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Post-harvest management of aflatoxin contamination in groundnut

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REVIEW ARTICLE

Abstract

Aflatoxin contamination in groundnut by *Aspergillus* section *Flavi* is a major pre- and post-harvest problem causing kernel-quality loss. Post-harvest aflatoxin contamination is caused initially by infestation of aflatoxigenic strains at the pre-harvest stage, resulting in reduced kernel quality after harvest. Improper handling of pods and storage methods after harvest lead to high moisture and ambient temperatures, directly causing aflatoxin contamination. In this review, we report the extent of post-harvest contamination along the groundnut value chain in the Kolokani, Kayes, and Kita districts of Mali in West Africa. Groundnut kernels and paste samples were collected from retailers in selected markets from December 2010 to June 2011, and aflatoxin B₁ (AFB₁) content was estimated. Aflatoxin was significantly higher in groundnut paste than in kernels. Kolokani recorded the highest toxin levels in both kernels and groundnut paste compared with the other districts. Overall, AFB₁ levels in kernels and paste increased during storage at the market level in the three districts and were above permissible levels (>20 µg/kg). The effect of weather factors on post-harvest contamination and the reasons for aflatoxin build-up in Mali are discussed. This paper also highlights different management tools for reducing post-harvest aflatoxin contamination, such as post-harvest grain handling, post-harvest machinery, physical separation, storage methods and conditions, disinfestation, detoxification, inactivation, filtration, binding agents, and antifungal compounds. Post-harvest management options and enhanced use of good agricultural practices for mitigating this problem in Mali are also presented.

Keywords: *Aspergillus* spp., groundnut value chain, groundnut paste, Mali, post-harvest control

1. Introduction

Post-harvest deterioration in groundnut (*Arachis hypogaea* L.) is largely due to mould development, especially by the *Aspergillus* section *Flavi* group of fungi. Aflatoxins produced by *Aspergillus flavus*, *Aspergillus parasiticus*, and others in this group are secondary metabolites with carcinogenic, estrogenic, teratogenic, and immunosuppressive effects (Klich *et al.*, 2009). Contamination of groundnuts by these fungi occurs at both pre- and post-harvest stages, leading to aflatoxin contamination. However, aflatoxin contamination occurs more during post-harvest than during pre-harvest conditions (Wild and Hall, 2000). Improper management practices and adverse climatic conditions at harvest and after harvest are predisposing factors for post-harvest aflatoxin contamination. Significant grain deterioration caused by moulds also occurs during storage because of prevailing ambient conditions. Maize is a staple cereal that

is also frequently contaminated with aflatoxins. Up to a 10-fold rise in aflatoxin levels was reported within 3 days when field-harvested maize was stored under high-moisture conditions (Hell *et al.*, 2008). It is assumed that aflatoxin build-up in groundnut pods can be similar to that of maize during storage. Aflatoxin contamination levels during post-harvest storage of groundnuts are alarming; for example, insect-damaged samples collected from farmers in Andhra Pradesh, India, have aflatoxin levels of >500 µg/kg (Waliyar *et al.*, 2003). Further, these aflatoxin levels increase in food during storage (Kaaya and Kyamuhangire, 2006).

According to the Food and Agricultural Organization (FAO), 25% of the world's food crops are significantly contaminated with mycotoxins (Boutrif and Canet, 1998). Because maize and groundnuts are dietary staples for the majority of the rural poor in sub-Saharan Africa (SSA), mycotoxin poisoning is common in this region

(Wagacha and Muthomi, 2008). Thus, there is a direct correlation between socioeconomic status and exposure to mycotoxins in SSA countries, with poor families experiencing significantly higher exposure (Wagacha and Muthomi, 2008). Conditions, such as excessive heat, high humidity, lack of aeration in stores, and insect and rodent damage, which are common in the tropics, including SSA, aggravate toxin accumulation (Hell and Mutegi, 2011). Of 550,000 to 600,000 new hepatocellular carcinoma cases reported worldwide annually, approximately 25,200 to 155,000 cases are attributed to aflatoxin exposure. Most of these are in SSA, Southeast Asia, and China and are caused by uncontrolled aflatoxin accumulation in food and hepatitis B virus infection (Liu and Wu, 2010). Aflatoxin contamination impacts disproportionately on the livelihoods of the rural poor, and mycotoxicoses-related economic losses are prevalent in African countries (Fellinger, 2006; Wu, 2004).

Increased food production coupled with reduced post-harvest losses is an ideal strategy for overcoming global hunger (Kimatu *et al.*, 2012). SSA is the only region in the world where food production continues to decline, remaining prone to famine and other vagaries. Reduction in post-harvest losses is one of the keys to improving profit. Moreover, post-harvest management is vital for increasing food availability without the need for additional resources (Kimatu *et al.*, 2012). However, the cost effectiveness, sustainability, and technical feasibility of pod-handling techniques should be evaluated with regard to local context and practices before devising strategies for post-harvest aflatoxin contamination. Post-harvest stages generally include cleaning, grading, transportation, storage, processing, packaging, and retailing at the market (Kimatu *et al.*, 2012). Some of the factors affecting aflatoxin contamination in food grains are harvesting, drying, and storage methods as well as moisture content, insect damage, and physical damage (Kaaya and Warren, 2005; Waliyar *et al.*, 2008). In general, adopting proper practices such as harvesting at right crop maturity stage followed by pod stripping soon after harvest, rapid drying, and cleaning of any extraneous matter including damaged pods and gynophores reduce aflatoxins after harvest prior to storage (Rahmianna *et al.*, 2007). Early harvesting and threshing of groundnuts is also recommended to reduce aflatoxin levels (Rachaputi *et al.*, 2002). Here we review the magnitude of the post-harvest aflatoxin contamination along the groundnut value chain in Mali, West Africa. Good post-harvest storage practices and other management options for minimising post-harvest aflatoxin contamination in groundnut are also critically reviewed.

2. Post-harvest aflatoxin contamination in groundnut in Mali

Adoption of proper post-harvest handling measures by small retailers/traders in SSA is difficult because of the prevalence of the informal/unorganised market system (Hell and Mutegi, 2011). This is aggravated by the lack of efficient market policy implementation mechanisms in most SSA countries. Open-air market systems are also prone to pod spoilage owing to the occurrence of abrupt rain storms that wets the pods. To determine the magnitude of post-harvest aflatoxin (aflatoxin B₁ (AFB₁)) contamination along the groundnut value chain in Mali, a study was undertaken by Waliyar *et al.* (unpublished results) from December 2010 to June 2011 in selected markets of the Kolokani, Kita, and Kayes districts. Groundnut kernel and paste samples were collected separately from 30 small retailers of each district for seven continuous months. The samples were collected from the same selected retailers starting from the crop harvest stage (December 2010) until June 2011¹. Kernel and paste samples (1 kg each) were taken at monthly intervals and used to estimate AFB₁ content with an indirect competitive enzyme-linked immunosorbent assay (Reddy *et al.*, 2001). Our studies indicated that in all three districts, AFB₁ levels were higher in groundnut paste than in groundnut kernels (Figure 1) and were above permissible levels (>20 µg/kg). The AFB₁ levels in kernels ranged from 105 to 226.3 µg/kg in the three districts, whereas those in paste ranged from 171.2 to 530.2 µg/kg. Among the districts, Kolokani recorded the highest AFB₁ levels in both paste and kernels over the other districts. This was followed by Kita with toxin levels of 168.2 (kernels) and 236.7 (paste) µg/kg (Figure 1). Month-wise AFB₁ analysis of kernel and paste samples from December 2010 to June 2011 indicated an increase in toxin levels after 7 months

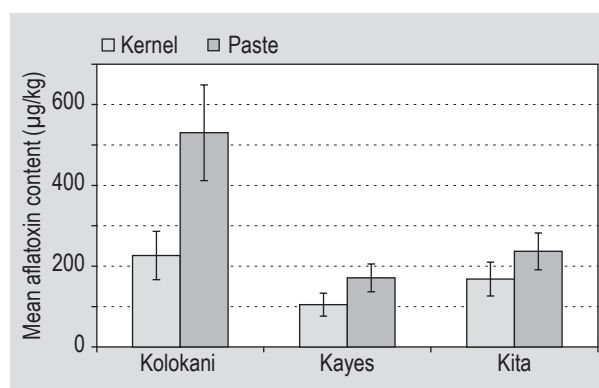


Figure 1. Mean aflatoxin B₁ (AFB₁) levels in market samples of groundnut kernels and paste in different districts of Mali, West Africa, during June 2011.

¹ Mali has one rainy season each year, usually beginning in June and ending in October. The study was designed to ensure visits to farmers' fields during storage time.

in all the markets of Kolokani, Kita, and Kayes. In general, the toxin levels of both kernels and paste increased with an increase in storage time at the trader level in Kolokani, Kayes, and Kita of Mali (Figure 2 and Figure 3).

3. Reasons for post-harvest aflatoxin contamination

From our studies, it is clear that AFB₁ levels are far above the permissible levels along the groundnut value chain in Mali. Market samples of kernel and paste in all three districts showed increased AFB₁ levels over 7 months of storage. This confirms that post-harvest aflatoxin contamination of groundnuts is a major problem in Mali and probably in most West African groundnut-producing countries. Particularly, the toxin levels were higher in groundnut paste than in kernels, and this can be attributed to the inferior quality of kernels used for preparing groundnut paste. Toxin build-up at the retailer end in groundnut samples from Kolokani, Kayes, and Kita can be attributed to improper storage of the primary source of pod stock by the farmers in their granaries. Data from Mali suggest that aflatoxin contamination could further increase while stored by wholesalers (F. Waliyar, unpublished results).

4. Management tools for post-harvest aflatoxin contamination in groundnut

In general, aflatoxin levels in food commodities increase with storage time (Kaaya and Kyamuhangire, 2006). Several strategies and practices for minimising qualitative and quantitative post-harvest losses in groundnut have been developed (Hell *et al.*, 2008). Some of these improved practices have been successfully implemented in SSA at the village level, such as in Guinea as demonstrated by reduced blood aflatoxin adducts (Turner *et al.*, 2005). Here we summarise different management tools for post-harvest contamination.

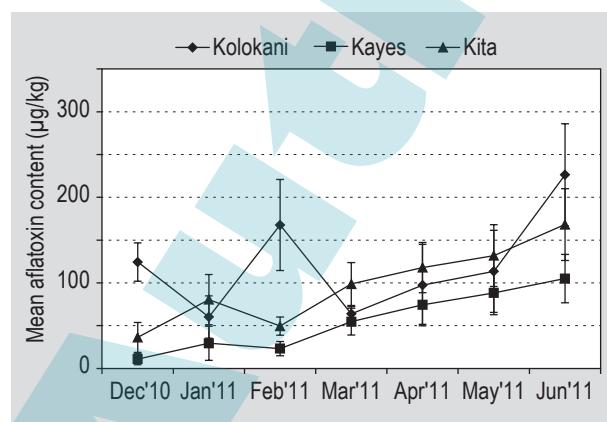


Figure 2. Mean aflatoxin B₁ (AFB₁) levels in groundnut kernels in three districts of Mali, West Africa, as measured in samples obtained from small retailers over 7 months during 2010 and 2011.

Post-harvest handling of pods

Because high grain moisture increases post-harvest moulding and aflatoxin contamination (Heathcote and Hibbert, 1978), proper drying of grains after harvest to ≤7% moisture levels is ideal to prevent growth of fungi, including aflatoxigenic strains (Dick, 1987). Inverted windrowing after harvest exposes pods to sunlight. It also enables increased air circulation, which facilitates rapid drying (Dickens and Khalsa, 1967). Devi and Hall (2000) confirmed that groundnut aflatoxin contamination could be reduced by inverted windrowing. A similar effect can be achieved by field drying of pods for a period of 4-6 days after windrowing to produce kernels free from aflatoxins (A'Brook, 1963). Research on post-harvest handling showed that dried pods have lower levels of aflatoxins than pods that were not dried. Both windrow and mat drying of pods are cost effective for controlling damage/ moulding and subsequent aflatoxin contamination (Attah *et al.*, 2007). Drying practices such as windrowing and immediate stripping with mat drying are cost effective for managing aflatoxin accumulation to within acceptable levels (Richard, 2000). Early uprooting, direct stripping, rapid drying, and cleaning of extraneous matter after harvesting at the appropriate time are necessary to reduce aflatoxin production in storage (Rahmianna *et al.*, 2007).

Use of post-harvest machinery

Post-harvest machines, such as threshers, dryers and shellers, help to increase yield and reduce post-harvest processing and drying time. As a result, they are often associated with decreased aflatoxin contamination in groundnuts (Paramawati *et al.*, 2006).

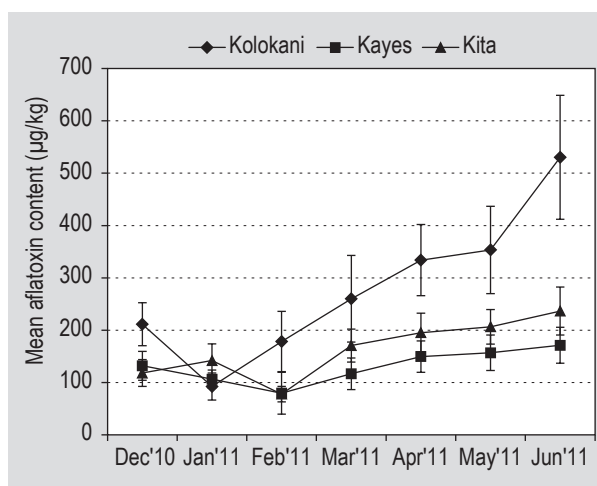


Figure 3. Mean aflatoxin B₁ (AFB₁) levels in groundnut paste in three districts of Mali, West Africa, as measured in samples obtained from small retailers over 7 months during 2010 and 2011.

Storage methods

In Africa, smallholder farmers traditionally store groundnuts in containers that are usually made of wood, bamboo, thatch, or mud placed on raised platforms and covered with thatch or metal roofing sheets. New storage practices, such as use of metal or cement bins offer an improvement over traditional storage methods. However, high cost and access to improved materials remain major constraints for their adoption by small-scale farmers (Hell and Mutegi, 2011). Polypropylene bags are now being used, but because these are not airtight, groundnut pods are still susceptible to fungal and aflatoxin contamination (Hell *et al.*, 2000; Udoh *et al.*, 2000). A major precaution in bag storage is to ensure that bags are clean when reusing them, especially when used for maize, rice, sorghum, beans, or cocoa. This is because reused bags often contain *A. flavus* spores (Awuah and Kpodo, 1996; Hell *et al.*, 2000). Grain moisture content, mould growth, aflatoxins, and free fatty acid content were significantly higher in pods stored in jute bags than in those stored in polyethylene-doubled jute bags (Bulaong and Dharmaputra, 2002). The use of hermetic triple-layer bags (PICS, Purdue Improved Crop Storage) for grain storage of several crops is gaining favour given their advantages over traditional storage devices (Hell *et al.*, 2010; Murdock *et al.*, 2003). These triple-layer bags are now being marketed in Africa (Ben *et al.*, 2009). Hermetic packaging could protect groundnuts from moulds and aflatoxin contamination (Paramawati *et al.*, 2006). Preliminary studies have indicated the efficacy of hermetic storage of groundnut pods using triple-layer polyethylene bags in minimising aflatoxins (H. Sudini, unpublished data). Use of small-scale metal silos also results in better-quality grains and less pesticide usage. Better storage allows farmers to set higher prices, particularly when grain is sold during the off-season, and directly contributes to income, rural development, and poverty reduction in SSA (Kimatu *et al.*, 2012).

Use of desiccants in storage

The use of desiccants to prolong groundnut seed viability during storage is also a good practice. Calcium chloride (CaCl₂) and silica gel are the most commonly used desiccants that help to maintain low seed moisture content, lower sugar content, and enhance seed germination. Other beneficial effects include increased field emergence and pod yield of the ensuing crop (Basave Gowda and Nanja Reddy, 2008).

Storage conditions

Moisture and temperature are the main factors that influence post-harvest contamination of stored commodities by *A. flavus* (Hell and Mutegi, 2011). Because groundnut is an oilseed crop and hygroscopic, the seeds absorb moisture

from the surrounding storage environment and lose viability (Ramamoorthy and Karivaratharaju, 1986). Kernel moisture of 7.5%, temperature of 10 °C, and relative humidity of 65% are optimal bulk storage conditions for groundnut, allowing storage of up to 1 year (Pattee and Young, 1982). Groundnut moisture content of >10% should be avoided to prevent mould growth (Diener and Davis, 1977). Awuah and Ellis (2002) reported that groundnuts dried to 6.6% moisture levels are free of fungi for 6 months regardless of the storage protectant used. These safe moisture levels are applicable to both unshelled and shelled groundnuts. The maximum moisture content for storage of unshelled groundnuts is 9%, higher than that for shelled groundnuts (7%). At these moisture levels, if the relative humidity is maintained at 70% and temperature at 25-27 °C, groundnuts can be stored for 1 year (Odogola, 1994; Waliyar *et al.*, 2007, 2008). Community-based intervention studies in Guinea, West Africa, with an emphasis on proper drying and storage conditions, achieved a significant reduction in mean aflatoxin levels in the villages where the intervention occurred (Turner *et al.*, 2005). It is also essential to maintain low moisture levels during storage, transportation, and sales by avoiding other moisture sources such as leaking roofs and condensation arising from inadequate ventilation (Wagacha and Muthomi, 2008). Biological activity during storage should be minimised to preserve grain quality by adequate drying to <10% moisture, elimination of insect activity (which increases moisture content through condensation of moisture resulting from respiration), low temperature, and inert atmosphere (Lanyasunya *et al.*, 2005; Turner *et al.*, 2005). Storing dry pods in airy, dry, and clean rooms reduces aflatoxin accumulation (Rahmianna *et al.*, 2007). Sanitation by cleaning storage units prior to loading new produce can result in reduced aflatoxin levels in grains (Hell *et al.*, 2000).

Physical separation

Kaaya and Warren (2005) showed that uprooting groundnuts with hand hoes results in considerable damage to shells and kernels, thus predisposing them to fungal infection during storage. Approximately 80% of aflatoxin contamination can be attributed to small, shrivelled seeds (Davidson *et al.*, 1982), mouldy and stained seeds (Fandohan *et al.*, 2005; Turner *et al.*, 2005), and damaged seeds (Hamid, 1997). Hence, sorting of kernels to remove discoloured or damaged/shrivelled pods is often recommended to minimise aflatoxin levels (Afolabi *et al.*, 2006; Awuah and Kpodo, 1996; Fandohan *et al.*, 2005; Park, 2002). Low-quality groundnuts have higher aflatoxin levels than high-quality groundnuts (Mutegi *et al.*, 2007). Sorting can be done by physical characteristics (colour, size, density) and by near-infrared reflectance (DeMello and Scussel, 2009). Floating and density segregation also reduces aflatoxins in storage units; kernels that float in tap water contain up to 95% aflatoxins (Kirskey *et al.*, 1989; Phillips *et al.*,

1994). Electronic sorting and hand-picking methods are also practiced to bring down aflatoxin levels significantly in shelled groundnuts (Dickens and Whitaker, 1975). Advances in sorting technologies, for example, infrared and UV sorting coupled with colour-detection technology, are now available to enable inspection of aflatoxin-contaminated products on a large scale (Womack *et al.*, 2014).

Smoking and chemical fumigation

Moisture content of grains can be reduced and mould infestation thereby minimised effectively by smoking grains during storage (Hell and Mutegi, 2011). The efficacy of smoking in reducing aflatoxin contamination of grains in farm storage was reported by Udoh *et al.* (2000). Ethylene oxide and methyl bromide are commonly used chemical fumigants that significantly reduce toxigenic moulds (Bankole *et al.*, 1996). However, some of the common fumigants also have adverse effects on human health. Some evidence has shown that inhalation exposure to ethylene oxide can increase the rate of miscarriage in female agricultural workers (ATSDR, 1990). Methyl bromide has also been phased out of many agricultural processes because of health concerns.

Antifungal compounds

Spray application of chemicals onto freshly harvested groundnut pods under field conditions reduces *A. flavus* invasion and aflatoxin contamination in kernels during storage (Bell and Doupnik Junior, 1971, 1972). Spraying 5% sodium ortho-phenylphenate (SOP) solution on moist in-shell groundnuts under field conditions and in bags effectively controlled the external fungal growth. However, SOP application was not effective for reducing aflatoxin contamination because it could not penetrate into the kernels (Fonseca *et al.*, 1992, 1994). Spraying antifungal materials from natural sources and chemical preservatives is a viable practice to prevent post-harvest aflatoxin contamination in groundnuts (Haciseferogullary *et al.*, 2005; Onyeagba *et al.*, 2004). Popular plant derivatives, such as cinnamon and clove oils, have shown significant inhibitory effects on growth and toxin production of *A. flavus* under experimental conditions (Bullerman *et al.*, 1977). Application of eugenol, the main antifungal active compound of clove, can be expensive. However, methyleugenol (4-allyl-1,2-dimethoxybenzene) can be a cost-effective derivative of eugenol and can be applied for post-harvest protection of groundnut pods and kernels from *A. flavus* and aflatoxin contamination when sprayed at 0.5% concentration (Sudhakar *et al.*, 2009).

Inactivation and detoxification of aflatoxins

Aflatoxins can be inactivated by physical methods, including cooking, roasting, frying, spray drying, and baking. Irradiation of unrefined groundnut oils by UV light destroys up to 85% of aflatoxins in 18 h and 40% of aflatoxins in 2 h (Choudhary and Kumari, 2010). Bright sunlight and gas-filled tungsten lamps also destroy aflatoxins in unrefined groundnut oils (Santha and Sreenivasamurthy, 1977).

Chemical detoxification by ozonation at various concentrations, temperatures, and times of exposure is also effective. AFB₁ was reduced in groundnuts by 77% with 10 min of ozonation at 75 °C (Proctor *et al.*, 2004). Ammonization also decontaminates produce containing aflatoxin when exposed for a long time at high temperature and high pressure (Gomaa *et al.*, 1997). Sodium bisulphite (Doyle and Marth, 1978), potassium bisulphite (Doyle *et al.*, 1982), and sodium chloride (Scott, 1984) are also reported to be effective chemical detoxifiers.

Filtration technique

Aflatoxins in oil can easily be separated by filtration. A single filtration can eliminate up to 90% of toxin from crude groundnut oil (Choudhary and Kumari, 2010). Basappa and Sreenivasamurthy (1979) developed a filter-pad system that can be adopted in oil mills to remove aflatoxins from crude oil.

5. Management options for post-harvest aflatoxin contamination in Mali

Community-based approaches through adoption of post-harvest intervention measures can be good options for minimising exposure to aflatoxins. Identification of low-cost technologies for post-harvest management of aflatoxins is a pre-requisite for such interventions. Use of low-technology approaches at the subsistence-farm level in SSA can substantially reduce the disease caused by aflatoxin exposure and its carcinogenic effects. Some of the important factors that lead to high aflatoxin risk in Africa are (1) lack of political commitment to mycotoxin research; (2) shortage of trained personnel and infrastructure; (3) limited awareness of both smallholder farmers and consumers on the negative effects of aflatoxins and the available strategies and technologies for mitigating aflatoxin contamination; and (4) prevailing climatic conditions (Hell and Mutegi, 2011). Possible intervention strategies for mycotoxin management in Africa can be broadly categorised into: (1) prevention of exposure to toxins; (2) decontamination; and (3) continuous surveillance and monitoring of moulds in contaminated food/feed. Preventive measures include the adoption of certain good agricultural practices (GAPs) in the field, during harvesting, storage, transportation, marketing, processing, and legislation etc. The GAPs

include early harvesting, proper drying of produce, physical separation, sanitation, proper storage, insect management, use of biocontrol agents, appropriate use of pesticides, decontaminants, and resistance breeding, etc. (Wagacha and Muthomi, 2008).

6. Conclusions

Not all GAPs for aflatoxin reduction are feasible for adoption at the farmer/trader level. Some GAPs suggested for post-harvest aflatoxin management that are feasible at the farmer/trader level in Mali are (1) lowering moisture content during storage (to $\leq 8\%$ moisture level); (2) adding preservatives to prevent insect infestation and fungal contamination during storage; (3) sorting of contaminated pods and kernels; (4) redrying of groundnut pods and kernels; (5) appropriate storage conditions to avoid favourable conditions for mould growth; (6) avoidance of rehumidification of pods; (7) detoxification of contaminated products; and (8) enhancing awareness of smallholders about available technologies and more broadly about aflatoxin contamination. Instead of relying on one or only a few options for reducing post-harvest aflatoxin contamination, conjunctive use of sustainable and cost-effective methods will be more effective for curbing the problem.

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