8 weeks after planting;RLOD= Root lodging; SLOD= Stalk lodging.

Table 40: Mean squares, Coefficient of Determination, Coefficient of Variation for agronomic traits measured on 10 white maize varieties evaluated in 5 locations in 2015.

SV	DF	PC	D50FL	D50SLK	ANSI	PHT	EHT	PLASP	EASP	PHARV	EHARV	HCOV	EROT	FWT
REP	2	37.61	5.94	1.79	0.74	513.74	64.10	36.22	1.48**	43.37*	88.91**	2.69	0.45	0.22
VAR(V)	10	162.39**	14.69	12.16*	2.62*	854.69*	625.43**	30.53	1.37**	126.08**	91.70**	1.32	2.18**	1.57**
LOC(L)	20	412.44**	472.80**	322.59**	7.75**	20389.83**	6187.07**	43.99	8.70**	487.27**	217.91**	6.14**	2.21**	10.03**
VXL	40	9.66	4.34	4.76	1.13	504.88	109.75	30.07	0.22	10.35	12.35	1.29	0.42	0.19
ERROR		13.35	8.85	6.40	1.31	460.28	244.13	32.33	0.30	11.28	14.12	1.24	0.57	0.25
TOTAL		32.48	19.83	14.28	1.50	1017.98	382.20	31.98	0.57	31.04	24.71	1.44	0.67	0.56
MEANS		16.27	57.75	59.32	1.67	168.71	80.41	1.02	3.00	3.00	2.45	13.24	2.45	1.57
R <sup>2</sup>		0.73	0.71	0.71	0.43	0.71	0.58	0.34	0.65	0.76	0.63	0.44	0.45	0.71
CV (%)		22.46	5.15	4.26	68.50	12.72	19.43	163.70	18.46	22.62	28.38	42.74	30.70	31.91

\*, \*\*= significant at probability level of 0.05 and 0.01 respectively; D50-FL= Days to flowering; D50-SILK= Days to 50% silking; ANSI= Anthesis-silking interval; PHT= Plant height; EHT= Ear height; PLASPECT=Plant aspect; EASP= Ear aspect; PHARV= Plant count at harvest; EHARV= Ear count at harvest (number of cobs); HCOV=Husk cover; EROT= Ear rot; FWT= Field weight.

#### 4.2.0 TEN YELLOW MAIZE PARENTS VARIETIES EVALUATED IN 5 LOCATIONS IN 2015

Table 41: Mean squares, Coefficient of Determination, Coefficient of Variation of stem borer traits measured on 10 yellow maize varieties evaluated in 5 locations in 2015.

SV	DF	PC	SBLF4	LF4	SBDH4	SBLF6	LFS6	SBDH6	SBLF8	LFS8	SBDH8	RLOD	SLOD	NOH	LOT
REP	2	7.04	43.12	3.86	0.12	36.76	7.01*	0.26	64.94**	4.34	0.40	15.73**	0.24	71.91	529.99
VAR(V)	10	140.20**	12.85	1.87	0.08	10.50	1.11	0.38	5.57	1.12	0.40	3.24	2.15*	37.80	1345.24
LOC(L)	4	475.78**	964.28**	106.36**	0.80**	917.28**	87.39**	1.27**	219.09**	18.02**	0.56*	32.47**	31.94**	1180.05**	32655.84**
VXL	40	6.68	20.60	1.49	0.14	19.04	1.62	0.37	9.98	1.76	0.13	2.49	2.17**	40.34	777.72
ERROR		6.92	21.92	1.69	0.15	19.21	1.59	0.29	11.73	2.40	0.22	3.0	1.04	44.28	745.79
TOTAL		26.66	45.37	4.34	0.16	41.65	3.79	0.34	16.83	2.60	0.22	3.78	2.14	71.16	1651.32
MEANS		17.07	4.12	2.35	0.12	4.21	2.54	0.16	3.35	2.55	0.12	0.9	1.19	8.13	112.05
R <sup>2</sup>		0.83	0.68	0.74	0.38	0.70	0.72	0.44	0.54	0.40	0.33	0.4	0.68	0.59	0.71
CV(%)		15.40	113.58	53.39	314.76	104.17	49.71	342.11	102.30	60.65	387.66	181.55	85.60	81.86	24.37

\*,\*\*=significant at probability level of 0.05 and 0.01 respectively. ns= not significant.

PC= Plant count at germination; SBLF4= Stem borer leaf feeding 4 weeks after planting; LFS4= Leaf feeding score 4 weeks after planting; SBDH4= Stem borer dead heart 4 weeks after planting; SBLF6= Stem borer leaf feeding score 6 weeks after planting; LFS6= Leaf feeding score 6 weeks after planting; SBDH6= stem borer dead heart 6 weeks after planting; SBLF8=Stem borer leaf feeding 8 weeks after planting; LFS8= Leaf feeding Score 8 weeks after planting.SBDH8=Stem borer dead heart 8 weeks after planting;RLOD= Root lodging; Stalk lodging; Number of holes; LOT=Length of tunnels.

**Table 42: Mean squares, Coefficient of Determination, Coefficient of Variation for agronomic traits measured on 10 yellow maize evaluated in 5 locations in 2015.**

SV	DF	D50_FL	D50_SLK	ANSI	PHT	EHT	PLASP	EASP	PHARV	EHARV	HCOV	EROT	FWT	MSV	MCT	GRWT	YLD
REP	2	24.99**	11.38	6.49**	2041.50**	662.38**	0.58	1.25*	24.20*	70.53	0.02	7.02	0.65	2.70**	29.98*	0.13	18850.52*
VAR	10	13.58*	10.61	1.17	1232.65**	330.13**	0.31	0.71	122.93**	141.08**	1.74**	3.51	0.92**	0.17	19.77**	0.67**	12501.56**
LOC	4	337.12**	230.58**	9.39**	18880.72**	3277.30**	3.50**	5.59**	453.84**	535.13**	1.95*	3.51	9.53**	8.72**	1177.09**	6.85**	743097.22**
VxL(VL)	40	324.88	6.31	0.91	197.43	108.78	0.24	0.54	8.47	36.65	0.47*	3.66	0.24	0.38	8.19	0.17	5155.73
ERROR		6.82	6.27	1.03	280.78	115.47	0.22	0.38	7.82	46.78	0.31	3.53	0.24	0.38	7.29	0.17	4607.15
TOTAL		15.96	12.15	1.28	795.40	212.59	0.32	0.58	25.94	62.13	0.47	3.61	0.51	0.58	38.64	0.37	24393.76
MEANS		57.52	58.98	1.53	170.92	83.09	3.01	2.75	16.41	15.95	2.88	2.57	1.63	2.17	21.09	1.25	1982.04
R <sup>2</sup>		0.72	0.6	0.47	0.77	0.64	0.54	0.56	0.80	0.51	0.57	0.36	0.69	0.58	0.88	0.70	0.88
CV		4.54	4.2	66.15	9.80	12.93	15.69	22.57	17.04	42.89	19.35	73.22	30.28	28.26	12.80	32.97	3.42

\*, \*\*= Significant at probability level of 0.05 and 0.01 respectively. D50-FL= Days to 50% flowering; D50-SILK= days to 50% silking; ANSI= Anthesis- silking interval; PHT= Plant height; EHT= Ear height; PLASP= Plant aspect;EASP= Ear aspect;PHARV= Plant count at harvest;EARV=Ear count or number of cobs at harvest;HCOV= Husk cover; EROT= Ear rot;FWT=Field weighth;MSV=Maize steak virus; MCT= Moisture content at harvest; GRWT=Grain weighth; YLD=Yield.

Variety was highly significant for plant count ( $p<0.01$ ), and significantly different for stalk lodging ( $p<0.05$ ), but not significant for stem borer leaf feeding 4 weeks after planting, leaf feeding score 4 weeks after planting, stem borer dead heart 4 weeks after planting, stem borer leaf feeding 6 weeks after planting, leaf feeding score 6 weeks after planting, stem borer dead heart 6 weeks after planting, stem borer leaf feeding 8 weeks after planting, stem borer dead heart 8 weeks after planting and number of holes ( $p<0.05$ ). Location was highly significantly different for plant count at germination, stem borer leaf feeding 4 weeks after planting, leaf feeding score 4 weeks after planting, stem borer leaf feeding 6 weeks after planting, stem borer dead heart 6 weeks after planting, stem borer leaf feeding 8 weeks after planting, leaf feeding score 8 weeks after planting, root lodging, stalk lodging, number of holes and length of tunnels ( $p<0.01$ ) but significantly different for stem borer dead heart at 8 weeks after planting ( $p<0.05$ ). However, variety by location (VxL) interaction was not significantly different for plant count at germination, stem borer leaf feeding 4 weeks after planting, leaf feeding score 4 weeks after planting, stem borer leaf feeding 6 weeks after planting, stem borer dead heart 6 weeks after planting, stem borer leaf feeding 8 weeks after planting, leaf feeding score 8 weeks after planting, stem borer dead heart at 8 weeks after planting, root lodging, stalk lodging, number of holes and length of tunnels ( $p<0.05$ ) (Table 41).

Variety was highly significantly different for plant height, ear height, plant count at harvest, ear count or number of cobs at harvest, husk cover, field weight, moisture content of grains at harvest, grain weight and yield per hectare ( $p<0.01$ ) but significantly different for days to 50% flowering ( $p<0.05$ ). Variety was not significantly different at days to 50% silking, anthesis-silking interval, plant aspect, ear aspect, ear rot and maize streak virus ( $p<0.05$ ). Location was highly significantly different for days to 50% flowering, days to 50% silking, anthesis silking interval, plant height, ear height, plant aspect, ear aspect, plant count at harvest, ear count or number of cobs at harvest, field weight, maize streak virus, moisture content of grains at harvest, grain weight and yield per hectare ( $p<0.01$ ) but significantly different for husk cover ( $p<0.05$ ). Location was however not significantly different for ear rot. Variety by location (VxL) interaction was not significantly different for days to 50% flowering, days to 50% silking, anthesis silking interval, plant height, ear height, plant aspect, ear aspect, plant count at harvest, ear count or number of cobs at harvest, field weight, maize streak virus, moisture content of grains at harvest, grain weight and yield per hectare ( $p<0.05$ ) but significantly different for husk cover ( $p<0.05$ ) (Table 42).

#### **4.2.1 GENOTYPE BY ENVIRONMENT INTERACTION OF TWENTY MAIZE VARIETIES EVALUATED FOR STEM BORERS IN 5 LOCATIONS IN 2015**

From the results of twenty varieties of maize that were evaluated for resistance to stem borers in five locations in 2015. Apart from stem borer screening parameters like Root lodging(RLD), stalk lodging(SLD), number of holes on the stems(NOH), length of stem tunnels (LOT), Stem borer leaf feeding(SBLF), leaf feeding score(LFS) etc. , agronomic characters were also taken.

Using Duncan Multiple Range Test for their means separation, for white maize varieties used, Plant count at germination were similar at Mokwa(13.56) and Kotangora(11.97) .The plant count were similar also at Ibadan and Kabba, but different from Mokwa, Kotangora, Ibadan and Kabba in Lugbe, Abuja (FCT). Days to flowering were similar in Mokwa, and Kotangora and Lugbe but differed in Ibadan and Kabba. The days to silking were also similar in Mokwa, Kotangora and Lugbe(54.59;56.09;55.36) but different in Ibadan and Kabba(58.42;64.02) Anthesis- silk interval was similar at Kotangora and Lugbe(1.94;1.70) but different in Mokwa, Ibadan and Kabba.(2.25;1.57;0.94). Plant heights were similar at Mokwa and Kotangora (145.31; 136.68) but different from that of Lugbe, Ibadan and Kabba.(183.61;178.55;196.76). Plant heights were also similar at Lugbe and Ibadan (183.61; 178.55) but different from Kabba (196.76). Ear heights at Mokwa (69.09) and Kotangora (66.13) are similar but from different from Ibadan, Kabba and Lugbe, Abuja (FCT) (96.99; 86.24; 86.36). Ear heights are similar at Lugbe and Kabba (96.99; 86.36) but different from Ibadan (86.24). The ear heights at Ibadan are different from other locations. Plant aspect was similar in all locations (Table 43).

Table 43: Means of agronomic and stemborerl parameter observed on 10 white maize genotypes evaluated in five locations in 2015

LOCATION	PC	D50FL	D50SLK	ANSI	PHT	EHT	PLASP	HSC	PHARV	EHARV	EASP	EROT	FWT	RLD	SLD	NOH	LOT
Mokwa	13.56c	54.59c	56.84c	2.26a	145.31c	69.09c	3.33a	2.13b	10.87c	10.53b	3.71a	2.56ab	0.84d	1.03b	0.50bc	1.13c	164.34a
Kotangora	11.97c	56.09c	58.03c	1.94ab	136.68b	66.13c	3.08a	2.79a	10.87c	10.48b	3.00bc	2.44abc	1.16c	0.42b	0.00c	6.63b	88.97c
Lugbe	16.55b	55.36c	57.06c	1.70ab	183.61b	96.99a	2.64a	2.18b	15.70b	14.90a	2.33d	2.30bc	2.22a	2.45a	2.06a	7.00b	130.39b
Ibadan	19.52a	58.42b	60.00b	1.57b	178.55b	86.24b	2.82a	2.76a	18.18a	15.33a	2.76c	2.82a	1.76b	0.97b	0.94b	6.60b	124.36b
Kabba	19.39a	64.09a	64.52a	0.94c	196.76a	86.36a	5.47a	3.17a	18.33a	14.70a	3.17b	2.15c	1.83b	0.76b	1.52a	16.60a	39.45d
Means	16.27	57.75	59.32	1.67	168.71	80.41	3.47	2.60	14.85	13.24	2.99	2.45	1.57	1.14	1.02	7.65	109.55
S.E(0.05)	0.45	0.35	0.30	0.10	2.51	1.54	0.44	0.09	0.44	0.39	0.06	0.06	0.06	0.12	0.11	0.58	4.04

Means with common letter were not significantly different at  $p < 0.05$

PC-Plant count; D50FL-Days to 50% flowering; D50SLK- Days to 50% silking; ANSI-Anthesis -Silking Interval; PHT-Plant Height; EHT- Ear Height; PLASP- Plant aspect; HSC- Husk cover; PHARV-Plant count at harvest; EHARV –Number of ears or cobs at harvest; EASP –Ear aspect; EROT- Ear rot; FWT-Field weight ; RLD- Root lodging; SLD-Stalk lodging; NOH-Number of holes; LOT= Length of tunnels.

Husk cover was similar in Mokwa and Lugbe, Abuja (FCT) (2.13; 2.18) but different from Kotangora, Ibadan and Kabba (2.79; 2.76; 3.17). The husk cover is similar in Kotangora (2.79), Ibadan (2.76) and Kabba (3.17). Plant count at harvest was similar at Mokwa (10.78) and Kotangora (10.87) but different from Lugbe, FCT (Abuja) (15.70), Ibadan (18.18) and Kabba (18.33). Plant count at harvest was similar at Ibadan (18.18) and Kabba (18.33) but different from Lugbe, FCT (Abuja) (15.70). The number of ears at harvest were higher and similar at Lugbe, FCT (Abuja)(14.90), and Ibadan(15.33) and Kabba(14.70) but different from Mokwa(10.53) and Kotangora(10.48) .The number of ears were similar at mokwa(10.53) and Kotangora(10.48) but lower than what is obtained at Lugbe(14.90), Ibadan(15.33) and Kabba(14.70). The ear aspect was similar at Kotangora (3.00) and Ibadan (2.76) but different from that of Mokwa (3.71) and Lugbe, Abuja (FCT) (2.33). The ear aspect was similar at both Kotangora (3.00) and Kabba (3.71) but was different from that of Mokwa (3.71) and Lugbe, Abuja (FCT) (2.33). The ear rot rating at Ibadan (2.82) and Kabba (2.15) were different. The ear rot rating at Mokwa (2.56) and Kotangora (2.44) were similar. The ear rot rating at Mokwa (2.56), Kotangora (2.44) and Lugbe (2.30) were similar. The ear rot at Kotangora (2.44) Lugbe (2.30) and Kabba (2.15) were similar. The ear rot rating at Mokwa (2.56), Kotangora (2.44) and Ibadan (2.82) were similar. The field weight was similar at Ibadan (1.76) and Kabba (1.83) but different from Lugbe, Abuja (FCT) (2.22), Kotangora (1.16) and Mokwa (0.84). The field weight at Lugbe, Abuja (FCT) (2.22) was different from that of Kotangora (1.16) and Mokwa (0.84). The field weight at Kotangora (1.16) was also different from that of Mokwa (0.84) and Lugbe, Abuja (FCT) (2.22) (Table 43).

Root lodging and stalk lodging were part of parameters used in determining the level of susceptibility or otherwise resistance of cereals to stem borers. Root lodging ratings were similar in Mokwa, Kotangora, Ibadan and Kabba (1.03; 0.42; 0.97; 0.76) but different from Lugbe, Abuja (FCT)(2.45). Stalk lodging was similar Mokwa and Kotangora (0.5; 0.0) but different from Lugbe, Abuja (FCT), Ibadan and Kabba (2.06; 0.94; 1.52). Stalk lodging was similar in Lugbe, FCT (Abuja) and Kabba (2.06; 1.52) but different from Ibadan (0.94). (Table 43)

The number of holes which is also normally used to determine the level of infestation with stem borers varies with various locations where the maize varieties were screened. The number of holes was similar at Kotangora (6.63), Lugbe (Abuja, FCT) (7.00b) and Ibadan. However, the



number of holes from these locations was different from that of Mokwa (1.13) and Kabba (16.60). The number of holes at Mokwa (1.13) was different from that of Kabba ((16.60). Length of tunnels is another parameter used to determine level of resistance or susceptibility of cereals to stem borers. Length of tunnels at Ibadan (124.36) and Lugbe (Abuja, FCT) ((130.39) were similar (based on DMRT) but different from that of Kabba (39.45), Kotangora (88.97) and Mokwa (164.3). The length of tunnels at Mokwa (164.3) was different from that of Kotangora (88.97). Length of tunnels at Kotangora (88.97) was different from that of Kabba (39.45) (Table 43).

Aflatoxin Syn W4 is the most susceptible variety using the root lodging with average of 2.00 plants and stalk lodging with average 1.60 plants. Whereas TZLComp 4 C4 was the most resistant using root lodging with average of 0.17 plant and stalk lodging of 0.42 plant. (Table 44)

TZL Comp 4C4 had bad husk cover with 3.58 while TZL Comp 4C3 with 2.10. TZL Comp 4C3 had the highest number of ears with average of 16.13 cobs while TZL Comp 4C4 with 6.17 cobs. Aflatoxin Syn W4 was the worst in terms of Ear rot with 3.50 on the scale of 5, while the best in term of ear rot is Obatanpa/TZL Comp 3C3 with rating of 2.07. The variety with the best field weight was 2.18 kg while the variety with the least field weight was Aflatoxin Syn W4 with average of 1.11kg. This is probably due to its susceptibility to ear rot. TZL Comp 4C3 had the highest number of plant count at harvest while TZL Comp 4C4 had the lowest number of plant count at harvest with average of 6.17 plants per plot. Aflatoxin Syn W4 had the worst ear aspect with score of 3.67 on the scale of 5. Obatanpa/TZL Comp 3C3 had the best cob with ear aspect rating of 2.53 (Table 45).

Tables 44 Means of agronomic and stemborer traits of white maize genotypes evaluated across 5 locations in 2015

	PC	D50FL	D50SILK	ANSI	PHT	EHT	PLASPECT	RLODGING	SLODGING
TZL Comp4C3	19.27a	59.60a	60.87a	1.27bc	177.07ab	84.87abc	2.70b	1.27abc	0.87ab
Obatanpa/TZL Comp3C3	18.53ab	57.8ab	58.73abc	0.93c	168.53ab	84.60abc	2.93ab	1.67ab	0.73ab
BR9943 DMSR	18.07ab	56.40b	57.87c	1.27bc	183.73a	92.67a	2.77b	1.73ab	1.07ab
Obatanpa/IWDC2Syn	17.60ab	56.27b	58.60bc	2.33a	159.47b	80.87abc	3.17ab	1.27abc	0.80ab
ACR06 TZL Comp3C4	17.27abc	57.40ab	59.27abc	1.87abc	165.67b	85.87ab	4.67a	0.93abc	1.07ab
Aflatoxin Syn W5	17.20abc	58.40ab	59.87abc	1.60abc	163.40b	80.93abc	3.03ab	1.20abc	1.53a
SynLDFo/ Obatanpa/ TZL Comp3C3*2	16.93abc	56.80b	58.67bc	2.00ab	165.40b	75.33bc	3.17ab	0.47c	1.20ab
Aflatoxin Syn W4	16.20abc	58.00ab	58.80abc	1.47abc	162.67b	72.00c	3.67ab	2.00a	1.60a
ACR06 TZL Comp4c4	15.47bc	58.07ab	59.67abc	2.00ab	175.53ab	72.67bc	2.71b	0.80bc	0.80ab
TZL Comp3c3DTC2	14.27c	58.47ab	60.20ab	1.73abc	161.00b	75.13bc	3.33ab	0.80bc	1.00ab
TZL Comp4C4	6.08d	58.17ab	60.17ab	2.00ab	174.50ab	79.42bc	2.96ab	0.17c	0.42b
Mean	16.27	57.75	59.32	1.67	168.71	80.41	3.47	1.14	1.02
S.E (0.05)	0.45	0.35	0.30	0.10	2.51	1.54	0.44	0.12	0.11

Means with common letter in each column were not significantly different at  $p>0.05$ .

PC=Plant count; D50FL=Days to 50% flowering; Days to 50% silking; Anthesis-Silk interval; PHT=Plant height; EHT=Ear height; PLASPECT= Plant aspect; RLODGING= Root Lodging; SLODGING= Stalk lodging.

Table 45: Means of agronomic traits of white maize genotypes evaluated across 5 locations in the year 2015

Variety	HSC	EHARV	EROT	FWT	PHARV	EASPECT
TZL Comp4C3	2.10b	16.13a	2.10b	2.18a	18.07a	2.70bc
Obatanpa/TZL Comp3C3	2.80ab	15.00ab	2.07b	1.88ab	16.67ab	2.53c
BR9943DMSR	2.63b	14.33abc	2.27b	1.84abc	16.53ab	2.97bc
Obatanpa/IWDC2Syn	2.53b	14.00abc	2.43b	1.45cdef	16.00ab	3.17b
ACRO6 TZL Comp3C4	2.37b	14.40abc	2.43b	1.73bcd	15.67ab	2.77b
Aflatoxin Syn W5	2.53b	14.40abc	2.57b	1.39def	15.33abc	3.07b
SynLDFo/ Obatanpa/TZL Comp3C3*2	2.57b	13.80abc	2.43b	1.36def	15.73ab	3.07b
Aflatoxin Syn W4	2.93ab	11.93bc	3.50a	1.11f	14.60bc	3.67a
ACRO6 TZL Comp4C4	2.20b	12.40bc	2.37b	1.65bcde	14.00bc	2.83bc
TZL Comp3C3 DTC2	2.60b	11.67c	2.30b	1.36def	12.80c	3.03b
TZL Comp4C4	3.58a	6.17d	2.54b	1.25ef	6.17d	3.13b
Mean	2.60	13.24	2.45	1.57	14.85	2.99
S.E (0.05)	0.09	0.39	0.06	0.06	0.44	0.06

Means with common letter in each column were not significantly different at  $p>0.05$ .

HSC=Husk cover; EHARV= Ears harvested; EROT= Ear rot; FWT= Field weight; PHARV= Plant count at harvest; EASPECT=Ear aspect.

Table 46: Means of entomological and pathological traits of white maize varieties evaluated across 5 locations in Nigeria in the year 2015

Variety	SBLF4	LFS4	SBDH4	SBLF6	LF6	SBDH6	SBLF8	LFS8	NOH	LOT	MSV
TZL Comp 4C3	7.07a	2.20a	0.07a	6.73a	2.67a	0.07a	2.67a	3.20ab	9.00a	130.47a	2.33a
Obantanpa/TZLComp 3C3	4.73ab	2.20a	0.07a	4.87ab	2.63a	0.07a	6.33a	3.57ab	7.00a	111.07ab	2.22a
BR 9943 DMSR	6.07a	2.73a	0.02a	6.80a	3.00a	0.20a	3.00a	3.90ab	8.400a	107.07b	2.11a
Obatanpa/ IWDC2 Syn	5.00ab	2.40a	0.02a	5.93a	2.97a	0.00a	2.97a	3.37ab	6.93a	104.00b	2.22a
ACR06 TZL Comp 3C4	5.13ab	2.33a	0.02a	5.06a	2.77a	0.07a	2.77a	3.17ab	10.67a	104.20b	2.00a
Aflatoxin Syn W5	5.60a	2.60a	0.00a	6.67a	2.97a	0.00a	6.47a	3.93a	6.00a	99.06b	2.44a
SynLDFO/Obatanpa/TZL Comp 3C3*2	5.13ab	2.67a	0.33a	6.27a	3.30a	0.13a	6.13a	3.87ab	6.87a	104.67b	2.44a
Aflatoxin Syn W4	3.67ab	2.20a	0.20a	3.67ab	2.67a	0.27a	2.67a	3.63ab	7.07a	108.00b	2.33a
ACR06 TZL Comp 4C4	4.33ab	2.60a	0.40a	3.33ab	3.70a	0.33a	2.70a	3.40ab	6.00a	115.73ab	2.00a
TZL Comp 3C3 DTC2	5.80a	2.60a	0.20a	6.67a	3.13a	0.20a	3.13a	3.47ab	7.53a	112.93ab	2.33a
TZL Comp4 C4	0.92b	1.31b	0.08a	1.30b	1.42b	0.08a	1.42b	2.38b	8.17a	106.25b	1.86a
Mean	4.91	2.36	0.18	5.31	2.76	0.13	5.13	3.46	7.65	109.55	2.22
S.E (0.05)	0.77	0.19	0.04	0.72	0.17	0.03	0.50	0.18	0.58	4.04	0.06

Means with common letter in each column were not significantly different at  $p>0.05$ .

SBLF4= Stem borer leaf feeding 4 WAP; LFS4=Leaf feeding score 4WAP; SBDH4= Stem borer dead heart 4WAP; SBLF6=Stem borer leaf feeding 6WAP; LF6=Leaf feeding score 6WAP; SBDH6=Stem borer dead heart 6WAP; SBLF8=Stem borer leaf feeding 8 WAP; LFS8= Leaf feeding score 8 WAP; NOH= Number of holes; LOT= Length of tunnel; MSV= Maize streak virus.

TZL Comp 4C4 had the least number of leaves with leaf feeding of 0.92 while the TZL Comp4C3 has the highest number of leaves with leaf feeding of 7.07 at 4 weeks after planting. BR 9943 DMSR had the highest feeding score of 2.73 while TZL had the lowest feeding score of 1.31 at four weeks after planting. There was no significant difference in terms of number of plant with stem borer dead heart at four weeks after planting but Aflatoxin Syn W5 had 0.00 while TZL Comp 4C4 had 0.08. BR 9943 DMSR had the highest number of plants with leaf feeding at 6 weeks after planting (6.80) while TZL Comp 4C4 had 1.30 at 6 weeks after planting. ACR06 TZL Comp 4C4 had the highest leaf feeding score at 6 weeks with after planting with 3.70 while TZL Comp 4C4 had the least feeding score at 6 weeks after planting with 1.42. There was no significant difference in the number of plants with dead heart at 6 weeks after planting but both Obatanpa/IWD C2 Syn and Aflatoxin Syn W5 had 0.00 while ACR06 TZL Comp 4C4 had the highest number of plants with dead heart of average of 0.33. Aflatoxin Syn W5 had average number of stem borer leaf feeding of 6.47 to rank highest at 8 weeks after planting while both TZL Comp 4C3 and Aflatoxin Syn W4 have average number of 2.67 plants with leaf feeding to rank lowest at 8 weeks after planting, although there was no significant difference in the number of plants with leaf feeding. However Aflatoxin Syn W5 had the highest leaf feeding score at 8 weeks after planting with average of 3.93 while ACR06 TZL Comp 3C4 had 3.17. There was no significant difference in the number of holes found on the stems of these tested maize varieties; ACR06 TZL Comp 3C4 had the highest with average of 10.67 while Aflatoxin Syn W5 and ACR 06 TZL Comp 4C4 both had the least number of holes with average of 6.0. TZL Comp 4 C3 had the highest length of tunnel with 130.47cm while Aflatoxin Syn W5 had the shortest tunnel of 99.06. There was no significant difference in resistance of the maize varieties to Maize streak virus. (Table 46)

Ibadan had the highest number of plant count at germination for the yellow maize varieties screened in 2015 with 19.58 while Kotangora had the lowest plant count with 10.56. Kotangora had the highest number of plants with leaf feeding at 4 weeks with 14.06 while Kabba had the least with average of 0.33. Kotangora had the highest feeding score of 5.63 at weeks after planting while Kabba had the least feeding score at 4 weeks after feeding with 1.09. Mokwa had 0.00 numbers of plants with dead heart at 4 weeks after planting while Kotangora had 0.41 to be the highest recorded. Kotangora had the highest average number of plants with leaf feeding at 6 weeks after planting with 13.94 while Kabba had the least with

1.30. Kotangora the highest leaf feeding score at 6 weeks after planting with 5.53 while Kabba had the least with leaf feeding score of 1.73. Kabba had 0.00 dead hearts at 6 weeks after planting while Kotangora had the highest number of plants with dead heart with average of 0.50 at 6 weeks after planting. Kotangora location had the highest number of plants with leaf feeding at 8 weeks after planting with 7.75, but Lugbe (Abuja, FCT) had the least average number of plants with leaf feeding 8 weeks after planting with 1.15. Kotangora had the highest leaf feeding score of 3.84 at 8 weeks after planting while Lugbe (Abuja, FCT) on the other hand has the least feeding score 8 weeks after planting with 1.94. Ibadan had highest average number of plants with dead heart at 8 weeks after planting with 0.30 while Kabba has 0.00. The highest root lodging was observed at Lugbe (Abuja, FCT) with average root lodging of 2.61 plants while the lowest was observed at Kotangora with average of 0.19. The stalk lodging at Kotangora was 0.00 while the highest stalk lodging was observed at Lugbe (Abuja, FCT) with average of 2.64 plants. The highest number of holes was observed at Kabba with average of 17.49 while the lowest number of holes was observed at Mokwa with 0.79. The longest length of tunnel was observed at Mokwa with mean of 136.24 cm while the shortest length of tunnel was recorded at Kabba with 51.83cm (Table 47).

Table 47: Means of stemborer traits of Yellow maize evaluated across 5 locations in Nigeria in 2015

Locations	PC	SBLF4	LFS4	SBDH4	SBLF6	LFS6	SBDH6	SBLF8	LFS8	SBDH8	RLOD	SLOD	NOH	LOT
Mokwa	18.73a	2.03b	1.45bc	0.00b	1.81a	1.85b	0.06b	1.69c	2.08b	0.21ab	1.09b	0.88c	0.79c	136.24a
Kotangora	10.56c	14.06a	5.63a	0.41a	13.94a	5.53a	0.50a	7.75a	3.84a	0.06ab	0.19b	0.00d	7.34b	113.22b
Lugbe, Abuja	16.88b	2.39b	1.93b	0.06b	1.79b	1.76b	0.03b	1.15c	1.94b	0.03b	2.61a	2.64a	7.70b	133.15a
Ibadan	19.58a	2.09b	1.70bc	0.12b	2.49b	1.91b	0.21b	3.70a	2.61b	0.30a	0.24b	0.79c	7.03b	118.55b
Kabba	19.42a	0.33b	1.09c	0.03b	1.30b	1.73b	0.00b	2.58bc	2.33b	0.00b	0.64b	1.61b	17.49a	51.83c
Mean	17.07	4.12	2.35	0.12	4.21	2.54	0.16	3.35	2.55	0.12	0.96	1.19	8.13	112.05
S.E(0.05)	0.40	0.53	0.16	0.03	0.50	0.15	0.05	0.32	0.13	0.04	0.15	0.11	0.66	3.21

Means with common letters in each column were not significantly different at  $p>0.05$ .

PC= Plant count; SBLF4= Stem borer leaf feeding 4WAP; LFS4= Leaf feeding Score 4WAP; SBDH4= Stem borer dead heart 4WAP; SBLF6= Stem borer leaf feeding 6WAP; LFS6= Leaf feeding Score 6WAP; SBDH6= Stem borer dead heart 6WAP; SBLF8=Stem borer leaf feeding 8 WAP; LFS8= Leaf feeding Score 8WAP; SBDH8=Stem borer dead heart 8WAP; RLODGING= Root lodging; SLODGING= Stalk lodging; NOH= Number of holes; LOT= Length of tunnels.

Kabba had the highest number of days to flowering with average of 61.97 days while Mokwa had the least with 54.30 days. Kabba location had the highest number of days to silking with average of 62.52 days while Mokwa site had the least days to silking with average of 56.36 days. Kabba location had the least Anthesis-Silking interval with average of 0.79 days while kotangora had the highest Anthesis-Silking interval of 2.09 days. Kabba had the highest average plant height with 202.12 cm while Kotangora had the shortest plant height with 134.53 cm. Kabba had the highest ear height with 95.94cm while Kotangora had the least ear height with 70.31 cm. Mokwa recorded the best plant aspect with 2.64 rating while Kabba had the worst plant aspect with rating of 3.29. The worst ear aspect was observed in Kotangora (3.40) while the best were observed both at Lugbe (Abuja, FCT) (2.42) and Ibadan (2.42). The highest plant count at harvest was noticed at Kabba with average of 19.55 while the lowest plant count was recorded at Kotangora with 10.28. Similarly the highest number of ears at harvest was 20.18 and was obtained at Kabba while the lowest number of ears was from Kotangora with average of 9.34. The best husk cover was observed at Lugbe (Abuja, FCT) with average rating of 2.55 while the worst was observed at Kabba with rating of 3.15. There was significant difference in the rating for the ear rot in all the locations. The highest field weight was observed at Kabba 2.30kg while the lowest field weight was observed at Kotangora with 0.81kg. The highest grain weight per plot was observed at Kabba with 1.96 Kg per plot, while the lowest grain weight was observed at kotangora with 0.65Kg Per plot. The highest yield was observed at Mokwa with 2164.57kg per hectare while lowest yield was observed at Ibadan with 1805.54kg per hectare (Table 48).

Provitamin yellow variety PVA Syn 6F2 had the highest number of plant count at germination with 19.13 while PVA Syn 3F2 had the least plant count at germination with mean of 8.00. The highest number of leaf feeding at 4 weeks after planting was observed on Aflatoxin Syn 2-Y with 5.80 while the lowest number of leaf feeding was observed on PVA Syn 3F2 with 2.80. There was a significant difference in the leaf feeding score but PVA Syn 19 F2 had the highest score with 2.87, PVA Syn 3F2 had the lowest feeding score with 1.80. There was no significant difference in the number of plants with dead heart 4 weeks after planting. Aflatoxin Syn 2-Y had the highest number of leaves with leaf feeding at 6weeks after planting while PVA Syn 3 F2 had the lowest number of leaves with leaf feeding with 2.67. Though there was no significant difference in the number of leaves with leaf feeding. There was also no significant difference in the leaf feeding score 6 weeks after planting. PVA



Syn 3F2 and PVA Syn 1F2 have no plant with stem borer dead heart 6 weeks after planting while BR 9928 DMRSR had the highest number of plant with stem borer dead heart average of 0.53. PVA Syn 10 F2 had the highest number of plants with leaf feeding 8 weeks after planting with 4.33 while PVA Syn 3F2 had the lowest average number of leaves with leaf feeding 8 weeks after planting with 2.53, although there was no significant difference in the number of leaves. There was no significant difference in the leaf feeding score 8 weeks after planting (Table 49).

PVA Syn 19F2 had the highest average number of plants (0.47) with stem borer dead heart at 8 weeks after planting while PVA Syn 6F2, ACR 91 SUWAN-1 SR-C1, PVA Syn 11F2, PVA Syn 3F2 and PVA Syn 1 F2 all had no plant with Stem borer dead heart 8weeks after planting. Resistant check BR 9928 DMRSR had the highest average days to flowering with 59.47 days while Aflatoxin Syn 2-Y had the lowest average number of days to flowering with 56.60. Resistant check BR 9928 DMRSR had the highest average days to silking with 61.00 days while Aflatoxin Syn 2-Y had the lowest days to silk with 58.20 days (Table 49).

Table 48: Means of agronomic traits of Provitamin A yellow maize evaluated in Nigeria across five locations in 2015

Locations	PC	D50_FL	D50_SLK	ANSI	PHT	EHT	PLASP	EASP	PHARV	EHARV	HCOV	EROT	FWT	GRWT	YLD
Mokwa	18.73a	54.30d	56.36d	2.06a	169.94b	80.79c	2.64c	2.88b	17.06b	17.46ab	2.76cd	2.03a	1.58b	1.20b	2164.57a
Kotangora	10.56c	55.00d	57.03cd	2.09a	134.53c	70.31d	3.42a	3.40a	10.28c	9.34c	2.86bc	2.67a	0.81c	0.65c	1839.94d
Lugbe,Abuja	16.88b	56.67c	57.94c	1.36b	172.00b	89.76b	2.80bc	2.42c	16.15b	16.00b	2.55d	2.67a	1.82b	1.18b	1805.54e
Ibadan	19.58a	59.58b	60.97b	1.36b	174.91b	78.27c	2.91b	2.42c	18.85a	16.55b	3.06ab	2.91a	1.60b	1.20b	2082.98b
Kabba	19.42a	61.97a	62.52a	0.79c	202.12a	95.94a	3.29a	2.62bc	19.55a	20.18b	3.15a	2.56a	2.30a	1.96a	1991.36c
Mean	17.07	57.52	58.98	1.53	170.92	83.09	3.01	2.75	16.41	15.95	2.88	2.57	1.63	1.25	1982.04
S.E(0.05)	0.40	0.31	0.27	0.09	2.20	1.14	0.04	0.06	0.40	0.62	0.05	0.15	0.06	0.04	12.39

Means with common letter in each column were not significantly different at  $p>0.05$ .

D50FL= Days to 50% flowering; D50 SILK= Days to 50% silking; ANSI= Anthesis-silk interval; PHT= Plant height; EHT= Ear height; PLASPECT= Plant aspect; EASPECT = Ear aspect; PHARV= Plant count at harvest; EHARV= Ear count at harvest; HCOV= Husk Cover; EROT= Ear rot; FWT=Field weight; GWT= Grain weight; YLD=yield.

Table 49: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of yellow maize evaluated across 5 locations in Nigeria in 2015

Varieties	PC	SBLF4	LFS4	SBDH4	SBLF6	LFS6	SBDH6	SBLF8	LFS8	SBDH8	D5OFL	D5OSILK
PVA syn 6 F2	19.13a	4.67a	2.67a	0.07a	4.67a	2.53a	0.07b	4.13a	2.53a	0.00b	56.80c	58.47b
PVA syn 9 F2	18.73a	3.53a	2.00a	0.13a	4.00a	2.40a	0.07b	3.73a	3.03a	0.33ab	57.33bc	58.67b
PVA syn 10 F2	18.67a	4.67a	2.60a	0.13a	5.13a	2.73a	0.33ab	4.33a	2.57a	0.07b	56.80c	58.47b
PVA syn 19 F2	18.00ab	5.33a	2.87a	0.20a	4.93a	2.83a	0.13ab	3.33a	2.47a	0.47a	56.93bc	58.27b
ACR 91 SUWAN-1 SR -C1	18.00ab	4.33a	2.67a	0.07a	4.00a	2.27a	0.07b	2.93a	2.17a	0.00b	56.67c	58.47b
PVA syn 17 F2	17.93ab	3.13a	2.00a	0.20a	3.40a	2.40a	0.20ab	2.67a	2.33a	0.13ab	57.80abc	59.13ab
Aflatoxin syn 2-Y	17.87ab	5.80a	2.73a	0.07a	5.60a	2.80a	0.13a	3.27ab	2.47a	0.07b	56.60c	58.20b
PVA syn 11 F2	17.87ab	4.20a	2.60a	0.20a	4.47a	3.00a	0.20ab	2.93a	2.87a	0.00b	57.27bc	58.73b
BR 9928 DMRSR	16.93ab	3.60a	2.27a	0.20a	3.67a	2.27a	0.53a	3.13a	2.40a	0.27ab	59.47a	61.00a
PVA syn 3 F2	16.13b	2.80a	1.80a	0.07a	2.67a	2.17a	0.00b	2.53a	2.33a	0.00b	58.07abc	59.33ab
PVA syn 1 F2	8.00c	3.21a	2.00a	0.00a	3.71a	2.50a	0.00b	3.86a	2.92a	0.00b	59.07ab	60.07ab
Mean	17.07	4.12	2.35	0.12	4.21	2.54	0.16	3.35	2.55	0.12	57.52	58.98
S.E (0.05)	0.40	0.53	0.16	0.03	0.50	0.15	0.05	0.32	0.13	0.04	0.31	0.27

Means with common letter in each column were not significantly different at  $p>0.05$ . PC=Plant count; SBLF4=Stem borer leaf feeding 4WAP; LFS4= Leaf feeding score 4WAP; SBDH4= Stem borer dead heart 4WAP; SBLF6= Stem borer leaf feeding 6WAP; LFS6=Leaf feeding score 6WAP; SBDH6= Stem borer dead heart 6WAP; SBLF8= Stem borer leaf feeding 8WAP; LFS8= Leaf feeding score 8WAP; SBDH8= Stem borer dead heart 8 WAP; D5OFL= Days to 50% flowering; D5OSILK = Days to 50% silking;

PVA Syn 1 F2 and PVA Syn 19 F2 had the worst ear aspect with 3.00 while ACR 91 SUWAN-1 SR-C1 had the best ear aspect of 2.40. In terms of ear rot, PVA Syn 9 F2 had the worst ear rot with average rating of 3.87 while PVA Syn 10 F2 had the best ear with rot rating of 2.17. Aflatoxin Syn 2-Y had the highest field weight per plot with average of 1.97kg per plot while PVA Syn 1 F2 has lowest average field weight per plot with 1.06kg per plot. PVA Syn F9 had the highest average number of holes on the stems with 11.0 while BR 9928 DMRSR had the least number of holes on the stem with average number of 6.13, although the difference were not statistically significant. PVA Syn 10 F2 had the highest yield per hectare with 2033.70 kg/ha while BR 9928 DMRSR had the least yield with 1934.81Kg/ha. Aflatoxin Syn 2-Y had the highest grain weight per plot with 1.51kg/plot while PVA Syn 1 F2 had the least grain weight per plot with 0.75kg/plot. Aflatoxin Syn 2-Y had the highest length of tunnel with 125.40 cm while PVA Syn 3 F2 has the least length of tunnel with 96.40 cm. There was no significant difference in the resistance to maize streak virus among the varieties (Table 50).

Table 50: Means of different agronomic and entomological data observed and separated by Duncan Multiple Range Test of yellow maize evaluated across 5 locations in 2015.

Varieties	EASPECT	EROT	FWT	NOH	YLD	GWT	LOT	MSV
PVA syn 6 F2	2.70ab	2.50ab	1.67abc	8.87a	2011.92ab	1.26ab	96.87c	2.22a
PVA syn 9 F2	2.70ab	3.87a	1.73abc	11.00a	1966.83bc	1.39ab	115.87abc	2.22a
PVA SYN 10 F2	2.63ab	2.17b	1.59abc	8.53a	2033.70a	1.23ab	117.60abc	2.33a
PVA syn 19 F2	3.00a	2.77ab	1.61abc	9.60a	1989.23abc	1.27ab	100.27bc	2.00a
ACR 91 SUWAN- 1 SR – C1	2.40b	2.20b	1.84ab	8.40a	1957.98bc	1.44ab	121.50ab	2.00a
PVA syn 17 F2	2.87ab	2.67ab	1.83abc	6.93a	1984.52abc	1.46ab	113.53abc	2.33a
Aflatoxin syn 2-Y	2.50ab	2.33ab	1.97a	9.60a	1989.23abc	1.51a	125.40a	2.22a
PVA syn 11 F2	2.90ab	2.67ab	1.53bc	7.53a	1992.64abc	1.12b	119.36abc	2.33a
BR 9928 DMRSR	2.97a	2.63ab	1.59abc	6.13a	1934.81c	1.15b	115.29abc	2.00a
PVA syn 3 F2	2.57ab	2.20b	1.41cd	6.21a	1984.78abc	1.16ab	96.40c	2.22a
PVA syn 1 F2	3.00a	2.21b	1.06d	6.50a	1956.65bc	0.75c	111.77abc	2.00a
Mean	2.75	2.57	1.63	8.13	1982.04	1.25	112.05	2.17
SE	0.06	0.15	0.06	0.66	12.39	0.04	3.21	0.04

Means with common letter in each column were not significantly different at  $p>0.05$ . EASPECT= Ear aspect; EROT= Ear rot; FWT=Field weight; NOH= Number of holes; YLD=Yield; GWT=Grain weight; LOT=Length of tunnel; MSV= Maize streak virus.

#### 4.2.2 SURVEY OF STEM BORERS IN TROPIC HUMID ECOLOGY OF SOUTHWEST NIGERIA AND KWARA STATE

During this survey, it was discovered that the enormity of problem of stem borers and ear borers on maize in the Tropic humid ecology especially in the South West Nigeria have been down played or underestimated. Despite all the other factors that favour maize production in South Western Nigeria, it can only be profitable if the maize is harvested green, otherwise stem borers will render it unprofitable. Alternatively Agricultural engineers need to construct appropriate cheaper and effective driers that can handle maize drying if harvested at physiological maturity. Southern Guinea Savannah, Northern Guinea Savannah and Sudan Savannah are probably better for maize production especially if the maize is to be left on the field till harvest time. Although with the great improvement to agriculture brought by Agricultural engineering, Maize can be harvested at physiological maturity and dried using drying engine. This is due to the fact that all the borers' complex and *Sitophilus zeamais* and *Sitophilus oryzae* combine to render the harvest almost useless at the time of harvest as the survey and screening of materials done on the field in the course of this project revealed.

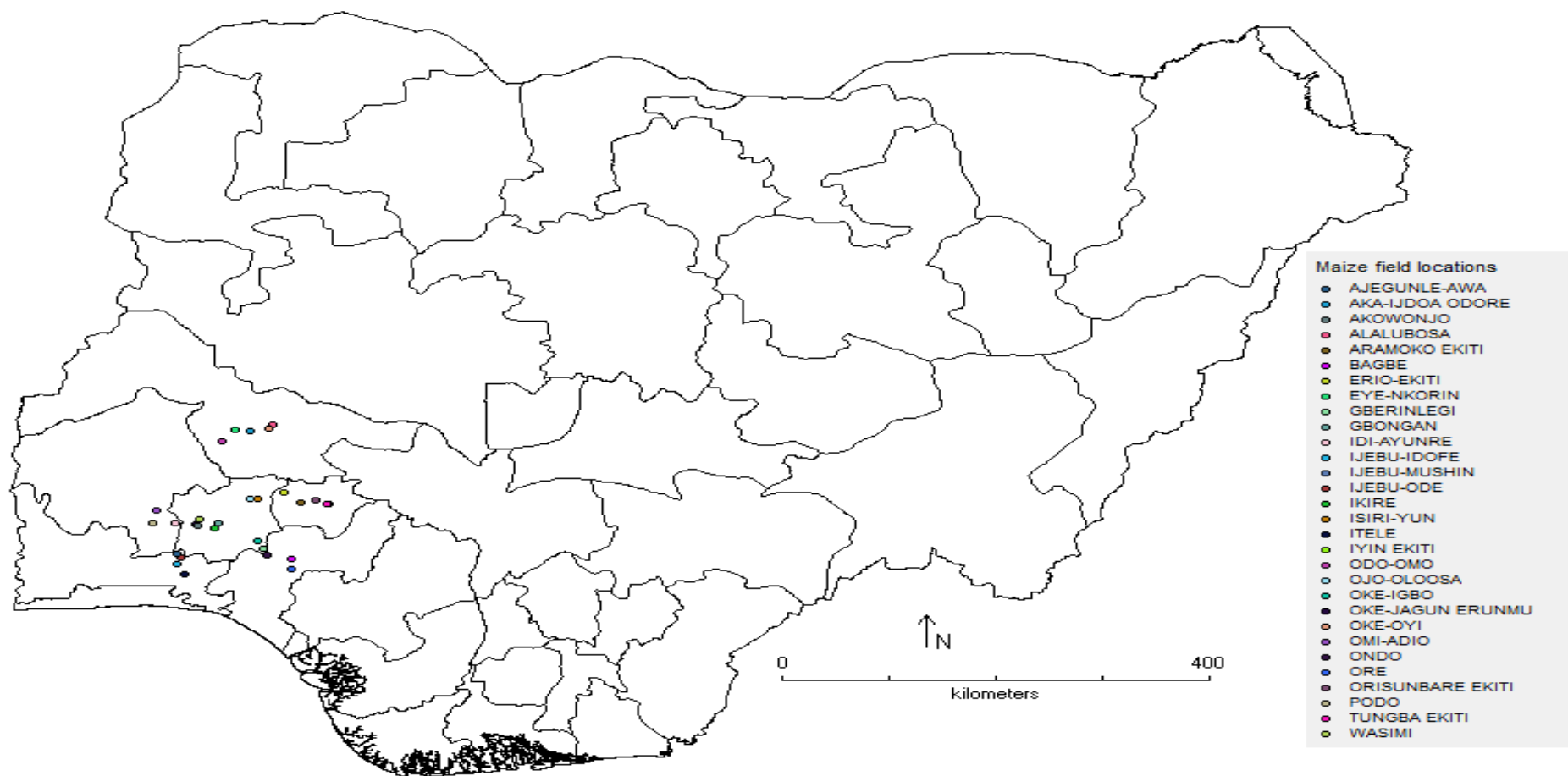
The survey also revealed that the stem borer's complex causes lodging; both root and stalk lodging in the places surveyed. The stalk lodging is more prevalent. The stalk lodging generally took place between 50cm to the tassel of the maize depending on the height of the maize. This further increases the loss of farmer by stem borers complex found in South Western Nigeria. The tassels were seriously affected by the stem

borers in all the places surveyed. This is probably caused by *Busseola fusca* which is known to migrate to the top of maize whorl before boring down the stem. The stem tunneling by the stem borers accompanied with rain and wind causes the root and stalk lodging of the maize plant.

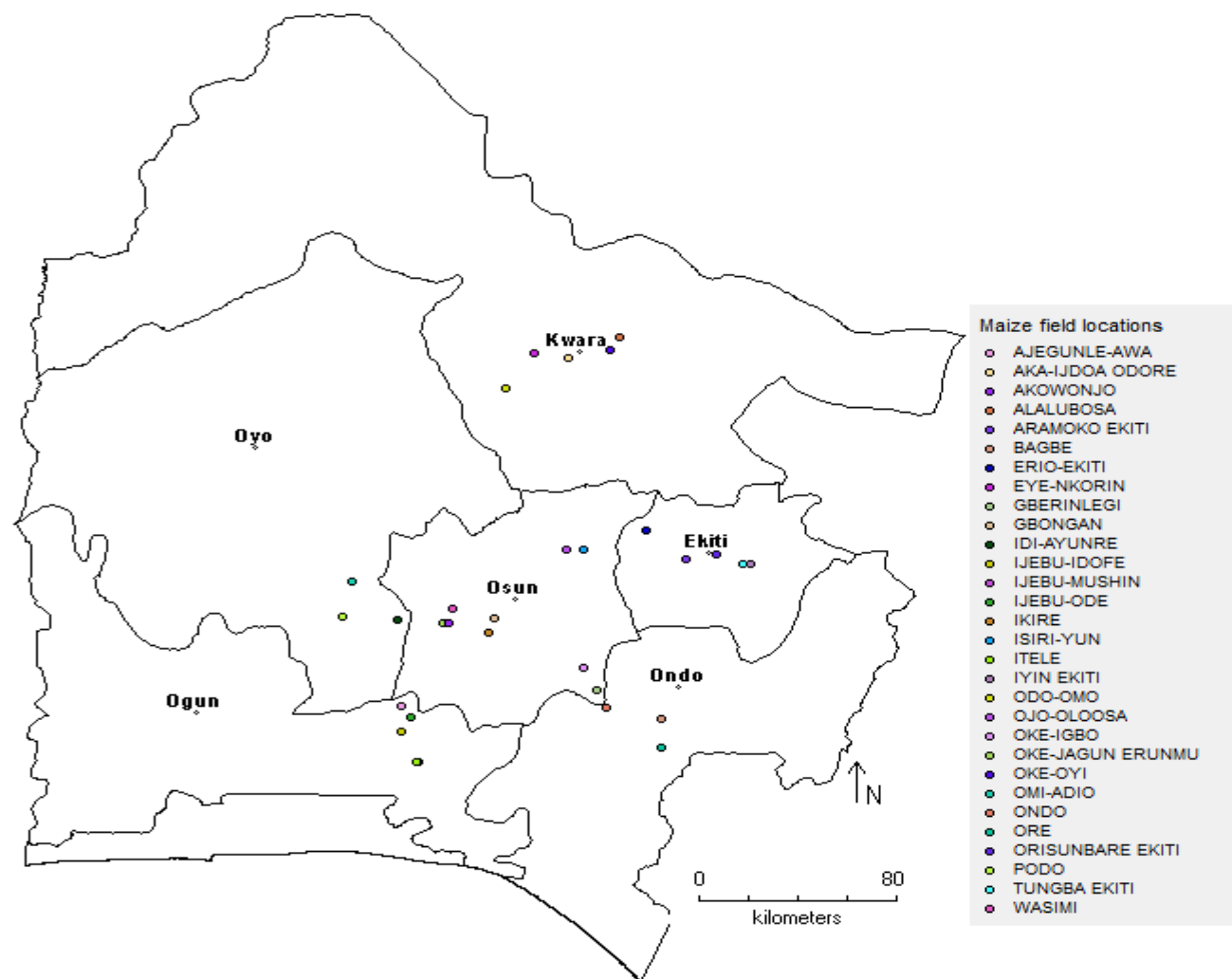
Table 51 B: **ANALYSIS OF DATA COLLECTED FROM THE SURVEY**

State	Altitude (m)	% Infestation	Stborer. Complex	Cropping system	
Oyo	559.24 <sup>d</sup>	9.40 <sup>b</sup>	4.40 <sup>c</sup>	1.20 <sup>b</sup>	KEY: %INFESTATION=AVERAGE % INFESTATION IN EACH STATE; STEM BORER COMPLEX= DIFFERENT STEM BORERS FOUND IN EACH STATE; CROPPING SYSTEM: SOLE=1, INTERCROPPING =2, MAIZE WITH FEW STANDS OF OTHER CROPS=1.5
Kwara	1213.20 <sup>b</sup>	44.00 <sup>a</sup>	3.60 <sup>c</sup>	1.30 <sup>b</sup>	
Ondo	677.58 <sup>cd</sup>	42.00 <sup>a</sup>	4.00 <sup>c</sup>	1.30 <sup>b</sup>	
Ogun	205.60 <sup>e</sup>	54.00 <sup>a</sup>	4.60 <sup>bc</sup>	1.60 <sup>ab</sup>	
Osun	863.50 <sup>c</sup>	40.00 <sup>a</sup>	5.60 <sup>ab</sup>	2.000 <sup>a</sup>	
Ekiti	1602.40 <sup>a</sup>	36.00 <sup>a</sup>	5.80 <sup>a</sup>	1.60 <sup>ab</sup>	

Fig 1:Map of Nigeria showing surveyed sites.







**Fig 2: Map of South West Nigeria showing surveyed sites.**

### 4.2.3 SURVEY OF STEM BORERS IN OYO STATE

The survey methods ensure that various part of the farms were covered. Estimate of infestation was made at different parts of the farm. Five farms were surveyed in each state. In Oyo state, the farm surveyed includes Omi-Adio Long. E003°46.569' and Lat. N07°23.900' and Altitude 592.1m. The percentage of stem borer infestation level was 8%. The natural enemies found on the farm includes Ichneumonid Wasp and Ants. The stem borer observed includes *Coniesta ignifusalis*, *Busseola fusca*; *Eldana saccharina*, *Mussidia nigrivenella* and *Sesamia calamistis*. Others include *Scirpophaga sp* and *Maliarpha separatella*. The cropping system on the farm was sole maize.

The next place surveyed was podo located on longitude E003°52.085' and Latitude N07°18.631' with the altitude at 480.2m. The percentage stems borer infestation was 3%. The cropping system on the farm was sole maize. Other insects observed on the farm were *Sitophilus zeamais*. During the field experiments it was discovered that *Sitophilus zeamais* also attacks maize stems. Other insects observed on the farm were Aphids. The maize stems of the farm observed was hard and dry. The next farm surveyed in Oyo state was at Idi-Ayunre. Location of the farm was at Longitude E003°51.805' and Latitude N07°13.803' and Altitude of 403.6m. The percentage infestation level of stems borer was 16%. Other insects found include Ants and green aphids.

The next farm surveyed was at Oke-Jagun Erunmu. Location was on Longitude E004°03.776' and Latitude N07°27.007' and height of 683.1m and percentage level of infestation was 12%. Others insect observed were *Sitophilus zeamais*. It will be observed that *Sitophilus zeamais* is a field to store pest. The infestation of maize grains start from the field.

The survey was also extended to Akowonjo, where the maize on that farm was intercropped with cassava. It was not clear whether the cropping system affected the level of infestation by stem borers. The location of farm was Longitude E 004°03.858' and Latitude N07°25.108' and height 637.6m. The level of infestation was 8%. Other insects observed were ants.

The average level of infestation in Oyo State was 9.4%, the highest being 16% at Idi Ayunre and the lowest infestation level being 3% at Podo. Bosque-Perez et. al., (1992) also observed average infestation level of up to 17% in Southern Guinea Savannah.

#### 4.2.4 SURVEY OF STEM BORERS IN KWARA STATE

The survey of stem borers in Kwara state was done on July 17, 2014. Five fields were also surveyed in Kwara state. The first field was at Odo-Omo (Longitude E 004°25.288') and Latitude N08°20.577' and Altitude 1197m above the sea level. The same method of assessment was used. Every part of the farm was surveyed to ensure unbiased assessment of the farm. The number of plants infested with stem borers compared with the total number of the plants sampled was used to estimate the percentage stem borer infestation. The estimated level of stem borer infestation there was 80%. The farmer that owns the farm was interviewed to know whether he was aware of the stem borers. The farmer confessed to know about the stem borer infestation. When asked of the solution he has been employing to tackle the problem of stem borer infestation. He said they were just looking up to God and praying concerning the problem! Other insects observed there were aphids and *Sitophilus zeamais*.

Another farm surveyed was at Aka-Ijoba Odore. The farm was at Longitude E 004°40.243'. There were few stands of sorghum in the maize. The stem borers found here include *Sesamia calamistis*, *Busseola fusca* and other stem borers. The level of infestation was estimated at 80%. Government storage silo was not far from the farm. Other insects found on the farm include beetles.

The next farm surveyed was at Alalubosa. The farm was located at Longitude N004°40.938' and Latitude E08°33.599' and at elevation 1048m above the sea level. The estimated stem borer level was 20%. The farm here has special form of intercrop. The maize was intercropped with vegetables. Probably, the intercrop reduces the level of stem borer infestation or probably the level of infestation with stem borer is low in the area.

The next farm surveyed was at Oke-Oyi. The farm was located at Oke-Oyi Longitude N 004°43.633' and Latitude N08°35.284' and elevation 1255m above the sea level. The level of infestation there was also 20%.

The level of infestation with stem borers in Kwara state during the survey was between 20% to 80%. The average levels of infestation was 44%. This also agreed with other scientists who observed infestation level of as high as average of 47% (Bosque-Perez et. al., (1992)).

#### 4.2.5 SURVEY OF STEM BORERS IN ONDO STATE

Survey of stem borers infestation was carried out on July 22, 2014. The first farm surveyed was at Oke-igbo in the border of Osun and Ondo State. The farm was located at Longitude E004°44.214' and Latitude N 07°09'367' and altitude of 800m above the sea level. The level of infestation of stem borers in this maize farm was 40%.

The next farm surveyed was at Gberinlegi Longitude E 004°46.273 and Latitude N07°8.064' and elevation 786.7m above the sea level. Maize farm there was intercropped with cassava. The level of stem borer infestation on maize farm was 40%. Other insects observed on the farm include cricket, aphids and ants.

The next farm surveyed was at Ondo town. The farm surveyed was located at Longitude E004°50.140' and Latitude N07°01.197' and elevation 699.9m above the sea level. The level of infestation there was 10%. Other insects observed on the farm were aphids and ants.

The next farm surveyed was at Bagbe which was located at Longitude E004°50.887' and latitude N06°58.207' and altitude of 8013m above the sea level. The level of infestation here was high and was estimated to be 80%. Others insects observed on the farm were aphids and ants. The next farm surveyed was at Ore. The farm was located at E004°53.707 and Latitude N06°45.540' and elevation 300m above the sea level. Natural enemies of stem borers observed here were ants. The level of infestation on maize farms surveyed in Ondo state varied from 10% to 80% while the average level of infestation was estimated at 42%

#### 4.2.6 SURVEY OF STEM BORERS IN OGUN STATE

The survey of stem borer infestation was carried out in Ogun state on July 23, 2014. The first farm surveyed was at Ijebu-Ode at Longitude N0650.722' and Latitude N0650.722' and altitude 336.8m above sea level. The level of stem borer infestation on the farm was 50%. This is probably due to the carry over of eggs and diapausing larvae from the previous seasons. The cropping system on the farm was maize/ cassava intercrop. The natural enemies of stem borers observed on the farm were ants. The stem borers that were predominant on the farm were *Busseola fusca* and *Mussidia nigrivenela*.

The second farm surveyed was at Ijebu- Mushin Esure. The farm was located at Longitude E00359.678' and Latitude N0647.190' and altitude of 210.5m above sea level. The level of infestation observed was 50%. The natural enemies of the stem borer observed on the

farm were ants.

The third farm surveyed was at Itele. The farm was located at Longitude E00359.675' and N0647.188' and altitude 133.3m above sea level. The cropping system on the farm was maize\ cassava intercrop. The level of infestation was higher and it was estimated as 80% a larvae of parasitoid was observed on a dead stem borer on the farm. The larvae of parasitoid was suspected to be *Cotesia sesamia*. (*Braconidae*)

The fourth farm surveyed was at Ijebu-Idofe. The farm was located at Longitude E00356.636' and Latitude N0655.178' and 182.0m. The percentage infestation level was 50% The natural enemies of stem borers observed there were ants.

The fifth farm observed in Ogun state was at Ajegunle Awa. The farm was located at Longitude E00355.703'and Latitude N0659.353' and altitude 165.4m above the sea level. The level of infestation at Ajegunle Awa was 40%. The infestation level range from 40% to 80% in Ogun state during the survey, while the average level of infestation was 54%.It was observed in Ogun state as in other places surveyed that most infested plants break at a short distance below the tassel or any other point on the stem.

#### **4.2.7 SURVEY OF STEM BORERS IN EKITI STATE**

The survey of stem borers was carried out in Ekiti state on July 30, 2014. The first farm surveyed was at Erio- Ekiti. The location was at Longitude E00501.002' and Latitude N0743.520' and altitude 1744m above sea level. The level of infestation on the farm was 20%.

The second farm surveyed was at Aramoko-Ekiti. The location of the farm was at Longitude E00503.436' and Latitude N0742.129' and altitude 155m above the sea level .The cropping system on the farm was maize intercropped with cassava and cocoyam. The level of infestation was 50%.

The third farm surveyed was at Orisunbare Ekiti. The farm was located at Longitude E00504.791' and Latitude N00741.263' and altitude of 1598m above the sea level. The cropping system on the farm was maize intercropped with cocoyam. The level of infestation was 50%.

The fourth farm surveyed was at Iyin- Ekiti. The farm was located at Longitude E005°04.791' and Latitude N07°40.167' and altitude 1716m above the sea level. The cropping system on the farm was maize intercropped with cocoyam. The level of infestation with stem borers on the farm was estimated to be 40%. Aphids were also found in the farm.

The fifth farm surveyed was at Tungba Village in Ekiti state. The location of the farm was at Longitude E005°11.719' and Latitude E007°40.159 and altitude 1408m. The level of infestation with stem borers here was estimated to be 20%. The farmers found on the farm were interviewed to know their view on the stem borers infestation. The farmers said the small birds normally visit the maize infested with stem borers. They also said stem borers normally infest the maize towards time of maturity.

The range of stem borer infestation at the time of this survey was 20% with average level of infestation of 36%.

#### **4.2.8 SURVEY OF STEM BORERS IN OSUN STATE**

The survey of stem borers infestation was done on August 1<sup>st</sup>, 2014. The first farm surveyed was at Ikire. The location of the farm was at Longitude E004°11.900' and Latitude N007°23.093' and the altitude was 695.2m above the sea level. The cropping system on the farm was maize intercropped with cassava. The insects observed on the farm were ants. The level of infestation on the farm was 40%. Other insects found were ants which were natural enemies.

The second farm surveyed was at Wasimi. The farm was located at Longitude E04°16.136' and Latitude N07°26.361' and altitude of 705.1m above the sea level. The level of infestation was 30%. The cropping system on the farm was maize intercropped with cassava, yams, and sweet potato. Ants which were natural enemies were found on the farm.

The third farm surveyed was at Gbongan. The farm was located at Longitude E04°20.457' and latitude N07°2228.025' and altitude 803.1m. The cropping system on the farm was maize intercropped in cocoyam and cassava. The natural enemies found on this farm were ants called *Psilochalcis Soudanensis* in the family chalcididae. The level of infestation with stem borer on the farm was found to be 30%.

The fourth farm surveyed was at Ojo-Oloosa. The farm was located on Longitude E004°31.748' and Latitude N07°33.811' and altitude 859m above sea level. The cropping system on the farm was maize intercropped with cassava and cocoyam. The level of stem borer infestation on the farm was 30%. The natural enemies of stem borers found on the farm were also ants.

The fifth farm surveyed were at Isiriynn. The farm was located at Longitude E004°41.389' and Latitude N07°34.765' and altitude 1255m above the sea level. The cropping system on the farm was maize intercropped with cocoyam and okra (*Abelmoschus callei*). The level of infestation of the farm was 60%. The stem borers found on this farm include *Busseola fusca*, *Sesamia calamistis*, *Mussidia nigrivenella*.

**Fig 3: Unidentified maize stem borer raised from larvae collected from maize stem at Wasimi, Osun State.**

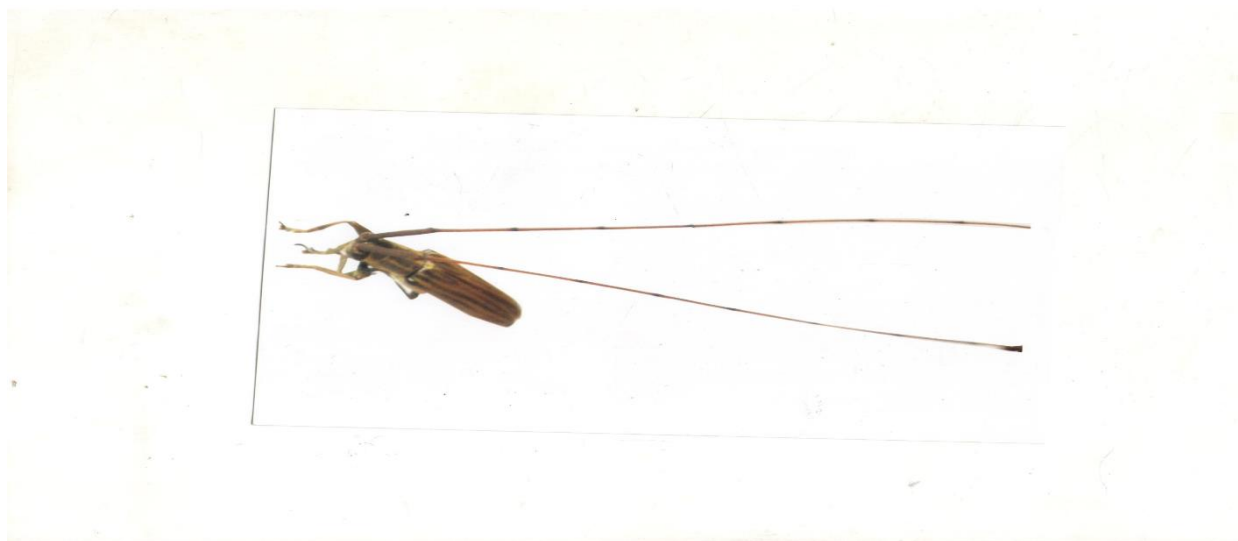


#### **4.2.9 IDENTIFICATION OF THE COLEOPTERA**

The photograph of the coleoptera was sent to Dr Michael Gates (Research Entomologist) with United States Department of Agriculture (USDA) and a Resource person with Smithsonian Institution (without giving him background of the insect). He and his friend Stephen identified it as Long horn beetle. When Long horn beetles was checked up in Wikipedia to know their identity. It was discovered they were wood borers common in United States of America, Asia and United Kingdom. The other stem borer that was found to be beetle among the borers collected from Moor plantation, Ibadan was different from the above beetle.



Fig 4: Another maize stem borer beetle found in Moor Plantation, Apata, Ibadan



#### **4.3.0 MOLECULAR ANALYSIS OF STEM BORERS COLLECTED.**

The basic chemical composition of DNA is the same. DNA is a polymer of only four different types of nucleotides (i.e nucleotides containing Adenine, Guanine, Thymine and Cytosine), the only difference being in the proportion of A + T to G +C among species of organisms. The choice of methods of extraction depends on a number of factors such as the required quantity, the molecular weight of the DNA required, the time and the expense of the method and the intended application of the DNA extract. It also depends on the source of DNA. All methods of cellular components, however involve:

- (a) Disruption and lysis of the starting material.
- (b) Removal of proteins and other contaminants.
- (c) Recovery of DNA or precipitation of DNA.

It is always important to purify DNA in a manner that prevents degradation of DNA, yielding the highest quality and quantity.

##### **4.3.1 (A) LYSIS OF THE CELL WALL/CELL MEMBRANE**

The general procedure to achieve lysis of cell walls/membrane, while minimizing DNA shearing can involve chemical or mechanical means or a combination of both. There are two

main detergents that are used to disrupt cell walls depending on the protocol. Sodium dodecyl sulphate (SDS) or Cetyl trimethylammonium bromide (CTAB). EDTA protects the released DNA from endogenous nuclease. NaCl<sub>2</sub> stabilizes the DNA.

#### 4.3.2 (B) SEPARATION OF DNA FROM NATURALLY OCCURRING CONTAMINANTS

- Polysaccharides and proteins can be removed from the DNA extracts by denaturation and precipitation using Chloroform and Isoamylalcohol.
- RNA can be removed by using RNase A enzyme.
- Polyphenolic can be removed by using  $\beta$ mercaptoethanol, ascorbic acid, bovine serum albumin(BSA), sodium azide and PVP 40 etc. The separation is achieved by centrifugation process thereby separating the DNA in solution from cell debris.

#### 4.3.3 (C) DNA PRECIPITATION

- Two main methods of precipitating DNA are used
- 70% ethanol with sodium acetate (with sodium hydroxide/acetic acid)(90 $\mu$ l 3M sodium acetate and 600  $\mu$ l ice cold absolute ethanol. Mix well by inversion of the tubes and leave at -20°C for 1 hour (preferably overnight) to precipitate DNA).

#### 4.3.4 DNA QUANTIFICATION

The extracted DNA is then quantified using nanodrop spectrophotometer or loading on 1% agarose gel or using both methods.

The stem borers used for extraction were larvae in the last instar stage and some pupae. They were first preserved in the ethanol before taken to Bioscience Centre (IITA, Ibadan) for extraction. Using Qiagen kit, total genomic DNA was extracted from the samples (Moyal et al 2011; Nowaczyk et al 2008; Moyal and Le Ru 2006).

The integrity and purity of DNA was checked by loading on 1% agarose gel electrophoresis (Sambrook et al 1989; Nowaczyk et al 2008).



Fig 5: Water Bath

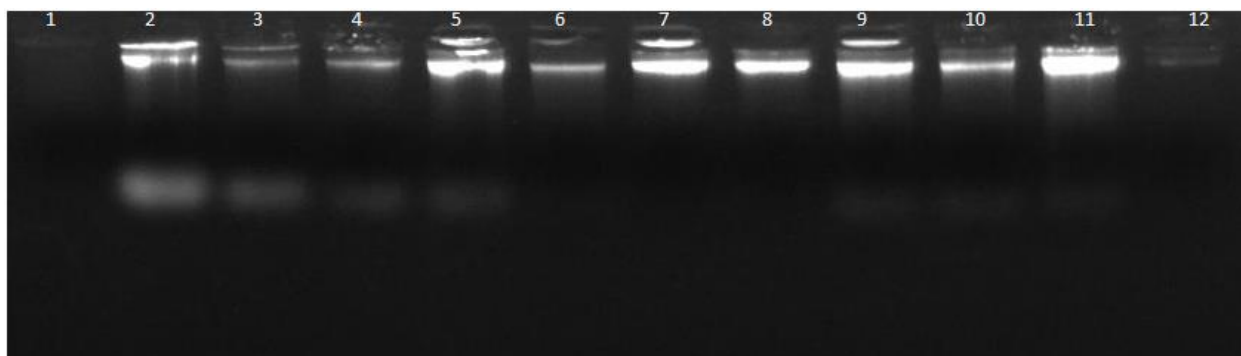


Fig 6: Electrophoregram for DNA quantification using 1% agarose gel

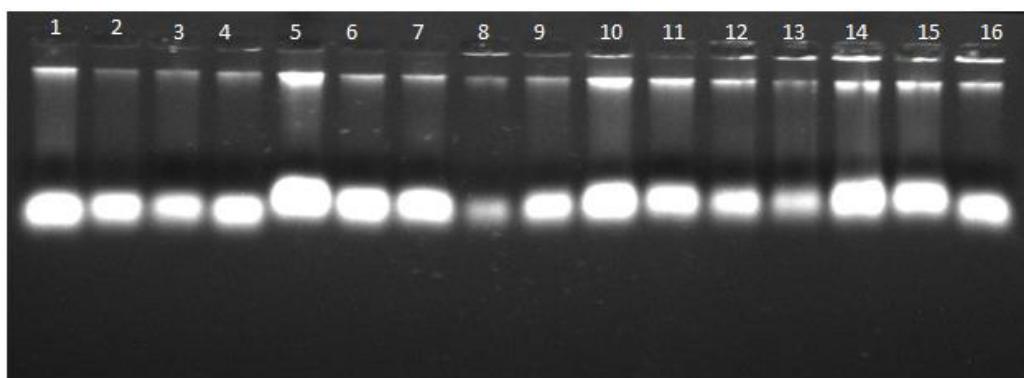


Fig 7: Picture of Gel band of DNA loaded on 1% Agarose

#### 4.3.5 OPTIMISING

Since more than one primer was used for amplification, the reactions were optimized. Four samples were optimized with four primers to ensure the reactions will work. This is to ensure reagents, time, efforts and resources are not wasted. The result was as given below.

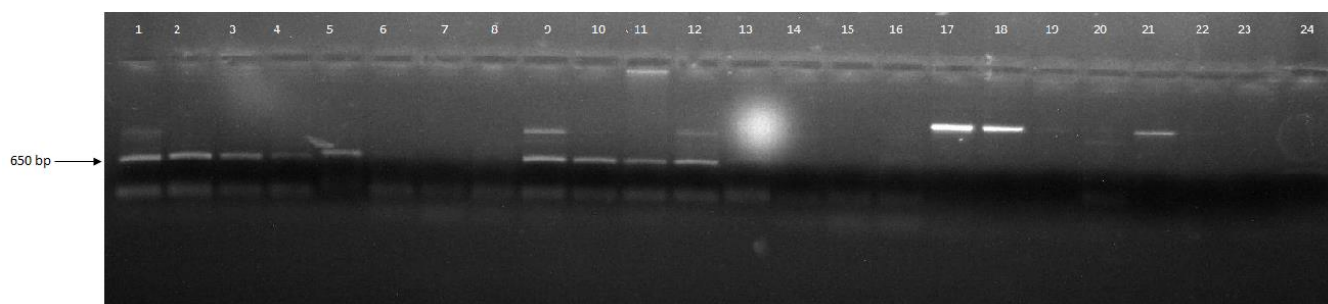


Fig 8: Picture of Optimising

#### 4.3.6 POLYMERASE CHAIN REACTION (AMPLIFICATION)

Cytochrome c oxidase, sub unit II (COX 2 gene) of the mitochondrial gene was amplified by Polymerase Chain Reaction(PCR) using the touch down method: Initial denaturation at 94°C for 5 minutes, followed by nine cycles of denaturation at 94°C for 30 seconds, annealing at 65°C for 30 seconds and extension at 72°C for 30 seconds. This was followed by another thirty four cycles of denaturation at 93°C for 15 seconds, annealing at 55°C for 20 seconds. This was followed by final extension at 72°C for 5 minutes and later left at 10°C until PCR products was needed. Some scientists have used genomic DNA from Mitochondrial genes such as Cytochrome *b* (*Cyt b*) (915/949 nucleotides(nt)), Cytochrome *c* oxidase, subunit 1(COXI) (890/894/925nt) and nuclear gene such as gene coding for the pheromone binding protein2 (PBP2) (685nt) and also 12S RNA(290nt) ( Moyal and Le Ru (2006); Moyal et. al (2011a); Moyal et. al (2011b); Moyal (2015)).The reaction mixture contained 10×PCRbuffer, 25mM MgCl<sub>2</sub>, 5pMol forward primer, 5pMol reverse primer, DMSO, 2.5Mm DNTPs, Taq 5µ/µl DNA and H<sub>2</sub>O per 10µl reaction mixture. The primers used were: CP1 (5'-GATGATGAAATTTTGGATC-3') [modified from Harry *et al.* (1998); Moyal and Le Ru (2013); Mehdi et al (2015); Ongamo(2008); TRs (5'-TCTATCTTATGTTTTCAAAAG-3') (Simon *et al.* 1994); Moyal and Le Ru (2013); Mehdi et al (2015); CP1 (5'-GATGATGAAATTTTGGATC-3')(modified from Harry et al., 1998); Moyal and Le Ru(2013); Mehdi et al (2015); Ongamo et al (2008); Tser (5'-TATTTCTTTATTATGTTTTCAAAAC-3') (Simon et al., 1994); Ongamo et al (2008); 16SAA (5'-ATGCTWCCTTTGCACRGTCAGATACYGCGGC-3') ( Chai and Du (2012) ); 16SBB (5'-CTTATCGAYAAAAAAGWTTGCGACCTCRATGTTG-3'), ( Chai and Du (2012) ); LP01 (5'-TGATTAGCTCCACAAATTTCTGAACATTGACC-3'), ( Chai and Du (2012) ) LP02 (5'-WACACCAGTTCATATTTDAACCAGAATGATATT-3') (Chai and Du (2012)).

The amplicons from the reaction was loaded on 1.5% agarose gel and gel pictures were as below. The ladder used is 1Kb plus ladder from Thermofisher Scientific.

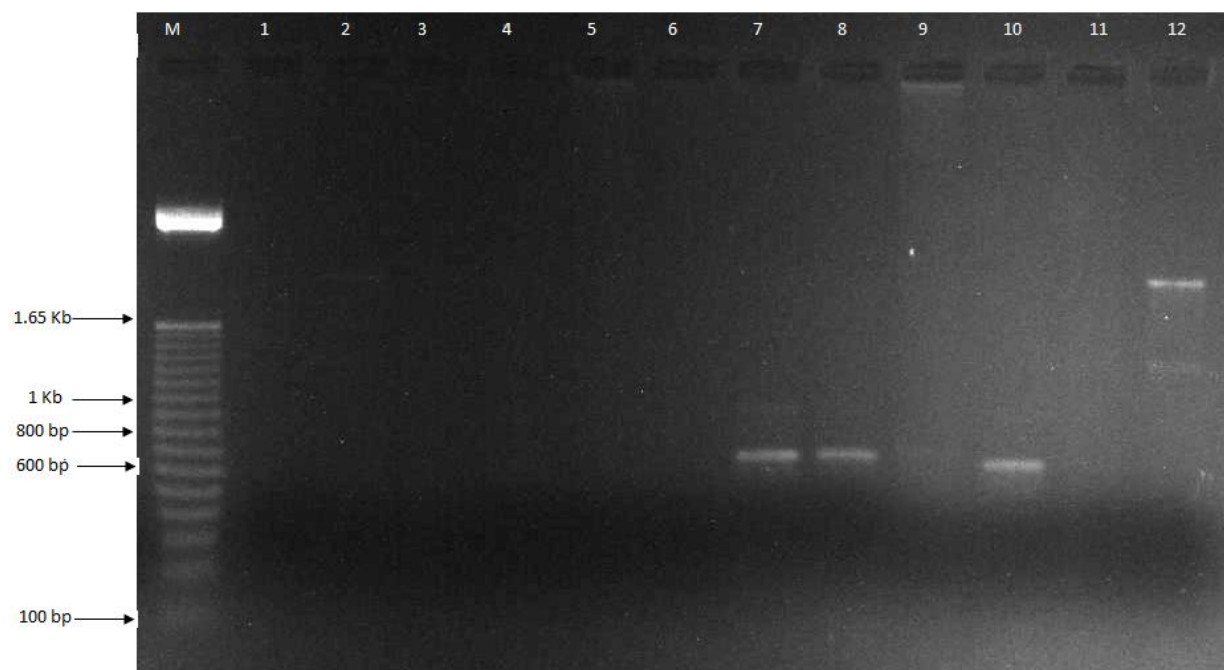
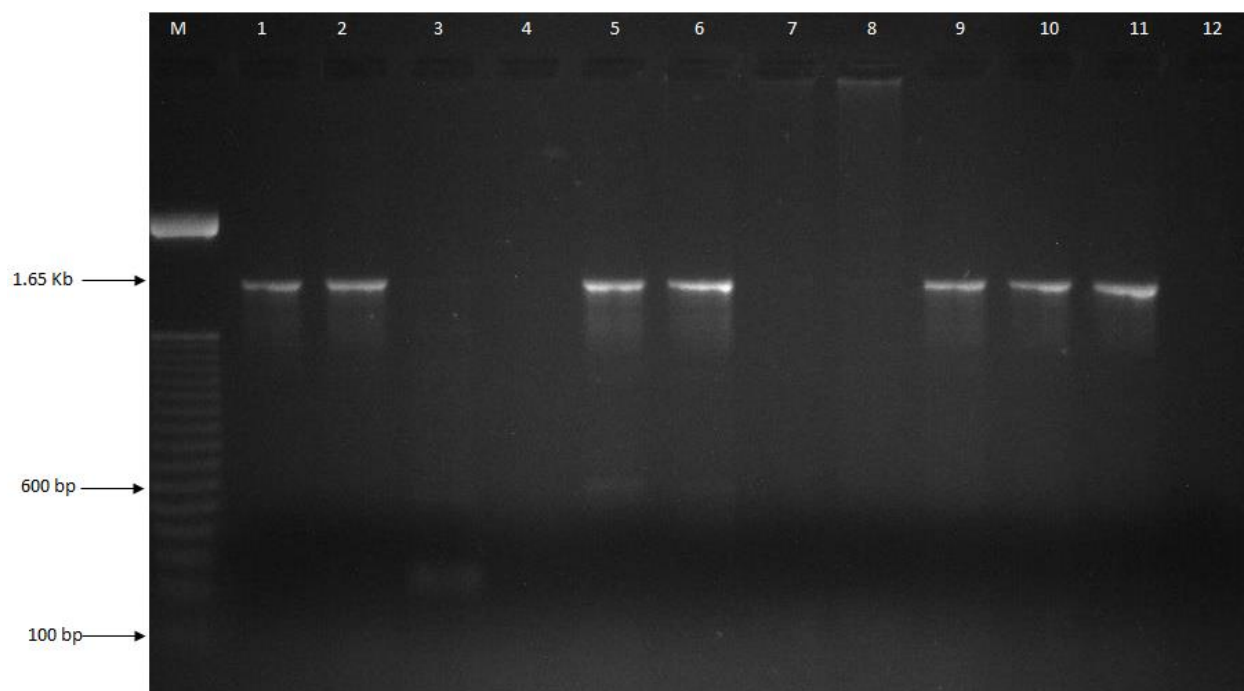
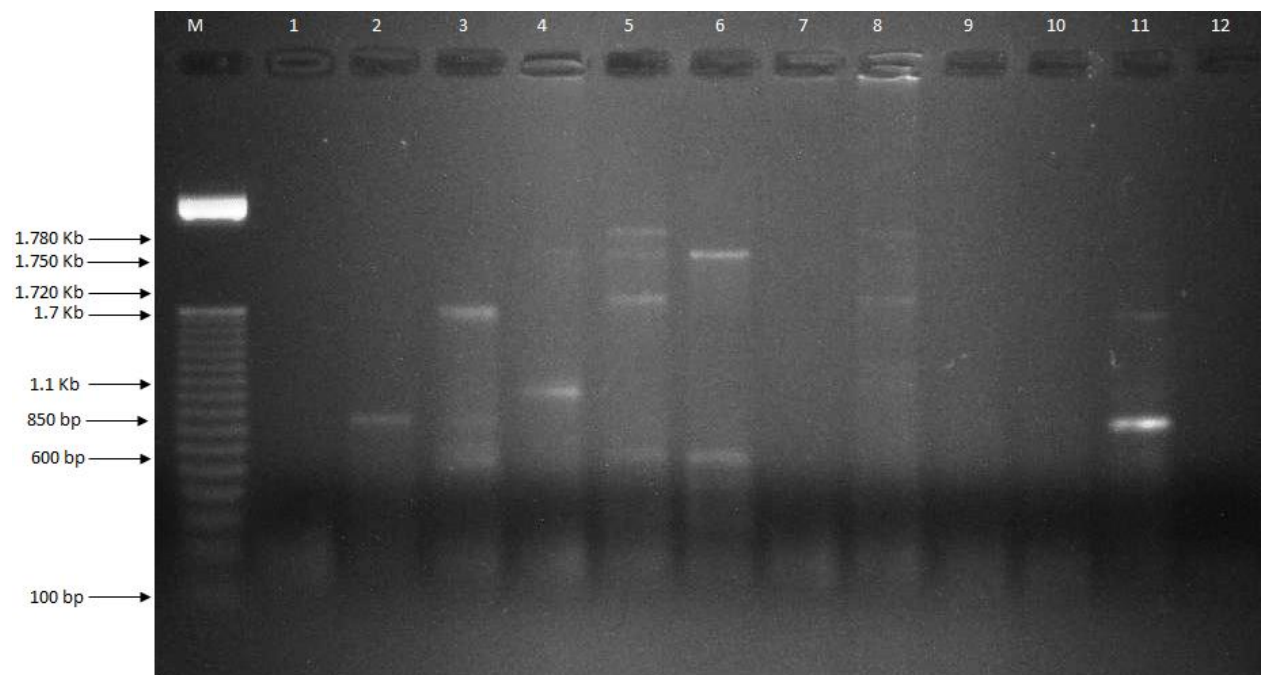


Fig 9: Gel Band of DNA Amplified with CP1 and TRs Primers

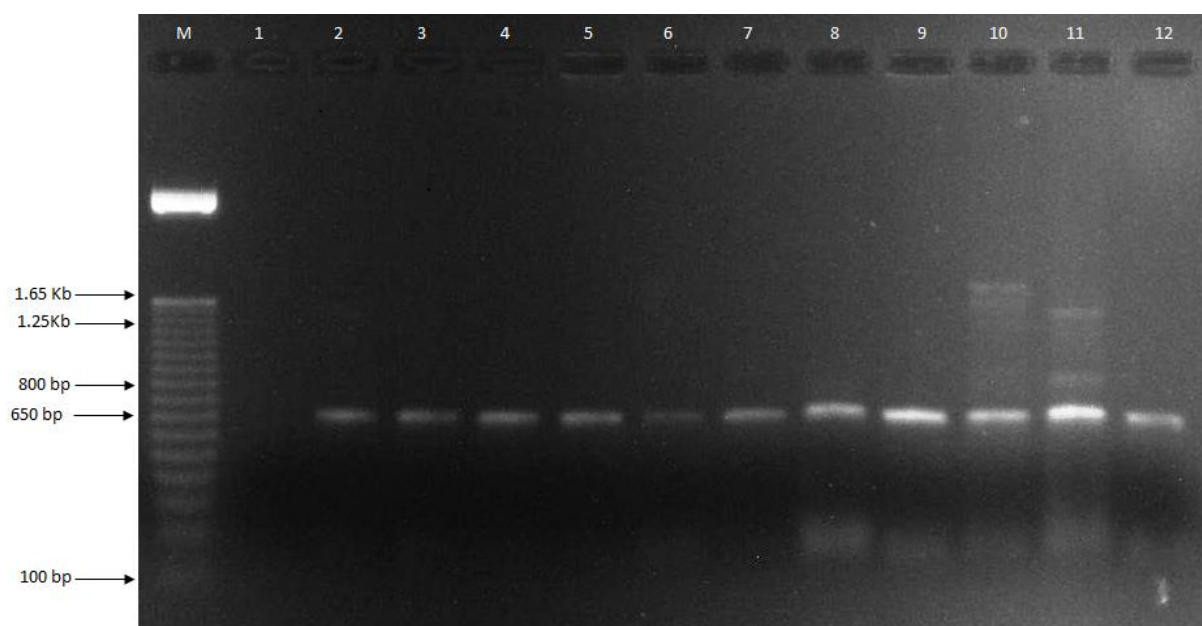


**Fig 10: Gel Band of DNA Amplified with CP1 and TSer Primers**



**Fig 11: Gel Band of DNA amplified with SAA and SBB Primers**





**Fig 12: Gel Band Amplified with LP 01 and LP 02 Primers**



**Fig 13: GEL ELECTROPHORESIS APPARATUS**

**Fig 14: GEL DOCUMENTATION SYSTEM**



Polymerase Chain Reaction was devised by Karis Mullis (Mullis 1990). It is an in vitro method for amplifying specific DNA sequences. Starting with trace amounts of a particular nucleic acid sequence from any source, PCR enzymatically generate millions of copies of a specific DNA sequence. This automated process by passes the need to use bacteria for amplifying DNA. This mimics what happens in nature. DNA can replicate itself during cell division and is the only material that is passed on from parents to offspring. A DNA strand is held together by weak hydrogen bonds between complementary base pairs. These bonds can break easily, causing the DNA to separate or unzip into two half-strands. Free nucleotides present in the nuclear material then arrange themselves along each half strand. Hydrogen bonds form between the complementary bases, resulting into two identical DNA strands. The starting DNA is called template DNA. The primers anneal to the region to be amplified and extended and then amplified. The ladder used during gel electrophoresis of this work was 1kb plus from Thermo Scientific. The primer SAA and SBB amplified seven(7) samples, while primer CP1 and TRs amplified four (4) samples. The primer CP1 and TSer amplified seven (7) samples, whereas the primers LP01 and LP02 amplified eleven (11) samples. The most effective primers out of the four pair of the forward and reverse primers were LP01 and LP 02 for the amplification of the DNA of the stem borers, while the least effective of the pairs of the primers were CP1 and TRs (Table 51) . Using CP1 and TRs primers, the base pairs of the amplified genes ranged from 600 to 1650bp, whereas the base pairs for the stem borers DNA using CP1 and TSer primers ranged from 600 to 1650bp. The base pairs of the stem borers amplified using 16SAA and 16SBB primers ranged from 600 to 1780bp. The base pairs of the stem borers amplified using LP 01 and LP 02 primers ranged from 600 to 1650 bp. (Table 52).

Table 52: Analysis showing Samples amplified with primers

Sample	Primers			
	SAA&SBB	CPI & TRs	CPI & Tser	LP01 & LP02
1	-	-	+	-
2	+	-	+	+
3	+	-	-	+
4	+	-	-	+
5	+	-	+	+
6	+	-	+	+
7	-	+	-	+
8	+	+	-	+
9	-	-	+	+
10	—	+	+	+
11	+	-	+	+
12	—	+	-	+

Table 53: Analysis showing the primers and sequence with number of samples amplified by the primers and the amplicon size.

Primers	Sequence (5' -3')	No of sample amplified	Amplicon size (bp)
SAA&SBB	F 32	7	600-1780bp
	R 34		
CPI & TRs	F 19	4	600-1650bp
	R 21		
CPI & Tser	F 19	7	600- 1650bp
	R 25		
LP01 & LP02	F 32	11	650- 1650bp
	R 32		

### **4.3.7 SEQUENCING**

The principles of DNA replication were used by Sanger et al. (1974) in the development of the process now known as Sanger dideoxy sequencing. This process takes advantage of the ability of DNA polymerase to incorporate 2', 3'-dideoxynucleotides, nucleotide base analogs that lack the 3'-hydroxyl group essential in phosphodiester bond formation. Sanger dideoxy sequencing requires a DNA template, a sequencing primer, DNA polymerase, nucleotides (dNTPs), dideoxynucleotides (ddNTPs), and reaction buffer. Four separate reactions are set up, each containing radioactively labeled nucleotides and either ddA, ddC, ddG, or ddT. The annealing, labeling, and termination steps are performed on separate heat blocks. DNA synthesis is performed at 37 °C, the temperature at which the T7 DNA polymerase used has the optimal enzyme activity.

DNA polymerase adds either a deoxynucleotide or the corresponding 2', 3'-dideoxynucleotide at each step of chain extension. Whether a deoxynucleotide or a dideoxynucleotide is added depends on the relative concentration of both molecules.

When a deoxynucleotide (A, C, G, or T) is added to the 3' end, chain extension can continue. However, when a dideoxynucleotide (ddA, ddC, ddG, or ddT) is added to the 3' end, chain extension terminates. Sanger dideoxy sequencing results in the formation of extension products of various lengths terminated with dideoxynucleotides at the 3' end.

### **4.3.8 ELECTROPHORESIS**

The extension products are then separated by electrophoresis. During electrophoresis, an electrical field is applied so that the negatively charged DNA fragments move toward the positive electrode. The speed at which a DNA fragment moves through the medium is inversely proportional to its molecular weight. This process of electrophoresis can separate the extension products by size at a resolution of one base.

### **4.3.9 Applied Biosystems Automated DNA Sequencing**

Applied Biosystems fluorescence-based cycle sequencing system is an extension and refinement of Sanger dideoxy sequencing. Applied Biosystems automated DNA sequencing generally follows this flow:

1. Template preparation 2. Cycle sequencing 3. Purification after cycle sequencing 4. Capillary electrophoresis 5. Data analysis

#### **4.4.0 Cycle Sequencing Process Overview**

Like Sanger sequencing, fluorescence-based cycle sequencing requires a DNA template, a sequencing primer, a thermal stable DNA polymerase, nucleotides (dNTPs), dideoxynucleotides (ddNTPs), and buffer. But unlike Sanger's method, which uses radioactive material, cycle sequencing uses fluorescent dyes to label the extension products and the components are combined in a reaction that is subjected to cycles of annealing, extension, and denaturation in a thermal cycler. Thermal cycling the sequencing reactions creates and amplifies extension products that are terminated by one of the four dideoxynucleotides. The ratio of deoxynucleotides to dideoxynucleotides is optimized to produce a balanced population of long and short extension products.

#### **How Extension Products are labeled**

Automated cycle sequencing procedures incorporate fluorescent dye labels using either dye-labeled dideoxynucleotide triphosphates (dye terminators) or dye-labeled primers (dye primers). Both chemistries use four different dyes. Because each dye emits a unique wavelength when excited by light, the fluorescent dye on the extension product identifies the 3' terminal dideoxynucleotide as A, C, G, or T.

#### **Dye Terminator Chemistry**

With dye terminator chemistry, each of the four dideoxynucleotide terminators is tagged with a different fluorescent dye. One reaction is performed, containing the enzyme, nucleotides, and all dye-labeled dideoxynucleotides. The products from this reaction are injected into one capillary.

## DENATURATION ANNEALING EXTENSION PRODUCTS

### Dye Primer Chemistry

With dye primer chemistry, four separate tubes of sequencing primer are each tagged with a different fluorescent dye. Four separate reactions are performed, each containing the enzyme, nucleotides, and a specific dye-labeled sequencing primer, and either A, C, G, or T dideoxynucleotides. The products from these four reactions are then combined and injected into one capillary. However the one used for this work is

### Cycle Sequencing Kits

Applied Biosystems cycle sequencing kits available for both dye primer and dye terminator chemistries:

- BigDye® Terminator v1.1 and v3.1 Cycle Sequencing Kits
- dGTP BigDye® Terminator v1.0 and v3.0 Cycle Sequencing Kits
- dRhodamine Terminator Cycle Sequencing Kits
- BigDye® Primer Cycle Sequencing Kits.

However the one used for this work was Big Dye® Terminator v1.1 and v3.1 cycle sequencing kits.

### Modified DNA polymerase

The cycle sequencing reaction is directed by highly modified, thermally stable DNA polymerases. These enzymes have been carefully selected to allow incorporation of dideoxynucleotides, to process through stretches of G-C-rich and other difficult sequences, and to produce uniform peak heights. The modified DNA polymerases are also formulated with a pyrophosphatase to prevent reversal of the polymerization reaction (pyrophosphorolysis).

### Emission Spectra of Fluorescent Dyes

The fluorescent dyes used in BigDye® terminators, BigDye® primers, and dRhodamine terminators have narrower emission spectra and less spectral overlap than the rhodamine dyes used in previous sequencing kits. As a result, the dyes produce less noise.

## **Capillary Electrophoresis**

Historically, DNA sequencing products were separated using polyacrylamide gels that were manually poured between two glass plates. Capillary electrophoresis using a denaturing flowable polymer has largely replaced the use of gel separation techniques due to significant gains in workflow, throughput, and ease of use. Fluorescently labeled DNA fragments are separated according to molecular weight. Because you do not need to pour gels with capillary electrophoresis, you can automate DNA sequence analysis more easily and process more samples at once.

### **4.4.1 Capillary Electrophoresis Process Overview**

During capillary electrophoresis, the extension products of the cycle sequencing reaction enter the capillary as a result of electrokinetic injection. A high voltage charge applied to the buffered sequencing reaction forces the negatively charged fragments into the capillaries. The extension products are separated by size based on their total charge. The electrophoretic mobility of the sample can be affected by the run conditions: the buffer type, concentration, and pH; the run temperature; the amount of voltage applied; and the type of polymer used. Shortly before reaching the positive electrode, the fluorescently labeled DNA fragments, separated by size, move across the path of a laser beam. The laser beam causes the dyes on the fragments to fluoresce. An optical detection device on Applied Biosystems genetic analyzers detects the fluorescence. The Data Collection

Software converts the fluorescence signal to digital data, then records the data in a \*.ab1 file. Because each dye emits light at a different wavelength when excited by the laser, all four colors, and therefore all four bases, can be detected and distinguished in one capillary injection.

### **4.4.2 DNA Sequencing Data Analysis Process Overview**

Data analysis software processes the raw data in the \*.ab1 file using algorithms and applies the following analysis settings to the results:

- Multicomponent analysis – Each fluorescent dye emits its maximum fluorescence at a different wavelength, but there is some overlap in the emission spectra. Thus a signal



generated primarily in one color channel may yield a lower signal in an adjacent color channel. Multicomponent analysis separates the four different fluorescent dye signals into distinct spectral components by mathematically filtering fluorescence signal from dyes with emission spectra overlap.

- Basecalling – The selected basecaller processes the fluorescence signals, then assigns a base to each peak (A, C, G, T, or N). If the KB™ basecaller is used, it also provides per-base quality value predictions, optional mixed base calling, and automatic identification of failed samples.
- Mobility shift correction – The mobility file corrects electrophoretic mobility changes imposed by the presence of different fluorescent dye molecules associated with differently labeled reaction extension product. The mobility file also corrects for the differences between the dye-to-nucleotide relationships in the raw data and the analyzed data.
- Quality value determination (QV) – If the KB basecaller is used for analysis, the software assigns a QV for each base. The QV predicts the probability of a basecall error. For example, a QV of 20 predicts an error rate of 1%. The quality prediction algorithm is calibrated to return QVs that conform to the industry standard relationship established by the Phred software. If your pipeline involves analysis with Phred software to assign QVs after the data is basecalled, you can simplify your workflow and use the KB basecaller instead. The KB basecaller can perform basecalling and assign QVs. Then, you can generate phd.1 or \*.scf files using the KB basecaller to integrate with your downstream pipeline.

In summary PCR product was purified using the Qiagen purification kit (Moyal et al 2011a; Moyal and Le Ru 2006; Moyal et al 2011b; Moyal 2015; Chai and Du 2012). Sequencing reactions were carried out using the Sanger dideoxy method (Sanger et al 1977). This was done by performing DNA sequencing reaction using an ABI PRISM® Big Dye® Terminator v3.1 cycle sequencing kit (Applied Biosystems), cleaned using ethanol/EDTA precipitation (Ongamo et al. 2008), and finally, sequences were run and detected on ABI 3130×L automated capillary sequencer (Genetic Analyzer) Moyal et al 2011a; Moyal et al 2011b; Esfandiari et al 2015; Moyal and Le Ru 2006; Moyal 2015). Result of sequencing is as below:

## RESULTS OF MOLECULAR STUDY

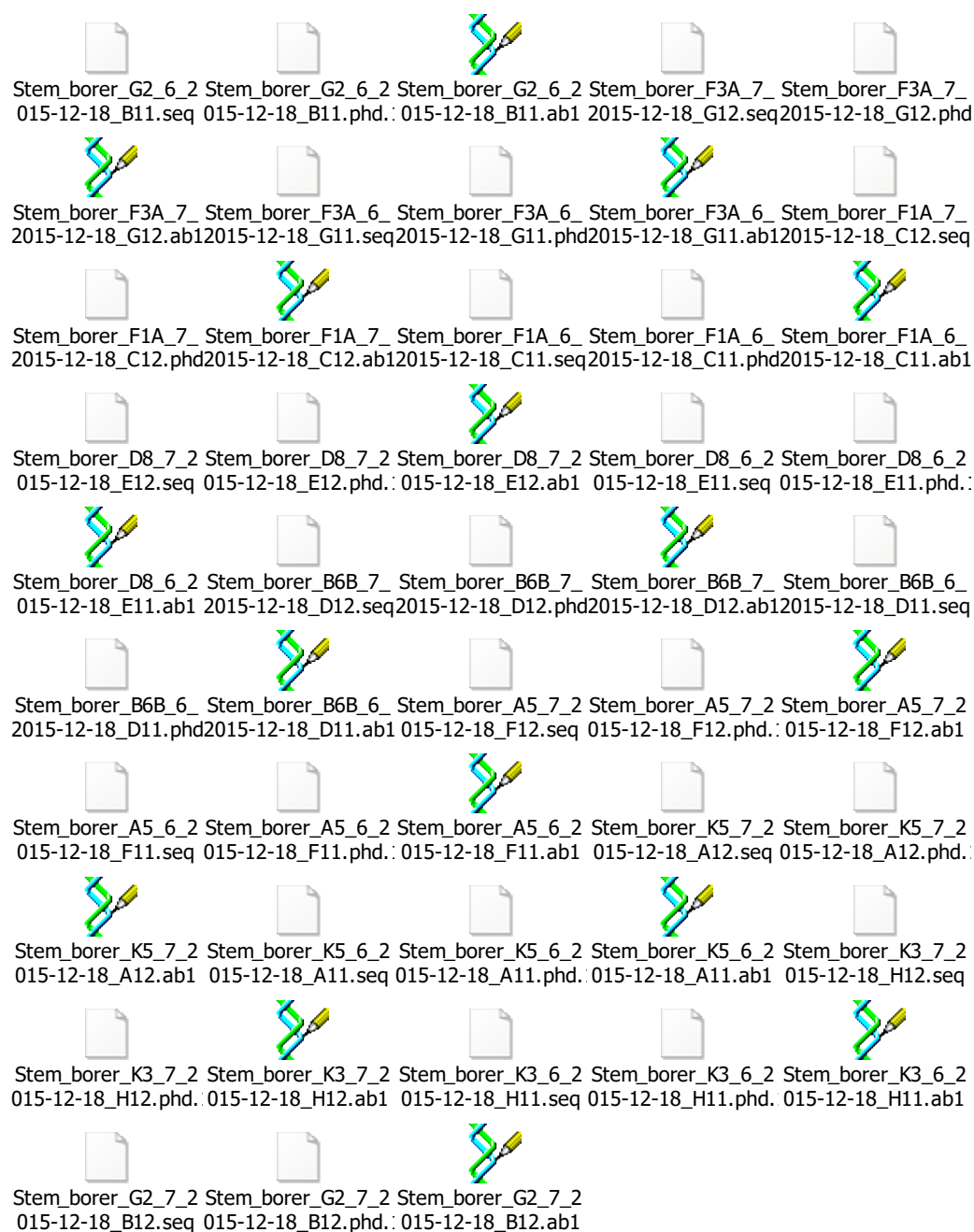


Fig 15: Result of sequencing



Fig 16: ABI 3130 xL GENETIC ANALYZER OPEN



Fig 17: ABI 3130×L GENETIC ANALYZER CLOSED

DNA:

Nucleotide Code: Base:

A.....Adenine

C.....Cytosine

G.....Guanine

T .....Thymine

R.....A or G

Y.....C or T

S.....G or C

W.....A or T

K.....G or T

M.....A or C

B.....C or G or T

D.....A or G or T

H.....A or C or T

V.....A or C or G

N.....any base

. or -.....gap

#### 4.4.3 NUCLEOTIDE BASES OF THE STEM BORERS ANALYSED

Stem\_borer\_A5\_6\_2015-12-18\_F11 (295 nucleotides)

CGGTTTCAAGCGGGCGAGTGTATATGAGGTGGGTTTAGATTTAATCGACCTGGAT  
TAGCATCTACTTTTACCCCTAAAGATGGGATTGTTTCATGAGTGGATTACATCTGT  
AGCTGTTACTATAATTCGAATTTGATTATTTATTGGTAAAATAATACGATTATCTA  
CATCTAAAAGTCGAAAATTATTATTTTTTAAATCTTTTGAGGGAATTATGTAAGA  
ATCAAATTCTACATTATTAATAATCAGAGTATTCGTATCTTCAATATCATTCTGGTT  
CAATATGAACTGGTGTTA

Stem borer sample “Stem\_borer\_A5\_6\_2015-12-18\_F11” has 295 nucleotides and was identified to be *Chilo orichalcociliellus*.

Stem\_borer\_A5\_7\_2015-12-18\_F12 (291 nucleotides)

GCGAGTCCAGGGTTGAATGTTAAGAGAGCTTTTTAGAGTCTTCAACCTACCCTAA  
CATGAACTTTTAACCTAATTTTCGACTTTTAGATGTAGAGAATCGTATTATTTTAC  
CAATAAATAATCAAATTCGAATTATAGTAACAGCTACAGATGTAATCCACTCATG  
AACAAATCCCATCATTAGGGGTAAAAGTAGATGCTAATCCAGGTCGATTAAATCA  
AACAAATTTTCTTATCAATCGCCCTGGAATTTTTTATGGTCAATGTTTCAGAAATTT  
GTGGAGCTAATCAAA

Stem borer sample “Stem\_borer\_A5\_7\_2015-12-18\_F12” has 291 nucleotides and was also identified to be *Chilo orichalcociliellus*.

Stem\_borer\_B6B\_6\_2015-12-18\_D11 (294 nucleotides)

CACATATCAAGGGGCCGGATTTTAATGAGATTAGTTTGGTTAAGGCGACCAGGGT  
TAGCATCTACTTTTACTCCTAAAGCTGGAATAGTTCAGGAATGGATTACATCTGT  
GGCTGTAATAAAATTCGAATTTGATTATTTATTGGTAAAATAATTCGATTATCTA  
CATCTAATAGGCGAAAATTATTAGAAGAAAGTTCATTTCTAGAGATTATGTAGGA  
ATCAAATTCAATATTAATAAAATCTGAATATTCATATCTTCAATATCATTCTGGTT  
CAATATGAACTGGTGTT

Stem borer sample “Stem\_borer\_B6B\_6\_2015-12-18\_D11” has 294 nucleotides and was identified to be *Eldana saccharina*.

Stem\_borer\_B6B\_7\_2015-12-18\_D12 (290 nucleotides)

TAAATTCCTGAGTCAGTTTAATTTGAGATCGGTTTGGTTATTAGTGGTCTCTTCAA  
TCTACTTTCTTCTATAATTTTCGCCTATTAGATGTAGATAATCGAATTATTTTACC

AATAAATAATCAAATTCGAATTTTAGTTACAGCCACAGATGTAATCCATTCCTGA  
ACTATTCCAGCTTTAGGAGTAAAAGTAGATGCTAACCCTGGTCGCCTTAACCAAA  
CTAATTTTTTTATTAATCGCCCTGGAATTTTTTATGGTCAATGTTTCAGAAATTTGT  
GGAGCTAATCAA

Stem borer sample “Stem\_borer\_B6B\_7\_2015-12-18\_D12” has 290 nucleotides and was identified to be *Eldana saccharina*.

Stem\_borer\_D8\_6\_2015-12-18\_E11 (294 nucleotides)

AAAAATTCTTCCGTTGATTTTTATTGGTTTTATTTAATGGAGTAAACCAGGGTTAG  
AATCTACTTTTACTCCTAAAGCTGGAATAGTTCAGGAATGGATTACATCTGTGGC  
TGTAATAAAATTCGAATTTGATTATTTATTGGTAAAATAATTCGATTATCTACAT  
CTAATAGGCGAAAATTATTAGAAGAAAGTTCATTTCTAGAGATTATGTAGGAATC  
AAATTCAATATTAAAAAAATCTGAATATTCATATCTTCAATATCATTCTGGTTCA  
ATATGAACTGGTGTAA

Stem borer sample “Stem\_borer\_D8\_6\_2015-12-18\_E11” has 294 nucleotides and was identified to be *Eldana saccharina*.

Stem\_borer\_D8\_7\_2015-12-18\_E12 (290 nucleotides)

GGAGGCCCGGGCTTTTTTAGTGGAGGGGAGTTGGGTCTTAAACCTGGCTAGAATG  
ACTTGTTTCCAAAGTTTCGCCTATGAGAGGTAGGTAATCGTATGATTTTACCAAT  
AAATATCCAAATTCGAATTTTAGTTGCAGCCACAGATGAAATCCATTCCTGAACT  
ATTCCAGCTTTAGGAGTAAAAGTAGATGCTAACCCTGGTCGCCTTAACCAAACTA  
ATTTTTTTATTAATCGCCCTGGAATTTTTTATGGTCAATGTTTCAGAAATTTGTGGA  
GCTTAATCAAAA

Stem borer sample “Stem\_borer\_D8\_7\_2015-12-18\_E12” has 290 nucleotides and was identified to be *Eldana saccharina*.

Stem\_borer\_F1A\_6\_2015-12-18\_C11 (295 nucleotides)

TAAATTCCGGGGTTAGTTTAGTACTGGTTTAATTTGGTTTAGGCGACCAGGGTTA  
GCATCTACTTTTACTCCTAAAGCTGGAATAGTTCAGGAATGGATTACATCTGTGG  
CTGTAACTAAAATTCGAATTTGATTATTTATTGGTAAAATAATTCGATTATCTACA  
TCTAATAGGCGAAAATTATTAGAAGAAAGTTCATTTCTAGAGATTATGTAGGAAT  
CAAATTCAATATTAAAAAAATCTGAATATTCATATCTTCAATATCATTCTGGTTCA  
ATATGAACTGGTGTAAAA

Stem borer sample “Stem\_borer\_F1A\_6\_2015-12-18\_C11” had 295 nucleotides and was identified as *Eldana saccharina*.

Stem\_borer\_F1A\_7\_2015-12-18\_C12 (294 nucleotides)

AAAATGGCTTGGAGAGTTGTTGGGGGTATCGAATTGGATTCTCGAACTGTTTAGA  
ATGAACTTTCTTCTAAAATTTTCGCCTATTGGTGTAGATAATCGAATGTTTTTACC  
AATAAAAAATCAAATTCGAATTTTAGTGACAGCCACAGATGTAATCCATTCTGA  
ACTATTCCAGCTTTAGGAGTAAAAGTAGATGCTAACCCTGGTCGCCTTAACCAAA  
CTAATTTTTTTTATTAATCGCCCTGGAATTTTTTATGGTCAATGTTTCAGAAATTTGT  
GGAGCTAATCAAAAATT.

Stem borer sample “Stem\_borer\_F1A\_7\_2015-12-18\_C12” had 294 nucleotides and was identified as *Eldana saccharina*.

Stem\_borer\_F3A\_6\_2015-12-18\_G11 (280 nucleotides)

TAAAAAGGTAATTTTTTTTATGTTAAGGCAACCAGGGTTAGCATCTACTTTTACTCC  
TAAAGCTGGAATAGTTCAGGAATGGATTACATCTGTGGCTGTAATAAAATTCGA  
ATTTGATTATTTATTGGTAAAATAATTCGATTATCTACATCTAATAGGCGAAAATT  
ATTAGAAGAAAGTTCATTTCTAGAGATTATGTAGGAATCAAATTCAATATTAATA  
AAATCTGAATATTCATATCTTCAATATCATTCTGGTTCAATATGAACTGGTGTTAA  
A

Stem borer sample “Stem\_borer\_F3A\_6\_2015-12-18\_G11” had 280 nucleotides and was identified as *Eldana saccharina*.

Stem\_borer\_F3A\_7\_2015-12-18\_G12 (296 nucleotides)

GAAAAATTCCTAGGGACGGTGTGGTTTCGGATCAATTTTCGATTCCTAGTAGTCTC  
TAGAATGACTTTCTTCTAATAATTTTCGCCTATTAGATGTAGATAATCGAATTATT  
TTACCAATAAATAATCAAATTCGAATTTTAGTTACAGCCACAGATGTAATCCATT  
CCTGAACTATTCCAGCTTTAGGAGTAAAAGTAGATGCTAACCCTGGTCGCCTTAA  
CCAACTAATTTTTTTTATTAATCGCCCTGGAATTTTTTATGGTCAATGTTTCAGAAA  
TTTGTGGAGCTAATCAATA

Stem borer sample “Stem\_borer\_F3A\_7\_2015-12-18\_G12” had 296 nucleotides and was identified as *Eldana saccharina*.

Stem\_borer\_G2\_6\_2015-12-18\_B11 (293 nucleotides)

GAAATTCCCGGGTTTCGAATAGTACTGGAGTAATTTGATTTAGGCGACCAGGGTTA  
GCATCTACTTTTACCCCTAAAGCTGGAATAGTTCAGGAATGGATTACATCTGTGG  
CTGTAATAAAATTCGAATTTGATTATTTATTGGTAAAATAATTCGATTATCTACA  
TCTAATAGGCGAAAATTATTAGAAGAAAGTTCATTTCTAGAGATTATGTAGGAAT  
CAAATTCAATATTAATAAAAAATCTGAATATTCATATCTTCAATATCATTCTGGTTCA  
ATATGAACTGGTGTTA



Stem borer sample “Stem\_borer\_G2\_6\_2015-12-18\_B11” had 293 nucleotides and was identified as *Eldana saccharina*.

Stem\_borer\_G2\_7\_2015-12-18\_B12 (294 nucleotides)

AAATAAGGTCATGTGCGGTTTGGTGGGGATGAAATTGAATCTTAATAATCTCTAG  
AATGAACTTTCTTCTAATAATTTTCGCCTATTAGATGTAGATAATCGAATTATTTT  
ACCAATAAATAATCAAATTCGAATTTTAGTTACAGCCACAGATGTAATCCATTCC  
TGAAC TATTCCAGCTTTAGGGGTAAAAGTAGATGCTAACCCCTGGTCGCCTTAACC  
AAACTAATTTTTTTTATTAATCGCCCTGGAATTTTTTTATGGTCAATGTTTCAGAAATT  
TGTGGAGCTAATCAAAA

Stem borer sample “Stem\_borer\_G2\_7\_2015-12-18\_B12” had 294 nucleotides and was identified as *Eldana saccharina*.

Stem\_borer\_K5\_6\_2015-12-18\_A11 (294 nucleotides)

GATTTTCGCCTGGCCGGTTATTGAAAAAATGGGTTTGATTTAGCATCCTGGATTTG  
CATCTACTTTAACTCCTAAGGATGGAATAGTTCAAGAGTGAATAACATCTGTAGC  
AGTAACTAAAATTCGAATTTGATTATTTAAAGGTAAAATAATTCGATTATCAACA  
TCTAAAAGACGAAAATTATTGGATGATATTTTCATTGGTGGGGATTATATAGGAGT  
CAAATTCAATTTTATTGAAATCTGAATATTCATAACTTCAATATCATTCTGGTTCA  
ATATGAACTGGTGTAAA

Stem borer sample “Stem\_borer\_K5\_6\_2015-12-18\_A11” has 294 nucleotides and was identified as *Sesamia calamistis*.

Stem\_borer\_K5\_7\_2015-12-18\_A12 (294 nucleotides)

GATGGGGCAAGGGGGTTTTTGGGAGGGATTGTATTTTGACTCCTCTATAATGCCC  
ACCAATGAAATATCATCCAATAATTTTCGTCTTTTAGATGTTGATAATCGAATTAT  
TTTACCTTTAAATAATCAAATTCGAATTTTAGTTACTGCTACAGATGTTATTCACT  
CTTGAAC TATTCCATCCTTAGGAGTTAAAGTAGATGCAAATCCAGGACGTTTAAA  
TCAAAC TAAATTTTTTCATTAATCGTCCTGGTATTTTTTATGGTCAATGTTTCAGAAA  
TTTGTGGAGCTAATCA

Stem borer sample “Stem\_borer\_K5\_7\_2015-12-18\_A12” had 294 nucleotides and was identified as *Sesamia calamistis*.

#### 4.4.4 PHYLOGENETIC TREES

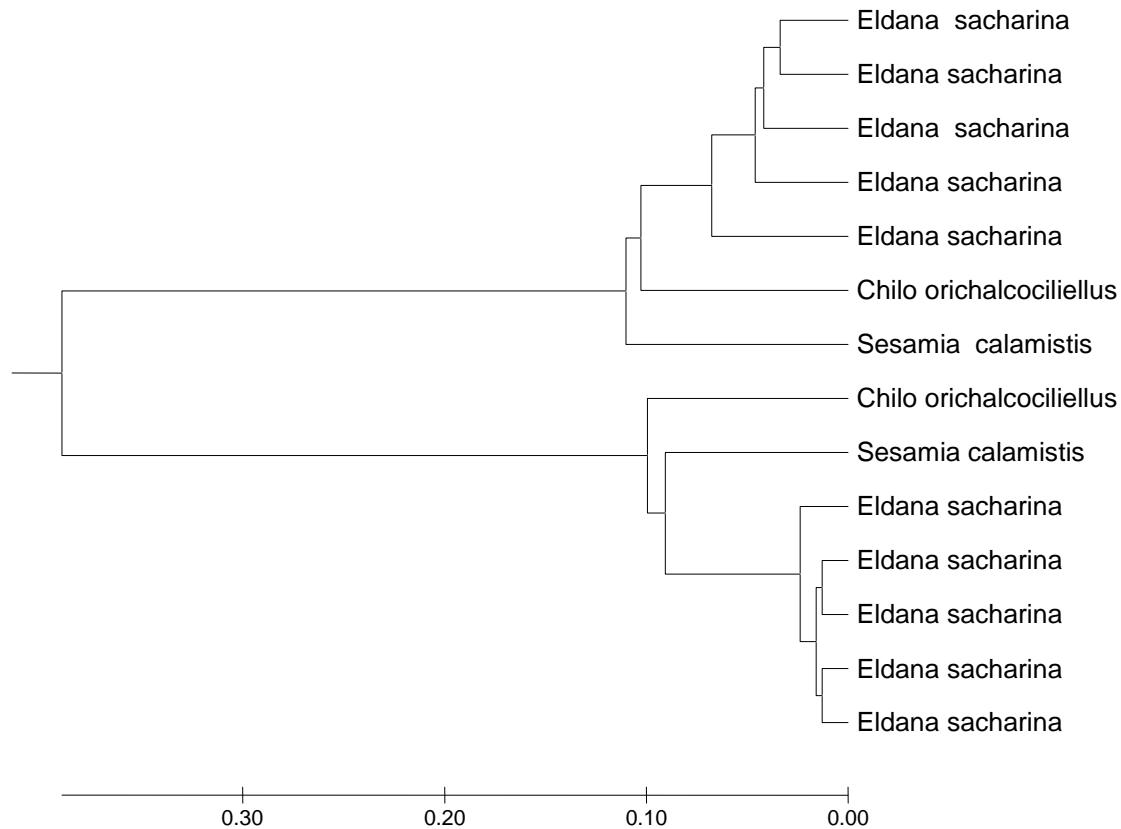


Figure18: Evolutionary relationships of taxa

The evolutionary history was inferred using the UPGMA method (Sneath and Sokal 1973). The optimal tree with the sum of branch length = 1.43733580 is shown. The tree is drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the phylogenetic tree. The evolutionary distances were computed using the Maximum Composite Likelihood method (Tamura et. al., 2004) and are in the units of the number of base substitutions per site. The analysis involved 14 nucleotide sequences. All positions containing gaps and missing data were eliminated. There were a total of 235 positions in the final dataset. Evolutionary analyses were conducted in MEGA7 (Kumar et. al., 2016).



Figure 19: Evolutionary relationships of taxa

The evolutionary history was inferred using the UPGMA method (Sneath and Sokal 1973). The optimal tree with the sum of branch length = 1.43733580 is shown. The tree is drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the phylogenetic tree. The evolutionary distances were computed using the Maximum Composite Likelihood method (Tamura et. al., 2004) and are in the units of the number of base substitutions per site. The analysis involved 14 nucleotide sequences. All positions containing gaps and missing data were eliminated. There were a total of 235 positions in the final dataset. Evolutionary analyses were conducted in MEGA7 (Kumar et. al., 2016).

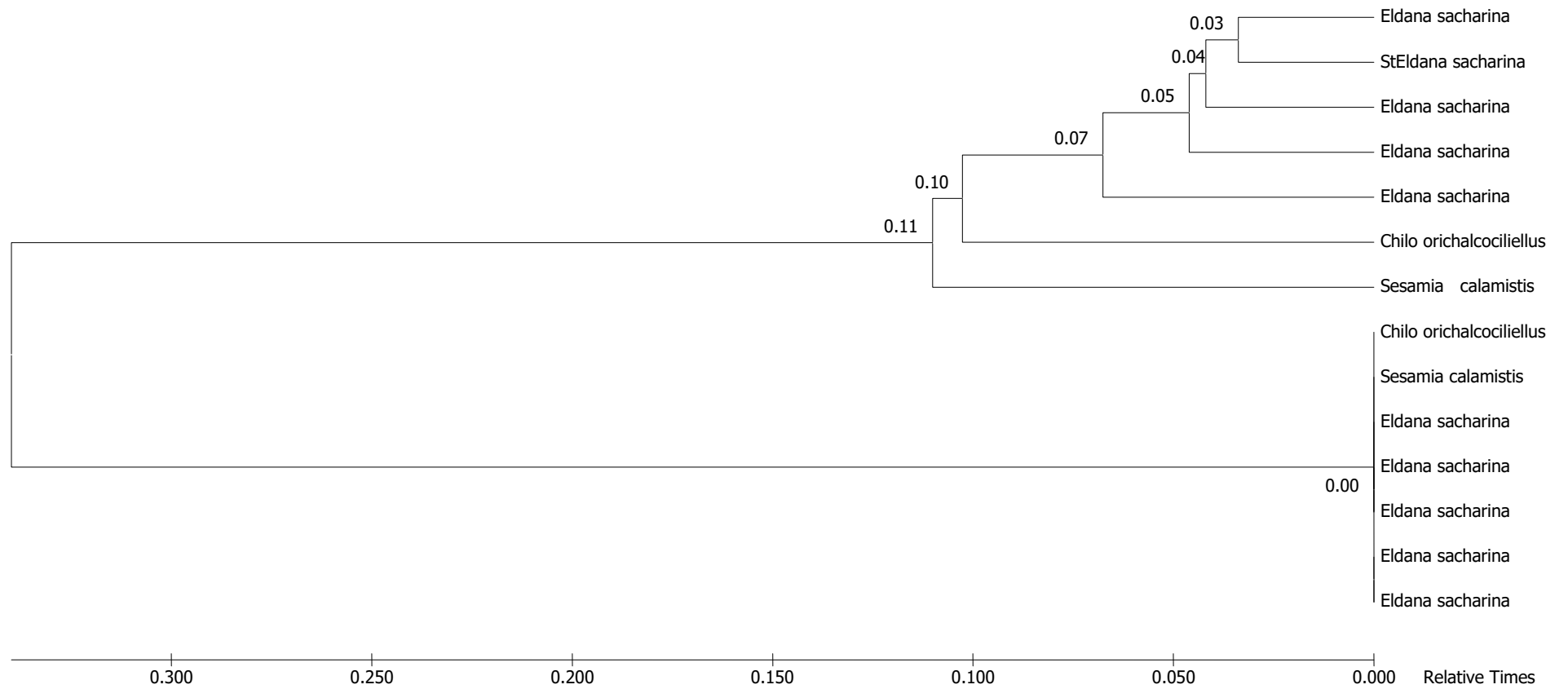


Figure 20: Evolutionary relationships of taxa (timetree)

A timetree inferred using the Reltime method (Tamura et.al, 2012) and estimates of branch lengths inferred using the UPGMA method (Sneath and Sokal 1973). The analysis involved 14 nucleotide sequences. All positions containing gaps and missing data were eliminated. There were a total of 235 positions in the final dataset. Evolutionary analyses were conducted in MEGA7 (Kumar et.al, 2016).

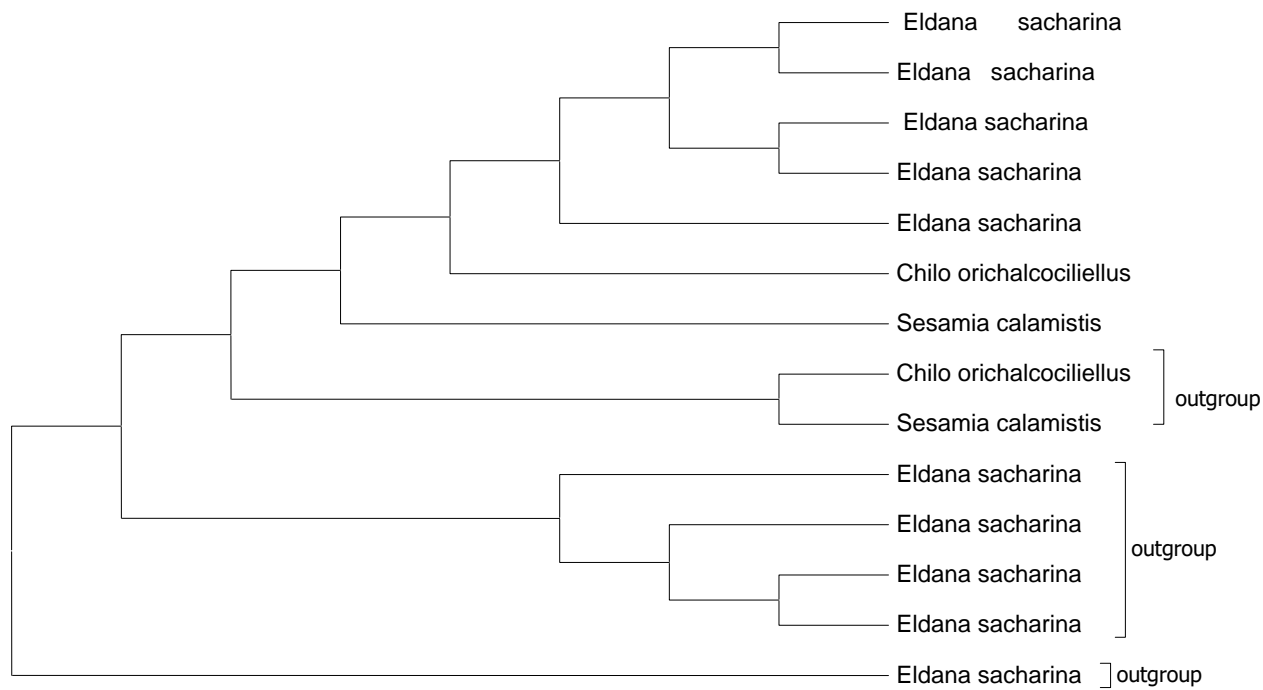


Figure 21: Maximum Parsimony analysis of taxa

The evolutionary history was inferred using the Maximum Parsimony method. Tree #1 out of 6 most parsimonious trees (length = 278) is shown. The consistency index is 0.787770 (0.753138), the retention index is 0.898100 (0.898100), and the composite index is 0.707496 (0.676393) for all sites and parsimony-informative sites (in parentheses). The MP tree was obtained using the Subtree-Pruning-Regrafting (SPR) algorithm (Nei and Kumar 2000) with search level 0 in which the initial trees were obtained by the random addition of sequences (10 replicates). The analysis involved 14 nucleotide sequences. All positions containing gaps and missing data were eliminated. There were a total of 235 positions in the final dataset. Evolutionary analyses were conducted in MEGA7 (Kumar et.al. 2016).







Figure 23: Maximum Parsimony analysis of taxa

The evolutionary history was inferred using the Maximum Parsimony method. The consensus tree inferred from 6 most parsimonious trees is shown. Branches corresponding to partitions reproduced in less than 50% trees are collapsed. The consistency index is 0.787770 (0.753138), the retention index is 0.898100 (0.898100), and the composite index is 0.707496 (0.676393) for all sites and parsimony-informative sites (in parentheses). The MP tree was obtained using the Subtree-Pruning-Regrafting (SPR) algorithm (Nei and Kumar 2000) with search level 0 in which the initial trees were obtained by the random addition of sequences (10 replicates). The analysis involved 14 nucleotide sequences. All positions containing gaps and missing data were eliminated. There were a total of 235 positions in the final dataset. Evolutionary analyses were conducted in MEGA7 (Kumar et. al., 2016).

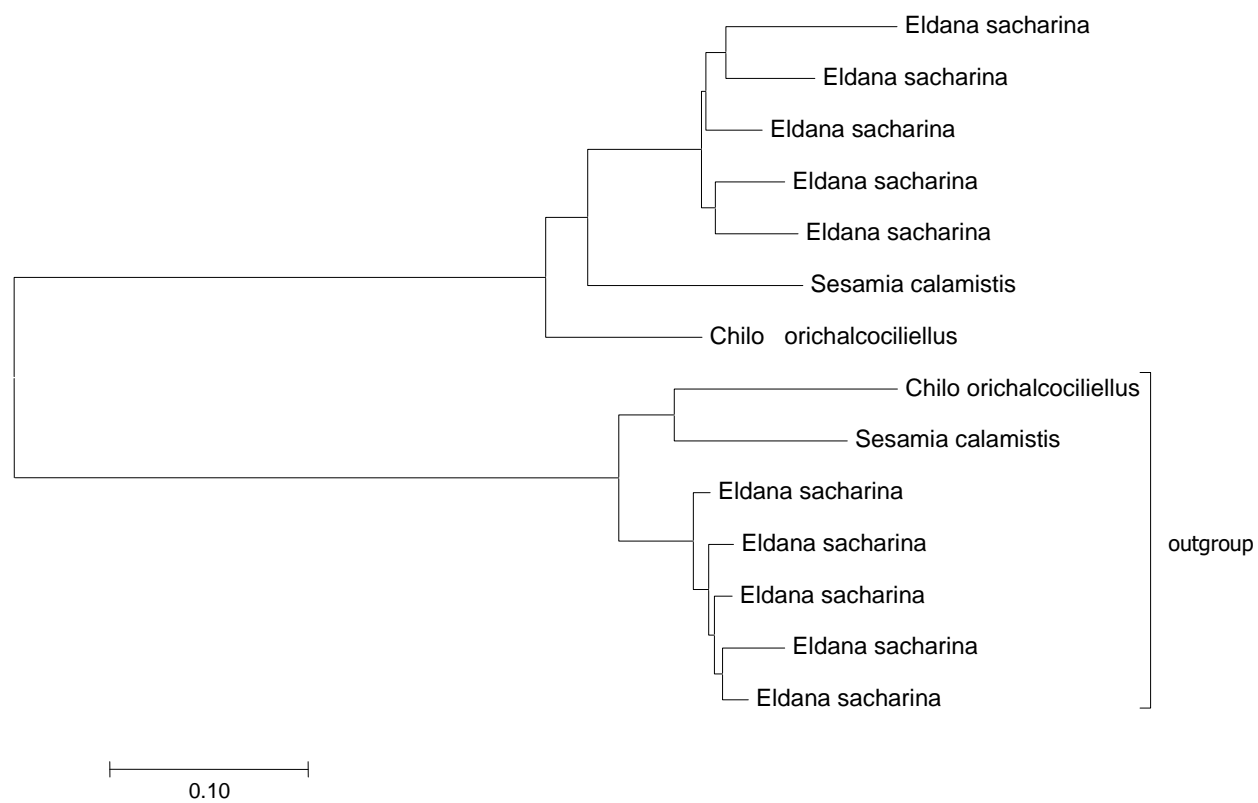


Figure 24: Evolutionary relationships of taxa

The evolutionary history was inferred using the Minimum Evolution method (Rzhetsky and Nei 1992). The optimal tree with the sum of branch length = 1.44690300 is shown. The tree is drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the phylogenetic tree. The evolutionary distances were computed using the Maximum Composite Likelihood method (Tamura et. al., 2004) and are in the units of the number of base substitutions per site. The ME tree was searched using the Close-Neighbor-Interchange (CNI) algorithm (Nei and Kumar 2000) at a search level of 1. The Neighbor-joining algorithm (Saitou and Nei 1987) was used to generate the initial tree. The analysis involved 14 nucleotide sequences. All positions containing gaps and missing data were eliminated. There were a total of 235 positions in the final dataset. Evolutionary analyses were conducted in MEGA7 (Kumar et. al., 2016).

Table 54: Estimates of Evolutionary Divergence between Sequences

Sesamia calamistis														
Chilo {outgroup}	0.837													
C. orichalcociliellus	0.214	0.815												
E. sacharina {outgroup}	0.763	0.188	0.688											
Eldana sacharina	0.203	0.884	0.205	0.766										
Eldana sacharina {outgroup}	0.805	0.217	0.746	0.044	0.780									
Eldana sacharina	0.264	0.906	0.234	0.813	0.132	0.820								
Eldana sacharina {outgroup}	0.777	0.200	0.710	0.026	0.707	0.054	0.788							
Eldana sacharina	0.229	0.856	0.204	0.784	0.102	0.770	0.131	0.770						
Eldana sacharina {outgroup}	0.777	0.206	0.730	0.026	0.742	0.044	0.820	0.026	0.780					
Eldana sacharina	0.208	0.891	0.198	0.752	0.077	0.776	0.152	0.739	0.092	0.759				
Eldana sacharina {outgroup}	0.766	0.183	0.675	0.031	0.733	0.049	0.798	0.026	0.770	0.044	0.723			
Eldana sacharina	0.203	0.828	0.186	0.736	0.068	0.719	0.126	0.756	0.082	0.745	0.091	0.739		
S. calamistis {outgroup}	0.749	0.199	0.760	0.172	0.810	0.195	0.887	0.178	0.806	0.183	0.828	0.178	0.806	

Table 55: Test of the Homogeneity of Substitution Patterns between Sequences

Sesamia calamistis		0.000	0.770	0.000	0.000	0.123	0.574	0.000	0.111	0.000	0.021	0.153	0.000	0.000
C. orichalcociliellus {outgroup}	1.000		0.711	0.149	0.000	0.370	0.511	0.187	0.000	0.187	0.000	0.409	0.000	0.009
C. orichalcociliellus	0.004	0.064		0.106	0.340	0.145	0.115	0.149	0.017	0.319	0.115	0.009	0.191	0.340
Eldana sacharina {outgroup}	1.000	0.148	0.282		0.000	0.013	0.379	0.000	0.000	0.013	0.000	0.009	0.000	0.000
Eldana sacharina	1.000	1.000	0.040	1.000		0.000	0.443	0.000	0.030	0.000	0.000	0.000	0.000	0.000
Eldana sacharina {outgroup}	0.256	0.038	0.282	0.304	1.000		0.694	0.000	0.000	0.000	0.000	0.000	0.000	0.000
Eldana sacharina	0.016	0.112	0.198	0.122	0.004	0.044		0.485	0.149	0.745	0.251	0.447	0.374	0.477
Eldana sacharina {outgroup}	1.000	0.112	0.246	1.000	1.000	1.000	0.106		0.000	0.004	0.000	0.004	0.000	0.000
Eldana sacharina	0.192	1.000	0.340	1.000	0.204	1.000	0.082	1.000		0.000	0.000	0.000	0.000	0.000
Eldana sacharina {outgroup}	1.000	0.110	0.162	0.258	1.000	1.000	0.064	0.392	1.000		0.000	0.034	0.000	0.000
Eldana sacharina	0.358	1.000	0.182	1.000	1.000	1.000	0.032	1.000	1.000	1.000		0.000	0.000	0.000
Eldana sacharina {outgroup}	0.284	0.010	0.378	0.318	1.000	1.000	0.108	0.378	1.000	0.130	1.000		0.000	0.000
Eldana sacharina	1.000	1.000	0.080	1.000	1.000	1.000	0.002	1.000	1.000	1.000	1.000	1.000		0.000
Sesamia calamistis {outgroup}	1.000	0.408	0.142	1.000	1.000	1.000	0.122	1.000	1.000	1.000	1.000	1.000	1.000	

The probability of rejecting the null hypothesis that sequences have evolved with the same pattern of substitution, as judged from the extent of differences in base composition biases between sequences (Disparity Index test, Kumar and Gadagkar 2001). A Monte Carlo test, Kumar (500 replicates) was used to estimate the P-values (Kumar and Gadagkar 2001), which are shown above the diagonal. P- Values smaller than 0.05 are considered significant. The estimates of the disparity index per site are shown for each sequence pair below the diagonal. The analysis involved 14 nucleotides sequences . All positions containing gaps and missing data were estimated. There were a total of 235 positions in the final dataset. Evolutionary analyses were conducted in MEGA 7(Kumar et.al. 2016).

**Table 56: NUCLEOTIDE COMPOSITION**

Domain: Data

	T(U)	C	A	G	Total
Sesamia calamistis	37.8	15.3	29.6	17.3	294.0
C. orichalcociliellus	38.0	12.5	31.2	18.3	295.0
C. orichalcociliellus	32.3	17.2	34.4	16.2	291.0
Eldana sacharina	34.7	12.9	34.4	18.0	294.0
Eldana sacharina	37.9	16.9	30.0	15.2	290.0
Eldana sacharina	37.4	11.6	35.4	15.6	294.0
Eldana sacharina	32.6	17.4	28.8	21.2	288.0
Eldana sacharina	35.9	12.2	34.2	17.6	295.0
Eldana sacharina	34.7	15.6	31.3	18.4	294.0
Eldana sacharina	35.8	11.8	36.6	15.8	279.0
Eldana sacharina	35.5	17.2	31.1	16.2	296.0
Eldana sacharina	34.5	13.3	34.5	17.7	293.0
Eldana sacharina	35.0	15.6	32.7	16.7	294.0
Sesamia calamistis	35.0	12.2	34.7	18.0	294.0
Avg.	35.5	14.4	32.8	17.3	292.2

Table 57: Maximum Likelihood Estimate of Substitution Matrix

	A	T/U	C	G
A	-	10.23	3.89	7.83
T/U	8.81	-	5.83	4.03
C	8.81	15.31	-	4.03
G	17.11	10.23	3.89	-

Each entry is the probability of substitution ( $r$ ) from one base (row) to another base (column). Substitution pattern and rates were estimated under the model (Tamura and Nei 1993). Rates of different transitional substitutions are shown in bold and those of transversionsal substitutions are shown in italics. Relative values of instantaneous  $r$  should be considered when evaluating them. For simplicity, sum of  $r$  values is made equal to 100, the nucleotide frequencies are A = 32.67%, T/U = 37.93%, C = 14.44%, and G = 14.95%. For estimating ML values, a tree topology was automatically computed. The maximum Log likelihood for this computation was -1489.944. The analysis involved 14 nucleotide sequences. All positions containing gaps and missing data were eliminated. There were a total of 235 positions in the final dataset. Evolutionary analyses were conducted in MEGA7 (Kumar et. al., 2016).



Table 58: Maximum Likelihood fits of 24 different nucleotide substitution models

Model	#Param	BIC	AICc	InL	Invariant
T92+G	28	3181.232983	3010.96899	-1477.235492	n/a
T92+G+I	29	3189.331626	3013.004726	-1477.235492	0
HKY+G	30	3193.654306	3011.265748	-1475.34751	n/a
T92+I	28	3197.729271	3027.465279	-1485.483636	0.180310138
TN93+G	31	3199.171931	3010.722965	-1474.057001	n/a
HKY+G+I	31	3201.752949	3013.303983	-1475.34751	0
GTR+G	34	3202.697991	2996.075317	-1463.672067	n/a
T92	27	3204.679021	3040.479184	-1493.007832	n/a
TN93+G+I	32	3207.276629	3012.768508	-1474.060029	0.00001
HKY+I	30	3210.413084	3028.024526	-1483.726899	0.182201636
GTR+G+I	35	3210.796654	2998.118586	-1463.672077	0
TN93+I	31	3215.773011	3027.324046	-1482.357542	0.187187956
HKY	29	3217.821291	3041.49439	-1491.480324	n/a
TN93	30	3224.428573	3042.040015	-1490.734644	n/a
GTR+I	34	3230.034833	3023.412159	-1477.340488	0.186865675
GTR	33	3237.403952	3036.837927	-1485.074369	n/a
JC+G	26	3279.831487	3121.697052	-1534.633387	n/a
K2+G	27	3284.459911	3120.260073	-1532.898277	n/a
JC+G+I	27	3287.93013	3123.730293	-1534.633387	0
K2+G+I	28	3292.558699	3122.294706	-1532.89835	0.00001
JC+I	26	3293.400823	3135.266388	-1541.418054	0.17733153
JC	25	3297.771329	3145.703542	-1547.652629	n/a
K2+I	27	3298.302953	3134.103115	-1539.819798	0.175859728
K2	26	3302.419298	3144.284863	-1545.927292	n/a

<b>Gamma</b>	<b>R</b>	<b>Freq A</b>	<b>Freq T</b>	<b>Freq C</b>
0.995807789	0.723172355	0.353039514	0.353039514	0.146960486
0.99585481	0.72317206	0.353039514	0.353039514	0.146960486
0.977866702	0.724717429	0.32674772	0.379331307	0.1443769
n/a	0.698214743	0.353039514	0.353039514	0.146960486
0.931524012	0.731667198	0.32674772	0.379331307	0.1443769
0.977871869	0.724717348	0.32674772	0.379331307	0.1443769
0.862891171	0.728506371	0.32674772	0.379331307	0.1443769
n/a	0.679328809	0.353039514	0.353039514	0.146960486
0.924624313	0.731801738	0.32674772	0.379331307	0.1443769
n/a	0.697537833	0.32674772	0.379331307	0.1443769
0.862584306	0.728597033	0.32674772	0.379331307	0.1443769
n/a	0.706975493	0.32674772	0.379331307	0.1443769
n/a	0.679809205	0.32674772	0.379331307	0.1443769
n/a	0.681975197	0.32674772	0.379331307	0.1443769
n/a	0.562823858	0.32674772	0.379331307	0.1443769
n/a	0.565463158	0.32674772	0.379331307	0.1443769
1.322304572	0.5	0.25	0.25	0.25
1.335441278	0.668827982	0.25	0.25	0.25
1.322334831	0.5	0.25	0.25	0.25
1.33549776	0.668827049	0.25	0.25	0.25
n/a	0.5	0.25	0.25	0.25
n/a	0.5	0.25	0.25	0.25
n/a	0.65600592	0.25	0.25	0.25
n/a	0.654860693	0.25	0.25	0.25

Freq G	A=>T	A=>C	A=>G	T=>A	T=>C	T=>G	C=>A	C=>T	C=>G	G=>A	G=>T	G=>C
0.146960486	0.09	0.04	0.07	0.09	0.07	0.04	0.09	0.16	0.04	0.16	0.09	0.04
0.146960486	0.09	0.04	0.07	0.09	0.07	0.04	0.09	0.16	0.04	0.16	0.09	0.04
0.149544073	0.1	0.04	0.07	0.09	0.07	0.04	0.09	0.18	0.04	0.15	0.1	0.04
0.146960486	0.1	0.04	0.07	0.1	0.07	0.04	0.1	0.16	0.04	0.16	0.1	0.04
0.149544073	0.1	0.04	0.09	0.09	0.05	0.04	0.09	0.14	0.04	0.19	0.1	0.04
0.149544073	0.1	0.04	0.07	0.09	0.07	0.04	0.09	0.18	0.04	0.15	0.1	0.04
0.149544073	0.15	0.01	0.09	0.13	0.06	0.04	0.02	0.15	0.03	0.2	0.11	0.03
0.146960486	0.1	0.04	0.07	0.1	0.07	0.04	0.1	0.16	0.04	0.16	0.1	0.04
0.149544073	0.1	0.04	0.09	0.09	0.05	0.04	0.09	0.14	0.04	0.19	0.1	0.04
0.149544073	0.1	0.04	0.07	0.09	0.07	0.04	0.09	0.17	0.04	0.15	0.1	0.04
0.149544073	0.15	0.01	0.09	0.13	0.06	0.04	0.02	0.15	0.03	0.2	0.11	0.03
0.149544073	0.1	0.04	0.08	0.09	0.05	0.04	0.09	0.14	0.04	0.18	0.1	0.04
0.149544073	0.1	0.04	0.07	0.09	0.06	0.04	0.09	0.17	0.04	0.15	0.1	0.04
0.149544073	0.1	0.04	0.08	0.09	0.06	0.04	0.09	0.15	0.04	0.17	0.1	0.04
0.149544073	0.14	0.03	0.07	0.12	0.06	0.04	0.07	0.15	0.03	0.15	0.11	0.02
0.149544073	0.14	0.03	0.06	0.12	0.06	0.04	0.07	0.16	0.03	0.14	0.11	0.02
0.25	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08
0.25	0.07	0.07	0.1	0.07	0.1	0.07	0.07	0.1	0.07	0.1	0.07	0.07
0.25	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08
0.25	0.07	0.07	0.1	0.07	0.1	0.07	0.07	0.1	0.07	0.1	0.07	0.07
0.25	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08
0.25	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08
0.25	0.08	0.08	0.1	0.08	0.1	0.08	0.08	0.1	0.08	0.1	0.08	0.08
0.25	0.08	0.08	0.1	0.08	0.1	0.08	0.08	0.1	0.08	0.1	0.08	0.08

#### 4.4.5 Evolutionary analysis

Models with the lowest BIC Scores (Bayesian information Criterion) are considered to describe the substitution pattern the best. For each model, AICc value (Akaike information criterion, corrected), Maximum Likelihood value (lnL), and the number of parameters (including branch lengths) are also presented (Nei and Kumar 2000). Non-uniformity of evolutionary rates among sites may be modeled using a discrete Gamma distribution (+G) with 5 rate categories and by assuming that a certain fraction of sites are evolutionarily invariable (+I). Whenever applicable, estimates of gamma shape parameter and/ or the estimated fraction of invariant sites are shown. Assumed or estimated values of transition/transversion bias (R ) are shown for each model as well. They are followed by nucleotide frequencies (f) and rates of base substitutions (r) for each nucleotide. Relative values of instantaneous should be considered when evaluating them. For simplicity, sum of r values is made equal to 1 for each model. For estimating ML values, a tree topology was automatically computed. The analysis involved 14 nucleotide sequences. All positions containing gaps and

missing data were eliminated. There were a total of 235 positions in the final data set. Evolutionary analyses were conducted in MEGA 7(Kumar et al., 2016).

Abbreviations:GTR: General Time Reversible; HKY: Hasagawa-Kishino-Yana; TN93: Tamura-Nei; T92: Tamura 3-parameter; K2: Kimura 2-parameter; JC: Jukes- Cantor:

## **CHAPTER FIVE**

### **5.0 DISCUSSIONS**

#### **5.1 VARIETAL PERFORMANCE FOR GRAIN YIELD AND STEM BORER RESISTANCE**

The criteria which were used to classify resistant or susceptible maize varieties in these trials were stem borer leaf feeding, leaf feeding scores, dead hearts, stem tunneling and overall plant damage such as stalk lodging, root lodging, plant aspect and number of holes per plant (Ajala and Saxena, 1994; Ajala *et al.*, 1995; Aroga and Ajala, 2007). However other agronomic parameters were taken because they are important to farmers and the plants with desirable agronomic characteristics hold greater potential (Leuschner, 1989).

##### **5.1.1 PERFORMANCE OF ELITE WHITE MAIZE VARIETIES**

From the varieties screened in 2010 there were no significant differences for characters that can be used to classify them as resistant or susceptible (Tables 6-10). This is probably due to the fact most of these maize varieties are from IITA with same source of gene pool and background. The major breeding effort of IITA during the period when these maize were bred was to make them resistant to diseases and pests, stem borers in particular, therefore most of them had assumed same level of resistance to diseases and pests including stem borers (Kamara 2004; Olosunde 2015).

When the criteria for classifying resistant and susceptible cultivars was compared to yield (Tables 7), the yield of susceptible and the resistant were not significantly different, even though Sammaz 17 seems susceptible while resistant check BR 9943 DMRSR seems resistant to stem borer. Although with Nwanze and Leuschner (1989) submitted that stem tunneling does not appear to be correlated with grain yield, hence other parameters may be considered in addition to stem tunneling for determining stem borer resistance.

However Sammaz 19, TZM 104, TZM 106, TZM 108 and resistant check can be classified as tolerant using leaf feeding scores 4 weeks after planting and some other parameters. However, the resistant character does not translate to yield (Table 7). This is probably due to the fact that TZM 104 and TZM 106 were mid- altitude materials with little adaptation to hot

humid ecology Nwanze and Leuschner (1989) concluded that stem tunneling on fully expanded internodes is not important, therefore stem tunneling may not correlate with grain yield. Also Olosunde (2015) also suggested that land races should not be neglected as they can be sources of valuable traits for maize improvement.

### **5.1.2 TEN YELLOW MAIZE VARIETIES EVALUATED AT NACGRAB IN 2012**

Most of the ten yellow maize varieties were significantly different with respect to stem borer resistance parameters probably because they were from different populations (Table 11-14). Most of the varieties screened in 2010 were maize materials improved by IITA specifically for yield potential, diseases and pests resistance especially stem borers, distinct germplasm complexes adapted to different agro-ecosystems and development of hybrid maize (Kamara *et al* 2004, Olosunde 2015). Only few of the maize material evaluated in 2010 were land races which were also part of the land races evaluated in 2012. Land races are farmers' adapted varieties which have not been improved. However some of the land races also were not significantly different for some of the parameters such as root lodging, stalk lodging, plant aspect and number of holes per plant, leaf feeding score at 7 weeks and 8 weeks after planting, stem borer dead heart 7 weeks and 8 weeks after planting. But TZM 1327, based on number of holes, and stem borer leaf feeding 8 weeks after planting can be said to be tolerant (Tables 13-14).

### **5.1.3 TEN WHITE MAIZE VARIETIES SCREENED AT NACGRAB 2012**

The varieties were not significantly different for most of the parameters assessed in evaluating maize for stem borer resistant such as leaf feeding 7 weeks after planting, stem borer dead heart 7 weeks after planting, leaf feeding score 7 weeks after planting, leaf feeding score 8 weeks after planting and stem borer dead heart 8 weeks after planting (Table 19). This agrees with the submission of Ajayi (1989) on the attempt to find source of resistant to stem borers in sorghum. He stated that borer infestation is highly variable from year to year and as a result of mixture of *Sesamia* and *Busseola*, interpretation of data becomes difficult (Ajayi 1989). Moreover if the infestations have been solely that of *Sesamia calamistis*, there would not be leaf feeding. However, the white maize varieties were significantly different for stem borer leaf feeding 8 weeks after planting. Other scientists also found some maize genotype resistant or tolerant to stem borers such as *Chilo partellus*, *E. saccharina* and *S. calamistis* (Bosque-Perez *et al.*, 1989; Bosque-Perez and Mareck, 1990;

Ajala *et al.*, 1995; IITA 2000) and the criteria used for these classifications were leaf feeding scores, dead hearts, stem tunneling and overall plant damage (Ajala and Saxena 1994; Ajala *et al.*, 1995) (Table 15-19). This study also adopted same criteria.

By using Duncan Multiple Range Test TZM 112 ranked most tolerant or resistant to stem borer using root lodging and stalk lodging (Table 18).

#### **5.1.4 PERFORMANCE OF PARENT LINES OF WHITE HYBRID MAIZE EVALUATED IN NACGRAB IN 2014.**

The varieties were significantly different for stalk lodging (Tables 20). Ajala and Saxena (1994) and Ajala *et al* (1995) also reported resistant and susceptible maize lines using overall plant damage.

Using Duncan Multiple Range Test, Aflatoxin Syn W5 and ACR 06 TZL Comp4 C4 appeared borer resistant and compare favourably with BR 9943 DMRSR (Table 23). There was no significant difference among the ten varieties using other parameters that are normally used in screening for resistant cultivars such as length of tunnels stem borer leaf feeding 4 weeks after planting, root lodging and stalk lodging (Tables 22 &23).

#### **5.1.5 PERFORMANCE OF PARENT LINES OF YELLOW CROSSES EVALUATED IN 2014**

Just like the white maize parents screened in 2014, varieties were not significantly different for many of the parameters studied. However, variety was significantly different for stalk lodging (Tables 24 & 27) while PVA Syn 1F2 was the most resistant compared to BR 9928 DMRSR using stalk lodging ratig (Table 27).

Also, using number of holes, PVA Syn 11F2 is similarly the most resistant variety while using length of tunnels placed PVA 9 F2 to be the most resistant variety (Table 28).

### **5.1.6 F1 OF THE WHITE MAIZE HYBRID EVALUATED IN 2014**

Like the parents, the crosses were not significantly different for the most of the stem borer parameters used in assessing stem borer damage. Ige (2014) had earlier reported that genotypes as well as breeding eras were not significantly different for all the stem borer damaged traits. He observed that sources due to genotypes within era 1 and era 2 as well as era 1 vs era 2 were similar for all the stem borer damage traits (Ige 2014).

The crosses were similar for the entire stem borer damage trait. However TZL Comp4 C4 and Aflatoxin Syn W4 show level of promise using stalk lodging (Table 32).

### **5.1.7 F1 OF THE YELLOW MAIZE HYBRID SCREENED IN 2014**

Yellow maize crosses were not significantly different from one another with respect to stem borer damage traits (Ige 2014). But root lodging in PVA Syn 1F2, PVA Syn 10F2 and PVA Syn 6F2 shows levels of resistance that compare favourably with resistant check BR 9928 DMRSR (donor parent). Also using the stalk lodging, PVA Syn 19F2 and PVA syn 11 F2 show high level of resistance to stem borer just like resistant BR 9928 DMRSR check (Table 38).

### **5.1.8 GENOTYPE BY ENVIRONMENT INTERACTION IN TEN WHITE MAIZE PARENTS EVALUATED IN 5 LOCATIONS IN 2015**

Locations effect was not significantly different for plant aspect but significantly different for plant count at germination, days to 50% flowering, days to 50% silking, anthesis- silking interval, plant height of harvest, ear height, root lodging, stalk lodging, husk cover, plant count at harvest, ear count at harvest, ear aspect, ear rot, field weight, stem borer leaf feeding 4 weeks after planting, stem borer dead heart 4 weeks after planting etc. but variety (Genotype) by environment (Locations) interaction were not significant for plant count at germination, days to 50% flowering, days to 50% silking, anthesis-silking interval, plant height, ear height, plant aspect, root lodging, plant count at harvest, ear count at harvest, ear aspect, ear rot, field weight, stem borer leaf feeding 4 weeks after planting, leaf feeding score 4 weeks after planting, stem borer dead heart 4 weeks after planting, stem borer leaf feeding 6 weeks after planting, leaf feeding score 6 weeks after planting, stem borer dead heart 6 weeks after planting, stem borer leaf feeding 8 weeks after planting and leaf feeding score 8 weeks after planting. Genotype by environment interaction was however significant for stalk



lodging and stem borer dead heart 8 weeks after planting. Ige (2014) also observed that genotype by environment interaction was not significantly different for maize agronomic traits he tested for. However, Oyekunle *et al.*, (2016) reported significant genotype by environment interaction for some of the agronomic traits of some maize hybrids and varieties. Other researchers also observed significant response of maize genotype to variable environmental conditions (Olosunde 2015; Badu-Apraku 2003; Ewool 2004) (Tables 39-40 and Tables (43-46).

#### **5.1.9 GENOTYPE BY ENVIRONMENT INTERACTION OF TEN YELLOW MAIZE PARENTS EVALUATED IN 5 LOCATIONS IN 2015**

The maize genotype were significantly different for agronomic traits such as days to 50% flowering, plant height, ear height, stalk lodging, husk cover, plant count at harvest, ear count at harvest, field weight, grain moisture content at harvest, yield and grain weight. Location also was also significant for all the agronomic traits including stem borer resistant traits. However, genotypes (varieties) by environment (location) interaction were not significant for most of the agronomic traits except stalk lodging and husk cover.

The fact that the maize genotypes tested showed significant variability in agronomic traits especially those for stem borer resistance suggests that the germplasm can be used for breeding programme to develop resistance to stem borer (especially *Sesamia calamistis* and other African stem borers). This report agrees with the observations of other researchers will that breeding using improved germplasm will lead to various types of maize open pollinated varieties (OPV), hybrids and/ or inbred lines for desirable characters (Lucchin *et al.*, 2003; Pressoir and Berthaund 2004; Badu Apraku *et al.*, 2003; Badu-Apraku 2006; Badu-Apraku 2007; Sokolov and Gushva 1997; Ilarslan *et al.*, 2002; Sanchez *et al.*, 1993; Doebley *et al.*, 2005, Azar *et al.*, 1997; Nisam-uddim *et al.*, 2010; Sansern *et al.*, 2010; Waqar *et al.*, 2007; Sampoux *et al.*, 1989; Oyekunle *et al.*, 2016).

#### **5.2.0 MEANS OF AGRONOMIC TRAITS OF WHITE MAIZE VARIETIES SEPARATED BY DUNCAN MULTIPLE RANGE TEST (DMRT) IN 2015 EVALUATION FOR EACH LOCATION.**

The agronomic traits differ from location to location as stated earlier (Oyekunle 2016). The fact that each location responds differently in terms of agronomic traits especially those used to determine susceptibility or resistance to stem borers show probably effect of seasons

on stem borer infestation, or the measure of how endemic a location is to stem borer infestation or types of strains of stem borers or combinations of stem borer types in those locations (Dike et al 2002) (Table 43).

#### **5.2.1 MEAN PERFORMANCE OF AGRONOMIC TRAITS IN WHITE MAIZE VARIETIES SEPARATED BY DUNCAN MULTIPLE RANGE TEST (DMRT) IN 2015 EVALUATION IN EACH LOCATION.**

Each maize exhibits different phenotypic characteristics as expression of genotypes when screened in the five locations. These agronomic traits separated by DMRT shows that the maize genotypes were similar for some agronomic traits while different for some traits. However, using stem borer resistant traits, such as root lodging and stalk lodging stem borer leaf feeding four (4) and six (6) weeks after planting, leaf feeding score six (6) and eight (8) weeks, TZL Comp4C4 appears a promising genotype that can be recommended for planting as stem borer tolerant or resistant genotype as seen in (Tables 44-46) This is in agreement with Ajayi (1989) who reported result of USAID JP 26 project in which hundreds of Sorghum lines were screened for stem borer but 26 varieties were found resistant.

#### **5.2.2 MEANS OF ANOVA OF YELLOW MAIZE EVALUATED ACROSS 5 LOCATIONS IN 2015 SEPARATED BY DMRT**

Agronomic traits of these yellow maize varieties differ from one location to the other locations because of their responses to climatic factors such as sunshine, temperature, edaphic factors, rainfall and humidity. Other researchers also discovered the effect of environments on the phenotypic expression of genotypes (Badu - Apraku 2007; Olosunde 2015) (Tables 47-48).

#### **5.2.3 MEANS OF ANOVA FOR AGRONOMIC TRAITS OF YELLOW MAIZE EVALUATED ACROSS FIVE LOCATIONS IN 2015 WITH VARIETIES RANKED BY DMRT**

The varieties exhibited variations in agronomic traits tested for the yellow maize varieties considering number of holes and length of tunnels which are two of traits that show how resistant or susceptible a variety or genotype is to stem borers, PVA Syn 3F2 is resistant

stem borers. Kamara (2004) had also reported resistant varieties or genotypes to stem borers using the same criteria (Tables 49-50).

#### 5.2.4 SURVEY OF FARMERS' MAIZE FIELDS

This present survey was carried out between July and August of 2014 in South West Nigeria and Kwara state. This was to survey the occurrence, abundance and diversity of stem borers' infestation in South West Nigeria and Kwara state in Tropic Humid ecology. Bowden (1976) and Moyal and Le Ru (2006) recommended studies in the wild environment in order to get a better insight into the ecology and the way of controlling these pests. Hence the needs for field survey become imperative.

The survey shows *Busseola fusca* to be more common than *Sesamia calamistis* contrary to the general belief that *Sesamia calamistis* is more common in Southern Nigeria than *Busseola fusca*. The reason for this is not known. However, Appert (1970) also affirmed that *Busseola fusca* is considered by some authors to be the most abundant stem borer complex that was found in all the surveyed areas. The species was first described by Ragonot in 1888, and since then it has been recorded on numerous plant species, including maize, cotton, cocoa, lima beans (*Phaseolus lunatus*), the shea butter tree (*Vitellaria paradoxa* (*Syn Butyrospermum parkii*), *Mucuna sp*, *Canavalia Sp* and *Sphenostylis stenocarpa* (Moya and Trans 1991b). Plant breeders may wish to concentrate more effort in breeding for resistant crop genotypes to combat the devastating effect of these species due to its wide distribution and ability to survive on many crops. This pest infects fruiting structures of mature plants and continues feeding on the stored products (Bordat and Renand, 1987). Infestation on maize starts in the field; female moth lay their eggs on the silks and husk leaves. Eggs hatch in 5-7 days and young larva feed within the silk channel for a few days before reaching the grain. Developing larvae feed on the distal portion of the maize ear and tunnel through the grain, causing extensive damage and often consuming the embryos in a way that is not superficially visible. Only close inspection reveals the degree of damage. Pupation takes place within the tunnels or on the surface of the grain, and the pupae are surrounded by a silky cocoon. Damage to the grains continues during storage, hence, *M. nigrivenella* can be regarded as both a field and a storage pest; although no reproduction occurs in the store. Preliminary observation indicates that the pest can survive in the stored cobs for up to 8 weeks, even at a grain moisture content of 12-15% (O.Bolaji, N.A Bosque –Perez and M.Ivbijaro, unpublished data).

Surveys have been conducted by IITA scientists in farmer's field in Benin, Ghana, Ivory Coast and Nigeria to establish the Geographical distribution, host plant range and natural enemy complex of this species in West Africa. *Mussidia nigrivenella* has been found in every country and ecological zone from the forest to the Northern Guinea Savannah but is rare in the Sudan Savannah. Survey from Benin indicate that each larvae causes an average, 4% ear damage, five larvae per infested ear are often found (F. Schulthess, unpublished). Although this pest may appear to be minor in these ecology for now, it is equally important that breeding programme against it is initiated at various breeding institution before it becomes endemic.

The time this survey was carried out was towards the time of harvest, *Mussidia nigrivenella* was abundant. Though the survey was not particularly for *Mussidia nigrivenella*, but this species of ear borer or stem borer seems to be abundant. Surveys in South Western Nigeria demonstrated this borer to be the most abundant pest of maize at the time of harvest (O.Bolaji, N.A Bosque-Perez and M. Ivbijaro unpublished data). In studies conducted at IITA, Ibadan, maize varieties with a short husk- tip extension and loose husk leaves were found to be more severely infested by *M. nigrivenella* than those with good husk cover; additionally, the abundance of this ear borer was found to increase with delayed harvesting (O.Bolaji and N, A Bosque-Perez, unpublished data). Therefore prompt harvesting is recommended.

Harris (1962) described the biology and distribution of the pearl millet stem borer, *Coniesta ignefusalis* (Lepidoptera: Pyralidae) in Nigeria. His survey revealed that the insect was a pest in virtually all the millet producing areas in Nigeria (Dike et al 2002). Dike et al (2002) also carried out three surveys in 1995 and 1996 to survey the incidence of millet stem borer *Coniesta ignefusalia* in farmers' field in Nigeria. *Coniesta ignefusalis* was found to be pest virtually in all the surveyed areas (Dike et al 2002).

During the past thirty five years, surveys have been conducted by scientists from various National and International Institutes in several countries of West Africa to obtain information on the abundance, species composition and relative importance of maize borer up to 8 weeks after planting onwards in Southern Nigeria from August to November of 1985 and 1986 (Harris 1962; Dike et al., 2002). *Mussidia nigrivenella* was found in all sites, while *Busseola fusca* was found in forest/savannah transition zone location. Other encountered stem borers include *Coniesta ignefusalis* and *Cryptophlebia Sp.*

Additional survey of borers and their natural enemies were carried out by International Institute of Tropical Agriculture (IITA) Scientists in Nigeria during 1991 and 1992 (Bosque-perez et al., 1995 unpublished data). *Sesamia calamistis* and *Eldana saccharina* was the most commonly encountered pest of maize: additionally, *Busseola fusca*, *Coniesta ignefusalis*, and *Mussidia nigrivenella* in 1991 and 1992 respectively.

The average level of infestation in Oyo state was 9.4%, the highest being 16% at Idi-Ayunre and the lowest infestation level being 3% at Podo. The level of infestation with stem borers in Kwara state during the survey ranged from 20% to 80% with average level of infestation being 44%. The level of infestation of Maize farms surveyed in Ondo State varied from 10% to 80% while the average level of infestation was estimated at 42%. The infestation level in Ogun state during the survey range from 40% to 80% while the average level of infestation was 50%. The range of stem borer infestation at the time of survey in Ekiti State was 20% to 50% with average level of infestation of 36%. The range of Maize stem borer infestation found in Osun State was between 30% to 60% with average being 38%. These results agreed with submission of Bosque-pereze et al (1995) that the percentages of plant with borer damage in individual field varied according to year and ecological zone with a maximum of 17% for both Southern Guinea and Northern Guinea Savannah in 1991 and up to 30% in the Northern and 47% in the Southern Guinea Savannah in 1992. Sithole (1989) submitted that the infestation in farmers field in Zimbabwe varies from 15-40%, while in Southern Africa generally, according to Sithole, the infestation ranges from 30-70% in subsistence farmers' fields but average less than 30% on commercial fields. These findings are therefore suggesting regular survey of these pests so as to initiate the current and appropriate control strategies adaptable for enhancing higher yields.

The stem borers found in all the states however include *Sesamia calamistis*, *Sesamia inferens*, *Busseola fusca*, *Eldana saccharina*, *Diatraea saccharalis*, *Diatraea lineolata*, *Diatrea grandiosella*, *Maliarpha separatella*, *Scirpophaga Sp*, *Coniesta ingnefusalis* and *Mussidia nigrivenella* and *Cryptophlebia Sp*. One or two or more of these stem borers were found on single plant during the survey (Seshu Reddy 1989). The survey revealed enormity of the challenge posed to farmers by stem borers complex in maize production.

Stem borers was found virtually in all places surveyed. This agreed with the observation of Dike et al., (2002) during their survey of stem borers in millet producing areas of Nigeria. However all the stem borers found on the maize were either in family *noctuidae*

or *pyralidae* in the order *Lepidoptera*, except an unusual borer found in Wasimi, Osun state which when raised to adult was discovered to be a beetle( Order: Coleoptera) and not a moth. There has not been any report in Nigeria of any stem borer attacking maize except moth from the family *noctuidae* or *pyralide*.

The only place where there was a report of stem borer different from moths was in U.S.A and the stem borer was on Soya bean, (*Dectes texanus, texanus*). The soya bean stem borer found in U.S.A was also a beetle but it is different from one found at Wasimin, Osun state and Moor Plantation, Ibadan, Oyo state. Entomologists may need to assist in proper identification, classification and biology of these new pests for effective control on crops.

### 5.2.5 LONGHORN BEETLES

The first beetle was identified to be Long horn beetle from family Cerambycidae. The longhorn beetles (Cerambycidae; also known as long-horned or longhorn beetles or longicorns) are a cosmopolitan family of beetles typically characterized by extremely long antennae which are often, as long as or longer than the beetle's body. In various members of the family, however, the antennae are quite short (e.g., *Neandra brunnea*) and such species can be difficult to distinguish from related beetle families such as the Chrysomelidae. The family is large, with over 26,000 species described, slightly more than half from the Eastern hemisphere. Several are serious pests. The larvae, called roundheaded borers, bore into wood, where they can cause extensive damage to either living trees or untreated lumber (or, occasionally, to wood in buildings; the old house borer, *Hylotrupes bajulus*, is a particular problem indoors). A number of species mimic ants, bees, and wasps, though a majority of species are cryptically colored. The rare titan beetle (*Titanus giganteus*) from northeastern South America is often considered the largest (though not the heaviest, and not the longest including legs) insect, with a maximum known body length of just over 16.7 cm (6.6 in). The scientific name of this beetle family goes back to a figure from Greek mythology: after an argument with nymphs, the shepherd Cerambos was transformed into a large beetle with horns.

As with many large families, different authorities have tended to recognize many different subfamilies, or sometimes split subfamilies off as separate families entirely (e.g., Disteniidae, Oxypeltidae and Vesperidae); there is thus some instability and controversy regarding the constituency of the Cerambycidae. There are few truly defining features for the group as a

whole, at least as adults, as there are occasional species or species groups which may lack any given feature; the family and its closest relatives, therefore, constitute a taxonomically difficult group, and relationships of the various lineages are still poorly understood.(Wikipedia 2017).

The second beetle was identified to be Longhorn beetles but from Sub family Lamiinae. According to literature, all the members of this subfamily are xylophagous and phytophagous (Ozdikmen and Caglar 2004; Suksawat Ponpiniji et al 2011). The larval develop in plant tissues. Adaptation to such a large variety of host plants has resulted in tremendous variation in the behavior and ecology of these borers. Many species are important pests of forests, plantations and street trees. Different species attack various types of trees and shrubs. A few attack living trees or branches (Suksawat Ponpiniji et al 2011). However some are beneficial insects through their role as insect pollinators on some plant species (Gutowski 1990; Tasen 2001; Suksawat Ponpiniji et al 2011).

This survey reveals the need for resistance with poly genes in the maize to survive the onslaught of stem borers complex of tropic humid ecology of South- West Nigeria. In fact green maize may be more profitable in tropic humid ecology of South- Western Nigeria due to stem borers complex. Farmers can easily harvest their maize before the onslaught of maize stem borers, ear borers and field to store pest like *Mussidia nigrivenella*, *Sitophilus zeamais* and other insect pests that rain avalanche of attacks on maize during physiological maturity. Alternatively resistant genotypes may be adopted while integrated pest management had been globally recommended.

## **5.2.6 MOLECULAR ANALYSIS**

Bioinformatics is computational molecular biology. Bioinformatics is broadly defined as the development and application of computational tools to acquire, store, organize, retrieve, and analyze large amount of biological information. It is a new field that was born out of the need for high powered computational ability to help organize, analyze, and store biological information. The primary types of information involved in bioinformatics are DNA and protein sequence data. Sequence alignment is the prerequisite of virtually all forms of sequence analysis ranging from search, to assembly, and to phylogenetics. Various algorithms have been developed to produce optimal alignment. Two examples of widely used open access softwares, are BioEdit (Hall, 1999), and MEGA (Tamura et. al.,2007). Another

one used for analysis in this work is FinchTV. The origin of bioinformatics can be traced to the development by Sanger and Coulson (1975) in which they used a technique for rapid sequencing of nucleic acids. This technique was improved upon and automated by Sanger et al (1977) and Maxam and Gilbert (1977).

Once DNA sequencing became technologically simple and automated, massive numbers of gene sequences were generated. Public databases were created to hold information and allow everyone to use it. Such public database in the United States of America is called GenBank which is administered by the National Centre for Biotechnology Information (NCBI) and contains billions of nucleotide bases in millions of sequence records from thousands of different organisms (microbe, plant, insects and animal species). The public database in Japan is the Data Bank of Japan (DDBJ) and in Europe is the European Molecular Biology Laboratory (EMBL). All of these databases are cooperative systems and the data contained in them can be accessed using the following web addresses: Genbank:- <http://www.ncbi.nlm.nih.gov/web/search/index.html>; EMBL:-<http://www.ebi.ac.uk/ebidocs/embl-db/abi/topembl.html>; DDBJ: - <http://www.ddbj.nig.ac.jp>. Sequence comparison is essential for understanding evolutionary relationship between genes. The most common and widely used similarity search tool is BLAST (Basic Local Alignment Search Tool (Ye et al. 2006). BLAST is a set of programs used to compare a nucleotide or protein query sequence to all of the available sequence databases. Phylogenetic analysis is the basis of taxonomical and evolutionary studies. Phylogenetic analysis is performed to cluster multiple sequences based on genetic distances. All of these were adopted in this study for molecular identification and grouping of the stem borers collected during the field survey.

### **5.2.7 PHYLOGENETIC TREE OF STEM BORERS SEQUENCE**

The sequences identify the stem borers as *Chilo orichalcociliellus*, *Eldana saccharina* and *Sesamia calamistis*.

#### ***Chilo orichalcociliellus***

The pest is very important and common in East Africa, the Indian subcontinent and the Far East, but not in West Africa. *Chilo orichalcociliellus* was originally described by Strand in 1911 in the genus *Diaraea*. Bleszynski (1962) put it into synonymy with *Diatraea argyrolepida* (Hampson) and later published (Bleszynski, 1970) a detailed taxonomic description and stated that it was easily distinguishable from other species of *Chilo* by



characters of male and female genitalia. Bleszynski (1970) recorded this species from Kenya, Madagascar, Malawi, South Africa, Tanzania and Zimbabwe and noted that it does not occur in West Africa. But it is now found in Nigeria in West Africa as Maize pest belonging to the family pyralidae. This is probably being reported authentically in Nigeria for the first time. The implication is that farmers in West Africa should be warming up for means of controlling them. The current climate change and one of its attendant problems is emergence of new pests where they were not known before. It may also be due to movement of cereals genetic resources or germplasms across the world especially maize germplasms.

### ***Eldana saccharina***

This stem borer occurs throughout Africa south of the Sahara. It belongs to the family pyralidae (galleriinae) in the order Lepidoptera. The newly hatched larvae feed on the leaves, usually boring into the midrib. Fully grown larvae bore into the stems and cause dead hearts. The larvae hang down by means of silken threads and are sometimes blown onto neighbouring plants. Eldana was described by Walker in 1865. It is well characterized, easily recognizable and unlikely to be confused with any of the other stem borers of African cereals. The species occurs throughout tropical Africa from 15°N to 30°S and has been recorded in Nigeria, Chad, Sierra Leone, Ghana, Burundi, Kenya, Mozambique, Rwanda, Somalia, Tanzania, Uganda, Zaire and South Africa. It is found on Sugar cane, maize, Sorghum, rice, millet and other cultivated cereals as well as sedges (*Cyperus* spp), its presumed original hosts (Maes 1998). Hence maize which is the staple of people in sub-saharan region should be properly protected against these common pest called stem borer.

### ***Sesamia calamistis***

This insect is widely distributed in Africa. Among the indigenous stem borers in Africa, *Sesamia calamistis* (Hampson) (Lepidoptera: Noctuidae) is one of the prominent ones. The hosts include maize (*Zea mays*), Sorghum (*Sorghum bicolor* (L.) Moench), Millet (*Pennisetum americanum*), Rice (*Oryza* sp) and other plants in the family gramineae (poaceae) (Ingram 1958; Bowden 1976; Ongamo et. al., 2008). It is a major pest in West Africa and Nigeria in particular (Moyal 1988; Bosque-Perez and Schulthess, 1998; Ongamo et. al., 2008). The larvae of *Sesamia calamistis* feeding cause stems breaking and chaffy panicles in sorghum, millet and rice. Panicles also can be attacked.

According to Tams and Bowden (1953), the ecological requirements of *Sesamia nonagrioides botanephaga* and *Sesamia calamistis* are different, *Sesamia nonagrioides botanephaga* being dominant species in forest and forest range areas, where there is a minimum rainfall of about 50 inches (127 cm) well distributed through the year and therefore a relatively short and less severe dry season. *Sesamia calamistis* is more common, at least in West Africa, in savannah areas that have a well marked dry season. However the two species can occur in the same field at the same time (Holloway, 1998). The species is known from maize, rice, sorghum and sugar cane. Among the wild hosts are *P. Purpureum*, *S. arundinaceum*, *Rottboelia exaltata* and *Chasmopodium afzelii* (Tams and Bowden, 1953). This species was equally found in the fields surveyed in South West Nigeria and Kwara State indicating the need to put in place effective control measure for profitable maize production in these ecology.

### 5.2.8 PHYLOGENETIC TREES

Fig. 32 shows evolutionary relationships of taxa. It shows two main groups. *Sesamia calamistis*, *Chilo orichalcociliellus* and *Eldana sacharina* belonging to the two groups or clades. A clade is a group of organisms that consists of a common ancestor and all its lineal and represents a single branch on the tree of life. Also the phylogenetic tree shows there are variations within each species. Fig. 33 also shows evolutionary relationship of taxa. The phylogenetic tree using their nucleotide bases show that the stem borers are related. Fig. 34 shows evolutionary relationship of taxa (time tree). The phylogenetic tree shows two groups. Though there are subgroups in the upper group. Fig. 35 shows maximum parsimony analysis of taxa. The phylogenetic tree shows three groups, though there are subgroups in the upper two groups. Fig. 36 also shows maximum parsimony of taxa. This analysis is also possible. The two groups are divided into subgroups. Fig. 37 also shows maximum parsimony analysis of taxa. The phylogenetic tree shows the relationship between all the stem borers. Fig. 38 also shows evolutionary relationship of taxa using minimum evolution method. There are two groups and each stem borer belongs to the two groups. The understanding of groups each of these pests belongs will provide plant breeders the opportunity of using the same/ similar procedure or pest traits to select for resistant genotypes of crops being affected by pests of common group in phylogenetic trees.

## **CHAPTER SIX**

### **CONCLUSIONS AND RECOMMENDATIONS**

#### **6.0 SUMMARY OF FIELD EXPERIMENTS**

Among the white and yellow maize varieties screened from 2010 to 2015 in order to identify sources of stem borer resistance were:-

From the twenty maize varieties screened in NACGRAB Ibadan in 2010, the varieties that were found tolerant to stem borers and can be seen as resistant donors in controlling stem borers were Sammaz 19, TZM 104, TZM 106, and TZM 108. Sammaz 19 was an improved variety while TZM 104, TZM 106 and TZM 108 were land races.

From ten yellow maize varieties screened at Ibadan in 2012, TZM 1327 can be said to be tolerant to stem borers. Also from ten white maize varieties screened at Ibadan in 2012, land race TZM 112 can be said to be promising as stem borer resistance donor.

The ten maize parents screened in 2014 at Ibadan were also screened in 5 locations including Ibadan, Mokwa, Kotangora, Kabba and Abuja in 2015. The possible maize resistance donors found among them include Aflatoxin Syn W5, ACR 06 TZLComp 4 C4 and TZL Comp 4 C4.

The ten yellow maize parents screened in 2014 at Ibadan were also screened in 5 locations including Ibadan, Mokwa, Kotangora, Kabba and Abuja in 2015. The possible resistance donors found among them include PVA Syn 1F2, PVA Syn 11F2, PVA Syn 9F2 and PVA Syn 3F2.

From the ten White F1 of maize crossed with the resistant check BR 9943 DMRSR and screened in 2014, the F1 showing level of promise to be resistant or tolerant to stem borers were TZL Comp 4 C4 and Aflatoxin Syn W4 using stalk lodging due to stem borers infestation.

From the ten yellow F1 of Yellow maize crossed with resistant check BR 9928 DMRSR screened in 2014, the F1 showing level of promise to be resistant to stem borers were PVA Syn 1F2, PVA Syn 10F2, PVA Syn 6F2 and PVA Syn 19F2.

### 6.1.1 SUMMARY OF FIELD SURVEYS

The goals of this study include (1) to survey the occurrence and the abundance and diversity of stem borers on maize in South Western Nigeria. (2) To collect and molecularly characterize populations of stem borers in South Western Nigeria. (3) To screen maize varieties (local and improved germplasm) to identify resistance donors among maize germplasm to *sesamia calamistis* and other stem borers.

Six states including Oyo State, Ondo State, Ogun State, Ekiti State, Osun State and Kwara State were surveyed to re-establish the occurrence, abundance and diversity of stem borers' infestation in South West and Kwara State in Tropic humid ecology. The survey shows *Busseola fusca* to be more common than *Sesamia calamistis* contrary to the general belief that *Sesamia calamistis* is more common in Southern Nigeria than *Busseola fusca*.

Also, the average level of infestation in Oyo State was 9.4%, the range being 3% to 6%. The average level of stem borer infestation in Kwara State was 44%, the infestation level ranging from 20% to 80%. The average level of infestation in Ondo State was 42%, the range being 10% to 80%. The average level of stem borer infestation was 50% in Ogun State while the infestation level range from 40% to 80%. The average level of infestation in Ekiti State was 36%, the range of infestation being from 20% to 50%. The average level of stem borer infestation in Osun State was 38%, the range of stem borer infestation being from 30% to 60%.

The stem borers collected from survey were analyzed by raising some to adults and some were subjected to DNA extraction, amplification and gene sequencing. The stem borers found can be categorized into three groups. The first group is the traditionally known stem borers in Nigeria on cereals such as *Sesamia calamistis*, *Sesamia inferens*, *Busseola fusca*, *Eldana saccharina*, *Diatraea saccharalis*, *Diatraea lineolata*, *Diatraea grandiosella*, *Maliarpha separata*, *Scirpophaga* sp, *Coniesta ignefusalis*, *Mussidia nigrivenella* and *Cryptophlebia* sp. These stem borers are moth in either family *noctuidae* or *pyralidae* in the order *lepidoptera*. The second group found during the survey includes those ones that though they are *lepidoptera*, it has not been reported in Nigeria. This is *Chilo orichalcociliellus*. It is very important and common in East Africa, India subcontinent and the Far East. It is now found in Nigeria.

The third group of stem borers found in this survey includes two different types of unidentified beetles raised to adult from borers collected from maize stems from Wasimi and Ibadan. The two beetles belong to the order coleoptera. They are all called long horn beetles found in the family Cerambycidae, in the subfamily Lamiinae.

### **6.1.2 CONCLUSIONS**

The status of insects and insect pests are dynamic. There are different strains of even traditionally known stem borers as Phylogenetic tree have shown. The result of molecular analysis confirms that some insects are cryptic species (insects that resemble each other very closely so that they cannot be separated on morphological grounds alone) especially lepidoptera. Molecular data and DNA barcoding helps in separating sibling species of insect pest. It has been used to separate putative single species into several species, estimate species richness, and indicates intraspecific variation or to delimits entities that are biologically and taxonomically distinct. This is so when mitochondrial cytochrome oxidase subunit -1 (CO1 or COX1) single gene region is sequenced (Wilson 2008; Samways et al., 2010; Janzen et al. 2005; Hebert et al., 2004). Therefore, there are new species of maize stem borers. Breeders cannot rely on single gene resistant maize varieties. Poly genes may be more appropriate. Also artificial screening using only one stem borer type in the screen house may not produce a truly stem borer resistant maize varieties. The resistant lines produced under this condition may break down in the presence of stem borer complex in the field.

Also among the local and improved germplasm screened during the period of this research, some white and yellow maize varieties such as Sammaz 19, TZM 104, TZM 106, TZM 108, TZM 1327, TZM 112, Aflatoxin Syn W5, ACR 06 TZL Comp 4 C4, TZL Comp4C4, PVA Syn 1F2, PVA Syn 11F2, PVA Syn 9F2, PVA Syn 3F2, PVA Syn 1F2, PVA Syn 10F2, PVA Syn 6F2 and PVA Syn 19 F2 shows level of resistance to stem borers and are promising varieties to be used in breeding programmes to get a stem borer resistant varieties or hybrids.

The use of resistant varieties is by far, the most promising control measure in reducing yield losses caused by stem borers for resource constrained African farmers and which may be enhanced by cultural practice (Adesiyun, 1983).

### **6.1.3 RECOMMENDATIONS**

Survey of the stem borers on maize in south west and other agro-ecology in Nigeria should be carried out to further establish level of occurrence and abundance and stem borers biotypes in Nigeria. Molecular tools and DNA barcoding and possibly Genome wide association studies (GWAS) should be explored in studying stem borer biotypes in Nigeria. Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) i.e CRISPR/cas9 system should be explored in addressing the menace of stem borers in cereals farm in Africa. There is need for entomologists in Nigeria to see how to characterize stem borers in Nigeria and deposit the data in international data bases such as Gen Bank of National Centre for Biotechnology Information NCBI (USA), DNA Data Bank of Japan (DDBJ) and European Molecular Biology Laboratory (EMBL). Breeding efforts should be geared towards getting resistant varieties of maize that can withstand stem borers complex currently found in the farmers field probably due to climate change. Africa should also look into having regional data base with headquarter in Nigeria.

Further studies should also be carried out on the change of status of field pests to ascertain pests that formerly their eggs, larva and pupa stages were not inside the cereals or legume stems but now make use of inside stems to complete their life cycle.

The breeders should explore production of more resistant maize varieties using these identified sources of stem borer's resistant germplasm.

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## APPENDIX

**TABLE 9: SURVEY OF STEM BORERS ON MAIZE IN SOUTH WESTERN NIGERIA  
AND KWARA STATE.**

STATE	FARM	TOWN	CROPPING SYSTEM	LONGITUDE	LATITUDE	ALTITUDE	%INFESTATION	NAME OF STEM BORERS FOUND	PA
Oyo state	A	Omi Adio	maize sole	E003° 46.569'	N07° 23.900'	592.1m	8%	<i>Chilo partellus</i>  <i>Maliarpha separatella</i>  <i>Scirpophaga sp</i>  <i>Mussidia nigrivenella</i>  Unidentified stem borer(a beetle)	
Oyo state	B	Podo	maize sole	E003° 52.085'	N07° 18.631'	480.2m	3%	<i>Sesamia calamistis</i>  <i>Sesamia inferens</i>  <i>Busseola fusca</i>  <i>Mussidia nigrivenella</i>	
Oyo state	C	Idi- Ayunre	maize sole	E003° 51.875'	N07° 13.903'	403.6m	16%	<i>Busseola fusca</i>  <i>Sesamia calamistis</i>	
Oyo state	D	Oke- Jagun Erunmu	maize sole	E004° 03.776'	N07° 27.007'	683.1m	12%	<i>Busseola fusca</i>  <i>Eldana saccharina</i>  <i>Mussidia nigrivenella</i>	
Oyo state	E	Akowonjo	Maize intercropped with cassava	E004° 03.858'	N07° 25.108'	637.6m	8%	<i>Eldana saccharina</i>  <i>Busseola fusca</i>  <i>Sesamia calamistis</i>  <i>Mussidia nigrivenella</i>	
Kwara state	A	Odo-Omo	maize sole	E004° 25.288	N08° 20.577'	1197m	80%	<i>Coniesta ignefusalis</i> <i>Scirpophaga sp</i> <i>Sesamia cretica</i> <i>Mussidia nigrivenella</i>	

Kwara state	B	Aka- ijoba odore	maize sole with few stand of sorghum	E004° 40.243'	N08° 33.282'	1122m	80%	<i>Busseola fusca</i> <i>Sesamia</i> <i>calamistis</i> <i>Coniesta</i> <i>ignefusalis</i> <i>Eldana</i> <i>sacharina</i>	
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Kwara state	C	Alalubosa	Maize intercropped with vegetable	E004° 40.938'	N08° 33.599'	1048m	20%	<i>Scirpophaga sp</i> <i>Sesamia calamistis</i> <i>Coniesta ignefusalis</i>		
Kwara state	D	OKE-Oyi	Maize sole	E004° 40.938'	N08° 35.284'	1255m	20%	<i>Scirpophaga sp</i> <i>Sesamia calamistis</i> <i>Coniesta ignefusalis</i>		
Kwara state	E	Eye-Nkorin	maize sole	E004° 30.373'	N08° 26.777'	1444m	20%	<i>Busseola fusca</i> <i>Sesamia calmistis</i> <i>Coniesta ignefusalis</i> <i>Mussidia nigrivenella</i>		
Ondo state	A	Oke-igbo	maize with few stands of cassava	E004° 44.214'	N07° 09.367'	800m	40%	<i>Mussidia nigrivenella</i> <i>Sesamia nonagrioides</i> <i>Sesamia calamistis</i> <i>Syanthodonini sp</i>		
Ondo state	B	Gberinlegi	maize intercropped with cassava	E004° 48.273'	N07° 08.064 <sup>1</sup>	786.m7	40%	<i>Antheraea pernyi</i> <i>Sesamia inferens</i> <i>Sesamia calamistis</i> <i>Busseola calamistis</i>		Skirt and House; crickets, ants
Oyo state	D	Oke- Jagun Erunmu	maize sole	E004° 03.776'	N07° 27.007'	683.1m	12%	<i>Busseola fusca</i>  <i>Eldana saccharina</i>  <i>Mussidia nigrivenella</i>		
Oyo state	E	Akowonjo	Maize intercropped with cassava	E004° 03.858'	N07° 25.108'	637.6m	8%	<i>Eldana saccharina</i>  <i>Busseola fusca</i>  <i>Sesamia calamistis</i>  <i>Mussidia nigrivenella</i>		Ants.

Kwara state	A	Odo-Omo	maize sole	E004° 25.288	N08° 20.577'	1197m	80%	<i>Coniesta ignefusalis</i> <i>Scirpophaga sp</i> <i>Sesamia cretica</i> <i>Mussidia nigrivenella</i>		Aphids ( <i>Rhopalosiphum maidis</i> ); <i>Sitophilus zeamais</i>
Kwara state	B	Aka- ijoba odore	maize sole with few stand of sorghum	E004° 40.243'	N08° 33.282'	1122m	80%	<i>Busseola fusca</i> <i>Sesamia calamistis</i> <i>Coniesta ignefusalis</i> <i>Eldana sacharina</i>		Beetles
Kwara state	C	Alalubosa	Maize intercropped with vegetable	E004° 40.938'	N08° 33.599'	1048m	20%	<i>Scirpophaga sp</i> <i>Sesamia calamistis</i> <i>Coniesta ignefusalis</i>		
Kwara state	D	OKE-Oyi	Maize sole	E004° 40.938'	N08° 35.284'	1255m	20%	<i>Scirpophaga sp</i> <i>Sesamia calamistis</i> <i>Coniesta ignefusalis</i>		
Kwara state	E	Eye-Nkorin	maize sole	E004° 30.373'	N08° 26.777'	1444m	20%	<i>Busseola fusca</i> <i>Sesamia calmistis</i> <i>Coniesta ignefusalis</i> <i>Mussidia nigrivenella</i>		
Ondo state	A	Oke-igbo	maize with few stands of cassava	E004° 44.214'	N07° 09.367'	800m	40%	<i>Mussidia nigrivenella</i> <i>Sesamia nonagrioides</i> <i>Sesamia calamistis</i> <i>Syanthodonini sp</i>		
Ondo state	B	Gberinlegi	maize intercropped with cassava	E004° 48.273'	N07° 08.064 <sup>1</sup>	786.m7	40%	<i>Antheraea pernyi</i> <i>Sesamia inferens</i> <i>Sesamia calamistis</i> <i>Busseola calamistis</i>		Skirt and House; crickets, ants

Ondo state	C	Ondo	Maize sole	E004° 50.140'	N07° 01.197'	699.9m	10%	<i>Busseola fusca</i> <i>Coniesta ignefusalis</i> <i>Sesamia calamistis</i> <i>Busseola fusca</i> <i>Mussidia nigrivenella</i>		Aphids (Rhopalosiphum maidis); ants
Ondo state	D	Bagbe	maize sole	E004° 50.887'	N06° 58.722'	801.3m	80%	<i>Busseola fusca</i> <i>Coniesta ignefusalis</i> <i>Sesamia calamistis</i> <i>Eldana saccharina</i> <i>Mussidia nigrivenella</i>		Aphids (Rhopalosiphum maidis); ants
Ondo state	E	Ore	maize sole	E004° 53.707'	N06° 45.540'	300m	40%	<i>Scirpophaga sp</i> <i>Coniesta ignefusalis</i>		Ants
Ogun state	A	Ijebu-ode	Maize intercropped with cassava.	E003° 56.761'	N06° 50.722'	336.8m	50%	<i>Busseola fusca</i> <i>Sesamia calamistis</i> <i>Sesamia inferens</i> <i>Coniesta calamistis</i> <i>Mussidia nigrivenella</i>		Ants
Ogun state	B	Ijebu- mushin Esure	maize sole	E003° 59.678'	N06° 47.190'	210.5m	50%	<i>Busseola fusca</i> <i>Sesamia calamistis</i> <i>Coniesta ignefusalis</i> <i>Eldana sacharina</i> <i>Mussidia nigrivenella</i>		Ants
Ogun state	C	Itele	Maize intercropped with cassava.	E003° 59.675'	N06° 47.188'	133.3m	80%	<i>Coniesta ignefusalis</i> <i>Sesamia calamistis</i> <i>Busseola fusca</i> <i>Eldana sacharina</i> <i>Mussidia nigrivenella</i>	Larva of cotesia sesamiae	

Ogun state	D	Ijebu-Idofe	Maize intercropped with cassava	E003° 56.636'	N06° 55.178'	182.0m	50%	<i>Sesamia calamistis</i> <i>Busseola fusca</i> <i>Eldana sacharina</i> <i>Ostrinia nubilalis</i> <i>Mussidia nigrivenella</i>		Ants
Ogun state	E	Ajegunlel-Awa	maize sole	E003° 55.703'	N06° 59.353'	165.4m	40%	<i>Scirpophaga sp</i> <i>Ostrinia nubilalis</i> <i>Mussidia nigrivenella</i>		
Osun state	A	Ikire	Maize intercropped with cassava	E004° 11,900'	N07° 23.093'	695.2m	40%	<i>Chilo partellus</i> <i>Busseola fusca</i> <i>Sesamia inferens</i> <i>Sesamia calamistis</i> <i>Mussidia nigrivenella</i>		Ants
Osun state	B	Wasinmi	Maize intercropped with cassava, yam and sweet potato.	E004° 16.136'	N07° 26.301'	705.1m	30%	<i>Sesamia calamistis</i> <i>Busseola fusca</i> <i>Chilo Sp</i> <i>Eldana sacharina</i> <i>Mussidia nigrivenella</i>		Ants

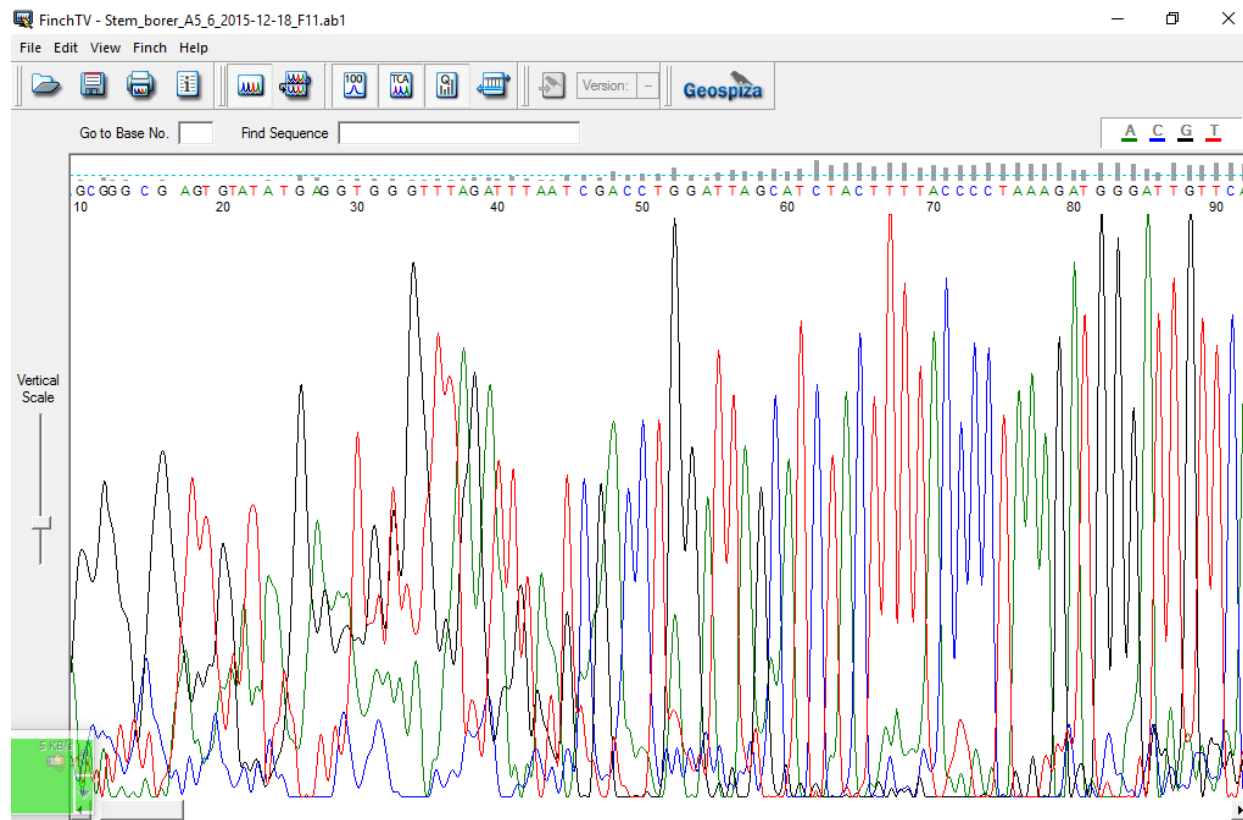
Osun state	C	Gbongan	Maize intercropped with cocoyam and cassava.	E004° 20.457'	N07° 26.301'	803.1m	40%	<i>Eldana saccharina</i> <i>Busseola fusca</i> <i>Diatraea saccharalis</i> <i>Diatraea lineolata</i> <i>Diatraea grandiosella</i> <i>Sesamia calamistis</i>	Ants
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Osun state	D	Ojo-Oloosa	Maize intercropped with cassava, and cocoyam.	E004° 31.748'	N07° 33.811'	859.1m	30%	<i>Antheraea pernyi</i> <i>Busseola fusca</i> <i>Sesamia calamistis</i> <i>Mussidia nigrivenella</i> <i>Coniesta ignefusalis</i>	Ants
Osun state	E	Isiriyun	Maize intercropped with cocoyam and okra.	E004° 41.389'	N07° 34.765'	1255m	60%	<i>Manga nubifera</i> <i>Manga melanodonta</i> <i>Manga fuliginosa</i> <i>Busseola quadrata</i> <i>Busseola fusca</i> <i>Sesamia calamistis</i> <i>Sesamia nonagrioides</i>	

# ANALYSIS OF VARIANCE OF STEM BORER SURVEY DATA

CHARACTER	SOURCE	Sum of Square	Df	Mean Square	F	Sig.
ALTITUDE	MODEL	6138232.295	5	1227646.459	46.219	100
	ERROR	637475.440	24	26561.477		
	SUM	6775707.735	29			
% Infestation	MODEL	5664.167	5	1132.833	2.912	0.034
	ERROR	9335.200	24	388.967		
	SUM	1499.367	29			
Stem Borer Complex	MODEL	19.067	5	3.813	5.867	0.001
	ERROR	15.600	24	0.650		
	SUM	34.667	29			
Cropping system	MODEL	2.200	5	0.440	2.200	0.088
	ERROR	4.800	24	0.200		
	SUM	7.000	29			





Stem\_borer\_A5\_6\_2015-12-18\_F11 (295 nucleotides)

CGGTTTCAAGCGGGCGAGTGTATATGAGGTGGGTTTAGATTTAATCGACCTGGATTAGCATCTACTTTTACCCCTAAAGATGGGATTGTTTCATGAGTGGATTAC  
 ATCTGTAGCTGTTACTATAATTCGAATTTGATTATTTATTGGTAAATAATACGATTATCTACATCTAAAAGTCGAAAATTATTATTTTTAAATCTTTTGAGGGAA  
 TTATGTAAGAATCAAATTCTACATTATTAATAATCAGAGTATTCGTATCTTCAATATCATTCTGGTTCAATATGAACTGGTGTTA

## BLAST Result for Stem\_borer\_A5\_6\_2015-12-18\_F11

NCBI Blast:Stem\_borer\_A5\_6\_2015-12-18\_F11

blast.ncbi.nlm.nih.gov/Blast.cgi

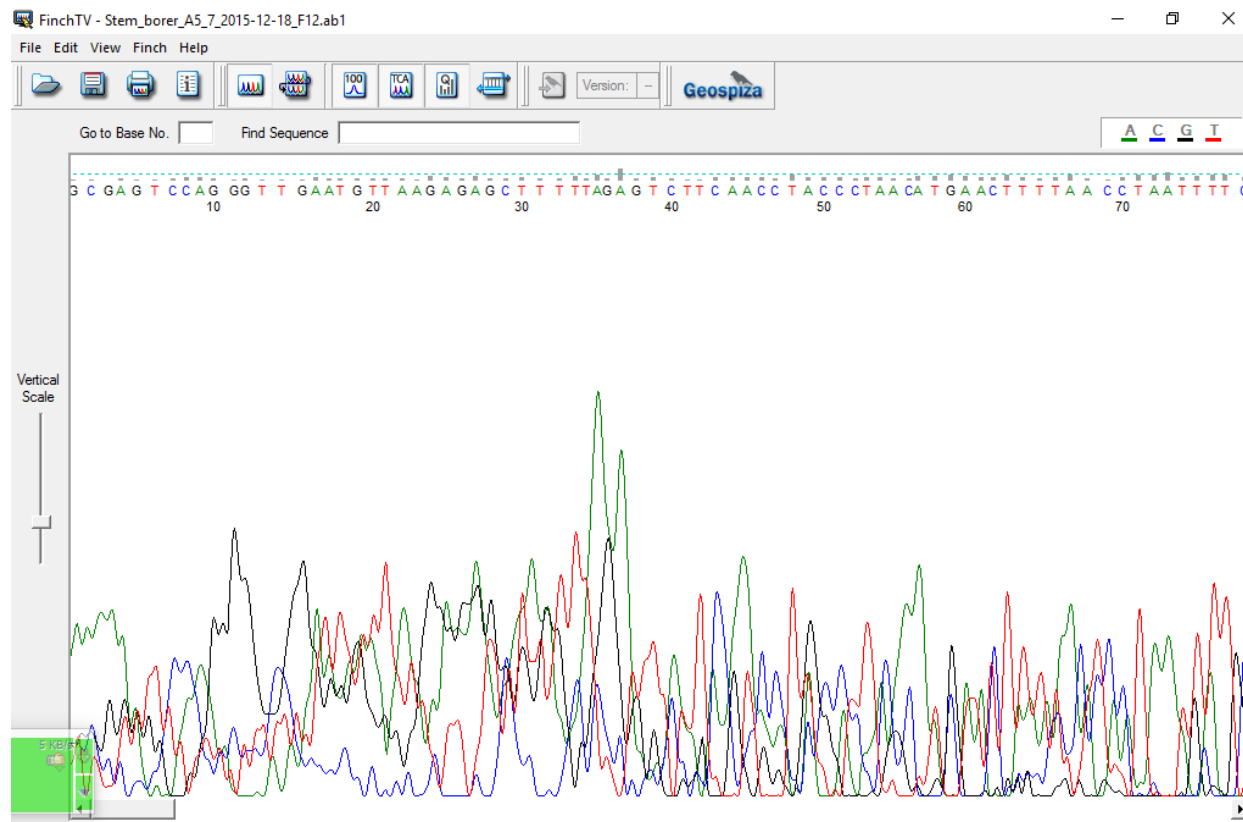
Sequences producing significant alignments:

Select: [All](#) [None](#) Selected:0

[Alignments](#) [Download](#) [GenBank](#) [Graphics](#) [Distance tree of results](#)

	Description	Max score	Total score	Query cover	E value	Ident	Accession
<input type="checkbox"/>	<a href="#">Chilo orichalcociliellus from Kenya cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	336	336	72%	2e-88	95%	<a href="#">AY320478.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate R-3 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, parti</a>	320	320	80%	1e-83	90%	<a href="#">JF798450.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate LA-1 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, part</a>	320	320	80%	1e-83	90%	<a href="#">JF798440.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate H-2 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, parti</a>	320	320	80%	1e-83	90%	<a href="#">JF798448.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate G-5 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, parti</a>	315	315	80%	6e-82	89%	<a href="#">JF798452.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate G-1 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, parti</a>	315	315	80%	6e-82	89%	<a href="#">JF798451.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate N-3 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, parti</a>	315	315	80%	6e-82	89%	<a href="#">JF798446.1</a>
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<input type="checkbox"/>	<a href="#">Chilo partellus isolate L-1 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, parti</a>	315	315	80%	6e-82	89%	<a href="#">JF798442.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate H-1 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, parti</a>	311	311	80%	7e-81	89%	<a href="#">JF798447.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate LA-5 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, part</a>	311	311	80%	7e-81	89%	<a href="#">JF798441.1</a>
<input type="checkbox"/>	<a href="#">Sphinx istar cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	311	311	80%	7e-81	89%	<a href="#">HQ677796.1</a>
<input type="checkbox"/>	<a href="#">Chilo infuscatellus from India cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	309	309	72%	3e-80	92%	<a href="#">AY320476.1</a>
<input type="checkbox"/>	<a href="#">Bombyx huttoni voucher BHUT20141201 mitochondrion, complete genome</a>	307	359	88%	9e-80	88%	<a href="#">KP216766.1</a>
<input type="checkbox"/>	<a href="#">Manduca sexta mitochondrion, complete genome</a>	307	307	84%	9e-80	88%	<a href="#">EU286785.1</a>

## Stem\_borer\_A5\_7\_2015-12-18\_F12



(291 nucleotides)

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NCBI Blast:Stem\_borer\_ X

blast.ncbi.nlm.nih.gov/Blast.cgi

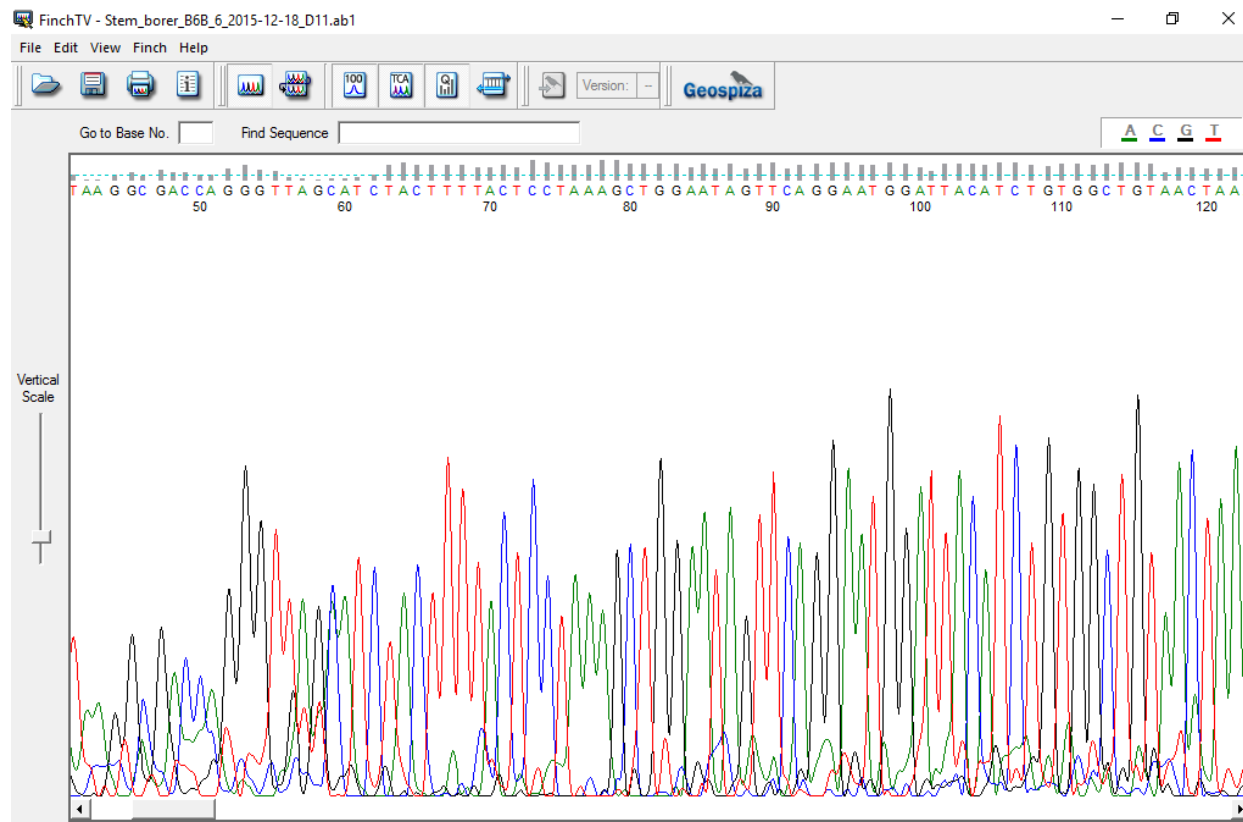
Sequences producing significant alignments:

Select: [All](#) [None](#) Selected:0

[Alignments](#) [Download](#) [GenBank](#) [Graphics](#) [Distance tree of results](#)

	Description	Max score	Total score	Query cover	E value	Ident	Accession
<input type="checkbox"/>	<a href="#">Chilo orichalcociliellus from Kenya cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	333	333	75%	2e-87	94%	<a href="#">AY320478.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate G-1 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, parti</a>	318	318	75%	5e-83	92%	<a href="#">JF798451.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate R-3 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, parti</a>	318	318	75%	5e-83	92%	<a href="#">JF798450.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate LA-1 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, part</a>	318	318	75%	5e-83	92%	<a href="#">JF798440.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate L-1 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, parti</a>	318	318	75%	5e-83	92%	<a href="#">JF798442.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate LA-5 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, part</a>	318	318	75%	5e-83	92%	<a href="#">JF798441.1</a>
<input type="checkbox"/>	<a href="#">Allanacstria cretica isolate FS-b-2038 cytochrome oxidase subunit I (COI) gene, partial sequence; tr</a>	318	318	75%	5e-83	92%	<a href="#">DQ351041.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus from Kenya cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	318	318	75%	5e-83	92%	<a href="#">AY320480.1</a>
<input type="checkbox"/>	<a href="#">Chilo auricilius from India cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	318	318	75%	5e-83	92%	<a href="#">AY320475.1</a>
<input type="checkbox"/>	<a href="#">Allanacstria cerisyi cytochrome c oxidase I (COI) gene, partial cds; tRNA-Leu gene, complete sequer</a>	318	318	75%	5e-83	92%	<a href="#">AF170869.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate G-5 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, parti</a>	315	315	75%	6e-82	92%	<a href="#">JF798452.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate H-2 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, parti</a>	315	315	75%	6e-82	92%	<a href="#">JF798448.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate N-1 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, parti</a>	315	315	75%	6e-82	92%	<a href="#">JF798444.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus from Zimbabwe cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	315	315	75%	6e-82	92%	<a href="#">AY320482.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus from South Africa cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	315	315	75%	6e-82	92%	<a href="#">AY320481.1</a>
<input type="checkbox"/>	<a href="#">Antheraea pernyi strain 731 mitochondrion, complete genome</a>	309	309	75%	3e-80	91%	<a href="#">KP881616.1</a>
<input type="checkbox"/>	<a href="#">Chilo partellus isolate H-1 cytochrome oxidase subunit II gene, partial cds; and tRNA-Lys gene, parti</a>	309	309	75%	3e-80	91%	<a href="#">JF798447.1</a>

## Stem\_borer\_B6B\_6\_2015-12-18\_D11



(294 nucleotides)

```
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ATCTGTGGCTGTAACATAAATTGCAATTTGATTATTTATTGGTAAAATAATTCGATTATCTACATCTAATAGGCGAAAATTATTAGAAGAAAGTTCATTTCTAGAG
ATTATGTAGGAATCAAATTCAATATTAATAAATACTGAATATTCATATCTTCAATATCATTCTGGTTCAATATGAACTGGTGTT
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NCBI Blast:Stem\_borer\_I X +

blast.ncbi.nlm.nih.gov/Blast.cgi

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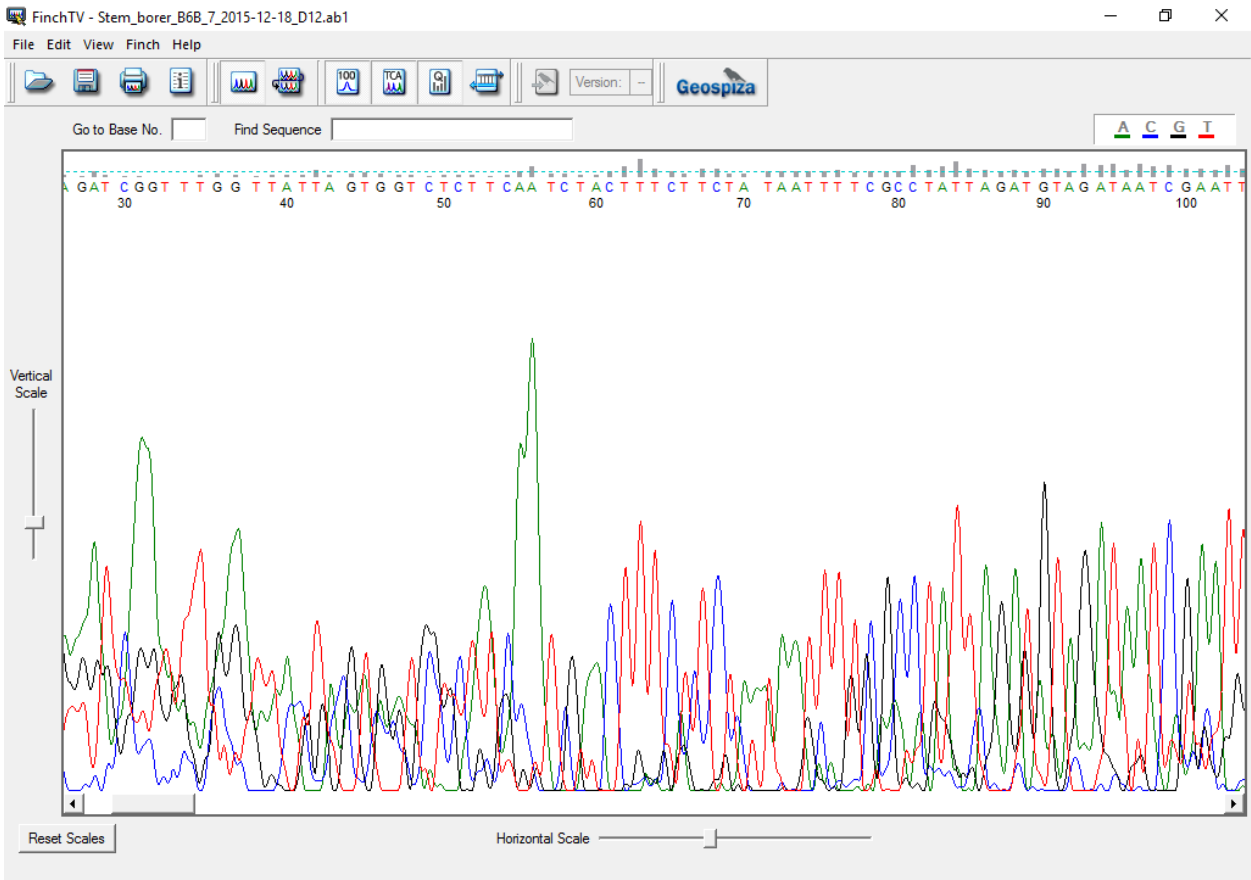
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[Alignments](#) [Download](#) [GenBank](#) [Graphics](#) [Distance tree of results](#)

	Description	Max score	Total score	Query cover	E value	Ident	Accession
<input type="checkbox"/>	<a href="#">Eldana saccharina from Kenya cytochrome oxidase subunit II (COII) gene, partial cds; mitochondri</a>	396	396	74%	2e-106	100%	<a href="#">AY320502.1</a>
<input type="checkbox"/>	<a href="#">Eldana saccharina from Zimbabwe cytochrome oxidase subunit II (COII) gene, partial cds; mitoch</a>	387	387	74%	1e-103	99%	<a href="#">AY320504.1</a>
<input type="checkbox"/>	<a href="#">Eldana saccharina from South Africa cytochrome oxidase subunit II (COII) gene, partial cds; mitoc</a>	372	372	74%	3e-99	98%	<a href="#">AY320503.1</a>
<input type="checkbox"/>	<a href="#">Noctuidae sp. VB-2010 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	315	315	82%	6e-82	89%	<a href="#">HQ677797.1</a>
<input type="checkbox"/>	<a href="#">Corcyra cephalonica mitochondrion complete genome</a>	313	313	86%	2e-81	87%	<a href="#">HQ897685.1</a>
<input type="checkbox"/>	<a href="#">Feltia jaculifera cytochrome oxidase subunit 1 (COI) and subunit 2 (COII) genes, complete cds, an</a>	313	313	82%	2e-81	89%	<a href="#">U60990.1</a>
<input type="checkbox"/>	<a href="#">Meroptera pravella voucher JBWM0363025 mitochondrion complete genome</a>	311	311	82%	7e-81	88%	<a href="#">MF073207.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria auranticella haplotype OS1 cytochrome oxidase subunit I (COI) gene, partial cds; tRNA-I</a>	311	311	82%	7e-81	88%	<a href="#">DQ295176.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria auranticella voucher Du02 cytochrome c oxidase subunit I (COI) gene, partial cds; tRNA-</a>	311	311	82%	7e-81	88%	<a href="#">DQ247736.1</a>
<input type="checkbox"/>	<a href="#">Hippotion celerio mitochondrial coxI (partial), tRNA-Leu and coxII (partial) genes, isolate 695934</a>	311	311	82%	7e-81	88%	<a href="#">AJ749424.1</a>
<input type="checkbox"/>	<a href="#">Hippotion celerio mitochondrial coxI (partial), tRNA-Leu and coxII (partial) genes, isolate 16132</a>	311	311	82%	7e-81	88%	<a href="#">AJ749422.1</a>
<input type="checkbox"/>	<a href="#">Feltia jaculifera voucher FSb152 cytochrome oxidase subunit I (COI) gene, partial cds; tRNA-Leu g</a>	307	307	82%	9e-80	88%	<a href="#">DQ792591.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa sp. Bahia-02 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	302	302	82%	4e-78	88%	<a href="#">KF625005.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria resinosella voucher AR386 cytochrome c oxidase I and cytochrome c oxidase II genes, r</a>	302	302	82%	4e-78	88%	<a href="#">JN162736.1</a>
<input type="checkbox"/>	<a href="#">Nemoria leptalea voucher MDLEP1139 cytochrome oxidase subunit II (COII) gene, partial cds; mit</a>	302	302	82%	4e-78	88%	<a href="#">EU151620.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria ponderosae voucher Du114 cytochrome c oxidase subunit I (COI) gene, partial cds; tRN</a>	302	302	82%	4e-78	88%	<a href="#">DQ247733.1</a>
<input type="checkbox"/>	<a href="#">Cnaphalocrocis medinalis mitochondrion complete genome</a>	300	300	90%	1e-77	85%	<a href="#">JQ647917.1</a>

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Stem\_borer\_B6B\_7\_2015-12-18\_D12 (290 nucleotides)

TAAATTCCTGAGTCAGTTTAATTTGAGATCGGTTTGGTTATTAGTGGTCTCTTCAATCTACTTTCTTCTATAATTTTCGCCTATTAGATGTAGATAATCGAATTATT  
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Stem\_borer\_B6B\_7\_2015-12-18\_D12

NCBI Blast:Stem\_borer\_B6B\_ NCBI Blast:Stem\_borer\_I

blast.ncbi.nlm.nih.gov/Blast.cgi

Sequences producing significant alignments:

Select: [All](#) [None](#) Selected: 0

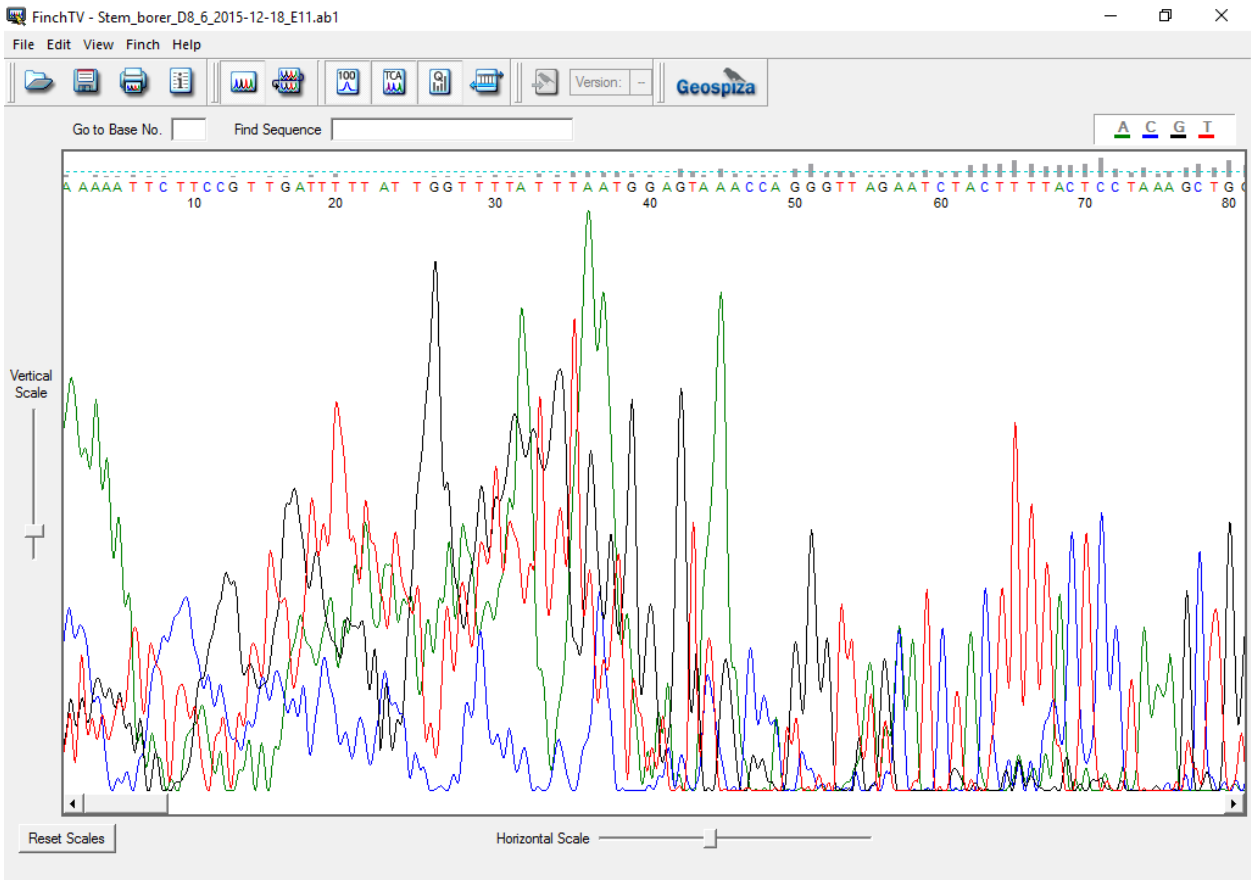
[Alignments](#) [Download](#) [GenBank](#) [Graphics](#) [Distance tree of results](#)

	Description	Max score	Total score	Query cover	E value	Ident	Accession
<input type="checkbox"/>	<a href="#">Eldana saccharina from Kenya cytochrome oxidase subunit II (COII) gene, partial cds: mitochondrial</a>	390	390	79%	9e-105	98%	<a href="#">AY320502.1</a>
<input type="checkbox"/>	<a href="#">Eldana saccharina from Zimbabwe cytochrome oxidase subunit II (COII) gene, partial cds: mitochon</a>	381	381	79%	5e-102	97%	<a href="#">AY320504.1</a>
<input type="checkbox"/>	<a href="#">Eldana saccharina from South Africa cytochrome oxidase subunit II (COII) gene, partial cds: mitoch</a>	378	378	79%	6e-101	97%	<a href="#">AY320503.1</a>
<input type="checkbox"/>	<a href="#">Spodoptera litura mitochondrion, complete genome</a>	316	316	75%	2e-82	92%	<a href="#">KF701043.1</a>
<input type="checkbox"/>	<a href="#">Spodoptera litura mitochondrion, complete genome</a>	316	316	75%	2e-82	92%	<a href="#">JQ647918.1</a>
<input type="checkbox"/>	<a href="#">Parnassius tenedius isolate FS-b-1784 cytochrome oxidase subunit I (COI) gene, partial sequence:</a>	313	313	75%	2e-81	92%	<a href="#">DQ351027.1</a>
<input type="checkbox"/>	<a href="#">Papilio protenor euprotenor isolate AB362 cytochrome oxidase subunit I gene, partial cds: tRNA-Le</a>	311	311	75%	7e-81	91%	<a href="#">KX557579.1</a>
<input type="checkbox"/>	<a href="#">Papilio protenor mitochondrion, complete genome</a>	311	311	75%	7e-81	91%	<a href="#">KY272622.1</a>
<input type="checkbox"/>	<a href="#">Astenoptycha cf. sphaltica AZ-2013 cytochrome c oxidase subunit II (COII) gene, partial cds: mitoc</a>	311	311	75%	7e-81	91%	<a href="#">KC315465.1</a>
<input type="checkbox"/>	<a href="#">Spodoptera litura mitochondrion, complete genome</a>	311	311	75%	7e-81	91%	<a href="#">KF543065.1</a>
<input type="checkbox"/>	<a href="#">Papilio maackii mitochondrion, complete genome</a>	311	311	75%	7e-81	91%	<a href="#">KC433408.1</a>
<input type="checkbox"/>	<a href="#">Papilio syfanius isolate Ps22 cytochrome oxidase subunit II (COII) gene, partial cds: mitochondrial</a>	311	311	75%	7e-81	91%	<a href="#">JF281184.1</a>
<input type="checkbox"/>	<a href="#">Papilio syfanius isolate Ps17 cytochrome oxidase subunit II (COII) gene, partial cds: mitochondrial</a>	311	311	75%	7e-81	91%	<a href="#">JF281179.1</a>
<input type="checkbox"/>	<a href="#">Spilosoma virginica cytochrome oxidase subunit II (COII) gene, partial cds: mitochondrial</a>	311	311	75%	7e-81	91%	<a href="#">HQ677812.1</a>
<input type="checkbox"/>	<a href="#">Papilio maackii voucher UASM 9900084 from Japan cytochrome oxidase subunit I (COI) gene, parti</a>	311	311	75%	7e-81	91%	<a href="#">AY457573.1</a>
<input type="checkbox"/>	<a href="#">Corcyra cephalonica mitochondrion, complete genome</a>	309	309	79%	2e-80	90%	<a href="#">HQ897685.1</a>
<input type="checkbox"/>	<a href="#">Noctuidae sp. VR-2010 cytochrome oxidase subunit II (COII) gene, partial cds: mitochondrial</a>	309	309	79%	2e-80	90%	<a href="#">HQ677779.1</a>

https://blast.ncbi.nlm.nih.gov/Blast.cgi#alnHdr\_315229721



Stem\_borer\_D8\_6\_2015-12-18\_E11



Stem\_borer\_D8\_6\_2015-12-18\_E11 (294 nucleotides)

AAAAATTCTTCCGTTGATTTTTATTGGTTTTATTTAATGGAGTAAACCAGGGTTAGAATCTACTTTTACTCCTAAAGCTGGAATAGTTCAGGAATGGATTACATCT  
GTGGCTGTAATAAAATTCGAATTTGATTATTTATTGGTAAAATAATTCGATTATCTACATCTAATAGGCGAAAATTATTAGAAGAAAGTTCATTTCTAGAGATT  
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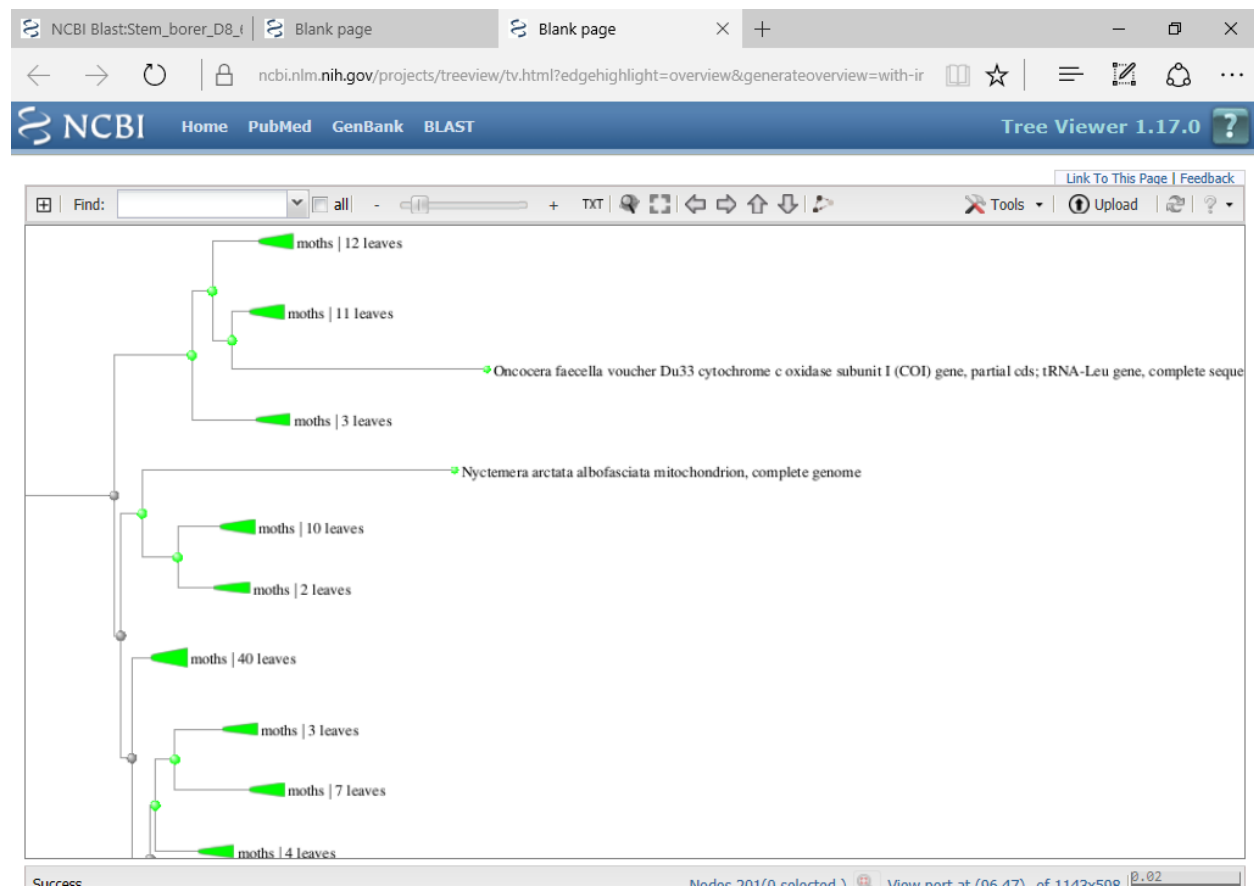
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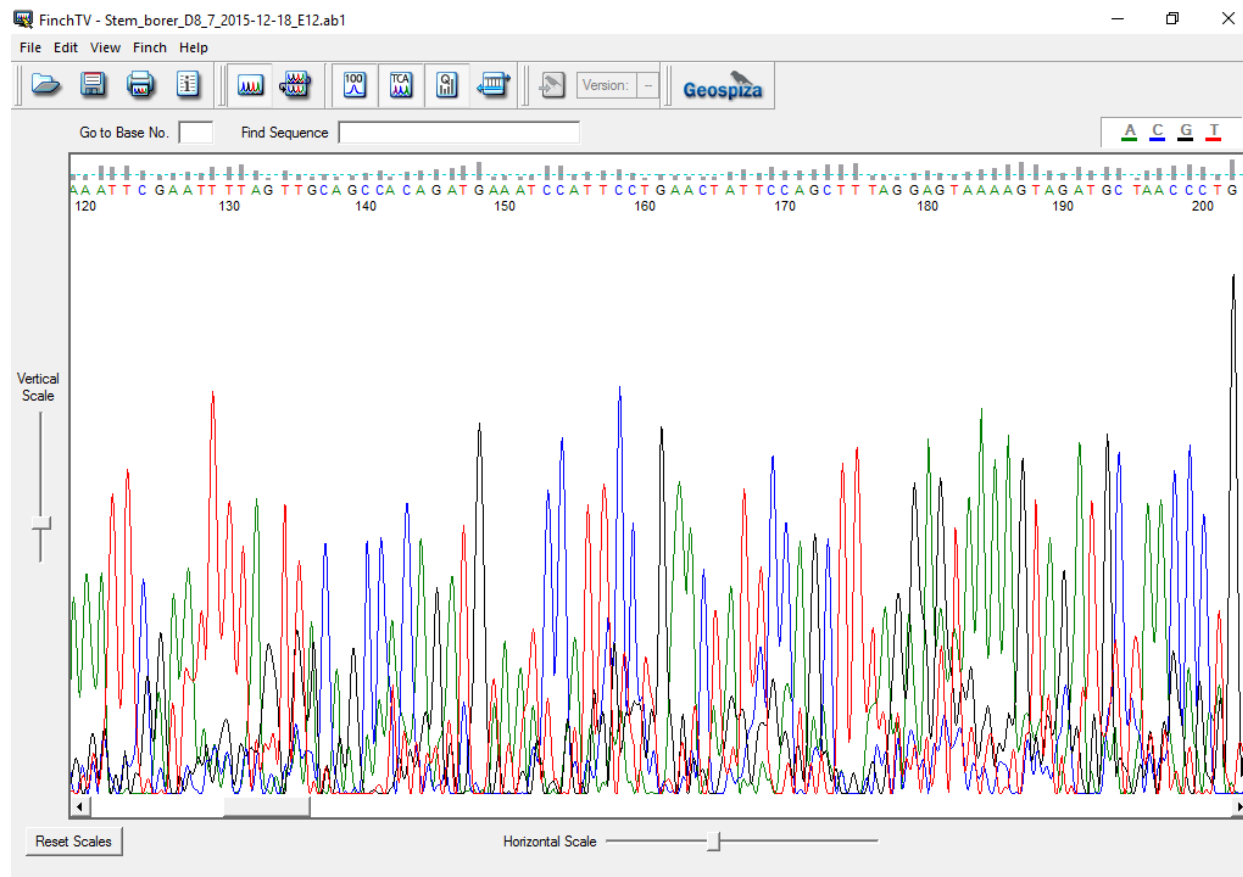
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	Description	Max score	Total score	Query cover	E value	Ident	Accession
<input type="checkbox"/>	<a href="#">Eidana saccharina from Kenya cytochrome oxidase subunit II (COII) gene, partial cds, mitochondrial</a>	358	358	68%	6e-95	99%	<a href="#">AY320502.1</a>
<input type="checkbox"/>	<a href="#">Eidana saccharina from Zimbabwe cytochrome oxidase subunit II (COII) gene, partial cds, mitochondrial</a>	349	349	68%	3e-92	99%	<a href="#">AY320504.1</a>
<input type="checkbox"/>	<a href="#">Eidana saccharina from South Africa cytochrome oxidase subunit II (COII) gene, partial cds, mitochondrial</a>	336	336	68%	2e-88	97%	<a href="#">AY320503.1</a>
<input type="checkbox"/>	<a href="#">Meroptera pravella voucher JBWM0363025 mitochondrion, complete genome</a>	297	297	84%	2e-76	87%	<a href="#">MF073207.1</a>
<input type="checkbox"/>	<a href="#">Corcyra cephalonica mitochondrion, complete genome</a>	295	295	87%	6e-76	85%	<a href="#">HQ897685.1</a>
<input type="checkbox"/>	<a href="#">Noctuidae sp. VB-2010 cytochrome oxidase subunit II (COII) gene, partial cds, mitochondrial</a>	293	293	80%	2e-75	87%	<a href="#">HQ677797.1</a>
<input type="checkbox"/>	<a href="#">Hippotion celerio mitochondrial coxI (partial), tRNA-Leu and coxII (partial) genes, isolate 695934</a>	291	291	76%	7e-75	89%	<a href="#">AJ749424.1</a>
<input type="checkbox"/>	<a href="#">Hippotion celerio mitochondrial coxI (partial), tRNA-Leu and coxII (partial) genes, isolate 16132</a>	291	291	76%	7e-75	89%	<a href="#">AJ749422.1</a>
<input type="checkbox"/>	<a href="#">Dichorda iridaria voucher MC02C301 cytochrome oxidase subunit II (COII) gene, partial cds, mitochondrial</a>	289	289	87%	2e-74	85%	<a href="#">EU151649.1</a>
<input type="checkbox"/>	<a href="#">Chlorochlamys chloroleucaria voucher MC02C205 cytochrome oxidase subunit II (COII) gene, partial cds, mitochondrial</a>	289	289	87%	2e-74	85%	<a href="#">EU151648.1</a>
<input type="checkbox"/>	<a href="#">Acanthopteryx pulvipennella cytochrome oxidase subunit II gene, partial cds, mitochondrial</a>	289	289	75%	2e-74	89%	<a href="#">AY527040.1</a>
<input type="checkbox"/>	<a href="#">Hyles livornicoides mitochondrial coxI gene (partial), tRNA-Leu and coxII gene (partial), isolate 3466</a>	286	286	75%	3e-73	88%	<a href="#">FN386575.1</a>
<input type="checkbox"/>	<a href="#">Nemoria leptalea voucher MDLEP1139 cytochrome oxidase subunit II (COII) gene, partial cds, mitochondrial</a>	286	286	75%	3e-73	88%	<a href="#">EU151620.1</a>



Stem\_borer\_D8\_7\_2015-12-18\_E12



Stem\_borer\_D8\_7\_2015-12-18\_E12 (290 nucleotides)

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ACCAATAAATATCCAAATTCGAATTTTAGTTGCAGCCACAGATGAAATCCATTCTCTGAAGCTATTCAGCTTTAGGAGTAAAAGTAGATGCTAACCCCTGGTCGCCT  
TAACCAAATAATTTTTTTTATTAATCGCCCTGGAATTTTTTATGGTCAATGTTTCAGAAATTTGTGGAGCTTAATCAAAA
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## Stem\_borer\_D8\_7\_2015-12-18\_E12

NCBI Blast:Stem\_borer\_I X +

blast.ncbi.nlm.nih.gov/Blast.cgi

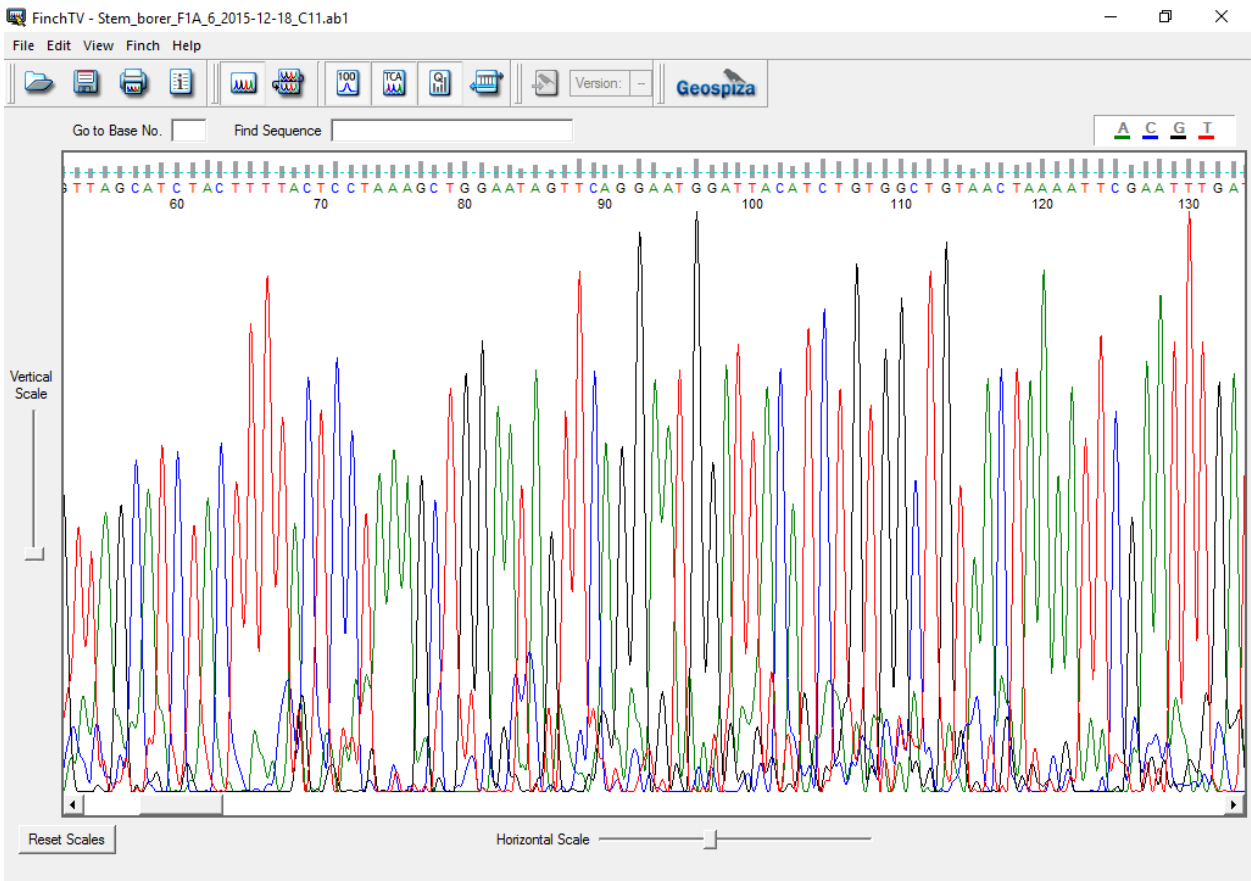
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	Description	Max score	Total score	Query cover	E value	Ident	Accession
<input type="checkbox"/>	<a href="#">Eidana saccharina from Kenya cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	324	324	76%	1e-84	93%	<a href="#">AY320502.1</a>
<input type="checkbox"/>	<a href="#">Eidana saccharina from Zimbabwe cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	315	315	76%	6e-82	92%	<a href="#">AY320504.1</a>
<input type="checkbox"/>	<a href="#">Eidana saccharina from South Africa cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	311	311	76%	7e-81	92%	<a href="#">AY320503.1</a>
<input type="checkbox"/>	<a href="#">Epitymbia alaudana cytochrome c oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	266	266	76%	3e-67	87%	<a href="#">KC315475.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa zea haplotype 6 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	264	264	75%	9e-67	88%	<a href="#">HQ677776.1</a>
<input type="checkbox"/>	<a href="#">Parmassius tenedius isolate FS-b-1784 cytochrome oxidase subunit I (COI) gene, partial sequence; full</a>	262	262	76%	3e-66	87%	<a href="#">DQ351027.1</a>
<input type="checkbox"/>	<a href="#">Syndemis sp. ms646bcrk cytochrome c oxidase subunit 2 (COII) gene, partial cds; mitochondrial</a>	260	260	76%	1e-65	87%	<a href="#">KY501341.1</a>
<input type="checkbox"/>	<a href="#">Syndemis sp. ms645bcrk cytochrome c oxidase subunit 2 (COII) gene, partial cds; mitochondrial</a>	260	260	76%	1e-65	87%	<a href="#">KY501340.1</a>
<input type="checkbox"/>	<a href="#">Syndemis sp. ms644bcrk cytochrome c oxidase subunit 2 (COII) gene, partial cds; mitochondrial</a>	260	260	76%	1e-65	87%	<a href="#">KY501339.1</a>
<input type="checkbox"/>	<a href="#">Syndemis sp. ms643bcrk cytochrome c oxidase subunit 2 (COII) gene, partial cds; mitochondrial</a>	260	260	76%	1e-65	87%	<a href="#">KY501338.1</a>
<input type="checkbox"/>	<a href="#">Syndemis sp. as02bcrk cytochrome c oxidase subunit 2 (COII) gene, partial cds; mitochondrial</a>	260	260	76%	1e-65	87%	<a href="#">KY501319.1</a>
<input type="checkbox"/>	<a href="#">Syndemis sp. as01bcrk cytochrome c oxidase subunit 2 (COII) gene, partial cds; mitochondrial</a>	260	260	76%	1e-65	87%	<a href="#">KY501318.1</a>
<input type="checkbox"/>	<a href="#">Syndemis sp. dr137bcrk cytochrome c oxidase subunit 2 (COII) gene, partial cds; mitochondrial</a>	260	260	76%	1e-65	87%	<a href="#">KY501309.1</a>
<input type="checkbox"/>	<a href="#">Syndemis sp. dr136bcrk cytochrome c oxidase subunit 2 (COII) gene, partial cds; mitochondrial</a>	260	260	76%	1e-65	87%	<a href="#">KY501308.1</a>
<input type="checkbox"/>	<a href="#">Syndemis sp. dr125bcrk cytochrome c oxidase subunit 2 (COII) gene, partial cds; mitochondrial</a>	260	260	76%	1e-65	87%	<a href="#">KY501299.1</a>
<input type="checkbox"/>	<a href="#">Syndemis sp. dr121bcrk cytochrome c oxidase subunit 2 (COII) gene, partial cds; mitochondrial</a>	260	260	76%	1e-65	87%	<a href="#">KY501295.1</a>
<input type="checkbox"/>	<a href="#">Dercas lycorias voucher D.lvc1 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	260	260	76%	1e-65	87%	<a href="#">KM669560.1</a>

Stem\_borer\_F1A\_6\_2015-12-18\_C11



Stem\_borer\_F1A\_6\_2015-12-18\_C11 (295 nucleotides)

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TGTGGCTGTAATAAAATTCGAATTTGATTATTTATTGGTAAATAATTCGATTATCTACATCTAATAGGCGAAAATTATTAGAAGAAAGTTCATTTCTAGAGATT  
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Stem\_borer\_F1A\_6\_2015-12-18\_C11

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blast.ncbi.nlm.nih.gov/Blast.cgi

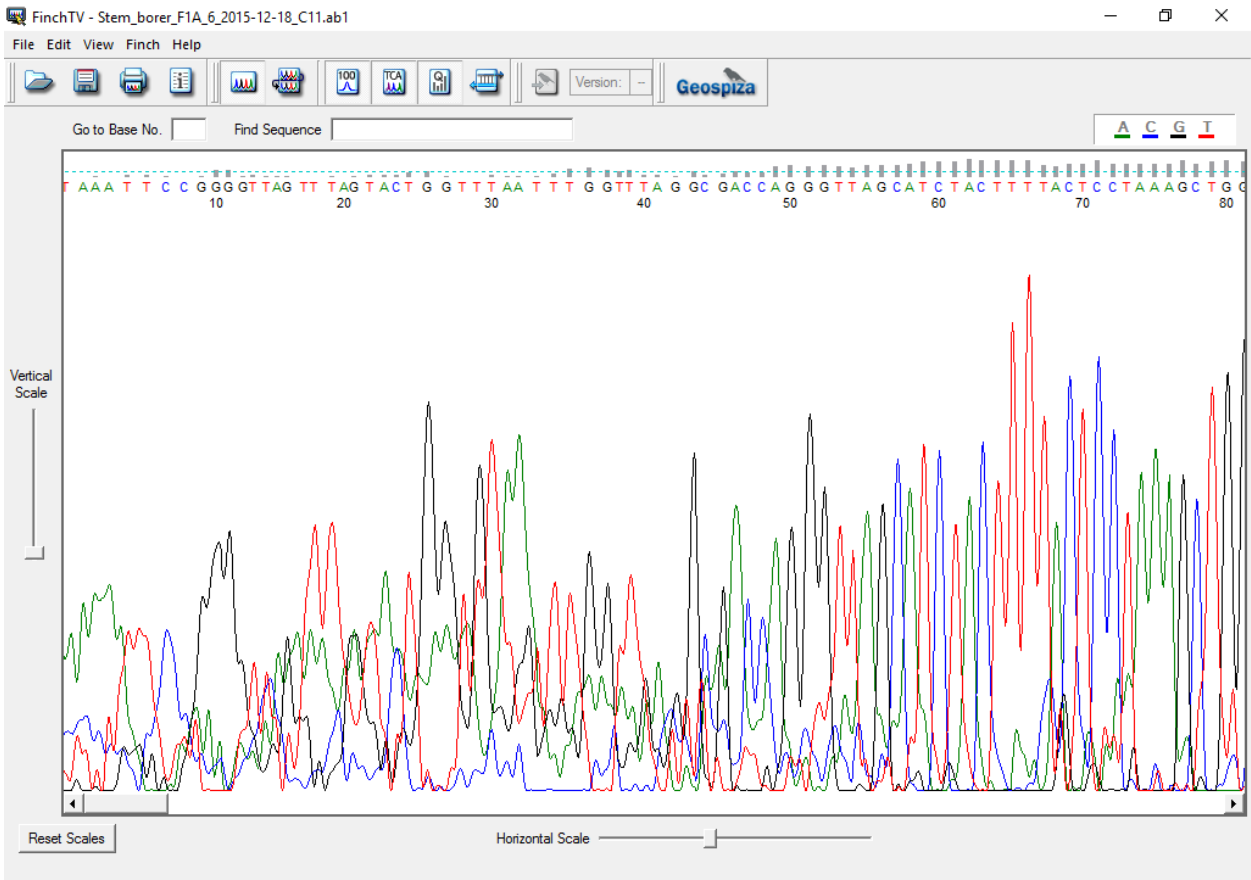
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Select: [All](#) [None](#) Selected:0

[Alignments](#) [Download](#) [GenBank](#) [Graphics](#) [Distance tree of results](#)

	Description	Max score	Total score	Query cover	E value	Ident	Accession
<input type="checkbox"/>	<a href="#">Eidana saccharina from Kenya cytochrome oxidase subunit II (COII) gene, partial cds; mitochondria</a>	385	385	73%	4e-103	99%	<a href="#">AY320502.1</a>
<input type="checkbox"/>	<a href="#">Eidana saccharina from Zimbabwe cytochrome oxidase subunit II (COII) gene, partial cds; mitochondria</a>	376	376	73%	2e-100	98%	<a href="#">AY320504.1</a>
<input type="checkbox"/>	<a href="#">Eidana saccharina from South Africa cytochrome oxidase subunit II (COII) gene, partial cds; mitochondria</a>	361	361	73%	5e-96	97%	<a href="#">AY320503.1</a>
<input type="checkbox"/>	<a href="#">Corcyra cephalonica mitochondrion complete genome</a>	313	313	84%	2e-81	88%	<a href="#">HQ897685.1</a>
<input type="checkbox"/>	<a href="#">Noctuidae sp. VB-2010 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	313	313	81%	2e-81	89%	<a href="#">HQ677797.1</a>
<input type="checkbox"/>	<a href="#">Hippotion celerio mitochondrial coxI (partial), tRNA-Leu and coxII (partial) genes, isolate 695934</a>	311	311	80%	7e-81	89%	<a href="#">AJ749424.1</a>
<input type="checkbox"/>	<a href="#">Hippotion celerio mitochondrial coxI (partial), tRNA-Leu and coxII (partial) genes, isolate 16132</a>	311	311	80%	7e-81	89%	<a href="#">AJ749422.1</a>
<input type="checkbox"/>	<a href="#">Meroptera pravella voucher JBWM0363025 mitochondrion, complete genome</a>	309	309	81%	3e-80	88%	<a href="#">MF073207.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria auranticella haplotype OS1 cytochrome oxidase subunit I (COI) gene, partial cds; tRNA-L</a>	309	309	81%	3e-80	88%	<a href="#">DQ295176.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria auranticella voucher Du02 cytochrome c oxidase subunit I (COI) gene, partial cds; tRNA-L</a>	309	309	81%	3e-80	88%	<a href="#">DQ247736.1</a>
<input type="checkbox"/>	<a href="#">Feltia jaculifera cytochrome oxidase subunit 1 (COI) and subunit 2 (COII) genes, complete cds, and</a>	302	302	81%	4e-78	88%	<a href="#">U60990.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria resinosella voucher AR386 cytochrome c oxidase I and cytochrome c oxidase II genes, p</a>	300	300	81%	1e-77	88%	<a href="#">JN162736.1</a>
<input type="checkbox"/>	<a href="#">Nemoria leptalea voucher MDLEP1139 cytochrome oxidase subunit II (COII) gene, partial cds; mito</a>	300	300	81%	1e-77	88%	<a href="#">EU151620.1</a>
<input type="checkbox"/>	<a href="#">Aqonopterix pulvipennella cytochrome oxidase subunit II gene, partial cds; mitochondrial</a>	300	300	80%	1e-77	89%	<a href="#">AY527040.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria ponderosae voucher Du114 cytochrome c oxidase subunit I (COI) gene, partial cds; tRNA</a>	300	300	81%	1e-77	88%	<a href="#">DQ247733.1</a>
<input type="checkbox"/>	<a href="#">Dichorda iridaria voucher MC02C301 cytochrome oxidase subunit II (COII) gene, partial cds; mito</a>	298	298	84%	5e-77	87%	<a href="#">EU151649.1</a>
<input type="checkbox"/>	<a href="#">Chlorochlamys chloroleucaria voucher MC02C205 cytochrome oxidase subunit II (COII) gene, parti</a>	298	298	84%	5e-77	87%	<a href="#">EU151648.1</a>

Stem\_borer\_F1A\_6\_2015-12-18\_C11





Stem\_borer\_F1A\_6\_2015-12-18\_C11

TAAATTCCGGGGTTAGTTTAGTACTGGTTTAATTTGGTTTAGGCGACCAGGGTTAGCATCTACTTTTACTCCTAAAGCTGGAATAGTTCAGGAATGGATTACATC  
TGTGGCTGTAATAAAATTCGAATTTGATTATTTATTGGTAAATAATTCGATTATCTACATCTAATAGGCGAAAATTATTAGAAGAAAGTTCATTTCTAGAGATT  
ATGTAGGAATCAAATTCATATTAATAAATCTGAATATTCATATCTTCAATATCATTCTGGTTCAATATGAACTGGTGTAATA

## Stem\_borer\_F1A\_6\_2015-12-18\_C11

NCBI Blast:Stem\_borer\_I X +

blast.ncbi.nlm.nih.gov/Blast.cgi

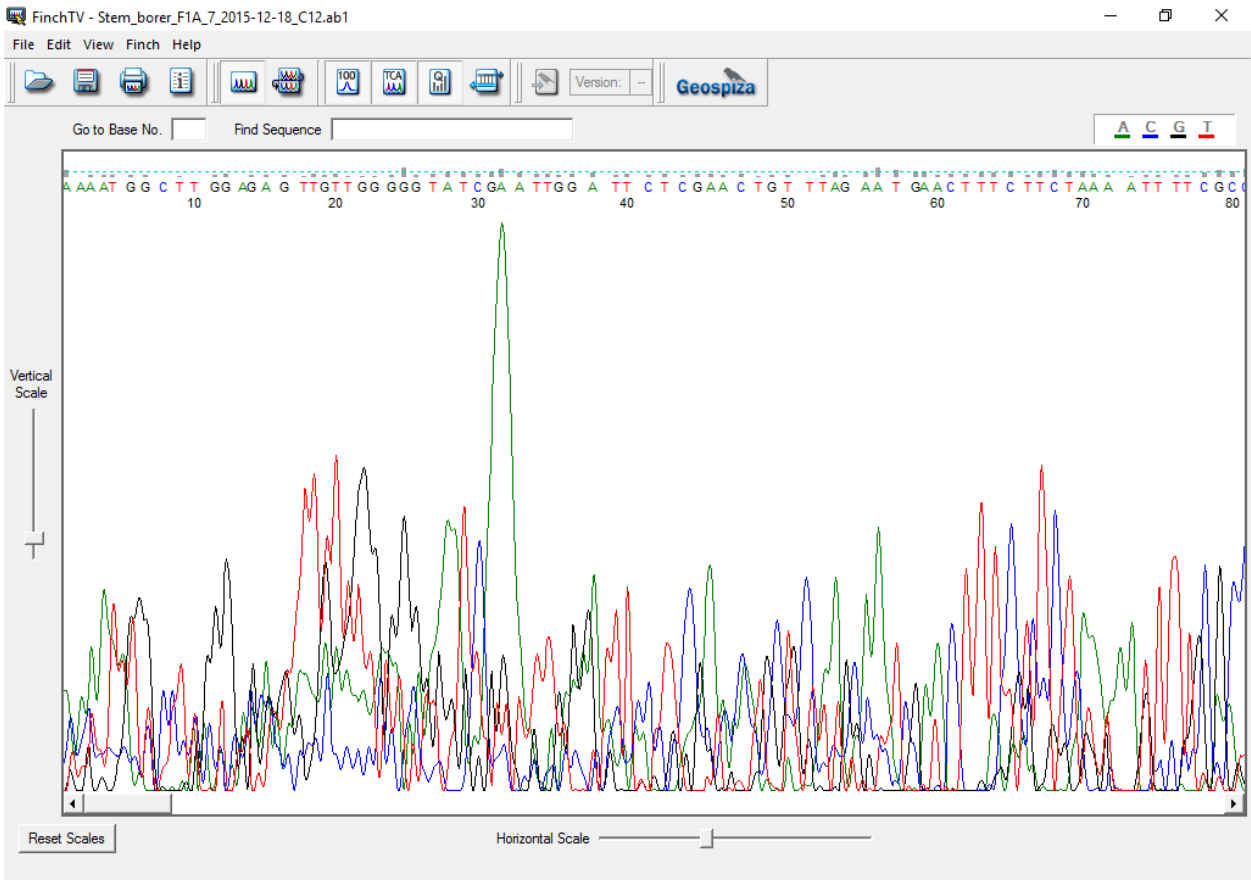
Sequences producing significant alignments:

Select: [All](#) [None](#) Selected:0

[Alignments](#) [Download](#) [GenBank](#) [Graphics](#) [Distance tree of results](#)

	Description	Max score	Total score	Query cover	E value	Ident	Accession
<input type="checkbox"/>	<a href="#">Eldana saccharina from Kenya cytochrome oxidase subunit II (COII) gene, partial cds; mitochondria</a>	385	385	73%	4e-103	99%	<a href="#">AY320502.1</a>
<input type="checkbox"/>	<a href="#">Eldana saccharina from Zimbabwe cytochrome oxidase subunit II (COII) gene, partial cds; mitochondria</a>	376	376	73%	2e-100	98%	<a href="#">AY320504.1</a>
<input type="checkbox"/>	<a href="#">Eldana saccharina from South Africa cytochrome oxidase subunit II (COII) gene, partial cds; mitochondria</a>	361	361	73%	5e-96	97%	<a href="#">AY320503.1</a>
<input type="checkbox"/>	<a href="#">Corcyra cephalonica mitochondrion, complete genome</a>	313	313	84%	2e-81	88%	<a href="#">HQ897685.1</a>
<input type="checkbox"/>	<a href="#">Noctuidae sp. VB-2010 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	313	313	81%	2e-81	89%	<a href="#">HQ677797.1</a>
<input type="checkbox"/>	<a href="#">Hippotion celerio mitochondrial coxI (partial), tRNA-Leu and coxII (partial) genes, isolate 695934</a>	311	311	80%	7e-81	89%	<a href="#">AJ749424.1</a>
<input type="checkbox"/>	<a href="#">Hippotion celerio mitochondrial coxI (partial), tRNA-Leu and coxII (partial) genes, isolate 16132</a>	311	311	80%	7e-81	89%	<a href="#">AJ749422.1</a>
<input type="checkbox"/>	<a href="#">Meroptera pravella voucher JBWM0363025 mitochondrion, complete genome</a>	309	309	81%	3e-80	88%	<a href="#">MF073207.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria auranticella haplotype OS1 cytochrome oxidase subunit I (COI) gene, partial cds; tRNA-L</a>	309	309	81%	3e-80	88%	<a href="#">DQ295176.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria auranticella voucher Du02 cytochrome c oxidase subunit I (COI) gene, partial cds; tRNA-L</a>	309	309	81%	3e-80	88%	<a href="#">DQ247736.1</a>
<input type="checkbox"/>	<a href="#">Feltia jaculifera cytochrome oxidase subunit 1 (COI) and subunit 2 (COII) genes, complete cds, and</a>	302	302	81%	4e-78	88%	<a href="#">U60990.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria resinosella voucher AR386 cytochrome c oxidase I and cytochrome c oxidase II genes, p</a>	300	300	81%	1e-77	88%	<a href="#">JN162736.1</a>
<input type="checkbox"/>	<a href="#">Nemoria leptalea voucher MDLEP1139 cytochrome oxidase subunit II (COII) gene, partial cds; mito</a>	300	300	81%	1e-77	88%	<a href="#">EU151620.1</a>
<input type="checkbox"/>	<a href="#">Aqonopterix pulvipennella cytochrome oxidase subunit II gene, partial cds; mitochondrial</a>	300	300	80%	1e-77	89%	<a href="#">AY527040.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria ponderosae voucher Du114 cytochrome c oxidase subunit I (COI) gene, partial cds; tRNA</a>	300	300	81%	1e-77	88%	<a href="#">DQ247733.1</a>
<input type="checkbox"/>	<a href="#">Dichorda iridaria voucher MC02C301 cytochrome oxidase subunit II (COII) gene, partial cds; mito</a>	298	298	84%	5e-77	87%	<a href="#">EU151649.1</a>
<input type="checkbox"/>	<a href="#">Chlorochlamys chloroleucaria voucher MC02C205 cytochrome oxidase subunit II (COII) gene, parti</a>	298	298	84%	5e-77	87%	<a href="#">EU151648.1</a>

Stem\_borer\_F1A\_7\_2015-12-18\_C12



Stem\_borer\_F1A\_7\_2015-12-18\_C12 (294 nucleotides)

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TTTACCAATAAAAAATCAAATTCGAATTTTAGTGACAGCCACAGATGTAATCCATTCTGAACTATTCCAGCTTTAGGAGTAAAAGTAGATGCTAACCTGGTCG  
CCTTAACCAAATAATTTTTTTTATTAATCGCCCTGGAATTTTTTATGGTCAATGTTTCAGAAATTTGTGGAGCTAATCAAAAATT

Stem\_borer\_F1A\_7\_2015-12-18\_C12

NCBI Blast:Stem\_borer\_F1A\_ NCBI Blast:Stem\_borer\_I\_ X

blast.ncbi.nlm.nih.gov/Blast.cgi

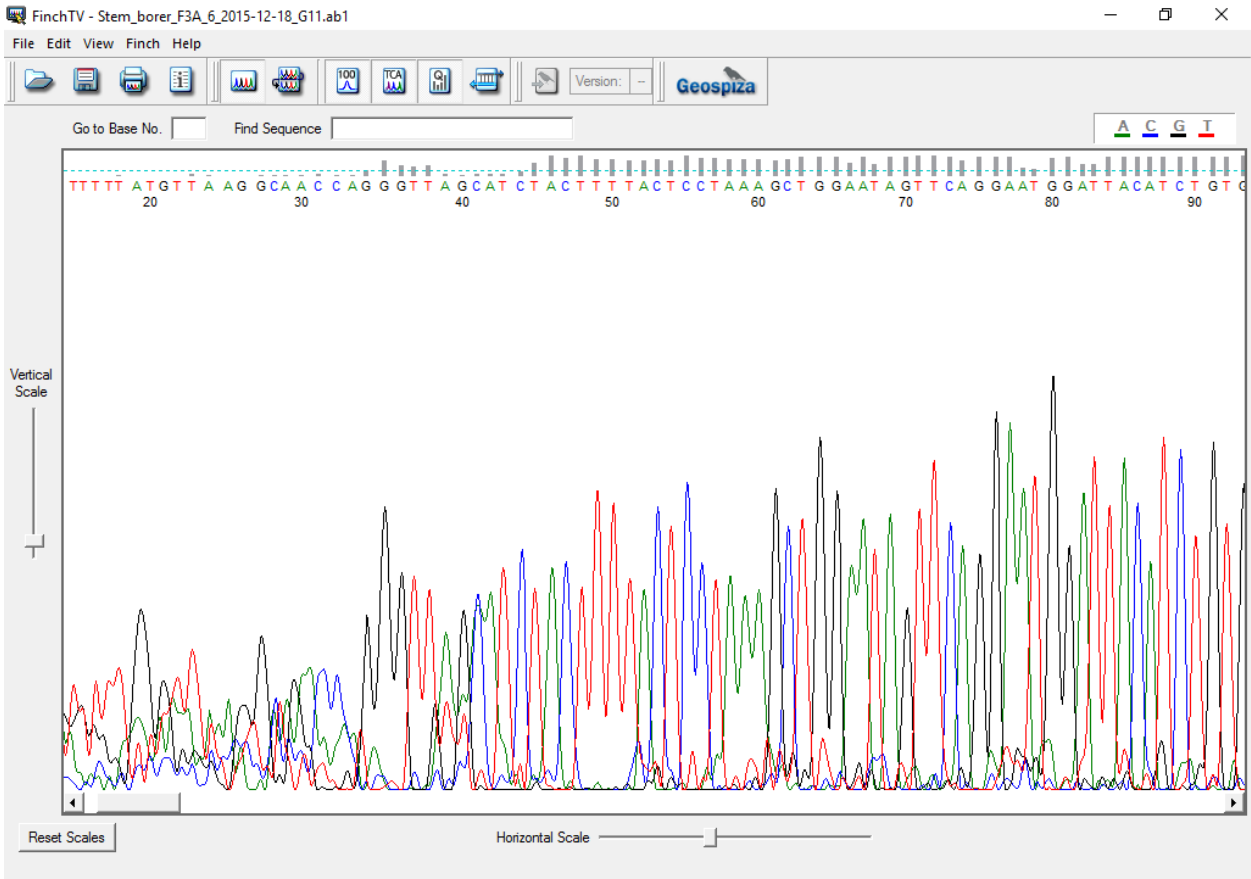
Sequences producing significant alignments:

Select: [All](#) [None](#) Selected:0

[Alignments](#) [Download](#) [GenBank](#) [Graphics](#) [Distance tree of results](#)

	Description	Max score	Total score	Query cover	E value	Ident	Accession
<input type="checkbox"/>	<a href="#">Eldana saccharina from Kenya cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	369	369	79%	3e-98	95%	<a href="#">AY320502.1</a>
<input type="checkbox"/>	<a href="#">Eldana saccharina from Zimbabwe cytochrome oxidase subunit II (COII) gene, partial cds; mitochond</a>	360	360	79%	2e-95	94%	<a href="#">AY320504.1</a>
<input type="checkbox"/>	<a href="#">Eldana saccharina from South Africa cytochrome oxidase subunit II (COII) gene, partial cds; mitochor</a>	356	356	79%	2e-94	94%	<a href="#">AY320503.1</a>
<input type="checkbox"/>	<a href="#">Noctuidae sp. VB-2010 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	295	295	81%	6e-76	88%	<a href="#">HQ677797.1</a>
<input type="checkbox"/>	<a href="#">Feltia jaculifera voucher FSb152 cytochrome oxidase subunit I (COI) gene, partial cds; tRNA-Leu qer</a>	295	295	81%	6e-76	88%	<a href="#">DQ792591.1</a>
<input type="checkbox"/>	<a href="#">Feltia jaculifera cytochrome oxidase subunit 1 (COI) and subunit 2 (COII) genes, complete cds, and t</a>	295	295	81%	6e-76	88%	<a href="#">U60990.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa punctigera mitochondrion, complete genome</a>	286	286	81%	3e-73	87%	<a href="#">KF977797.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa sp. Bahia-02 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	286	286	81%	3e-73	87%	<a href="#">KF625005.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa sp. Piaui-10 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	286	286	81%	3e-73	87%	<a href="#">KF624974.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa sp. Piaui-02 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	286	286	81%	3e-73	87%	<a href="#">KF624966.1</a>
<input type="checkbox"/>	<a href="#">Corcyra cephalonica mitochondrion, complete genome</a>	286	286	81%	3e-73	87%	<a href="#">HQ897685.1</a>
<input type="checkbox"/>	<a href="#">Parnassius tenedius isolate FS-b-1784 cytochrome oxidase subunit I (COI) gene, partial sequence; t</a>	286	286	74%	3e-73	89%	<a href="#">DQ351027.1</a>
<input type="checkbox"/>	<a href="#">Spodoptera litura mitochondrion, complete genome</a>	284	284	79%	1e-72	87%	<a href="#">KF701043.1</a>
<input type="checkbox"/>	<a href="#">Spodoptera litura mitochondrion, complete genome</a>	284	284	79%	1e-72	87%	<a href="#">JQ647918.1</a>
<input type="checkbox"/>	<a href="#">Spilosoma virginica cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	282	282	75%	4e-72	89%	<a href="#">HQ677812.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa sp. Bahia-07 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	280	280	81%	1e-71	86%	<a href="#">KF625010.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa sp. Piaui-21 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	280	280	81%	1e-71	86%	<a href="#">KF624985.1</a>

Stem\_borer\_F3A\_6\_2015-12-18\_G11



Stem\_borer\_F3A\_6\_2015-12-18\_G11 (280 nucleotides)

TAAAAAGGTAATTTTTTTATGTTAAGGCAACCAGGGTTAGCATCTACTTTTACTCCTAAAGCTGGAATAGTTCAGGAATGGATTACATCTGTGGCTGTAACATAAA  
 ATTCGAATTTGATTATTTATTGGTAAAATAATTCGATTATCTACATCTAATAGGCGAAAATTATTAGAAGAAAGTTCATTTCTAGAGATTATGTAGGAATCAAATT  
 CAATATTAATAAATCTGAATATTCATATCTTCAATATCATTCTGGTTCAATATGAACTGGTGTTAAA

Stem\_borer\_F3A\_6\_2015-12-18\_G11

NCBI Blast:Stem\_borer\_I X +

blast.ncbi.nlm.nih.gov/Blast.cgi

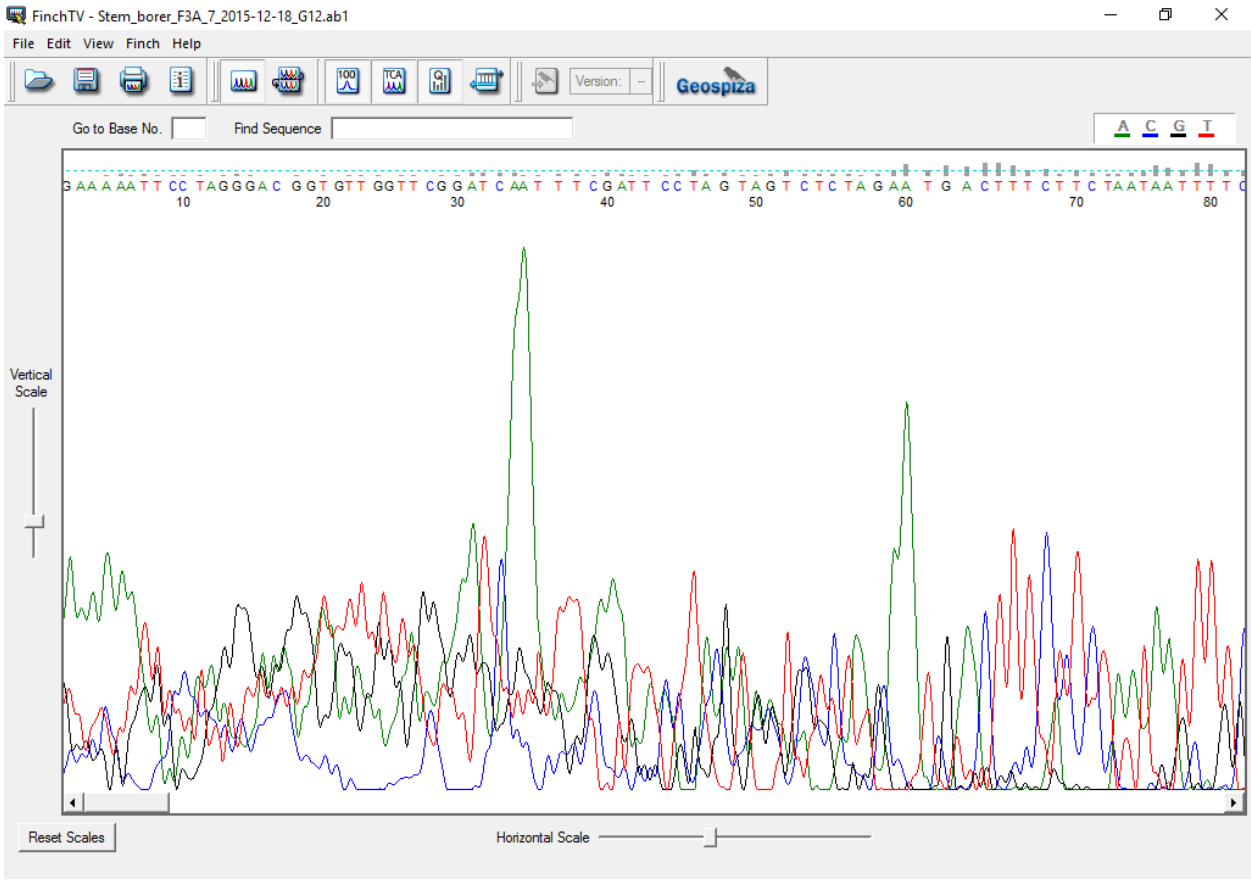
Sequences producing significant alignments:

Select: [All](#) [None](#) Selected:0

[Alignments](#) [Download](#) [GenBank](#) [Graphics](#) [Distance tree of results](#)

	Description	Max score	Total score	Query cover	E value	Ident	Accession
<input type="checkbox"/>	<a href="#">Eldana saccharina from Kenya cytochrome oxidase subunit II (COII) gene, partial cds; mitochondria</a>	374	374	75%	7e-100	99%	<a href="#">AY320502.1</a>
<input type="checkbox"/>	<a href="#">Eldana saccharina from Zimbabwe cytochrome oxidase subunit II (COII) gene, partial cds; mitochondria</a>	365	365	75%	4e-97	99%	<a href="#">AY320504.1</a>
<input type="checkbox"/>	<a href="#">Eldana saccharina from South Africa cytochrome oxidase subunit II (COII) gene, partial cds; mitochondria</a>	352	352	75%	2e-93	97%	<a href="#">AY320503.1</a>
<input type="checkbox"/>	<a href="#">Corcyra cephalonica mitochondrion, complete genome</a>	304	304	94%	1e-78	86%	<a href="#">HQ897685.1</a>
<input type="checkbox"/>	<a href="#">Hippotion celerio mitochondrial coxI (partial), tRNA-Leu and coxII (partial) genes, isolate 695934</a>	298	298	90%	4e-77	87%	<a href="#">AJ749424.1</a>
<input type="checkbox"/>	<a href="#">Hippotion celerio mitochondrial coxI (partial), tRNA-Leu and coxII (partial) genes, isolate 16132</a>	298	298	90%	4e-77	87%	<a href="#">AJ749422.1</a>
<input type="checkbox"/>	<a href="#">Meroptera pravella voucher JBWM0363025 mitochondrion, complete genome</a>	297	297	80%	2e-76	89%	<a href="#">MF073207.1</a>
<input type="checkbox"/>	<a href="#">Noctuidae sp. VB-2010 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	297	297	83%	2e-76	88%	<a href="#">HQ677797.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria auranticella haplotype OS1 cytochrome oxidase subunit I (COI) gene, partial cds; tRNA-Leu</a>	293	293	83%	2e-75	88%	<a href="#">DQ295176.1</a>
<input type="checkbox"/>	<a href="#">Aqonopterix pulvipennella cytochrome oxidase subunit II gene, partial cds; mitochondrial</a>	293	293	79%	2e-75	90%	<a href="#">AY527040.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria auranticella voucher Du02 cytochrome c oxidase subunit I (COI) gene, partial cds; tRNA-Leu</a>	293	293	83%	2e-75	88%	<a href="#">DQ247736.1</a>
<input type="checkbox"/>	<a href="#">Dichorda iridaria voucher MC02C301 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondria</a>	291	291	94%	6e-75	85%	<a href="#">EU151649.1</a>
<input type="checkbox"/>	<a href="#">Chlorochlamys chloroleucaria voucher MC02C205 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondria</a>	291	291	94%	6e-75	85%	<a href="#">EU151648.1</a>
<input type="checkbox"/>	<a href="#">Feltia jaculifera cytochrome oxidase subunit 1 (COI) and subunit 2 (COII) genes, complete cds, and</a>	291	291	83%	6e-75	88%	<a href="#">U60990.1</a>
<input type="checkbox"/>	<a href="#">Hyles livornicoides mitochondrial coxI gene (partial), tRNA-Leu and coxII gene (partial), isolate 3466</a>	289	289	79%	2e-74	89%	<a href="#">FN386575.1</a>
<input type="checkbox"/>	<a href="#">Nemoria leptalea voucher MDLEP1139 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondria</a>	289	289	79%	2e-74	89%	<a href="#">EU151620.1</a>
<input type="checkbox"/>	<a href="#">Hyles livornicoides mitochondrial coxI (partial), tRNA-Leu and coxII (partial) genes, isolate 23297</a>	289	289	79%	2e-74	89%	<a href="#">AJ749442.1</a>

Stem\_borer\_F3A\_7\_2015-12-18\_G12



Stem\_borer\_F3A\_7\_2015-12-18\_G12 (296 nucleotides)

GA AAAAATTCCTAGGGACGGTGTGGTTCGGATCAATTTTCGATTCTAGTAGTCTCTAGAATGACTTTCTTCTAATAATTTTCGCCTATTAGATGTAGATAATCGA  
ATTATTTTACCAATAAATAATCAAATTCGAATTTTAGTTACAGCCACAGATGTAATCCATTCTGAACTATTCCAGCTTTAGGAGTAAAAGTAGATGCTAACCCCTG  
GTCGCCTTAACCAAACTAATTTTTTTTATTAATCGCCCTGGAATTTTTTATGGTCAATGTTTCAGAAATTTGTGGAGCTAATCAATA

Stem\_borer\_F3A\_7\_2015-12-18\_G12

NCBI Blast:Stem\_borer\_F3A\_ NCBI Blast:Stem\_borer\_I X

blast.ncbi.nlm.nih.gov/Blast.cgi

Sequences producing significant alignments:

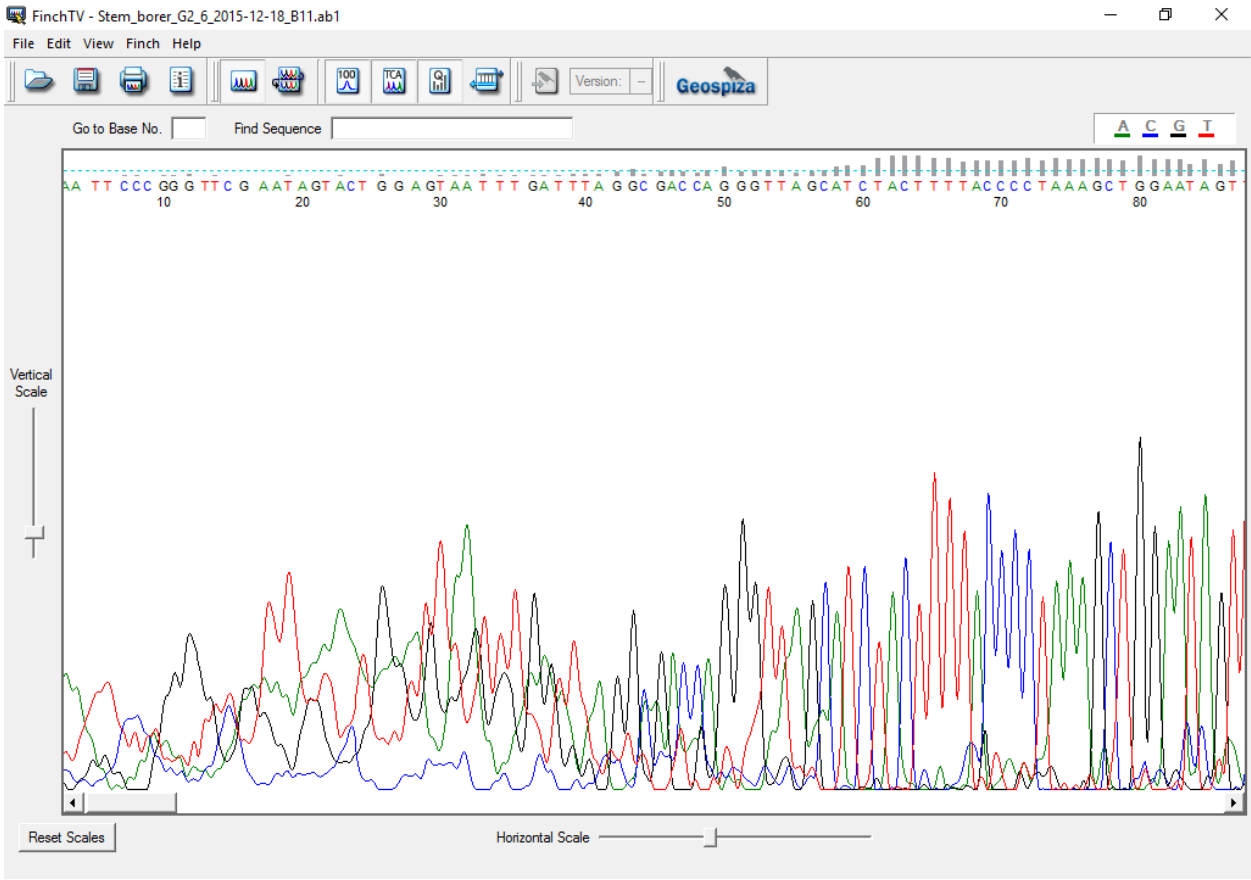
Select: All None Selected:0

Alignments Download GenBank Graphics Distance tree of results

	Description	Max score	Total score	Query cover	E value	Ident	Accession
<input type="checkbox"/>	<a href="#">Eldana saccharina from Kenya cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	416	416	87%	2e-112	96%	<a href="#">AY320502.1</a>
<input type="checkbox"/>	<a href="#">Eldana saccharina from Zimbabwe cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	407	407	87%	1e-109	95%	<a href="#">AY320504.1</a>
<input type="checkbox"/>	<a href="#">Eldana saccharina from South Africa cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	401	401	87%	5e-108	95%	<a href="#">AY320503.1</a>
<input type="checkbox"/>	<a href="#">Spodoptera litura mitochondrion, complete genome</a>	320	320	75%	1e-83	92%	<a href="#">KF701043.1</a>
<input type="checkbox"/>	<a href="#">Spodoptera litura mitochondrion, complete genome</a>	320	320	75%	1e-83	92%	<a href="#">JQ647918.1</a>
<input type="checkbox"/>	<a href="#">Noctuidae sp. VB-2010 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	318	318	79%	5e-83	90%	<a href="#">HQ677797.1</a>
<input type="checkbox"/>	<a href="#">Feltia jaculifera voucher FSb152 cytochrome oxidase subunit I (COI) gene, partial cds; tRNA-Leu gene</a>	318	318	79%	5e-83	90%	<a href="#">DQ792591.1</a>
<input type="checkbox"/>	<a href="#">Feltia jaculifera cytochrome oxidase subunit 1 (COI) and subunit 2 (COII) genes, complete cds, and</a>	318	318	79%	5e-83	90%	<a href="#">U60990.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa sp. Bahia-02 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	316	316	76%	2e-82	91%	<a href="#">KF625005.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa sp. Piaui-10 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	316	316	76%	2e-82	91%	<a href="#">KF624974.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa sp. Piaui-02 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	316	316	76%	2e-82	91%	<a href="#">KF624966.1</a>
<input type="checkbox"/>	<a href="#">Spilosoma virginica cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	316	316	75%	2e-82	91%	<a href="#">HQ677812.1</a>
<input type="checkbox"/>	<a href="#">Papilio protenor euprotenor isolate AB362 cytochrome oxidase subunit I gene, partial cds; tRNA-Leu</a>	315	315	75%	6e-82	91%	<a href="#">KX557579.1</a>
<input type="checkbox"/>	<a href="#">Papilio protenor mitochondrion, complete genome</a>	315	315	75%	6e-82	91%	<a href="#">KY272622.1</a>
<input type="checkbox"/>	<a href="#">Spodoptera litura mitochondrion, complete genome</a>	315	315	75%	6e-82	91%	<a href="#">KF543065.1</a>
<input type="checkbox"/>	<a href="#">Papilio maackii mitochondrion, complete genome</a>	315	315	75%	6e-82	91%	<a href="#">KC433408.1</a>
<input type="checkbox"/>	<a href="#">Papilio syfanius isolate Ps22 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	315	315	75%	6e-82	91%	<a href="#">JF281184.1</a>



Stem\_borer\_G2\_6\_2015-12-18\_B11



Stem\_borer\_G2\_6\_2015-12-18\_B11 (293 nucleotides)

GAAATTCCCGGGTTCGAATAGTACTGGAGTAATTTGATTTAGGCGACCAGGGTTAGCATCTACTTTTACCCCTAAAGCTGGAATAGTTCAGGAATGGATTACAT  
CTGTGGCTGTAATAAAATTCGAATTTGATTATTTATTGGTAAAATAATTCGATTATCTACATCTAATAGGCGAAAATTATTAGAAGAAAGTTCATTTCTAGAGAT  
TATGTAGGAATCAAATTCAATATTAATAAAAAATCTGAATATTCATATCTTCAATATCATTCTGGTTCAATATGAACTGGTGTTA

Stem\_borer\_G2\_6\_2015-12-18\_B11

NCBI Blast:Stem\_borer\_F3A\_ NCBI Blast:Stem\_borer\_F3A\_ NCBI Blast:Stem\_borer\_1 X +

blast.ncbi.nlm.nih.gov/Blast.cgi

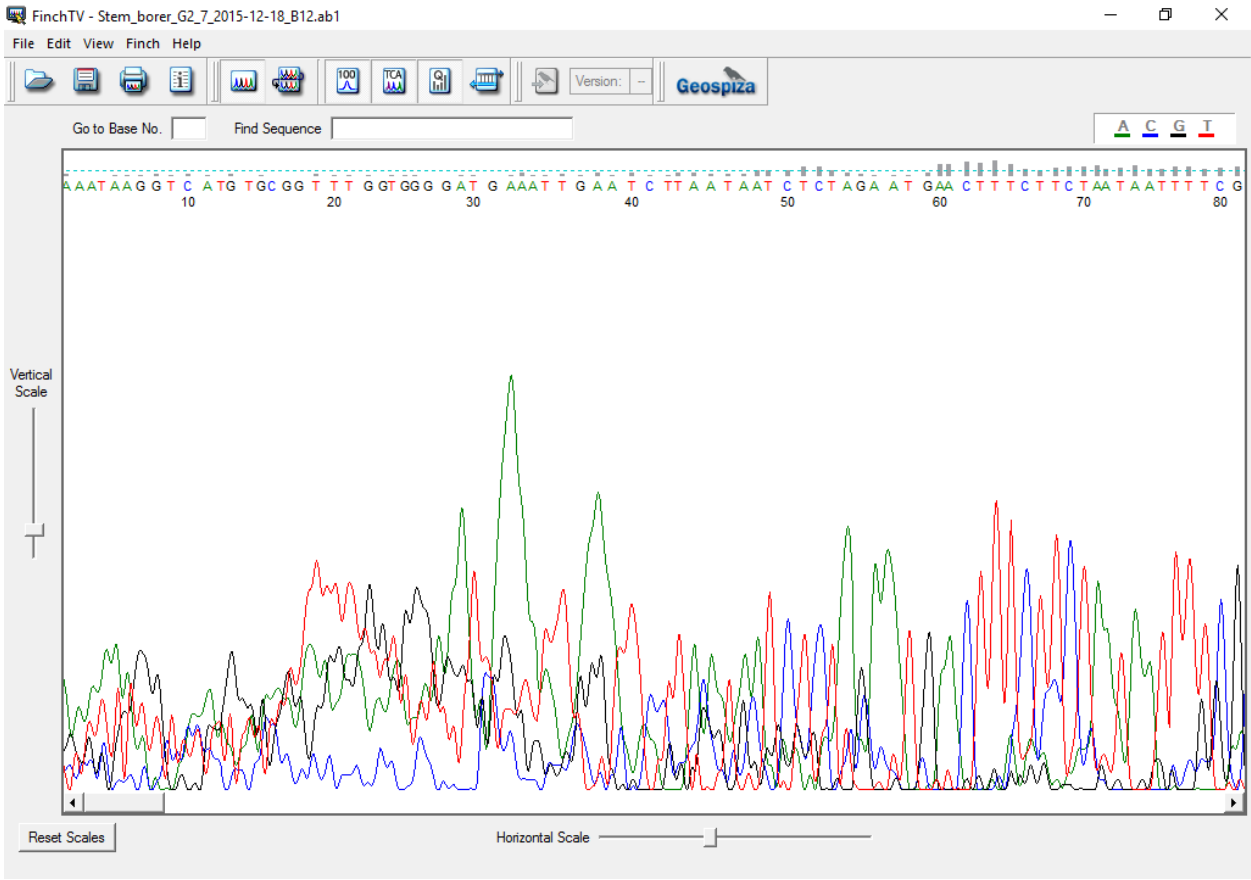
Sequences producing significant alignments:

Select: [All](#) [None](#) Selected:0

[Alignments](#) [Download](#) [GenBank](#) [Graphics](#) [Distance tree of results](#)

	Description	Max score	Total score	Query cover	E value	Ident	Accession
<input type="checkbox"/>	<a href="#">Eidana saccharina from Zimbabwe cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrion</a>	374	374	74%	7e-100	98%	<a href="#">AY320504.1</a>
<input type="checkbox"/>	<a href="#">Eidana saccharina from Kenya cytochrome oxidase subunit II (COII) gene, partial cds; mitochondria</a>	374	374	74%	7e-100	98%	<a href="#">AY320502.1</a>
<input type="checkbox"/>	<a href="#">Eidana saccharina from South Africa cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrion</a>	360	360	74%	2e-95	97%	<a href="#">AY320503.1</a>
<input type="checkbox"/>	<a href="#">Corcyra cephalonica mitochondrion, complete genome</a>	322	322	84%	4e-84	89%	<a href="#">HQ897685.1</a>
<input type="checkbox"/>	<a href="#">Meroptera pravella voucher JBWM0363025 mitochondrion, complete genome</a>	316	316	81%	2e-82	89%	<a href="#">MF073207.1</a>
<input type="checkbox"/>	<a href="#">Noctuidae sp. VB-2010 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	311	311	81%	7e-81	89%	<a href="#">HQ677797.1</a>
<input type="checkbox"/>	<a href="#">Chlorochlamys chloroleucaria voucher MC02C205 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrion</a>	307	307	84%	9e-80	88%	<a href="#">EU151648.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria auranticella haplotype OS1 cytochrome oxidase subunit I (COI) gene, partial cds; tRNA-L</a>	307	307	81%	9e-80	88%	<a href="#">DQ295176.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria auranticella voucher Du02 cytochrome c oxidase subunit I (COI) gene, partial cds; tRNA-L</a>	307	307	81%	9e-80	88%	<a href="#">DQ247736.1</a>
<input type="checkbox"/>	<a href="#">Dioryctria ponderosae voucher Du114 cytochrome c oxidase subunit I (COI) gene, partial cds; tRNA-L</a>	307	307	81%	9e-80	88%	<a href="#">DQ247733.1</a>
<input type="checkbox"/>	<a href="#">Nemoria festaria voucher EG03B829 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrion</a>	304	304	91%	1e-78	86%	<a href="#">EU151624.1</a>
<input type="checkbox"/>	<a href="#">Nemoria caerulea voucher EG03B845 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrion</a>	304	304	91%	1e-78	86%	<a href="#">EU151623.1</a>
<input type="checkbox"/>	<a href="#">Synchlora frondaria voucher MC02C279 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrion</a>	302	302	81%	4e-78	88%	<a href="#">EU151647.1</a>
<input type="checkbox"/>	<a href="#">Oncocera faecella voucher Du33 cytochrome c oxidase subunit I (COI) gene, partial cds; tRNA-Leu</a>	302	302	81%	4e-78	88%	<a href="#">DQ247727.1</a>
<input type="checkbox"/>	<a href="#">Hippotion celerio mitochondrial coxI (partial), tRNA-Leu and coxII (partial) genes, isolate 695934</a>	302	302	80%	4e-78	88%	<a href="#">AJ749424.1</a>
<input type="checkbox"/>	<a href="#">Hippotion celerio mitochondrial coxI (partial), tRNA-Leu and coxII (partial) genes, isolate 16132</a>	302	302	80%	4e-78	88%	<a href="#">AJ749422.1</a>
<input type="checkbox"/>	<a href="#">Agonopterix pulvipennella cytochrome oxidase subunit II gene, partial cds; mitochondrial</a>	300	300	80%	1e-77	89%	<a href="#">AY527040.1</a>

Stem\_borer\_G2\_7\_2015-12-18\_B12



Stem\_borer\_G2\_7\_2015-12-18\_B12 (294 nucleotides)

AAATAAGGTCATGTGCGGTTTGGTGGGGATGAAATTGAATCTTAATAATCTCTAGAATGAACTTTCTTCTAATAATTTTCGCCTATTAGATGTAGATAATCGAAT  
TATTTTACCAATAAATAATCAAATTCGAATTTTAGTTACAGCCACAGATGTAATCCATTCTGAACTATTCCAGCTTTAGGGGTAAAAGTAGATGCTAACCCCTGGT  
CGCCTTAACCAACTAATTTTTTTATTAATCGCCCTGGAATTTTTTATGGTCAATGTTGAGAAATTTGTGGAGCTAATCAAAA

Stem\_borer\_G2\_7\_2015-12-18\_B12

NCBI Blast:Stem\_borer\_i X +

blast.ncbi.nlm.nih.gov/Blast.cgi

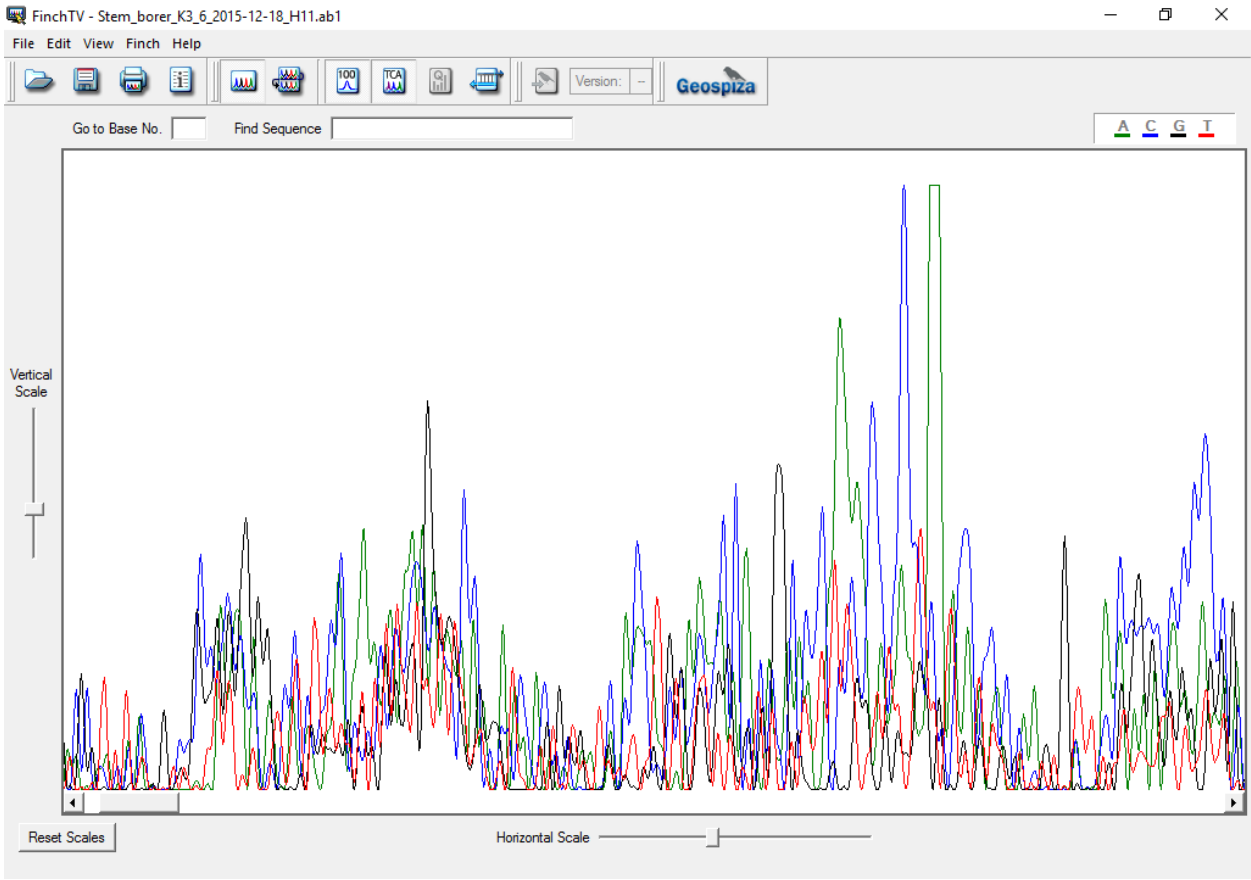
Sequences producing significant alignments:

Select: [All](#) [None](#) Selected:0

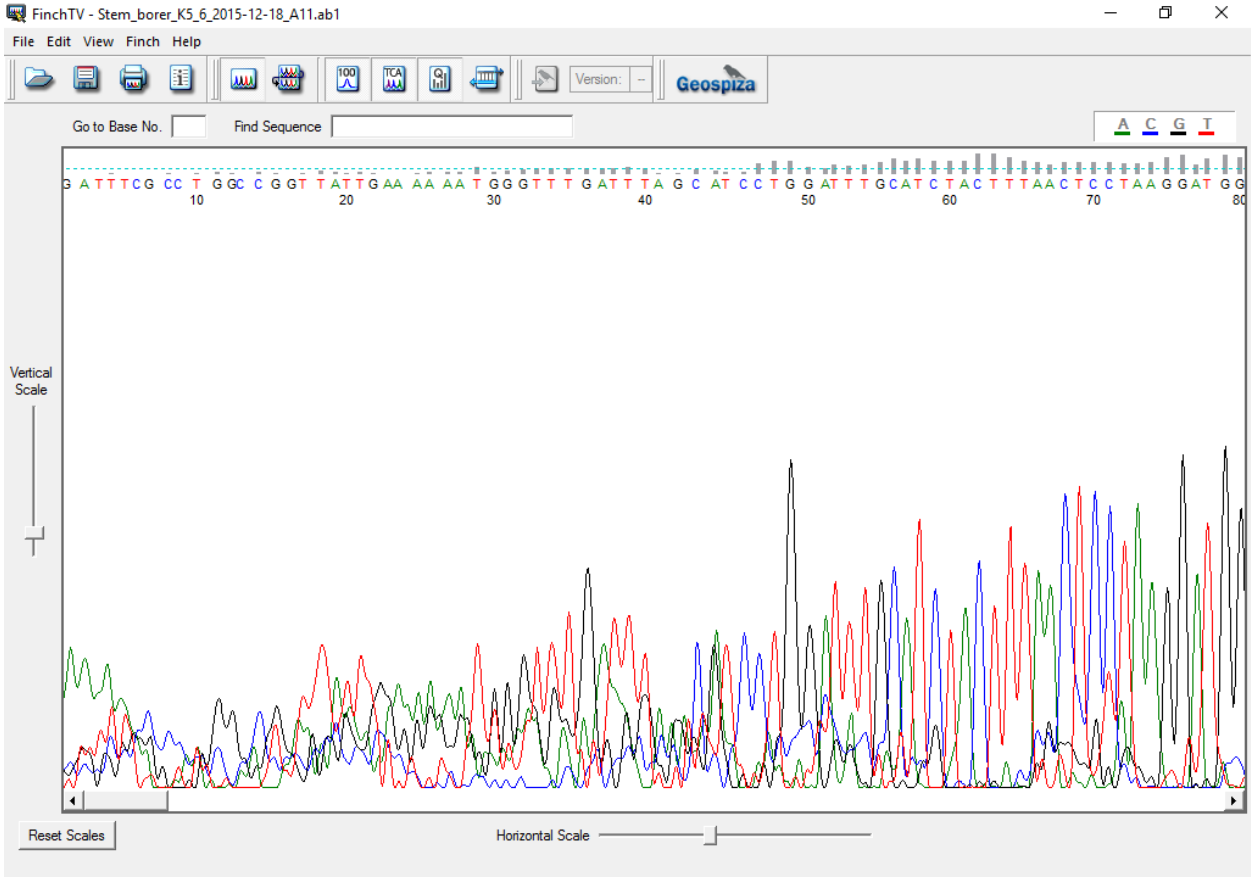
[Alignments](#) [Download](#) [GenBank](#) [Graphics](#) [Distance tree of results](#)

	Description	Max score	Total score	Query cover	E value	Ident	Accession
<input type="checkbox"/>	<a href="#">Eldana saccharina from Zimbabwe cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrion</a>	425	425	89%	5e-115	96%	<a href="#">AY320504.1</a>
<input type="checkbox"/>	<a href="#">Eldana saccharina from Kenya cytochrome oxidase subunit II (COII) gene, partial cds; mitochondria</a>	425	425	89%	5e-115	96%	<a href="#">AY320502.1</a>
<input type="checkbox"/>	<a href="#">Eldana saccharina from South Africa cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrion</a>	419	419	89%	2e-113	96%	<a href="#">AY320503.1</a>
<input type="checkbox"/>	<a href="#">Corcyra cephalonica mitochondrion, complete genome</a>	336	336	88%	2e-88	89%	<a href="#">HQ897685.1</a>
<input type="checkbox"/>	<a href="#">Feltia jaculifera cytochrome oxidase subunit 1 (COI) and subunit 2 (COII) genes, complete cds, and</a>	334	334	89%	6e-88	89%	<a href="#">U60990.1</a>
<input type="checkbox"/>	<a href="#">Nyctemera arctata albofasciata mitochondrion, complete genome</a>	329	329	89%	3e-86	88%	<a href="#">KM244681.1</a>
<input type="checkbox"/>	<a href="#">Feltia jaculifera voucher FSb152 cytochrome oxidase subunit I (COI) gene, partial cds; tRNA-Leu gene</a>	329	329	89%	3e-86	88%	<a href="#">DQ792591.1</a>
<input type="checkbox"/>	<a href="#">Vamuna virilis mitochondrion, complete genome</a>	325	325	89%	3e-85	88%	<a href="#">KJ364659.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa punctigera mitochondrion, complete genome</a>	325	325	89%	3e-85	88%	<a href="#">KF977797.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa sp. Bahia-02 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	325	325	89%	3e-85	88%	<a href="#">KF625005.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa sp. Piaui-10 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	325	325	89%	3e-85	88%	<a href="#">KF624974.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa sp. Piaui-02 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	325	325	89%	3e-85	88%	<a href="#">KF624966.1</a>
<input type="checkbox"/>	<a href="#">Spodoptera litura mitochondrion, complete genome</a>	325	325	89%	3e-85	88%	<a href="#">KF701043.1</a>
<input type="checkbox"/>	<a href="#">Spodoptera litura mitochondrion, complete genome</a>	325	325	89%	3e-85	88%	<a href="#">JQ647918.1</a>
<input type="checkbox"/>	<a href="#">Spilosoma virginica cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	325	325	89%	3e-85	88%	<a href="#">HQ677812.1</a>
<input type="checkbox"/>	<a href="#">Noctuidae sp. VB-2010 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	325	325	89%	3e-85	88%	<a href="#">HQ677797.1</a>
<input type="checkbox"/>	<a href="#">Helicoverpa zea haplotype 6 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	325	325	89%	3e-85	88%	<a href="#">HQ677776.1</a>

Stem\_borer\_K3\_6\_2015-12-18\_H11



Stem\_borer\_K5\_6\_2015-12-18\_A11



Stem\_borer\_K5\_6\_2015-12-18\_A11 (294 nucleotides)

GATTTGCCTGGCCGGTTATTGAAAAAATGGGTTTGATTAGCATCCTGGATTGTCATCTACTTTAACTCCTAAGGATGGAATAGTTCAAGAGTGAATAACATCTGTAGCAGTA  
ACTAAAATTCGAATTTGATTATTTAAAGGTAAAATAATTCGATTATCAACATCTAAAAGACGAAAATTATTGGATGATATTCATTGGTGGGGATTATATAGGAGTCAAATTCA  
ATTTTATTGAAATCTGAATATTCATAACTTCAATATCATTCTGGTTCAATATGAACTGGTGTTAAA

## Stem\_borer\_K5\_6\_2015-12-18\_A11

NCBI Blast:Stem\_borer\_I X +

blast.ncbi.nlm.nih.gov/Blast.cgi

Sequences producing significant alignments:

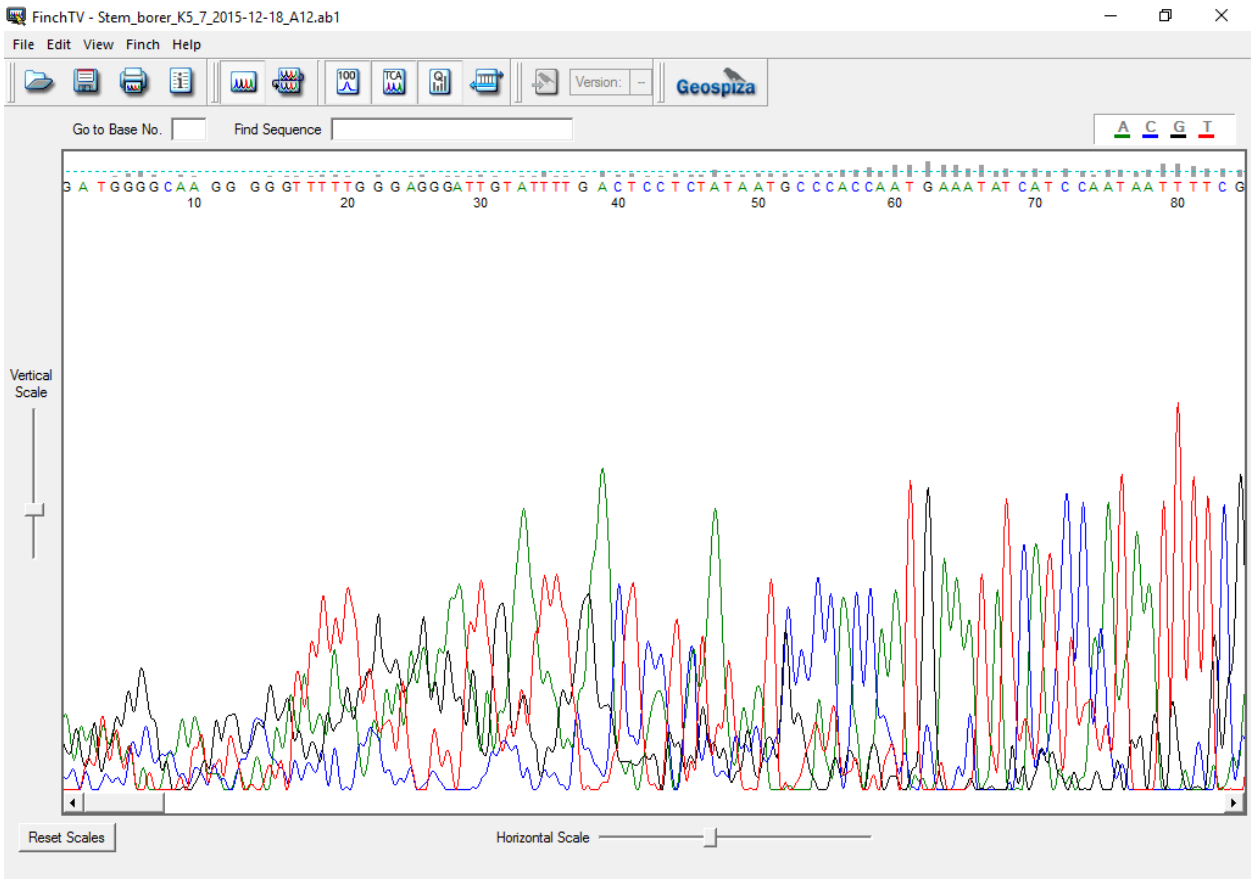
Select: [All](#) [None](#) Selected:0

[Alignments](#) [Download](#) [GenBank](#) [Graphics](#) [Distance tree of results](#)

	Description	Max score	Total score	Query cover	E value	Ident	Accession
<input type="checkbox"/>	<a href="#">Sesamia calamistis from Kenya cytochrome oxidase subunit II (COII) gene, partial cds; mitochondri</a>	374	374	80%	7e-100	95%	<a href="#">AY320510.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens mitochondrion, complete genome</a>	340	386	97%	1e-89	89%	<a href="#">JN039362.1</a>
<input type="checkbox"/>	<a href="#">Sesamia nonagrioides mitochondrial coi gene (partial), tRNA-Leu gene, and coi gene (partial), Ebr</a>	338	338	88%	5e-89	89%	<a href="#">AJ829718.1</a>
<input type="checkbox"/>	<a href="#">Sesamia nonagrioides mitochondrial coi gene (partial), tRNA-Leu gene, and coi gene (partial), The</a>	338	338	88%	5e-89	89%	<a href="#">AJ829717.1</a>
<input type="checkbox"/>	<a href="#">Sesamia nonagrioides mitochondrial coi gene (partial), tRNA-Leu gene, and coi gene (partial), Cop</a>	338	338	88%	5e-89	89%	<a href="#">AJ829716.1</a>
<input type="checkbox"/>	<a href="#">Sesamia nonagrioides mitochondrial coi gene (partial), tRNA-Leu gene, and coi gene (partial), Kato</a>	338	338	88%	5e-89	89%	<a href="#">AJ829715.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-Hyd(R) cytochrome oxidase subunit II (COII) gene, partial cds; mitochor</a>	336	336	89%	2e-88	89%	<a href="#">KT277733.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-blr(M) cytochrome oxidase subunit II (COII) gene, partial cds; mitochon</a>	336	336	89%	2e-88	89%	<a href="#">KT277732.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-Ko(M) cytochrome oxidase subunit II (COII) gene, partial cds; mitochon</a>	336	336	89%	2e-88	89%	<a href="#">KT277730.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-Al(M) cytochrome oxidase subunit II (COII) gene, partial cds; mitochon</a>	336	336	89%	2e-88	89%	<a href="#">KT277729.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-Gu(S) cytochrome oxidase subunit II (COII) gene, partial cds; mitochon</a>	336	336	89%	2e-88	89%	<a href="#">KT277728.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-Rr(M) cytochrome oxidase subunit II (COII) gene, partial cds; mitochon</a>	336	336	89%	2e-88	89%	<a href="#">KT277724.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-Pi(R) cytochrome oxidase subunit II (COII) gene, partial cds; mitochon</a>	336	336	89%	2e-88	89%	<a href="#">KT277722.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-Ar(M) cytochrome oxidase subunit II (COII) gene, partial cds; mitochon</a>	336	336	89%	2e-88	89%	<a href="#">KT277721.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-Km(R) cytochrome oxidase subunit II (COII) gene, partial cds; mitochon</a>	336	336	89%	2e-88	89%	<a href="#">KT277720.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-KU(Su) cytochrome oxidase subunit II (COII) gene, partial cds; mitochon</a>	336	336	89%	2e-88	89%	<a href="#">KT277719.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-PB(M) cytochrome oxidase subunit II (COII) gene, partial cds; mitochon</a>	336	336	89%	2e-88	89%	<a href="#">KT277716.1</a>



Stem\_borer\_K5\_7\_2015-12-18\_A12



Stem\_borer\_K5\_7\_2015-12-18\_A12 (294 nucleotides)

GATGGGGCAAGGGGGTTTTGGGAGGGATTGTATTTGACTCCTCTATAATGCCACCAATGAAATATCATCCAATAATTTTCGTCTTTTAGATGTTGATAATCGAATTATTTA  
CCTTTAAATAATCAAATTCGAATTTTAGTTACTGCTACAGATGTTATTCACCTTGAAGTATTCCATCCTTAGGAGTTAAAGTAGATGCAAATCCAGGACGTTTAAATCAAATAA  
TTTTTCATTAATCGTCCTGGTATTTTTATGGTCAATGTTGAGAAATTTGTGGAGCTAATCA

Stem\_borer\_K5\_7\_2015-12-18\_A12

NCBI Blast:Stem\_borer\_I X +

blast.ncbi.nlm.nih.gov/Blast.cgi

Sequences producing significant alignments:

Select: [All](#) [None](#) Selected:0

[Alignments](#) [Download](#) [GenBank](#) [Graphics](#) [Distance tree of results](#)

	Description	Max score	Total score	Query cover	E value	Ident	Accession
<input type="checkbox"/>	<a href="#">Sesamia calamistis from Kenya cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	428	428	88%	4e-116	97%	<a href="#">AY320510.1</a>
<input type="checkbox"/>	<a href="#">Sesamia nonagrioides botanephaqa from Iran cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	365	365	88%	4e-97	91%	<a href="#">AY320514.1</a>
<input type="checkbox"/>	<a href="#">Sesamia nonagrioides mitochondrial coi gene (partial), tRNA-Leu gene, and coi gene (partial), Kato</a>	365	365	88%	4e-97	91%	<a href="#">AJ829715.1</a>
<input type="checkbox"/>	<a href="#">Sesamia nonagrioides mitochondrial coi gene (partial), tRNA-Leu gene, and coi gene (partial), Ebr</a>	361	361	88%	5e-96	91%	<a href="#">AJ829718.1</a>
<input type="checkbox"/>	<a href="#">Sesamia nonagrioides mitochondrial coi gene (partial), tRNA-Leu gene, and coi gene (partial), Cop</a>	361	361	88%	5e-96	91%	<a href="#">AJ829716.1</a>
<input type="checkbox"/>	<a href="#">Lepidoptera sp. VB-2010 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	356	356	88%	2e-94	91%	<a href="#">HQ677818.1</a>
<input type="checkbox"/>	<a href="#">Lepidoptera sp. VB-2010 haplotype 2 cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	356	356	88%	2e-94	91%	<a href="#">HQ677801.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens from Wuhu cytochrome oxidase subunit II gene, partial cds; mitochondrial</a>	356	356	88%	2e-94	90%	<a href="#">EU240536.1</a>
<input type="checkbox"/>	<a href="#">Sesamia nonagrioides mitochondrial coi gene (partial), tRNA-Leu gene, and coi gene (partial), The</a>	356	356	88%	2e-94	90%	<a href="#">AJ829717.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-Hyd(R) cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	352	352	88%	2e-93	90%	<a href="#">KT277733.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-blr(M) cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	352	352	88%	2e-93	90%	<a href="#">KT277732.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-Ko(M) cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	352	352	88%	2e-93	90%	<a href="#">KT277730.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-Gu(S) cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	352	352	88%	2e-93	90%	<a href="#">KT277728.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-Rr(M) cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	352	352	88%	2e-93	90%	<a href="#">KT277724.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-Pi(R) cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	352	352	88%	2e-93	90%	<a href="#">KT277722.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-Ar(M) cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	352	352	88%	2e-93	90%	<a href="#">KT277721.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-Km(R) cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	352	352	88%	2e-93	90%	<a href="#">KT277720.1</a>
<input type="checkbox"/>	<a href="#">Sesamia inferens isolate SI-KU(Su) cytochrome oxidase subunit II (COII) gene, partial cds; mitochondrial</a>	352	352	88%	2e-93	90%	<a href="#">KT277719.1</a>

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2017-12-14

[illegible]

# T92+G

From\To	A	T	C	G
A	-	0.094337072	0.039269887	0.068420712
T	0.094337072	-	0.068420712	0.039269887
C	0.094337072	0.16436537	-	0.039269887
G	0.16436537	0.094337072	0.039269887	-

# T92+G+I

From\To	A	T	C	G
A	-	0.09433709	0.039269895	0.068420697
T	0.09433709	-	0.068420697	0.039269895
C	0.09433709	0.164365335	-	0.039269895
G	0.164365335	0.09433709	0.039269895	-

# HKY+G

From\To	A	T	C	G
A	-	0.101306279	0.038558079	0.069667968
T	0.087263021	-	0.067260741	0.039938052
C	0.087263021	0.176718748	-	0.039938052
G	0.152221678	0.101306279	0.038558079	-

# TN93 +G

From\To	A	T	C	G
A	-	0.10076151	0.038350735	0.085414878
T	0.086793769	-	0.054226221	0.039723288
C	0.086793769	0.142472262	-	0.039723288
G	0.186628037	0.10076151	0.038350735	-

# HKY+G+I

From\To	A	T	C	G
A	-	0.101306285	0.038558081	0.069667964
T	0.087263026	-	0.067260737	0.039938055
C	0.087263026	0.176718737	-	0.039938055
G	0.152221669	0.101306285	0.038558081	-

#### GTR+G

From\To	A	T	C	G
A	-	0.147069998	0.009325461	0.090471494
T	0.126682891	-	0.05771323	0.041483907
C	0.021104991	0.151633918	-	0.026258708
G	0.197676537	0.10522747	0.025351395	-

#### T+92

From\To	A	T	C	G
A	-	0.097077182	0.040410519	0.066139449
T	0.097077182	-	0.066139449	0.040410519
C	0.097077182	0.15888515	-	0.040410519
G	0.15888515	0.097077182	0.040410519	-

#### TN93+G+I

From\To	A	T	C	G
A	-	0.100752695	0.03834738	0.08544464
T	0.086786175	-	0.054212902	0.039719812
C	0.086786175	0.142437268	-	0.039719812
G	0.186693066	0.100752695	0.03834738	-

#### HKY+I

From\To	A	T	C	G
A	-	0.10310776	0.039243739	0.06824757
T	0.088814777	-	0.065889422	0.040648252
C	0.088814777	0.173115787	-	0.040648252
G	0.149118166	0.10310776	0.039243739	-

#### GTR+G+I

From\To	A	T	C	G
A	-	0.147061642	0.009321893	0.090484943
T	0.126675694	-	0.057710127	0.041482612
C	0.021096916	0.151625766	-	0.026258808
G	0.197705922	0.105224186	0.025351491	-

#### TN93+I

From\To	A	T	C	G
A	-	0.102388808	0.038970099	0.082610369
T	0.088195488	-	0.054323378	0.040364819
C	0.088195488	0.142727528	-	0.040364819
G	0.180500298	0.102388808	0.038970099	-

#### HKY

From\To	A	T	C	G
A	-	0.104317755	0.039704274	0.067293535
T	0.089857041	-	0.064968352	0.041125269
C	0.089857041	0.170695797	-	0.041125269
G	0.147033639	0.104317755	0.039704274	-

# TN93

From\To	A	T	C	G
A	-	0.104110573	0.039625418	0.07656178
T	0.089678578	-	0.057131644	0.041043591
C	0.089678578	0.150105877	-	0.041043591
G	0.167284377	0.104110573	0.039625418	-

# GTR

From\To	A	T	C	G
A	-	0.104110573	0.039625418	0.07656178
T	0.089678578	-	0.057131644	0.041043591
C	0.089678578	0.150105877	-	0.041043591
G	0.167284377	0.104110573	0.039625418	-

# JC+G

From\To	A	T	C	G
A	-	0.083333333	0.083333333	0.083333333
T	0.083333333	-	0.083333333	0.083333333
C	0.083333333	0.083333333	-	0.083333333
G	0.083333333	0.083333333	0.083333333	-

# K2+G

From\To	A	T	C	G
A	-	0.074902867	0.074902867	0.100194266
T	0.074902867	-	0.100194266	0.074902867
C	0.074902867	0.100194266	-	0.074902867
G	0.100194266	0.074902867	0.074902867	-



JC+G+I

From\To	A	T	C	G
A	-	0.083333333	0.083333333	0.083333333
T	0.083333333	-	0.083333333	0.083333333
C	0.083333333	0.083333333	-	0.083333333
G	0.083333333	0.083333333	0.083333333	-

K2+G+I

From\To	A	T	C	G
A	-	0.074902909	0.074902909	0.100194183
T	0.074902909	-	0.100194183	0.074902909
C	0.074902909	0.100194183	-	0.074902909
G	0.100194183	0.074902909	0.074902909	-

JC+I

From\To	A	T	C	G
A	-	0.083333333	0.083333333	0.083333333
T	0.083333333	-	0.083333333	0.083333333
C	0.083333333	0.083333333	-	0.083333333
G	0.083333333	0.083333333	0.083333333	-

JC

From\To	A	T	C	G
A	-	0.083333333	0.083333333	0.083333333
T	0.083333333	-	0.083333333	0.083333333
C	0.083333333	0.083333333	-	0.083333333
G	0.083333333	0.083333333	0.083333333	-

K2+1

From\To	A	T	C	G
A	-	0.075482822	0.075482822	0.099034356
T	0.075482822	-	0.099034356	0.075482822
C	0.075482822	0.099034356	-	0.075482822
G	0.099034356	0.075482822	0.075482822	-

K2

From\To	A	T	C	G
A	-	0.075535059	0.075535059	0.098929882
T	0.075535059	-	0.098929882	0.075535059
C	0.075535059	0.098929882	-	0.075535059
G	0.098929882	0.075535059	0.075535059	-

### ANALYSIS OF DATA COLLECTED FROM THE STEM BORER SURVEY

State	Altitude (m)	% Infestation	Stborer. Complex	Cropping system
Oyo	559.24± 51.55	9.40±2.18	4.40±0.24	1.20±0.20
Kwara	1213.20±67141	44.00±14.69	3.60±0.24	1.30±0.20
Ondo	677.58±96.25	42.00±11.14	4.00±0.55	1.30±0.20
Ogun	205.60±35.09	54.00±6.78	4.60±0.40	1.60±0.24
Osun	863.50±102.57	40.00±5.48	5.60±0.40	2.00±0.00
Ekiti	1602.40±60.49	36.00±6.78	5.80±0.20	1.60±0.24









