



# FEED THE FUTURE

The U.S. Government's Global Hunger & Food Security Initiative

## The Relationship Between Aflatoxin Exposure and Chronic Malnutrition in Nampula Province, Mozambique

### Webinar Transcript

#### Devyn Andrews

Good morning afternoon and evening, thank you all for joining today's webinar: The Relationship Between Aflatoxin Exposure and Chronic Malnutrition in Nampula Province, Mozambique. My name is Devyn Andrews and I'm the Communications Specialist for the Feed the Future Innovation Lab for Nutrition and I'll be your MC today. As more attendees are joining the webinar, I will begin by going over some of the housekeeping items.

I would like to direct all attendees to a few functions on this Zoom call. At the bottom of your screen, you should see a chat icon and a Q&A icon. Please use the chat feature to engage in relevant conversation with the other attendees. If you have a question for one of the panelists, please use the Q&A feature. Panelists will respond to questions in the Q&A as they're able. We have allotted the final 15 minutes of this webinar for Q&A, at which point the panelists will respond to any remaining questions from the audience. If you are experiencing technical difficulties, send a message in the chat to the panelists so that our technical support staff can work with you to help resolve them.

This webinar is being recorded and will be made available on the Innovation Lab for Nutrition and the USAID Advancing Nutrition websites. You can view previous recordings and slide decks on our websites. We will repeat these technical housekeeping items in the chat throughout the webinar as people may be joining in at later times. Before I introduce today's moderator, Shibani Ghosh, I'll give a brief introduction to the Nutrition Innovation Lab and our webinar series. We are a Feed the Future Innovation Lab supported by USAID Bureau of Resilience and Food Security, and we are active in supporting research and capacity building to build the evidence base around critical questions linked to agriculture, nutrition, and health.

As you can see from this map, we are active in both sub-Saharan Africa and South Asia, more information can be found on our website at [nutritioninnovationlab.org](http://nutritioninnovationlab.org). The Innovation Lab is a consortium led by the Freidman School of Nutrition Science and Policy at Tufts University. With U.S. university partners, including Purdue, Harvard Th Chan School of Public Health, Johns Hopkins School of Public Health, and Tuskegee University. In addition, we partner with government agencies in our host countries, UN agencies, local and international NGOs, as well as universities across the globe.

I'd now like to introduce today's moderator, Dr Shibani Ghosh. Dr Ghosh is a research associate professor at the Freidman School of Nutrition Science and Policy. She is also the associate director for the Feed the Future Innovation Lab for Nutrition, with experience working in the Middle East, sub-Saharan Africa, and South Asia. Her research interests are in understanding the role of agriculture and improving nutrition while ensuring health, assessing the diet and non-diet determinants of nutritional status of infants and young children, and testing interventions aimed at improving maternal and infant nutrition and growth. Dr Ghosh, over to you.

## Shibani Ghosh

Thank you, Devyn. And good morning, afternoon, and evening everybody joining from all over the world. I'm excited to be on this webinar and to introduce the illustrious group that we have here for presenting the findings of our study around aflatoxin exposure and chronic malnutrition in Nampula Province, Mozambique.

First and foremost, I'd like to introduce Maureen Malave, who is the nutrition team lead at USAID Mission in Mozambique, where she manages a multi sectoral nutritional portfolio focused on working with the government of Mozambique on specifically targeting childhood stunting. Maureen has been a critical part of the development, implementation, and dissemination of the aflatoxin study in Mozambique. Maureen has a master's in public health in a population and family health and forced migration from the Columbia University's Mailman School, and a BA in political science and Spanish from Villanova University.

Our next speaker is going to be Sofia Costa, who is the director of the Center for Studies and Health Services at the University of Lurio in Nampula Province, where she is also a lecturer and a researcher at the Faculty of Health Sciences. Sofia has completed her postgraduate studies in pediatric nutrition and is a nutritionist as well as the local co-principal investigator of our aflatoxin study in Mozambique.

Our next speaker will be Katie Appel, who is an assistant researcher for the Feed the Future Innovation Lab for Nutrition. Katie has an MS in Food Policy and Applied Nutrition from the Friedman School of Nutrition Science and Policy and has been with the Innovation Lab for Nutrition since 2016.

And finally co-moderating the Q&A session with me is Joao Salavessa, who is an associate professor at the University of Lurio, and also a coordinator of the large project of the African Development Bank at the University of Lurio, which focuses on skills development for agriculture and industry. Joao holds a PhD in food science and technology from the Universidade Técnica de Lisboa in Portugal, and he is the local principal investigator of this aflatoxin study that was conducted in Mozambique.

I'm really looking forward to the presentations that will be made by Sofia and Katie and followed by, I'm hoping, a very active Q&A session with all of you on this webinar. I'm going to hand this over to Sofia, excuse me, over to Maureen, who will give some opening remarks around the importance of the work that has been done in Mozambique around aflatoxins and malnutrition. Over to you, Maureen.

## Maureen Malave

Thank you, Dr Ghosh. Good morning, good afternoon everyone. I am really excited to be here today and to open up this exciting webinar. On behalf of the United States agency for International Development and the Feed the Future Innovation Lab for Nutrition, I am honored to extend my welcome to so many partners from the government of Mozambique, development organizations, civil society partners, colleagues, and friends.

It's a privilege to open this extremely relevant dissemination of findings from a study we conducted in Nampula Province that evaluated the relationship between aflatoxin exposure and stunting in children six to 59 months of age. USAID Mozambique is dedicated to supporting the government and people of Mozambique and seeking a secure, to secure, safe, affordable, and nutritious food for all. On behalf of the USAID Mission in Mozambique, my sincere thanks to the representatives of the government of Mozambique, UniLurio, to the National Institutes of Health, and the Association for Food and Nutrition Security, or ANSA, for their work on this study.

Every person's right to food and health is an unalienable and fundamental provision in the Plano de ação multissetorial para a redução da desnutrição crônica in Mozambique, otherwise known as the Plano RDC. It is reinforced by policies and strategies that cut across sectors. The US Government and the

USAID Global Food Security Strategy, as a path to self-reliance, support the right for food and highlight the importance of sustainably reducing hunger, malnutrition, and poverty by achieving agricultural-lead economic growth, resilient people and systems, and better nutrition, especially amongst women and children. Within this context, a key area that cuts across nutrition, food security and economic development is food safety. Challenges to food safety impact the health and economic vitality of millions of people in low- and middle-income countries around the world.

Problems relating to food safety contribute significantly to ill health, food borne disease outbreaks, and even mortality. These are dangers of particular concern in relation to poor households, especially for pregnant women, newborns and infants under the age of two. One major concern relating to food safety globally is the threat of mycotoxins. Mycotoxins are naturally occurring molds that are toxic to humans and livestock. More than 5 billion people in developing countries are estimated to be at risk for exposure to mycotoxins. The World Health Assembly and the US Government Global Nutrition Coordination Plan have committed to a 40% reduction in stunting in infants and young children under the age of five by the year 2030. Thus, a key action to achieve the target of reduction in stunting will require the application of new knowledge around mycotoxins control into practice across all susceptible crops and animal source foods. To be effectively-- to effectively and sustainably reduce the specter of mycotoxin exposure in Mozambique, we are collectively working towards informing sound policy underpinned by robust evidence and national research capacity, as well as a coordinated mycotoxins mitigation strategy.

As well as strategies involving multi sectoral and multi-level collaborations. Understanding the challenge of abating mycotoxins within the food system is critical in achieving the goals laid out in the Plano RDC. Indeed, it is key to Mozambique's achievement of the Sustainable Development Goals, since good health and nutrition represent fundamental platforms on which so many other Sustainable Development Goals are built. In addition to presenting the evidence and data from this study, this event will launch the study report which will be made available following the webinar. I look forward to engaging in this interactive session.

And now, I would like to hand it over to Sofia Costa. Thank you.

### Sofia Costa

Thank you, Maureen. Hi everyone, so I'll be presenting the first part of the results of our study about assessing the relationship of serum aflatoxin levels and stunting in children six to 59 months of age in ten districts of Nampula Province, Mozambique. Next, please. So, the study collaborators for the study were Universidade Lurio, the National Institute of Health in Mozambique, ANSA, INE, National Institute of Statistics, Nampula Central Hospital, University of Georgia and Tufts University. Next, please.

To give some aflatoxin background. The aflatoxin is a type of mycotoxin. It's produced by the mold aspergillus. Its colorless and odorless, although the mold is green, has a green color. But the aflatoxin itself has no color or odor. It contaminates staple crops such as maize, ground nuts, and cassava. It causes liver cancer and may be associated with stunting. The aflatoxin also may impair absorption of nutrients that are critical for a normal child growth and development. Next slide.

And as a research theme of neglected biological mechanisms, to contribute to a better understanding of the mycotoxin and stunting relationship. The Feed the Future Nutrition Innovation Lab has implemented several studies exploring the relationship to mycotoxins, with a focus on early life nutrition. This particular study is one of them, the aflatoxins study, the Gulu Cohort about HIV, food security in pregnancy in northern Uganda, The Birth Cohort Uganda, also in the north and south west Uganda, the study about aflatoxin in pregnancy and birth outcomes in, also in Uganda, and the aflacohort study that took place in Nepal. Next slide. And, to give some rationale about all these, the relationship about aflatoxin and growth and children, there are several studies that show some relations. So, there is a significant negative association between aflatoxin exposure and linear growth that was found in Benin

and Togo. Also, there's a study that shown that the presence of aflatoxin B1 in pregnancy in Ghana was associated with adverse birth outcomes. Also, in Uganda the aflatoxin levels in pregnancy were associated with adverse birth outcomes, and there was a study at the end birth outcomes, even at low levels of aflatoxin. Next slide.

Relationships were also observed in sub-Saharan Africa and South Asia. It was found, variability of the relationship in different populations, specifically in pregnancy in under twos and in under five years old. There was some conflicting evidence of the relationship in Nepal, and RCT on maize grain replacement in Kenya found that impact was founded primarily on younger children. Next slide.

About the studies specifically happening in Nampula. Nampula is known by having a high rate of stunting, so 55% of children under five years old are stunted in Nampula Province. The population is highly dependent on crops such as maize and groundnuts, and there were, it was done a previous assessment of crops, and soil levels in Nampula. And it was found that it was found aflatoxin contamination. Next slide.

About the study objectives. We intend to assess the mean serum aflatoxin in children in these two different ages, six to 23 months and 24 to 59 months of age. We wanted to examine the differences in serum aflatoxin mean by age group and also to enumerate the association between serum aflatoxin and linear growth to the Z score of height for age, adjusting for confounders. Next slide please.

About the methodology. It was a cross-sectional study with two groups defined, under two and two to five years old. We calculated the sample of 72 children per age group, plus 25% of attrition. It was the study was located in 10 districts of the Feed the Future zone of influence that are the green districts that you can see in the map. The sampling strategy. We... the population was proportional by districts and the enumeration areas within each district were selected randomly according to the 2017 census that the National institution, Institute of Statistics gave us. Inside each enumeration area, the households were also randomly selected. The study is representative of children aged six to 59 months of age, in the ten districts where the study took place. Next slide please.

About the ethical approval, sensitization and mobilization. The study was approved by UniLurio Bioethics Board and by the National Bio-ethics committee from Mozambique. And by the Institutional Review Board at Tufts University. The sensitization of Central Hospital and Provincial Department of Health and district officials, offices from all the ten districts took place different ways. About the provincial level, we had one on one interactions. Then we did visits to the 10 districts, and we did a mapping of the health facilities on each district previously to the study collection, to the data collection. Then right before the data collection, we had a launch event where, to which we invited representatives from each district. From the health departments of each district, and also from the Community radios of each district to support spreading the information that this study was going to take place in this districts. And also, we had a mobilization team that was sent in advance to, data collection team that intended to inform the population that this data was going to be collected and explain the reasons of the study. For that, when the data collection team arrives, they already have this information.

At data collection we undertake all the process of informed consent, and the caregivers were requested to participate in the study. Next slide please.

The household data collection took place at the household, and we had two main respondents for the questionnaire. The female caregiver, or the mother of selected child or children, and household head which usually is the husband. There were specific models for each of these respondents. For the female caregiver or the mother, we, the topics that were destined to for her to answer was the household identification and household member information, diet of caregiver and child or children, and the household food security, child feeding practices and health, water, hygiene and sanitation, and also social participation. For the household head, we had, they were supposed to answer the topics of agricultural production, socioeconomic characteristics, and access to durable goods. Next slide please.

Regarding the nutritional and biomarker data collection at the health facility. We had exclusion criteria, we did not enroll in the study children that were Severe Acute, that had Severe Acute Malnutrition, that we defined with the middle upper arm circumference below 11.5 centimeters. So, these children were not enrolled in the study. Children with severe anemia, with a hemoglobin below seven grams per deciliter did not have blood drawn. For the data collection about anthropometry. We collected weight, length or height, the MUAC, head circumference and knee to heel length. Also, we, the child had one venous blood draw by the technicians, by the Central Hospital technicians that were at the districts due to this activity, this data collection. And anemia assessment was done through the HemoCue 301 analyzer, and malaria was diagnosed with a rapid diagnostic test. We were, about referrals. So, we referred the children with Severe Acute Malnutrition, with anemia, and that were that were also diagnosed with malaria, to the health facility staff and the treatment was provided at the health facility. That was where all this anthropometry and blood collection took place. About some biomarker processing. The samples were processed and handled by the Central Hospital of Nampula technicians. They all, the samples were stored in the freezer in Nampula Central Hospital at minus 80 degrees Celsius, Celsius degrees. And then were transported to the United States. The serum aflatoxin was analyzed using HPLC through validated lab protocols, and this was done at University of Georgia in the United States. Next slide.

About data management calculations and analysis. Data were collected electronically, and then were transferred to a database at a Tufts server and cleaned daily. Data cleaning and sample weight computation were done. The variable generation and computation, we used anthropometric Z-scores computed using that WHO reference standards. The prevalence of stunting and underweight was determined. Also, we determined diet diversity, household food insecurity, and agricultural production and practices. Statistical analysis included descriptive statistics, bi-variate analysis, multi-variable analysis, ordinary least square and logistic regressions, and it was adjusted for sample weights and clustering. That was enumeration areas, the clusters. And to make findings, to make the findings representative of the children in the 10 districts where the study took place. Next.

About the study timeline. In 2017, in May took place the scoping visit and in November, the protocol development. In 2018 in February, it was the Tufts IRB submission. In March that we had some partner meetings and local IRB submission. In July, the health center identification at district levels and the cold chain assessment. In August, we had a supervisor training and the pretest, in October, the enumeration training and the launch event, and right after in November and December took place the data collection in all the 10 districts. In 2019 in March, the serum sample were, the serum samples were shipped to the United States. March to July, data cleaning took place. In March to August aflatoxin analysis took place. And in August we had the aflatoxin results. Next slide please.

About the sample size. We had enrolled and with household surveys completed, we had 493 children aged six to 23 months and 764 children 24 to 59 months, making a total of 1257. We had anthropometric measurements done in 419 children six to 23 months, 656 24 to 59 months, with a total of 1075. We had blood drawn from children, 317 children six to 23 months, 599 children 24 to 59 months, making a total of 916 blood, children with blood drawn. So, we had a total of complete records of 894. In which 311 were from children six to 23 months and 583 were from children 24 to 59 months of age. Next slide.

Okay, so now I'm passing to Katie to present the results, thank you.

[Katie Appel](#)

Thanks Sofia. So now I'll present what we found, including descriptive statistics and the results from linear and logistic regressions. Next slide.

About 90% of children in our study had a detectable level of aflatoxin in their serum sample. And, as you can see from this histogram, aflatoxin levels were highly skewed as is typical. The mean aflatoxin level was 7.7 picograms per milligram albumin, and due to the skewed nature of the data we calculated a geometric mean of 2.2 picograms per milligram albumin. Next slide.

As you can see from this table, the mean aflatoxin levels and rate of detectability that we found in the Mozambique study were similar to data from other studies conducted by the by the Innovation Lab, especially the study in Uganda. Next slide.

The first two objectives of the study were to assess the mean aflatoxin level of each age group and examine the difference in mean levels between the age groups. As we hypothesized, we found that children in the older age group had higher levels of aflatoxin than the younger age group. Children six to 23 months of age had a mean aflatoxin level of .24, while the mean level was .37 for children 24 to 59 months of age. The means were statistically significantly different, with the p value of .03. Next slide.

The prevalence of stunting was high in our sample, especially for children in the older age group, with over 50% of them being stunted. The prevalences we found were quite similar to those reported in the DHS and other reports from Nampula, and about 15% of children were underweight. We didn't report wasting as children with SAM were excluded from enrolling in the study. Next slide.

The prevalence of anemia and malaria were also quite high. About 80% of children in the study had anemia, with almost half having moderate anemia. More children in the younger age group had anemia than the older age group, and the younger children had more severe cases of anemia. Overall, 67% of children had malaria, with higher rates in the 24- to 59-month-old children. Next slide.

Looking at the diet, we found that less than 30% of caregivers met the minimum dietary diversity for women indicator. And almost half of children consumed four or more food groups to meet the infant and young child feeding minimum dietary diversity indicator. When it came to foods prone to aflatoxin contamination, 24-hour diet recalls found that about 35% of children consume maize, with little difference between observed between age groups.

A quarter of children consume groundnuts, with 17% of children 6 to 23 months of age and 27% of children 24 to 59 months of age. Consumption of cassava was most common, with two thirds of children consuming it. It also had the largest difference between age groups, where about half of children and the younger group consumed and three quarters of children in the older age group consumed cassava. Next slide.

At the household level about half of study households produce maize, and two thirds produce groundnuts. Next slide.

And finally, using the household food insecurity access scale, we found that less than a quarter of households were food secure, about 10% experiencing mild food insecurity, a quarter experiencing moderate food insecurity, and 44% of households were severely food insecure. Next slide.

The third objective of the study was to enumerate the association between aflatoxin and linear growth or stunting. In the entire sample, we found a significant relationship between stunting and aflatoxin, where a child was 60% more likely to be stunted with each unit increase in aflatoxin level. We adjusted for confounders including anemia and weight for height Z score, which accounted for a nutritional status of the child. The units of aflatoxin use in these models is log transformed aflatoxin, divided by a child's body weight in kilograms. We adjusted aflatoxin levels by body weight, because of their complex relationship in children, which I'll talk about more in a minute. Next slide.

The linear regressions also showed a significant and negative relationship between height or length for age Z score and aflatoxin, indicating that higher aflatoxin levels were associated with lower HAZ. Next slide.

Both logistic and linear regressions were also run on individual age groups, resulting in similar and significant findings in children 24 to 59 months of age. Models including only children in the younger age group were not significant, and this could be due to the fact that we did not meet our sample size, especially falling short in the children six to 23 months of age. Next slide.

As I mentioned before, the variable we used for aflatoxin exposure in the previous models were adjusted by child's body weight. And we did this because we observed different relationships between HAZ and aflatoxin in children with different weights. We explored these relationships by predicting the HAZ aflatoxin relationship in the entire study sample at three weights: high, mean, and low. Where high and low are two standard deviations above and below the mean weight of all children in the study respectively. The figure on the slide shows the predicted relationship of HAZ and aflatoxin at each weight and shows a positive association for children with a high weight, no relationship for children at a mean weight, and a negative relationship for children at low weights. Next slide.

Digging a little further, we also found that these relationships change depending on the age of the child. We ran the same predictions with an added specification for age, resulting in the figures on the slide. At six months of age, children with high, mean, and low weights all have positive associations between HAZ and aflatoxin. At 24 months, we see the same pattern as observed in the overall study. And at 59 months, the slopes of all lines become steeper and the association of HAZ and aflatoxin becomes negative for children with mean weight. From these figures, we can see that the relationship between HAZ and aflatoxin does not change much for children at high weights across the ages. However, the mean and low weights shift from a positive to negative association as the child gets older. Next slide.

Now we will look into some of the factors associated with aflatoxin levels. First is the diet. As mentioned before, the population in Nampula is highly dependent on maize, groundnuts, and cassava. The bar graph on the screen shows the percentage of children in the study who have consumed these foods in the past 24 hours. So, you can see that consumption of cassava is much more prominent in our study sample than maize or groundnuts. I do want to note, however, that these patterns vary by district, and some districts relied more heavily on maize than cassava. In relation to aflatoxin exposure, we found that overall consumption of ground nuts and cassava were significantly associated with increased aflatoxin levels. Only groundnut consumption by the older children was significantly associated with increased aflatoxin levels when the models were conducted on separate age groups. We did not find a significant relationship between aflatoxin exposure and maize consumption. Next slide.

Next, we looked at agriculture practices. Post-harvest practices around grain drying and storage are known to impact grain quality, and therefore potential mold development. Here, we will focus on two aspects of grain drying, the location of drying and the type of method used. I also want to note that we collected a substantial amount of information on storage practices; however, we did not see any significant relationships with the storage practices and child aflatoxin exposure. Regarding location, farmers dry their grain in the field after harvest, after bringing it in from the field, or both. And FAO technical brief on seed and grain storage systems emphasizes that grain must be dried rapidly, taking care to cover it if it rains, and to avoid over exposure to the sun. We hypothesize that grain dried only in the field does not meet these criteria, and therefore leads to greater risk for aflatoxin contamination. Concerning methods, there are many common methods used to dry crops and we categorized them into improved and unimproved groups. Methods in the improved category included using fans, placing grain on platforms or plastic sheets, or hanging grain under the roof or in the kitchen. These methods dry crops quickly, reduce the risk of infestation and moisture damage, and do not have high labor requirements. Unimproved methods include drying only in the field, spreading grain directly on dirt, cement, or brick floors, or drying grain on the roof. The bar graph here shows that these drying practices, describes these drying practices for maize in groundnut producers in the study.

A little over a third of the maize producers dried their crop out of the field, while two thirds of groundnut producers dried their crop out of the field. Only around 15% of maize or groundnut producers used improved drying practices. Next slide.

So, how did these drying practices relate to aflatoxin levels. Linear regressions found that children in maize producing households had lower aflatoxin levels if the household dried the maize out of the field, compared to only drying in the field. A relationship of similar magnitude was found with drying method, where children in households using improved drying practices had significantly lower aflatoxin levels than those in households that did not use improved drying practices. In addition to other covariates, these models were adjusted for intercropping, as it is known to reduce the risk of brain damage due to pest infestation. Next slide.

Similarly to maize, we found a significant relationship between aflatoxin levels and improved ground nut drying practices. Children in groundnut producing households that used improved drying practices had lower aflatoxin levels compared to children and households that did not use improved drying practices. We did not find a significant relationship between aflatoxin levels and groundnut drying location. Next slide.

In conclusion, we found ubiquitous exposure to aflatoxin in the study sample, as well as high prevalences of stunting, anemia, and malaria. We found that children are more likely to be stunted if they had high aflatoxin levels, and that relationship was stronger in the older age group. We also found that the relationship between aflatoxin and HAZ is complex, as it changes depending on the child's weight and age. Factors significantly associated with increased aflatoxin levels in children, included groundnut and cassava consumption, as well as poor maize and groundnut drying practices. Next slide.

Like any study we had some strengths and limitations. First, the study was designed in a way to be representative of all children under five in the 10 study districts in Nampula, and we used robust tools for data collection, anthropometry measurements, as well as collection of blood samples and aflatoxin analysis. Some limitations to our study include the inability to meet the sample size because of difficulties experienced during data collection. This could in turn be preventing us from finding significant associations between aflatoxin and HAZ or stunting in the younger age group. Finally, we did not collect data on agricultural practices related to cassava, due to overall questionnaire length and existing literature that reported low levels of aflatoxin in cassava. After seeing the strong association between serum aflatoxin levels and cassava consumption, it's important to understand the agricultural practices that could mitigate this relationship. Next slide.

From this study, a few future research areas have been identified. First, there are several ongoing initiatives targeting aflatoxin contamination in different crops in Mozambique. Agricultural and post-harvest interventions and policies that target aflatoxins may want to specifically consider health effects upon vulnerable populations, such as infants and young children. A better understanding of the exposure to aflatoxin in urban areas is needed, as our study focused primarily on rural areas. This includes examining the source of dietary exposure in the way of home production or market purchases. The study supports the need for enhanced multi sectoral collaboration between agriculture, health and nutrition sectors to mitigate the effects of aflatoxin and improve the health and wellbeing of vulnerable populations. And as I mentioned in the limitations of the study, the association between cassava and aflatoxin levels was not fully examined. We found a relationship with dietary consumption of cassava, but the link between cassava agricultural practices and serum aflatoxin levels warrants further investigation. Next slide.

And finally, we want to take this opportunity to let everyone know that our study report is finalized. Versions in English and Portuguese are available to download from our website. Thank you very much.



## Shibani Ghosh

Thank you, Sofia and Katie. I think there's been, I hope all our participants are looking at the Q&A box as well as the chat box. There's a lot of active interaction going on. And I do know that some of our panelists are responding to the questions as they have been coming along. So, I'd like to take this opportunity to also point out on a couple of things, that the study was very much a cross-sectional assessment of understanding what the situation is within the context of these 10 districts in Nampula Province, which have been the focus of USAID programs through the Feed the Future initiative. And so that's why we are in those, we were in those 10 districts of Nampula Province. And I think I want to sort of point out another point that Katie had made was that there wasn't a relationship or an association with storage practices. And often it could be because there isn't very much variability in the data that you do not see those relationships emerge and not because storage, post-harvest storage, isn't critical or important within the context of aflatoxin mitigation.

So, I think what I'm going to go forward now is to go through some of the Q&As and asks Joao, Sofia, and Katie to put on their video screens. I think we're set up in a way that doesn't allow us to all be in the panel at the, on the screen at the same time, but I request them to put their screens on. And I think the first question, I want to put it out to Katie and Sofia and ask you all, if you could respond to this. First of all, what could be the mechanisms of aflatoxin and chronic malnutrition? So, I don't know, Katie if you want to take a stab at that, or Sofia.

If you...

## Katie Appel

Sofia, do you want to take a stab first?

## Sofia Costa

No, you can go ahead, Katie. Thank you.

## Katie Appel

Umm. Honestly, I'm kind of drawing a blank.

## Shibani Ghosh

Okay, so I think maybe what I could say is that, from my understanding and for the colleague who wrote this request, asked this question. That there is an article that has been published by another colleague who works on aflatoxin around the proposed mechanism, the biological mechanism. And it's got to do with the gastrointestinal system and linked to even environmental enteric dysfunction. So, I believe the article is from Johns Hopkins University, and we're happy to share that link on our website. So, I think that might help understand that there is a proposed mechanism by which aflatoxin affects nutritional status and nutrient uptake in the gastrointestinal tract. But that isn't really, there isn't exactly a proven mechanism by which aflatoxin could affect nutritional status, and in this case, chronic malnutrition.

So, I think the question I would like to ask the... to Katie maybe, were there any reference ranges for anemia and, so how did you assess the data on anemia? Was there a connection between aflatoxin and anemia as well?

Katie Appel

I don't believe we did see an association, a significant association between aflatoxin and anemia levels. And we categorized the children into the WHO groups of non-anemia, mild, moderate, and severe based on the WHO groups.

Shibani Ghosh

Okay, and then a question for... around the maize and groundnuts. Did we collect groundnut samples and maize samples, did we analyze those? And if so, what did we find.

Sofia Costa

So, we didn't-- can I?

Shibani Ghosh

Yes, please Sofia.

Sofia Costa

We didn't analyze the foods itself, we analyzed just blood from the children. but we know from previous studies that these crops are usually contaminated by, with aflatoxin. So, we just analyzed the ingestion of these foods in the 24-hour recall and blood from the children. And correlated it, right. But no, we didn't analyze any foods. And maybe I'll pass it to Joao to add something.

Shibani Ghosh

Yes, please.

Joao Salavessa

We know that the presence of aflatoxins came from the mold development, and this development of the mold, it usually in the process of drying foods. So, when foods, when the crops are not efficiently dried, they start developing the mold. And if good practices are not used to dry the crops, the increase in the quantity of aflatoxins in the staples will be bigger. And for sure, as long as time goes by, gets older, all this is the crop, the probability of an increasing amount of aflatoxin presences gets bigger. Here in Mozambique, in Nampula, we have been talking about cassava. We didn't [inaudible], but the point is that here, the communities used to eat bitter cassava. And the bitter cassava, to be produced, it is ripe and the enjoy it with mold. So, we know that the content of aflatoxin will be bigger in this bitter cassava.

It is also the one that is easiest to grow, so it is also when more contribution for the contents of aflatoxin in the blood of the children. Thank you.

Shibani Ghosh

Thank you, Joao. And I think this question I'm going to pose to Katie. Katie, this is coming on specifically on what was the rationale of data collection in this particular period since it may affect aflatoxin contamination? and also, could you elaborate how you adjusted other confounding variables.

## Katie Appel

For the period is that meaning, like the time of year? Yes, okay. Most honestly, for the time of year it was when we were able to conduct a study before the rainy season started. And before the holidays, so we really... it was kind of between elections, which we wanted to avoid political turmoil in the area, and also, make it as safe as possible for the enumerators to avoid the rainy season. Because it can get quite tricky to travel around in Nampula during rainy season. And the other question was on confounders?

Yes, and so in the HAZ and aflatoxin models, we adjusted for malaria, weight for height Z-scores, age, and gender of the child. And the detectability of aflatoxin. And so, we adjusted for anemia and weight for height Z-scores as a way to represent the nutritional status of the child and adjust for that.

## Shibani Ghosh

Great. Thank you. I think the one question that I might just field is about that the case control study has its own limitation. And I agree. And it looks like you are conducting a number of studies in these areas-- is that a longitudinal study plan to further look into the relationship? So not specifically in Mozambique, but we, our studies and Uganda and Nepal have been longitudinal studies. And actually, we've presented the findings in different for as well as through webinars and some literature that is already published on the longitudinal relationships. And I think what we are finding, particularly in Nepal where we have very low levels of aflatoxin exposure in pregnancy, that that is associated with birth outcomes. And then we've done some longitudinal analysis around the relationship of aflatoxin exposure starting from three months of age to 24 months of age. So that was one of the questions that I thought I should address.

So, in turn, there is one question that that is about aflatoxin albumin adduct. We've mentioned that this is a reflection of long-term exposure to aflatoxin B1. How many 24-hour recalls did you conduct? So maybe Katie it might be good to clarify how aflatoxin albumin adduct is measured, and how that correlates to the 24 hour... the diet diversity and/or the measurement of intake metric.

## Katie Appel

Sure. Well, the aflatoxin in the blood that we measured is representative of the last like 90 days of the child's consumption. Because aflatoxin B1 has a half-life of 90 days in the blood. And so, we took serum samples and then...or, took blood samples and then centrifuged the serum and analyzed those. And we only did do one 24-hour recall. So.

## Shibani Ghosh

Yeah, so I think there are, there are clearly limitations here. I think it's very important to say that this is a cross sectional assessment, this was really very much to understand what the-- if there is a relationship within this context, within the context of what's going on in Nampula Province. And I believe that colleagues have done soil assessments just prior to the study occurring. And that the districts where we've been working in, that they have found aflatoxin contamination in the soil as well. So, there's a connection, there is a sort of pattern to these studies here. So, I think the question and maybe Katie again, this is something you might be able to answer. And so, what are the improve drying techniques practiced in this area? And I think, maybe not you so much as Joao maybe might be the right person for this, I apologize. So, Joao, do you want to take a stab at that?

Now you're on mute.

Joao Salavessa

Sorry Shibani. I was writing an answer and I didn't hear your question.

Shibani Ghosh

Oh, yeah. So, we have a question in the chat. The question is, what are the improved drying techniques practiced in this area? So do we have, in Nampula Province, what kind of improved drying techniques are practiced.

Joao Salavessa

Okay, so. I think that's, that's the main action to be taken. So, if we can train the communities in how to dry well the crops, and how they should preserve it to avoid the growth of the molds in crops, it will be for me the best approach and the most efficient one. Because, in general, we note in the, in the field, that they are not so, so many good practices during drying of crops. And they became very early contaminated. And I was answering for a colleague that asked me about the problem in the urban areas. The study was done in rural communities in the districts of the province of Nampula, but for sure, as the products go along the food chain, and if they are not preserved and, once this is a community issue, a problem that is always growing. Maybe the contents of aflatoxin in the blood of the children in the urban areas that are fed with the local crops, because in urban areas, we have a lot of the imports of maize, and maybe I want to believe that maize is controlled. And it is not so contaminated as the maize produced locally. And, of course, probably the contents in urban areas can be higher. It is something we must take care, and we can look into for further studies. That's it for now.

But I think another point is, it is for me important that communities start to diversify their diets. Because they eat always the same, the same food and the same kind of food that is contaminated. Because this is ripe food it is contaminated with aflatoxin. So, both the bad practices, drying, and then the enormous amount of contaminated food that is the base of the diet. So, for me, this is the two main points, and that's where we have to do something. Diversify the diet and simulate good practices for drying crops and for preserving. Thank you.

Shibani Ghosh

Thank you, Joao. And I think that this is something that has been raised by colleagues on the analysis that, you know, the controlling for diet diversity. But I think what we noticed was that the diet diversity was very poor, and there was an excessive reliance on maize groundnuts and cassava. And unfortunately, what we, in one of the limitations in our work was that we didn't actually collect agricultural practices on cassava because we, from our perspective, maize and groundnuts were the biggest offenders when it came to aflatoxin. So, this is another thing that is a lesson learned from our analysis, is that we should be looking at contamination in cassava as well. Because it is consumed in pretty large quantities, and in many southern African contexts.

I think one of the questions that has come up is, I feel like I should address it, in terms of the impact of body weight on the relationship of aflatoxin and stunting. The I think the norm, the norm around aflatoxins and outcomes of growth through the literature, whether it's animal nutrition or human nutrition, has been that body weight is also affected. That there is depressed body weight because of a high exposure to aflatoxin. And what we were finding is that what we're finding within the context of human nutrition is that there is, there is a lot of variability in body weight and how those relationships with height occur. And so, we are just starting to open this analytical pathway and I think it's very interesting. I don't have the answers, and I believe we've got some work to be done on why this sort of is occurring. And there is an age, so as the older versus younger children also have a differential

relationship when it comes to aflatoxin weight and height. So, something, something to follow up on. There is a question on whether they will be a long term follow up on the incidence of hepatocellular carcinoma. I think that is something that is not on the cards, but that probably is important to consider.

And I think we're running slowly out of time, I just wanted to make sure that we answer one more question, which is around aflatoxin and environmental enteropathy. And I think I'd like to say that I'm kind of going to say that we actually don't have a distinction in this study to make any comments about environmental enteropathy, but we have been looking at aflatoxin, other mycotoxins, and environmental enteropathy in a study that is being done in Nepal right now. So please keep an eye out for a webinar that will be specifically on that. On those findings. So, we've got two minutes left, and I just want to hand it back over starting with Sofia, then Katie and Joao. If you wanted to add, you know give your thoughts on where do you see the study taking us next within the context of research, but also programming around aflatoxins and nutrition, particularly stunting. So, Sofia?

If you have some final thoughts.

### Sofia Costa

Thank you. So yeah, I just want to say that there are several initiatives in Mozambique that focus on the aflatoxin, also work on them on the food, on these crops where to put it and how to deal with it after the... after bringing it in from the field. So, it would be important to work on that, on that area. Thank you.

### Katie Appel

Thanks everyone for attending and engaging in such thought-provoking discussion. I think we made it pretty clear that there's a lot more that we need to do to fully understand this relationship between aflatoxin and child growth, but you know, there are a lot of really good questions being asked. And it seems like we do have some clear actions that can be taken at least in the short term to mitigate the effects of aflatoxin on child growth. So, thank you all.

### Joao Salavessa

So, I would like to thank you all. And this is the first study we have done here about aflatoxin, and now we know it's an issue and that it is something we need to take care of. So, I think we have a lot of work to be done after this first conclusions. And let's continue searching for a better health for our communities, and for this region, thank you. Bye bye.

### Shibani Ghosh

And Maureen I don't want to put you on the spot, but I don't know if you have any 30 second thoughts?

### Maureen Malave

I just wanted to say thank you to everybody for participating in the webinar. I think this was really interesting, it's great to share this work that that we have done over the past couple years in Mozambique, and we are, through our multi sectoral nutrition programs, we work with local communities in Nampula on improved drying practices. And to educate the people who work in multisectoral about aflatoxins. So that's it and thank you all.

## Shibani Ghosh

Thank you so much, everybody, and thanks everyone I know there are some more comments and questions, but we really appreciate everybody joining us today, and thank you for the panelists. For such an engaging presentation and discussion. Thanks everybody.



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