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Screening of New Cocoa Types for Insect Infestation and Biochemical Analysis of the Stored Beans

W.A. Jonfia-Essien
School of Biosciences, University of Nottingham, Sutton Bonington Campus,
Loughborough, Leicestershire, LE12 5RD, UK

Abstract: New cocoa types, mainly hybrids, are being developed and selected by the Cocoa Research Institute of Ghana (CRIG) on the basis of their early establishment and high yields. It is, however, necessary to screen them for their attractiveness to insect pests and diseases to ensure that the materials given to farmers are not prone to insect pests and diseases, both in the field and during storage of the beans. This study reports on investigation on the susceptibility/tolerance in five hybrid cocoa types to four storage insect pests (both beetles and moths). It also reports on the effects of insect pests' infestation on the levels of free fatty acids (FFA), the percentage shell, sugar and fat contents. For all the five cocoa types tested, the level of insect infestation was minimal (0-1) by four months after their introduction into the beans. The results also showed that, *Lasioderma serricorne* (Fabricus), *Tribolium castaneum* (Herbst), *Ephestia cautella* (Walker) and *Corcyra cephalonica* (Stainton) had no adverse effects on the beans within the four-month experimental period. The fat content remained high whilst the FFA levels remained within acceptable limits. There was virtually no reduction in the sugar and shell contents of the cocoa beans.

Key words: Cocoa beans, fat content, FFA, *Lasioderma*, *Tribolium*, *Ephestia*, *Corcyra*

INTRODUCTION

Genetically improved varieties of Cocoa (mixed Amazon and Series II hybrids) were developed and released to Ghanaian farmers (Adu-Ampomah, 1996). To further improve on yield, the alternative hybrids were developed to augment the Series II hybrids (Adu-Ampomah and Sersah, 1988).

The increasing trend of the production levels of the alternative hybrids over the last nine years has shown that, their yields are relatively higher than most of the traditional Amelonado and Local Trinitario cocoa. It is however not certain whether these alternative hybrids can withstand insect attack in storage better than the traditional cocoa. In Ghana, dry cocoa beans in storage were monitored for insect pests from 1995 to 2000 and *Lasioderma serricorne*, *Tribolium castaneum*, *Cadra cautella* (*Ephestia cautella*) and *Corcyra cephalonica* were among the eleven species identified as most important (Jonfia-Essien, 2004).

Cocoa butter is the most valuable component of the bean. It has more influence on its physical and chemical properties in chocolate than all the ingredients and it is responsible for the different favourable characteristics, such as hardness at room temperature, brightness and fast and complete melting when placed in the mouth (Saldaña *et al*., 2002). Insect infestation during storage, however, results in the breakdown of the butter and this causes increases in free fatty acid (FFA) levels in the beans (Anonymous, 1970). The level of FFA in the beans must be less than 1.0% to meet the acceptable level of 1.75% in cocoa butter extracted from the beans (Anonymous, 1996).

It is suspected that, recent increases in the level of insect pests infestations during storage is probably related to the development of new hybrid types and this must be investigated.

The objectives of the present study are to:

- Assess the level of damage of stored cocoa beans of the five hybrid cocoa varieties caused by the four most important storage pests: (a) beetles: *Lasioderma serricorne* (Fabricus) and *Tribolium castaneum* (Herbst); (b) moths: *Ephestia cautella* (Walker) and *Corcyra cephalonica* (Stainton).
- Analyse the percentage shell, sugar and fat contents of each of the hybrid varieties of cocoa and relate them to the level of insect pest infestation.
- Assess the effect of the damage on free fatty acid levels in the stored cocoa beans.

MATERIALS AND METHODS

Dry cocoa beans of five new hybrid varieties (designated as HV1, HV2, HV3, HV4 and HV5) and traditional type (TCT) composed of mixed genotypes, were collected from the Cocoa Research Institute of Ghana (Table 1). The beans which were grown under

Table 1: Five cocoa types used in the study

Cocoa type	Variety	Genotype (♀ x ♂)	Yield (kg ha^{-1})
Amazon/Trinitario	HV_1	SC5 X K5	1,600
Inter Amazon Hybrids	HV_2	APA5 X PA7	1,250
(Amazon/Amazon)	HV_3	APA5 X K5	1,000
Amazon/Amelonado	HV_4	SPA10 X P30	1,500
Hybrids	HV_5	EET399 X P30	1,200
Traditional cocoa type	TCT	T85/799 X PA7	1,250
		T85/799 X T79/501	1,100
		T85/799 X Amelonado	1,350
		T 63/971 X Amelonado	-
		T60/887 X Amelonado	-

tropical conditions were harvested in November 2001 from the experimental farm of the Institute at Tafo, a town in the Eastern Region of Ghana. The harvested pods were broken to extract the beans, which are then fermented for a period of 6 days. After fermentation the beans were sun-dry on mats raised off the ground. The quality of the sample was improved by removing flat and broken beans after drying before the beans were bagged in jute sacks. The cocoa was then sent to the Research Department of the Quality Control Division at Tema, a town in the Greater Accra Region of Ghana in February 2002 to commence the experiment.

The dry cocoa beans were sieved to remove dirt and each variety was divided into four samples in triplicates of 500 g each and poured into proto-type jute sacks. Jute sacks were used so as to conform to the approved standard of storing cocoa beans. Cocoa has also been traditionally shipped in bags or sacks.

Laboratory reared storage insect pests: *Lasioderma serricorne* (Fabricus), *Tribolium castaneum* (Herbst), *Ephestia cautella* (Walker) and *Corcyra cephalonica* (Stainton) were used to investigate the level of insect infestation on the dry cocoa bean samples of the five selected hybrid varieties. The cocoa beans were stored for four months. The insect pests (10 each) were introduced into the dry cocoa beans and stored for four (4) months in a controlled environment at 30±2°C and relative humidity of 70±2%, based on the prevailing conditions at the cocoawarehouses in Ghana. After two months of storage, adult insect population in the new hybrid variety cocoa beans was increased to 10 using a fresh set from the main stock cultures.

Insect numbers were assessed once every month for the period of four months. The cocoa beans were examined for holes and any other physical damage. Percentage moisture and fat contents were determined using MM55 Plus equipment, while the free fatty acid was determined as a percentage by weight of the free fatty acid present, the molecular weight of which is assumed to be 282 corresponding to the oleic acid. An ADP 200 Polarimeter was used in the determination of percentage sugar (% sucrose) content.

Insect pests' infestation and the effect of storage on biochemical components of the dry cocoa beans were assessed by analysis of the beans before and after storage. Thus, paired data was generated from the experiments involving insect pests' infestation and biochemical components such as level of free fatty acids (FFA) percentage shell, sugar and fat contents. The data generated were subjected to analysis of variance using a randomised complete block design.

RESULTS

The results showed that insect numbers declined with time in the new hybrid variety cocoa beans during the experimental period (Fig. 1-4) unlike that of the

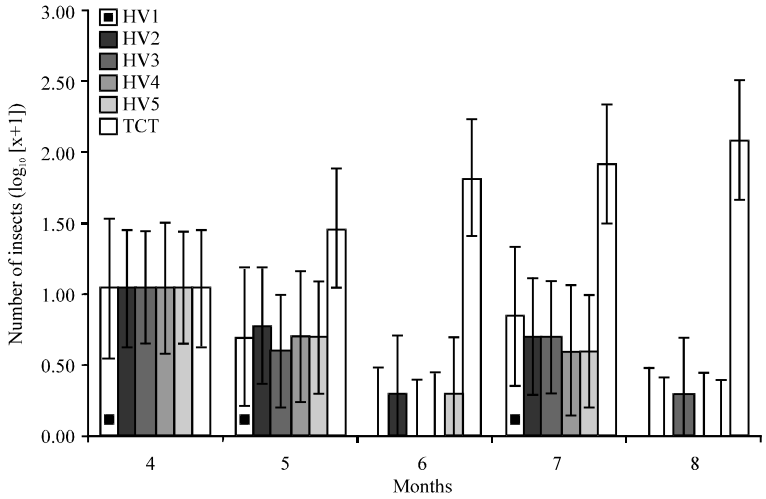

Fig. 1: Mean number of *Lasioderma serricorne* on different varieties of cocoa in storage. Fresh sets of insect pests were introduced into the new hybrid variety cocoa bean after the second month

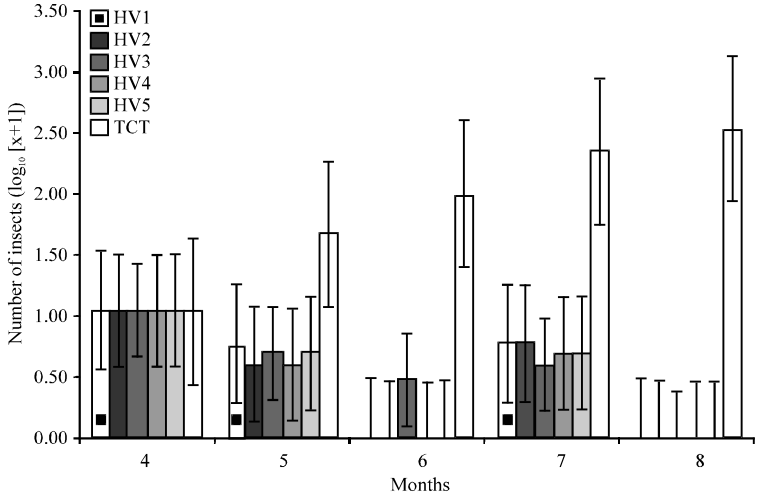

Fig. 2: Mean number of *Tribolium castaneum* on different varieties of cocoa in storage. Fresh sets of insect pests were introduced into the new hybrid variety cocoa bean after the second month

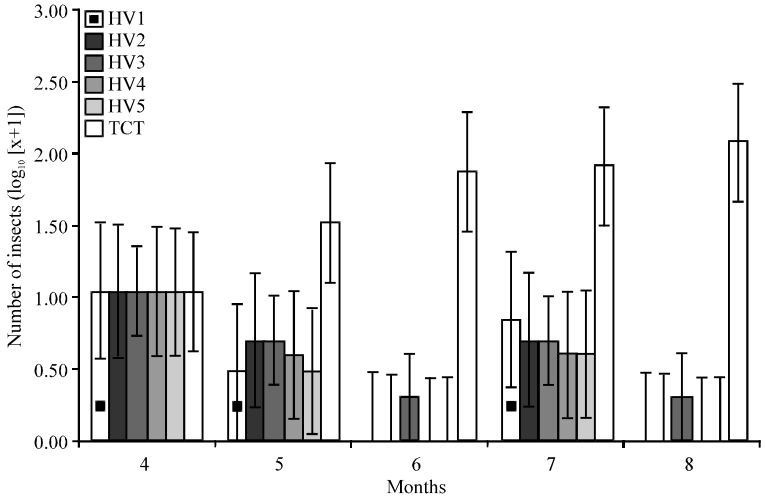

Fig. 3: Mean number of *Corcyra cephalonica* on different varieties of cocoa in storage. Fresh sets of insect pests were introduced into the new hybrid variety cocoa bean after the second month

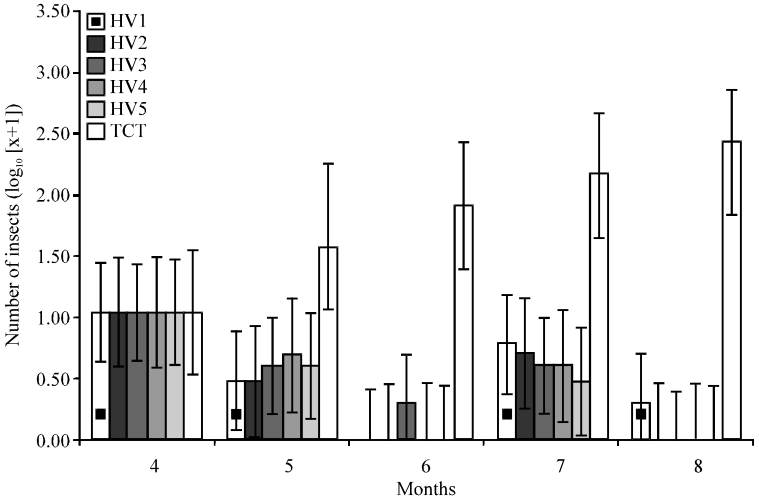

Fig. 4: Mean number of *Ephestia cautella* on different varieties of cocoa in storage. Fresh sets of insect pests were introduced into the new hybrid variety covoa bean after the second month

traditional type. By the end of the second month of storage (i.e., four months after harvest), almost all of the insects in the new hybrid variety cocoa beans were dead while that of the traditional type had increased. The insects introduced after the second month were also found dead by the end of the fourth month of storage (i.e. eight months after harvest). The percentage fat content was quite high (47.30-67.17%) in all the cocoa types over the period of four month (Fig. 5). On the other hand, percentage FFA was low (0.28-0.49%) in all the cocoa types but the levels increased slightly during the four-month storage period. In all cases, the highest percentage FFA (0.43-0.49%) was recorded after four months of storage (Fig. 6) except for HV3, which recorded the highest percentage FFA of (0.38%) after the third month of storage compared to 0.36% after four months.

There was no perceivable trend in the changes observed in the level of sugar content (Fig. 7). There was a general reduction in the shell content of all the hybrid varieties up to the second month, followed by increases in the third month, except for HV2, which continued to record a reduction in shell content (Fig. 8). In the forth month, the shell content of HV2 and HV4 increased but decreased that in HV1, HV3 and HV5. That of the traditional type decreased through the storage period.

During the four-month period, the moisture content declined every month in all the cocoa types (Fig. 9). The highest moisture content was 8.06% for HV4 recorded just before storage. The lowest moisture content was 5.17% recorded for HV1 and HV2 in the forth month of storage. The weight per bean was quiet high in all the new hybrid varieties (1.10-1.49 g) compared to the traditional type

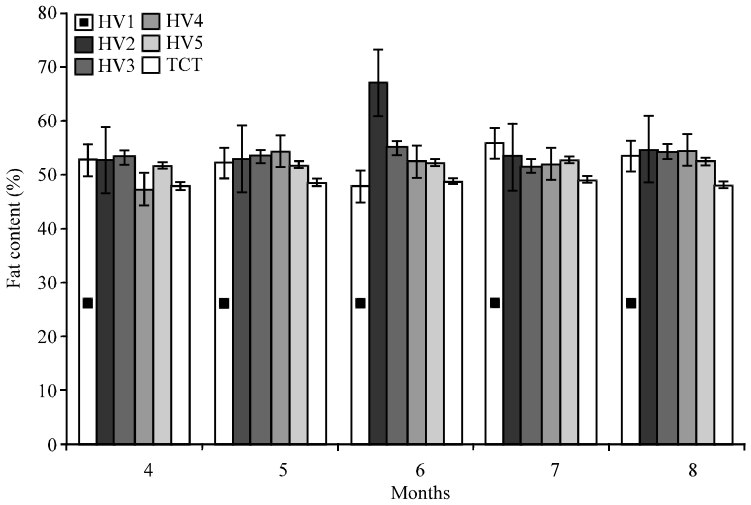

Fig. 5: Mean percentage fat content of different varieties of cocoa in storage

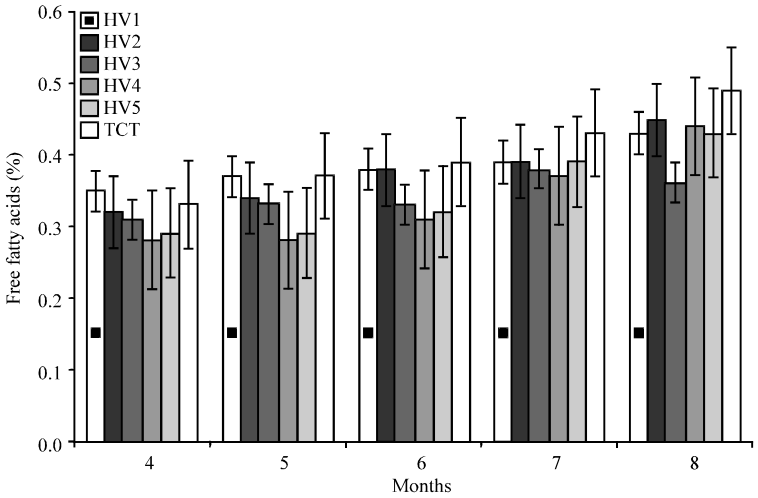

Fig. 6: Mean percentage free fatty acid of different varieties of cocoa in storage

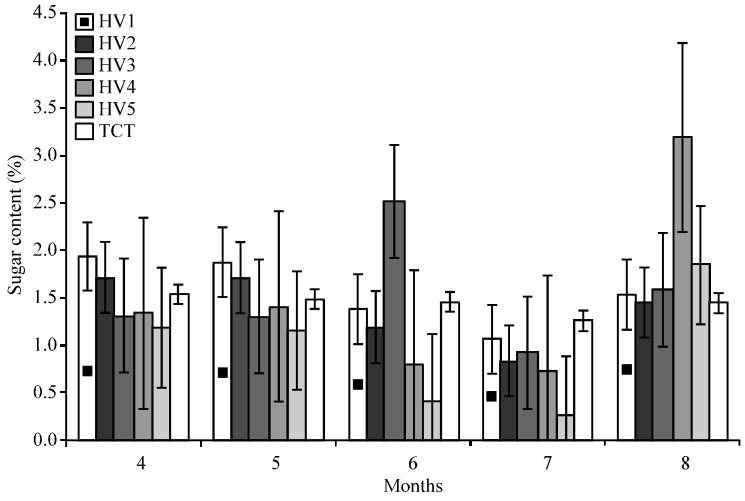

Fig. 7: Mean percentage of sugar content of different varieties of cocoa in storage

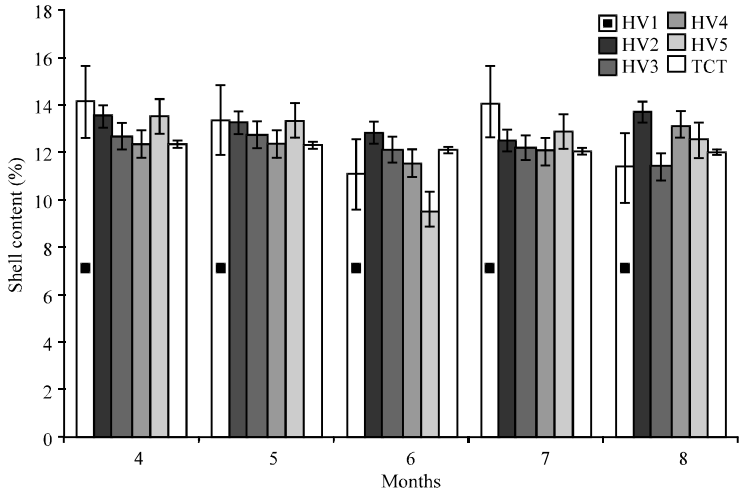

Fig. 8: Mean percentage shell content of different varieties of cocoa in storage

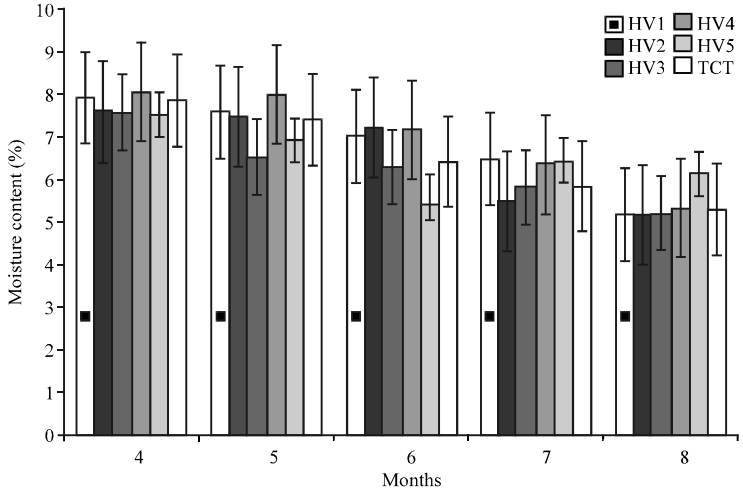

Fig. 9: Mean percentage moisture content of different varieties of cocoa in storage

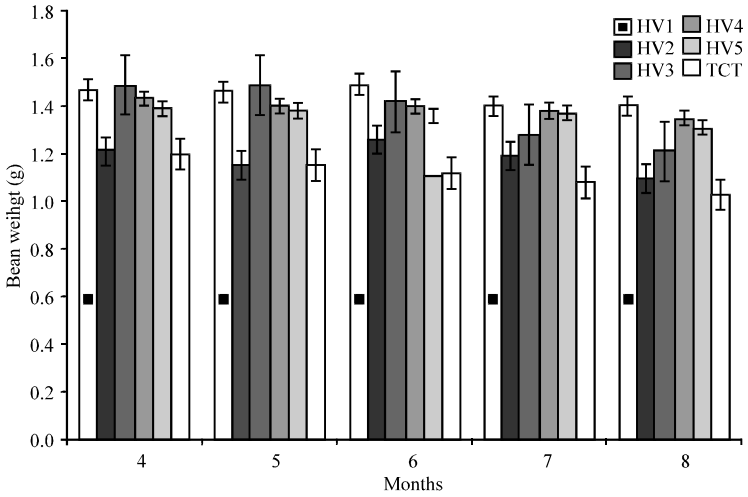

Fig. 10: Mean bean weight of different varieties of cocoa in storage

(1.03-1.20 g). However, HV2 generally recorded the lowest bean weight among the new hybrid variety cocoa beans before and throughout the storage period (Fig. 10).

DISCUSSION

Insect damage to the stored cocoa bean: All the four insect pests introduced into the new hybrid variety cocoa beans in storage did not survive probably, because they could not penetrate the cocoa beans. This probably explains why the usable nib of the hybrid variety cocoa beans remained intact after the four-month period of storage. The inability of the insect pests to cause damage, i.e., superficial evidence of deterioration, e.g., holed or broken beans from which losses may result (Boxall, 1986) to the stored beans of the new hybrid variety cocoa could be due to the thick shell of the beans and/or biochemical effects.

Some insects have been found not to be attracted to stored products at long range but to locate their food by making test burrows into anything soft enough to be bored (Hodges, 1994; Hodges *et al*., 1999) causing much damage. This was not the case of the beans of the new hybrid variety cocoa. It has been demonstrated that chemicals contribute to the location of host by prey (Velemir *et al*., 2001) but the feeding and multiplication could be influenced by different factors such as attractants, deterrents, different amount of chemical composition and biochemical changes. The insects inflicted damage on the traditional type mainly by direct feeding, which placed it at a disadvantage to the new hybrid variety cocoa beans. Their multiplication on the cocoa beans of the traditional type is a nuisance, for example, members of the genus *Tribolium* are known to produce toxic quinines (Mills and White, 1994).

Effect of insect pests on fat content and FFA level: It is known that insect damage to cocoa in storage results in mustiness, leading to mould formation and the break down of fat to free fatty acids in the beans. The insects did not survived on the beans of the new hybrid variety cocoa and consequently could not inflict appreciable damage to the beans. The level of insect damage was inadequate to effect changes in the fat content of the beans. The fat content in beans of the five cocoa types was statistically not significantly throughout the experimental period of four months ($p<0.01$).

Contrary to expectation, there was appreciable increase in the level of FFA throughout the period of storage ($p<0.01$). The increases were, however, not significant at 95% probability (i.e., $p<0.05$). Thus all the five hybrid cocoa types were good materials since the FFA levels recorded (<0.05%) remained within the acceptable limit of 1.0% (Anonymous, 1996). This suggests that other factors such as biochemical factors may also be responsible for the increases in FFA levels in stored cocoa beans.

The significant ($p<0.01$) reduction in the fat content and the significant ($p<0.01$) increase in the free fatty acids during storage could be attributed to the activities of the enzyme lipase, which is naturally present in raw cocoa (Minifie, 1989). The enzyme could have become active due to the changes in moisture content of the beans and the high temperatures of the storage environment, hence contributing to the observed phenomena. Also, it is an established fact that insect infestation during storage results in loss of usable nib in cocoa beans and

breakdown of the butter, the most valuable part of the beans. This could also result in an increase in the amounts of free fatty acids and changes in the flavour of the cocoa (BCCCA, 1996).

Sugar and shell contents: The fact that there were no appreciable changes in the sugar content and the shell of the beans further confirms that the five cocoa types were good materials. The sugar and the shell content of the beans were not affected by the insect activity. This could be attributed to several factors including the inability of the insects survive on the beans.

Moisture content and bean weight: For safe storage, the moisture content of cocoa beans should be between 6 and 7%. Above 8% there is danger of moulds developing on the beans, but below 5% the beans will be very brittle (Wood and Lass, 1989; Jonfia-Essien, 2004) and may disintegrate to give high levels of broken beans (Anonymous, 1996). The reduction in moisture content probably accounted for the significant reduction in the weight per bean ($p<0.01$), which also differed significant among the hybrids ($p<0.01$). Storage in a well managed controlled atmosphere for four months allowed gradual drying, resulting in a reduction in moisture content to the acceptable limit and eliminating any problem of mould development. This supports the earlier assertion (Jonfia-Essien, 2004) that prolonged storage results in moisture loss.

Beans of the new hybrid varieties and of the traditional type were both considered as main crop grade. The observed significant decrease in bean weight ($p<0.01$) for all the cocoa types over the storage period could be attributed to the gradual drying of the beans in storage. Genotypic effect may have contributed to the significant differences in the bean weight of all the cocoa bean types ($p<0.01$).

The higher bean weight of the new hybrid varieties compared to that of the traditional type implies that the beans of the new hybrid varieties were larger and heavier than those of the traditional type. These differences could be attributed to the significant differences in the weight of the nib ($p<0.01$), since the weight of the shell of the new hybrid varieties was not significantly different from that of the traditional type beans. The new hybrid varieties therefore fulfil one of the important quality requirements of cocoa manufacturing companies, i.e. the size and weight of the nib determine the level of the fat content or butter of cocoa beans, hence, the new hybrid varieties may contain higher fat content than the traditional type.

All of the five dry hybrid cocoa types tested were not susceptible to the four-test storage insects. The level of infestation as well as damage done to the cocoa beans, were very minimal. The fat content of the beans remained high in all the cocoa types to the end of the four-month period and the FFA levels remained within the acceptable limit. The sugar and shell contents of the different cocoa types were virtually the same and remained virtually unchanged during the experimental period. The significant weight loss recorded in the beans of all the cocoa types may be attributed to a reduction in moisture content. It is concluded that, all the five hybrid cocoa types tested are good materials with respect to storage insect damage and related problems.

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