Cocoa (*Theobroma cacao* L.) bean processing and storage conditions control for safe chocolate products

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Abstract

Cocoa beans can be infested during cultivation and storage by living organisms (insects: moths / cockroaches; mites; fungi) leading to spoilage/deterioration. Insect parts may be incorporated to chocolate products. Those chocolate contaminants, including their fragments/larvae/eggs, indicate low quality handling and storage of beans. This study evaluated cocoa beans handling/storage conditions in cocoa farms (48) and warehouses (12) located in Northeastern Brazil (Itabuna-Ilhéus regions - at Bahia state), during five months (March to July 2014). The parameters investigated were *storage facilities:* silos types & material (vertical or horizontal/wooden or metal/with or no thermometry); *environmental conditions and exposure* (rain precipitation/relative humidity-RH / temperatures); *cocoa bean batches* (humidity: moisture content-mc/water activity-a_w); presence of *living organisms* (insect/fungi toxins) and *cocoa bags materials* (raffia/jute / polyethylene layers) for green methods of prevention / control / decontamination recommendation. Of the farms and warehouse storage facilities evaluated, 67% did not have automatic environment control (i.e., aeration / TºC) and 33% applied high storage technology (control by aeration, refrigeration). Regarding external and internal warehouse conditions of r.h. and temperature, they reached a mean of 75% (60 min; 90 max) and 25ºC (20 min; 35 max), respectively during the period of study. Bag insect proliferation and fungi spoilage were registered in 50% of the total number of warehouses evaluated, leading to part of stored bags being discarded. Application of controlled atmospheres (vacuum / ozone / hermetic storage, through cocoon in-containers for export), was recommended as well as improvements in bagging material and temperature monitoring.

Keywords: cocoa, *Theobroma cacao*, light filth, storage, chocolate, insects, fungi control

1. Introduction

The early stages of cocoa bean processing is done in cocoa farms, where the characteristic chocolate flavor starts to develop. The post-harvest cocoa bean technology depends on the type of cocoa batches to be sold for chocolate making. Whether they are of quality (a) *Premium* (selected cocoa varieties – i.e., beans with high standards of tissue structure and characteristic aroma); (b) *Commercial* (beans of not highly controlled standards and sensory quality) or (c) *For export* (beans where flavor is not considered the most important contributors, usually utilized in formulation and cookery) (Opoku et al., 2010; Ceplac, 2014; Icco, 2014). After cocoa fruits are harvested, their seeds have the surrounding pulp extracted and the seeds undergo natural fermentation by yeast and bacteria. That process causes bean swelling and brownish color development (enzyme reaction on polyphenols content as pH reduces) and moisture content (mc) is high - ca. 60% (Ribeiro, 1996; Barel, 1997; Nachtigall, 1999). From that stage, the seeds are called beans (no germination capacity) and are sent for sun drying on cement platforms (mc: around 7.5%) (Hii et al., 2006; Efraim, 2009; Kreibich et al., 2014). Next, they are packaged in bags and sold for the internal market or exported.

Fungi proliferation due to climatic factors (rain/heat: high moisture/temperatures) that can lead to production losses up to 40% (Faleiro et al, 2004; Rubini et al., 2005; Scarpari et al., 2005; Mounjouenpou et al., 2008) must be controlled.

The presence of biological contaminants (insects / mites / rodents hair / fungi) in the final product (chocolate) is indication of lack of hygienic-sanitary control during the phases involving its production - from cocoa fruits (raw material) cultivation (harvest / transport), seed to bean transformation (fermentation / drying / storage / packaging) and selection /segregation at the industry reception for cocoa paste transformation (handling / processing / packaging) including the final product (processing / storage / transport) (Ribeiro, 1996; Nachtigall, 1999; Copetti et al., 2011). These contaminants are important carries of microorganisms (fungi, bacteria, viruses) and of their metabolites - toxins) (Scussel et al, 2011; Koerich de Souza, 2013; Kreibich et al., 2014; Savi et al., 2014). In addition, mites, which develop in stored products (especially those with high fat / protein content) can trigger allergic reactions (Lorini, 2012) and inappropriate packaging with improper storage conditions can enable them to be incorporated into the final products (Lorini, 2008; 2010).

The official cocoa classification for export per the Brazilian Agriculture Ministry Regulation number 161/88, stablishes standards for cocoa produced in the country as follows. *Type I:* fermented dry beans, maximum mc of 8%, characteristic aroma, no strange odors, free from foreign material, containing up to 110 beans/100 g, with some tolerated defects (mildewed, flattened & slates beans) up to 2% and insects up to 3%) and *Type II* (apart from being also fermented dry beans, with 8% mc, characteristic aroma, no strange odors, free from foreign material, containing up to 110 beans/100 g, with tolerable defects including moldy beans at a maximum of 2%, insects presence to a max of 6% and flattened beans max. 3% (mapa, 1988; Concex, 1988; FAO, 2014).

Considering the importance that cocoa beans and the chocolate production play in national and international trade, this paper aims to evaluate the safety of cocoa bean storage from the Itabuna-Ilheus region of Bahia, northeastern Brazil. The focus on their (a) processing steps (fruit seed extraction / fermentation / drying / handling) and (b) storage facilities *versus* their exposure to environmental conditions, in order to identify possible contaminants and recommend improviments.

2. Materials and Methods

2.1. Material

2.1.1. Samples: cocoa fruits and dry beans (60) of *Premium*, *Commercial* and *for Export* Types (24, 30 and 06, respectively), from the 2014 harvest season (March to July). They were from different cocoa producers *farms* (48) and *warehouses* (12) located at the Itabuna-Ilheus region of Bahia state, Northeastern Brazil (Figures 1 and 2).

2.1.2. Farms and warehouses: (a) farms – comprised of different buildings, including *(a.1) reception; (a.2) processing* area of fruit treatment (cut / cleaning), seed extraction (separation from pulp) and fermentation room; *(a.3) drying* areas; and *(a.4) storage* facility (*primary* storage); *(b) warehouses - (b.1) secondary storage* – with average storage capacity (185 bags /60 kg) for 30 days in austere conditions and control of hygiene and safety; *(b.2) central storage* - with different buildings including dry beans bags reception, cocoa classification and large storage space with capacity for 945 to 23000 bags (60 Kg each).

2.1.3. Equipment: (a) cocoa seed extraction - knives, wooden boxes (2 x 2 m width x height, respectively); *(b) fermentation* – wood and stainless steel fermentators (10 m^3) (with temperature control); *(c) drying* - cement platforms, electric ovens and hot air dryers

(tubular); *(d) packaging* - bag packaging machine; *(e) storage* - scales, sieves (10 x 20 mm, width x length), sample collector *(calador)*, impurities separator, waste/dust removal cleaners, conveyor belts, refrigeration equipment, moisture analyzer and mill.

2.1.4. Other materials: packaging material such as raffia (a synthetic material that mimics the polypropylene fiber), jute (*Corchorus capsularis* - a vegetable fiber with textile extraction), cotton (a textile, soft and hydrophilic, vegetable fiber), dimensions of 700 x 550 mm for height and width, respectively.

Figure 1 Cocoa (*Theobroma ca*cao L.) (a) mature fruits and (b) dry beans (post-fermentation & drying steps) from the Brazilian state of Bahia.

2.2. Methods

2.2.1. Sampling: cocoa beans (60) were collected from batches of farms (48) and warehouses (12), from the following cities (17): Camacã / Mascote / Arataca / Ilhéus / Firmino Alves / Ibirapitanga / Itacare / Uruçuca / Una / Belmonte / Buruare / Canasvieiras / Pau Brasil / Itabuna / Itape / Itapevi / Belmonte). All were located in the Bahia state main cocoa producers region (Figure 2). They were collected from pilled bags (500 g portions) utilizing cocoa sampling collector. After that, they were sent to the Food Microscopy and to the Mycotoxicology Food Contaminants laboratories at the UFSC to carry out the investigation on the: presence of living organisms (rodents / insect / mites / fungi) toxins, and humidity (mc / water activity - a_w).

2.2.2. Cocoa bean processing conditions: the whole processing steps were evaluated i.e., from the *(a) cocoa fruits seed extraction* - fruits (harvested mature) after cracked open and the seed separation from mucilaginous pulp (if insect/fungi infested); *(b) fermentation* - seeds to beans (no germination capacity) formation through bacteria fermentation (no fungi growth of which produce unpleasant taste or toxins - harmful to health); *(c) drying* – at this step, different technologies could be applied depending on the producer facilities / (mc/fungi evaluated) from sun light to hot air drier) and *(d) cocoa bags packaging* - whether carried out utilizing adequate/clean bags (capacity: 60 kg), proper material Type with jute / polyethylene / raffia or cotton, in different layers (impurities free) (Nachtigall, 1999; Hii et al., 2006; Beckett, 2008; Efraim, 2008; Copetti, 2011).

2.2.3. Storage evaluation: (a) facilities - the farms (primary storage) & warehouses (secondary and central storages) were evaluated for type of construction (vertical / horizontal stores), material (wooden / metal), whether they had temperature control (windows / thermometry cables) and aeration (equipment for regular air / with refrigeration). Also

whether the facilities had areas for dust cleaning procedures, and rodents/insects/mites or other vectors control; *(b) environmental conditions - (b.1) internal*: type of ventilation (natural / fan / refrigeration / other gases), time and method of mc control; *(b.2) external* - registration charts of daily/hour temperature checking (average/maximum/minimum), relative humidity (RH) and rain precipitation of the sampling storage site. The storage average time, was usually for a period of 30 to 90 days.

2.2.4. Living organisms possible presence in beans facilities: both, infestation and detection were evaluated *(a) infestation* - insect entrance/location was investigated throughout the whole process (at fruits opening / seed separation / during fermentation / drying / storage / packaging). Either, their presence indication of living organisms (rodents / hair / insect / egg / larvae / moth/fungi) and possible damage caused to cocoa beans. Also how the beans/bags were handled (whether loose - on the floor or in bags - on pilled/ pallets, also if close or far from the walls) and *(b) detection* - the living organisms presence/detection was investigated by checking through stereo microscopy the characteristic details by at different amplifications (beans damages by rodent / insects broken & fungi spoilage (Scussel, et al., 2014).

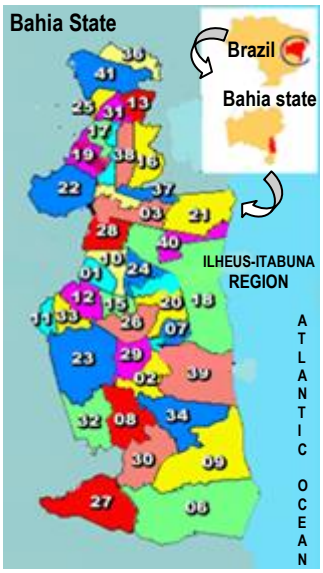

Figure 2 Itabuna-Ilheus cocoa beans (*Theobroma cacao* L.) producing region with its 46 sample collection sites at Bahia state, Brazil.

3. Results and Discussion

The evaluation of the whole cocoa bean production (seed fruit detaching ➡ fermentation ➡ drying ➡ bean bags storage) heady for chipping / commercialization, showed some points (mainly, at packaging and storage) of living organisms entrance/development that should be controlled/destroyed in order to avoid safety problems and solutions were recommended. Tables 1 & 2 and Figures 1 to 6 show sample characteristics, the processing and storage facilities, as well as environmental conditions that could influence the development of insects and fungi.

3.1. Cocoa bean processing conditions

It was observed that the main cocoa bean processing steps were similar among the different Bahia state main cocoa region beans producers. Producers harvest the fruits fully ripe and healthy and use selected (specific) varieties (Criolo). Most of them also inspect fruits for lesions or fungi injuries prior and after the washing step. *(a) Cocoa fruits seed extraction* - at the fruit opening (stainless steel knives) by trained staff and seeds extracted (staff also remove impurities

(shell fragments / placenta / defective or germinated seeds), seeds are inspected for insects/fungi injuries which can interfere on the next step - the fermentation. *(b) Fermentation* – at this step beans swell and turn into brown color as pH reduces. That process changes the bean characteristics and is the precursors of desired flavors are produced. At the end of fermentation, the bean MC is still high and ranges from 50 to 60% (Oetterer et al., 2006). *(c) Drying:* as expected, the drying process reduces MC to 7-8% and also the acidity (providing continuation of the chemical and biochemical transformations within the beans) (Lacey, 1991; Beckett, 2008). The drying process in the Bahia region is carried out in different sites and/or equipment: (c.1) sun (c.2) electric and/or (c.3) with hot air dryers reaching a maximum temperature of 55°C (allowing slow moisture release / evaporation). Regarding wood combustion/heat source driers, if smoke gets in contact with the beans, it can cause contamination by polyunsaturated aromatic hydrocarbons - PAHs (smoke produced and carcinogenic). In addition, if the temperature is too high, it also allows PAHs formation. Figure 3 shows top covered, on suspended tables (far from soil contact), safe drying process. Although under the sun (natural drying). As the post-harvest technology applied to cocoa bean depends upon the Type to be sold (*Premium / Commercial / for export*), the cultivation process, the fermentation type and the drying source applied also greatly influences the final flavor and cocoa quality Type. Some Brazilian farmers use firewood dryers, which provide smoke flavor to cocoa beans. That can cause PAHs (contamination) which in turn causes rejection by some export marketers. Regarding hygiene, from the 48 farms studied, 35 (73%) of them were commercial farms producing cocoa in conditions often had some lack of hygiene and good storage practices. *(d) Packaging* - in the different farms and warehouses surveyed, cocoa beans were stored, either in raffia, jute or other porous (to enable air exchange) material bags. Bags should be clean and free of odors. According to Decree n. 6268 (MAPA, 2007), that established the Technical Regulation for cocoa beans, the bags used in packaging should be of those appropriate materials and specifications. The bagged samples must be stored in containers with screens to allow breathing and bean protection against insect infestation. The mesh size of the screen must be small enough to prevent the entry of moths and larvae. Beans for export are packaged in 60 kg bags and Ceplac issues a report according to the classification Type and also a certificate accordingly (Ceplac, 2007). Vacuum & nitrogen, 3 layers material packaged beans are utilized mainly for export in several of the sites evaluated, as they are utilized also for Brazil nuts in the Amazon Region (Pacheco and Scussel, 2007).

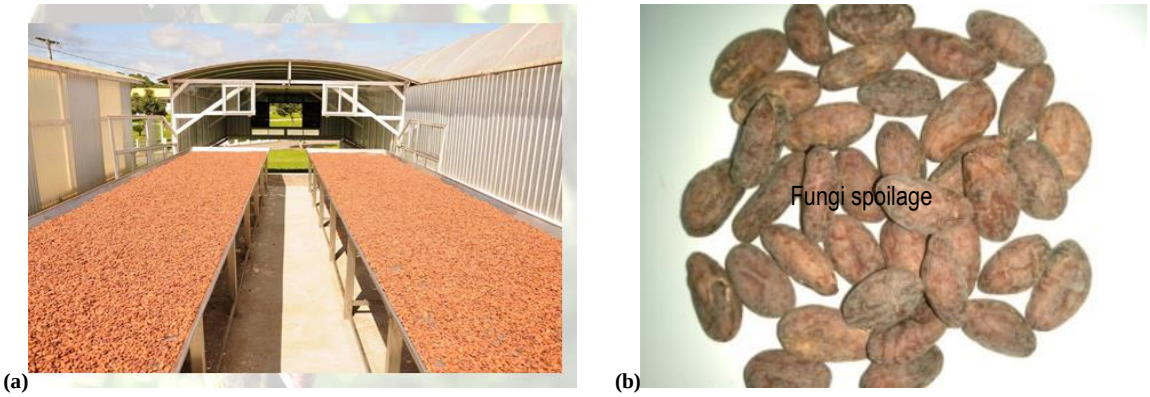

Figure 3 Cocoa beans (a) under proper sun drying procedure with protection (wall, ceiling, far from soil/floor contact) and (b) under improper drying allowing fungi proliferation (due to high humidity).

3.2. Cocoa bean farm & warehouse storage

Several cocoa warehouses (secondary & central storages) are located in cities near the producing cocoa farms (primary storage) (Figure 4). As soon as the bean bags reach the

warehouse they are weighed and quality checked (flavor, mc, damages, spoilage), prior to being stacked onto pallets for storage. For trade, they are transported on trucks either to the harbors for shipping (export) or distributed into the internal market. The USA market is the largest Brazilian cocoa importer.

(a) Facilities – the storage facilities of the Itabuna-Iheus region have quite broad differences. Although some of them could allow humidity/moisture absorption and thus worsen the quality for export, other storage facilities had floor, brick and walls are covered with wood or polyethylene/transparent to reduce indoor humidity; availability of ventilation windows (facing the direction of prevailing winds) protected by fine-mesh screens (to prevent insects entry); excellent sealing (to prevent rodents entry). Some facilities have their dimensions varying with length, width and walls height to minimum of 9, 7 and 2 m, respectively (Almeida et al., 2001). *(b) Conditions* - dry cocoa beans are hygroscopic, so they absorb moisture from the environment under high humidity conditions (FDA, 2014). It is recommended that maximum storage be 2 to 3 months in tropical countries conditions, to reduce exposure to high temperatures and humidities. If storage is longer, humidity should be strictly controlled, i.e., cocoa beans mc should be periodically checked and kept below 8% (CAC, 2013), which is done in the Itabuna-Ilheus region by most producers. In addition, the use of - bags in cocoon into containers - has been tested to inhibit the development of filamentous fungi mycotoxin producers during shipping. The storage performed on these farms has been carried out under air-conditioning (at 20 to 22°C). In that condition, their acidity decreases and beans indefinitely conserved (from mc initial: 50-60 %; to final: 7-8%). *(b.1) internal conditions* - from the 12 storage facilities visited, 33% (4) were considered suitable for beans storage. The warehouses initially assess the cocoa beans MC coming from farms (which cannot exceed 9% - otherwise they are rejected, or the price reduces). Samples with smoke odor also have reduced price. The temperature of the warehouse is controlled by air ventilation and the bags are distributed above the pallets to prevent contact with the warehouse floor and walls. *Pre-cleaning* – when cocoa beans are received (bags), they are submitted to pre-cleaning process, through a 3-stages sieve for impurities separation such as cocoa husks, sand and insects. The cocoa bean separated fraction then goes to the vacuum system to remove large and light density impurities such as pieces of bark, insect and strings, followed by passing through a magnetic and stones separators (Figure 5.b.2). *(b.2) external conditions* – although most of the cities in the South coast of Bahia has a humid tropical climate with average annual rainfall between 2000 and 2400 mm (well distributed throughout the year) summer is the period of higher precipitation. The annual temperature average is of 24.7°C, and in the winter period goes down only up to 21°C (Table 1). Only 4 of the 12 stores visited (33%) warehouses had control temperature, using air conditioning, in addition to controlling the RH. The Ceplac is responsible for providing technical assistance for the construction of warehouses. This assistance teaches how to control the attack of insects and rodents, how to maintain a good level of hygiene, such as exterminating pests and how to control humidity and temperature.

Figure 4 Cocoa beans warehouse facilities: (a) Primary storage, (b) Intermediary and (c) Central storage unity at Bahia state.

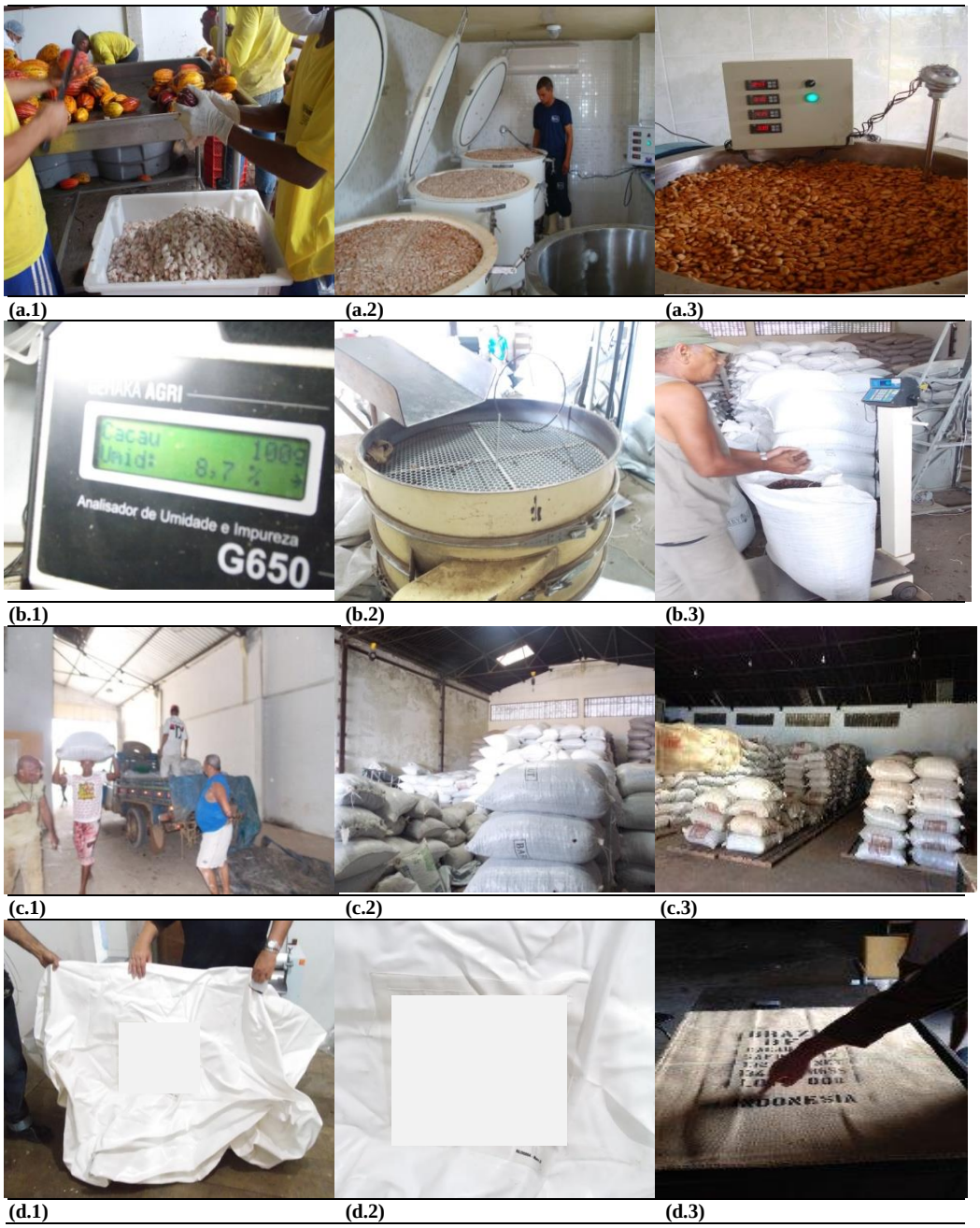

Figure 5 Cocoa beans (a) initial processing steps (a.1) seed fruit separation: (a.2) fermentation site / air-conditioned environment, (a.3) final fermentation step / temperature control (21 and 24ºC); (b) classification equipment: (c.1) humidity control meter, (c.2) three-stages sieves -cleaning/waste/size, (c.3) scales; (d) material applied for bags cocoon storage: (d.1) hermetic storage, (d.2) closer view, (d.3) jute bag.

3.3. Living organisms versus cocoa beans whole process & storage points

Foreign materials and/or impurities present in cocoa beans batches and in their storage environment surroundings can be responsible for whole batch *deterioration* as they can favor rodent / insects (egg/larvae) / mites / fungi proliferation. Also cocoa bean *mechanical* or *biological* damage can occur due to insect and rodent attack which promotes moisture absorption and facilitate fungi invasion / to the highly nutritious inner cocoa bean, leading to rapid fungi development thus mycotoxins production (Scussel, 2002). (a) *Infestation entrance / location* - as expected the main sites were close to the dispersing bean unities, also where

dust was accumulated in each processing step (fruits opening / seed separation / fermentation / drying / packaging /storage). *(b) Detection* - most of the farms segregate spoiled/damaged beans and check for possible living organisms detection, however we also checked their presence and beans damages produced through stereoscopy as shown in Figure 6 (cocoa beans *testae* damage / fungi / mites proliferation which are difficult to see by naked eye).

Table 1 Itabuna-Ilheus region climate parameters obtained during the cocoa bean study (2014 season)[a].

Parameter	Month				
	March	**April**	**May**	**June**	**July**
Temperature(°C)					
Average	25.9	25.3	24.1	22.7	22.0
[b] min.	22.4	21.8	20.9	19.7	18.9
[c] max.	29.4	28.7	27.5	26.4	25.8
Humidity					
Rain (mm)	216.9	204.7	144.5	200.6	200.5
RH[d] (%)	80.7	81.5	83.0	85.7	86.5
Days with rain	17	15	14	16	18
Sun light					
Hours sunshine	235.9	203.4	199.8	191.3	197.7

[a]March-July [b]minimum [m]aximum [d] relative humidity (National Institute of Meteorology, 2014)

3.4. Cocoa bean batch quality

The a_w of cocoa beans after drying should not exceed 0.7 to maintain quality and avoid fungal growth and mycotoxin production, besides indicated the chemical and physical stability if they are stored well. The mc of cocoa beans is reduced from 60-65 to 7-8% in the drying step. Some stores come to receive beans with higher mc however, in that case need to proceed to another drying step to keep bean safe (Ephraim, 2009). Based on these results the cocoa samples (60) evaluated in the current study, had a_w varying from 0.5964 to 0.8521 and 30 samples (50%) had higher levels (a_w: 0.7117 to 0.8521), still on the safe side. Levels higher than those will favor fungi growth and toxin production if toxigenic. There were also mc changes from 4.72 to 13:57% respectively in some of the storage evaluated. A total of 23 samples had high mc range (8.28 to 14.00%), thus favorable conditions for fungi development. Studies have reported toxigenic fungal species (*A. flavus* and *A. niger*) in dried cocoa beans from different growing areas and countries (Niles, 1981; Aroyeun et al., 2007; Mounjouenpou et al., 2008; Rahmadi and Fleet, 2008; Sanchez-Hervas et al., 2008; Copetti et al., 2011). From the visited 48 farms, 26 of them (54.2%) had production of cocoa Types I and II. Despite the superior quality required for Type III (for export), most of its production stays in the domestic market due to the high Brazilian chocolate production & consumption.

Recommendation: as fumigation is an effective insecticide treatment and not quite simple to apply, that procedure/step depends on the cocoa bean buyers to decide application. It may be carried out in the warehouses or in the cocoa containers. The fumigation consists of distributing tablets (2 cm) beneath the sacks (for as long as 20 sec). The amount is calculated per m^2 bags. After this step, the beans are covered with a canvas sheet during 72 hrs, only after that, it can be removed and the remaining gases vented off to the air. Other methods of insect/fungi control could be also by applying modified atmosphere (carbon dioxide, ozone), or also by keeping hermetic / sealed (as long as innovative facilities construction - no air entrance are adopted/developed) environment, thus preserving the contact with external environment / moist.

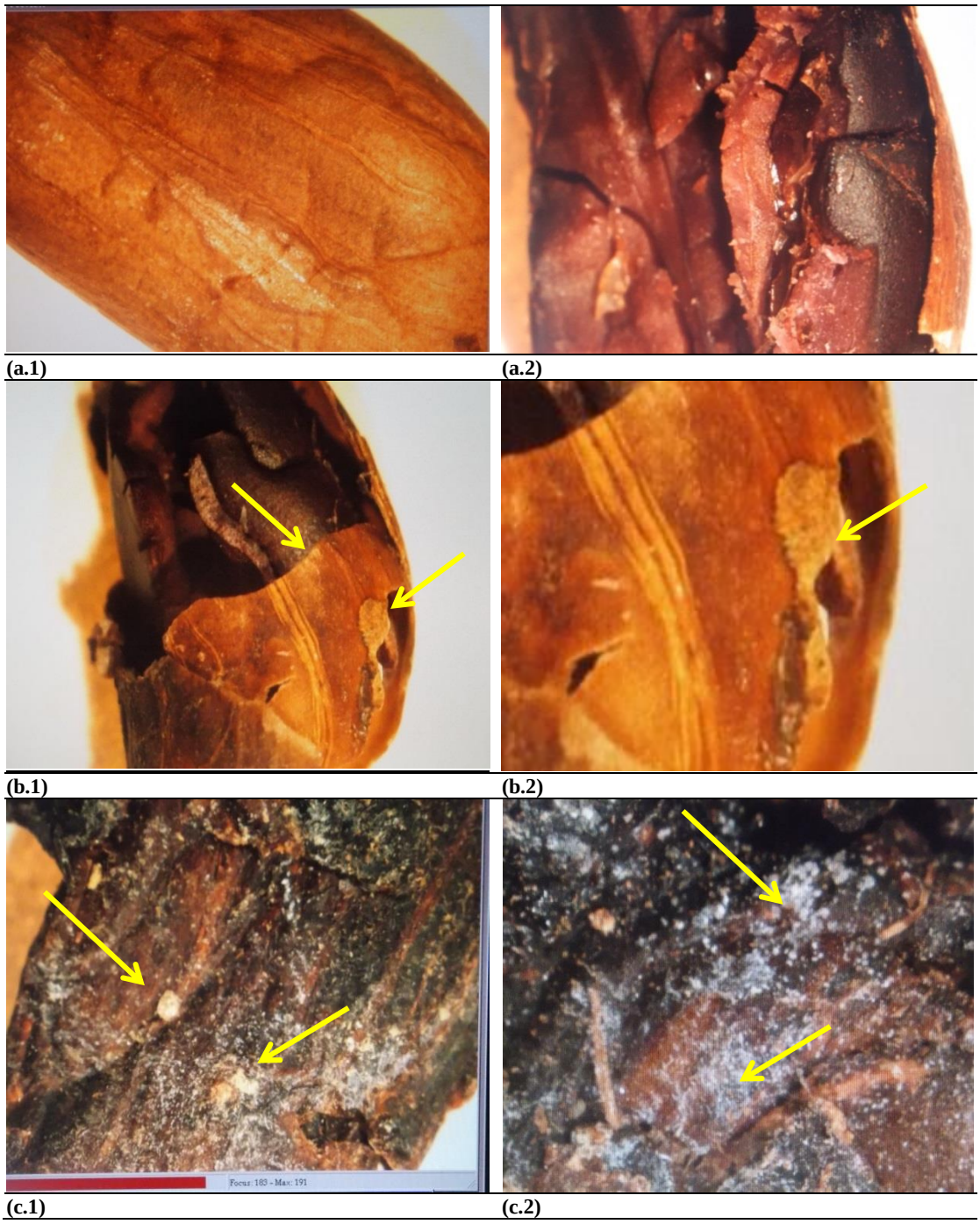

Figure 6 Stereo micrographs of cocoa beans (*Theobroma cacao* L.) (a) *Premium Type* [(a.1) intact *testae*, (a.2) cracked endosperm]; (b) *damaged testae* [(b.1) mechanic, (b.2) by insect]; (c) *spoiled* [(c.1) by fungi/mites, (c.2) fungi colonies].

Table 2 Cocoa farms (primary) & warehouses (intermediate & central) storage characteristics.

Storage				Cocoa bean		Drying process		Whole cocoa bean production
Type	Capacity (bags*Kg) Size (m^2)	Conditions	Time (days)	Type	Sellection	Heat	°C	
I - COCOA FARm* (primary n=48)								
185 (11.100)	100	Cement platform No rodents control No dust control Basic storage	30	Premium Commercial Export	Premium type Cocoa Export	Sun Oven	55	Fruits harvesting Breaking of fruits Fermentation Drying & Storage
II - WAREHOUSE (intermediate n=9)								
945 (56.700)	300	Cement platform Bags on pallets Storage devices No rodent & dust control (10%)	30-40	Premium Commercial Export	High moisture Fungi spoilage Color alteration Bad odors Light filths	NA**	NA	Reception Selection Weighing Screening Storage
III - WAREHOUSE (central n=3)								
23.000 (1.380.000)	1.500	Cement platform Bags on pallets Storage devices (100%) Windows with screens(100%) Controlled atmosphere (30%)	90	Premium Commercial Export	High moisture Fungi spoilage Color alteration Bad odors Light filths	NA	NA	Reception Selection Weighing Screening Storage/shipping

* 60 kg ** not applicable

4. Conclusions

The whole cocoa bean production process (from the different producers of Itabuna-Ilheus region) showed some points (mainly, at packaging and storage) of living organisms entrance/proliferation that should be controlled in order to avoid safety/quality problems for final product (either cocoa beans, paste or chocolate). A constant concern on maintaining the quality of cocoa to ensure the best processing conditions, storage and of their final products should be a goal to be achieved in the short term. In addition to a program of sustainable agriculture, living organism contamination of raw materials (fruits/seeds) through crop conservation methods and control (traps/fumigation/controlled atmospheres). That will result in the reliability and safety of the end buyer, adding value to the commercial product, enabling the national, international and export economy, as well as reducing losses due to low quality products. From the visited farms (48), only 13 farms (27%) produce premium cocoa. The whole process is important as the varieties utilized to get the following characteristics - beans have a fruity, flowering, wooden and/or nut aroma (from the Criollo and Trinitario varieties) whose aroma influence depends on the steps from fruit harvesting to the processing final stages.

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