

## Occurrence of Tomato Ringspot Virus Infecting Tomato Plants in Northern Nigeria

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### Abstract

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A field survey to identify Tomato ringspot virus (ToRSV) in the Sudan savanna region (Jigawa, Kano and Gombe) of Nigeria was conducted during the 2017 and 2018 dry seasons. Three Local Government Areas (LGAs) per State were chosen on the basis of tomato cultivation history from which 3 farms each were sampled. A total of 2160 tomato leaves were randomly sampled from all fields (n=9) and specifically tested against ToRSV by the TAS-ELISA technique. The results obtained established the level of spread of ToRSV disease in the three states surveyed. The Kaltungo LGA of Gombe State recorded significantly ( $P \leq 0.05$ ) the highest ToRSV incidence (29%), followed by Akko LGA (23.5%), whereas the least virus incidence (17.5%) was recorded in Yamaltu-Deba LGA. In Jigawa State, the highest virus incidence of 19.5% was recorded at Kazaure LGA, followed by Kirikasama LGA (16.6%), whereas the Hadejia LGA had the least incidence of 10.4%. In Kano State, ToRSV incidence was highest (31.1%) in Kura LGA, followed by Bagwai LGA (21.1%), whereas Garun Mallam LGA recorded the lowest virus incidence (16%). This is the first report of ToRSV on tomato in Nigeria. Informing tomato farmers on the effective management of this yield-limiting virus in the region is essential.

**Keywords:** Detection, distribution, Nigeria, Tomato ringspot virus, TAS-ELISA.

### Introduction

Nematode-transmitted polyhedral viruses (Nepoviruses) consist of about 46 viruses that cause economically important diseases in annual, perennial, and woody cultivated plants (Šneideris *et al.*, 2012). The Tomato ringspot virus (ToRSV, genus: *Nepovirus*, family: *Comoviridae*) is reported to naturally infect about 285 plant species in 159 genera belonging to 55 families (Edwardson & Christie, 1997; OEPP/EPPO, 2018), causing diseases of economic importance (Šneideris *et al.*, 2012) including field-grown tomato as by Hajiabadi *et al.* (2012).

ToRSV can be transmitted through vegetative propagation, sap inoculation, seeds and pollen (CABI, 2022; PEPP/EPPO, 2016; 2018; Sastry, 2013). Furthermore, its transmission by *Xiphinema* spp. in soil (Tomlinson, 2014) and the prevalence of alternative weed species (Abraham *et al.*, 2021b; Sastry, 2013;) complicates its management and poses a serious threat to quarantine authorities globally (Šneideris *et al.*, 2012). ToRSV causes significant economic yield losses (>50%) in many perennial fruit crops, ornamentals and horticultural crops worldwide (Basso *et al.*, 2016; CABI, 2022; Griesbach, 1995; Stace-Smith, 1996). Monitoring and early detection of plant virus diseases in the field by serology and DNA-based techniques are necessary steps to curb their spread and facilitate effective integrated management practices (Martinelli *et al.*, 2015; Mehetre *et al.*, 2021; Pappas *et al.*, 2021). The occurrence of some economically important tomato viruses including Tomato aspermy virus, Tomato leaf curl virus, Tomato mosaic virus, Tomato yellow leaf curl virus and Tomato bushy stunt virus have been well established in Nigeria (Abraham *et al.*,

2019a; 2019b; 2020; 2021a; 2022) Although ToRSV has been reported to be widespread infecting a wide range of plant hosts (OEPP/EPPO, 2018), in Nigeria, however, it has only been reported on weed species (Abraham *et al.*, 2021b) with no information on the occurrence and spread of the virus on tomato plants, particularly in the northern region where the bulk of tomato production (83.8%) lies in the country (NAERLS & FMARD, 2021). Therefore, this survey was conducted to investigate the incidence and distribution of ToRSV in northern part of Nigeria.

### Materials and Methods

#### Field survey and sample collection

A field survey was conducted to determine the incidence and spread of ToRSV infecting tomato plants in the Sudan savanna region of Nigeria during the 2017 and 2018 dry seasons. Three states (Gombe, Jigawa and Kano States) are among the major and leading commercial tomato producing states in Nigeria (GEMSA4, 2016). Three Local Government Areas (LGAs) in each State (Gombe: Akko, Yamaltu-Deba and Kaltungo, Jigawa: Hadejia, Kazaure and Kirikasama; Kano: Garun Mallam, Kura and Bagwai) were surveyed based on their tomato production records from which three farms each were surveyed. Forty tomato leaf samples from each farm (n=2160) were randomly collected in 5 quadrants (4 x 4 m<sup>2</sup>) in a diagonal pattern as described by Kashina *et al.* (2002). Data including virus disease symptoms, farm size, cultural practices, coordinates, surrounding crops, source of seed, sanitary status of the surveyed fields were documented (Table 1). Each of the samples collected was preserved over calcium chloride in a labelled sample bottle and conveyed to

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the virology laboratory unit of Crop Protection Department, Ahmadu Bello University Zaria for further analyses. Samples were maintained at 4°C until used for testing.

### Serological assay

All the samples collected in both years were assayed against ToRSV using the triple antibody sandwich enzyme-linked Immunosorbent assay (TAS-ELISA) method following the standard procedure recommended by the supplier (DSMZ, Braunschweig, Germany). ELISA reading was made 1 hr after adding the substrate using an ELISA plate reader Uniequip (Martinseed, Germany) at 405 nm wavelength. The positive control (ToRSV infected tomato leaves sourced from DSMZ, Germany) and the negative control (a healthy tomato leaves cultivated in a screen house) were included in each plate. A test sample was considered positive when the absorbance value is two times greater than that of the negative control (Abraham *et al.*, 2021a). Average ELISA results on ToRSV incidence (%) for the two years were calculated as the percentage of the positive sample count per farm divided by the total number of examined samples.

### Data analysis

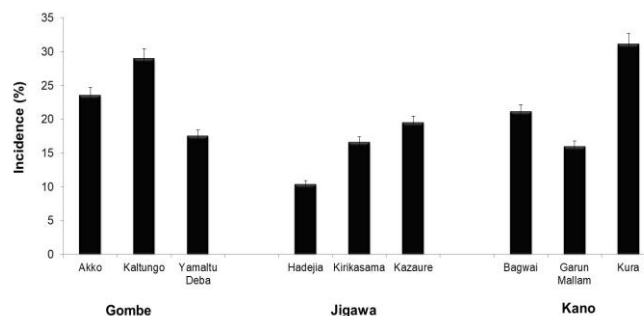
Data collected on the incidence of ToRSV was subjected to analysis of variance (ANOVA). The mean variations were separated at a 5% probability level using either the least significant difference (LSD) or by making a plot of the standard error (SE) of means (Gomez & Gomez, 1984).

## Results

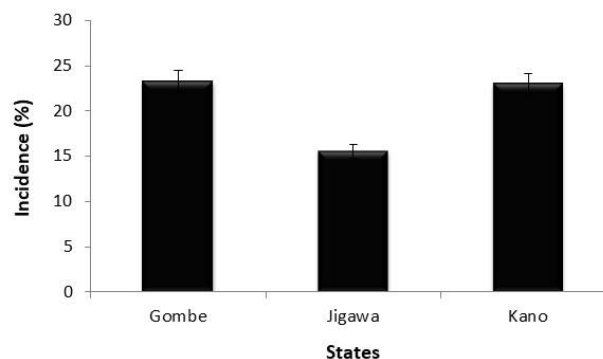
The ELISA results obtained showed that in Gombe State, significantly ( $P \leq 0.05$ ) highest incidence of ToRSV was recorded in Kaltungo LGA (29%) followed by Akko LGA (23.5%), whereas the lowest virus incidence (17.5%) was recorded in Yamaltu-Deba LGA (Figure 1). In Jigawa State, the highest virus incidence of 19.5% was recorded in Kazaure LGA, followed by Kirikasama LGA (16.6%), whereas Hadejia LGA had the least incidence of 10.4% (Figure 1). Kura State was shown to have significantly ( $P \leq 0.05$ ) the highest ToRSV incidence (31.1%) followed by Bagwai (21.1%) whereas Garun Mallam State recorded the least virus incidence (16%) in Kano State (Figure 1). ToRSV incidence was significantly ( $P \leq 0.05$ ) higher in Gombe (23.3%) and Kano (23%) States compared to Jigawa state with 15.5% (Figure 2).

## Discussion

The present study established that ToRSV occurs naturally infecting field-grown tomato in the Sudan savannah region of Nigeria. OEPP/EPPO, (2018) had earlier documented the prevalence of ToRSV in six continents which include Africa (Egypt, Togo and Tunisia), Asia (China, India, Iran, Japan, Jordan, South Korea, Oman, Pakistan, Taiwan), Australia (Australia, Fiji, New Zealand), Europe (Slovenia, Croatia, Poland, France, Belarus, Lithuania, Serbia, Slovakia, Turkey, Russia), North America (Mexico, USA, Canada) and South America (Brazil, Chile, Columbia, Peru, Venezuela).



**Figure 1.** Incidence of tomato ringspot virus in three local government areas each of Gombe, Jigawa and Kano States during the 2017 and 2018 dry seasons. The standard error of means were represented by bars at a 5% level of probability.



**Figure 2.** Mean incidence of tomato ringspot virus in Gombe, Jigawa and Kano States during the 2017 and 2018 dry seasons. The standard error of means was represented by bars at a 5% level of probability.

The prominent virus symptoms observed across all the surveyed fields irrespective of tomato variety grown and state, include chlorosis, necrosis, leaf curling, mosaic, chlorotic spot, stunting and malformation of leaves. Some of the virus disease symptoms observed in the field could be due to infection by other tomato viruses which were not considered in the present study. The prevalence of ToRSV in the study areas could be attributed to varying factors. For example, by our interaction with the local farmers during the survey, the majority were unaware of virus diseases and in most cases mistaken their symptoms for nutritional deficiencies. The inability of these farmers to distinguish biotic disease symptoms from nutrient deficiencies may have led to the inappropriate application of control measures to prevent the spread of ToRSV disease which may lead to significant crop yield losses as observed by Liu *et al.* (2020), Mishra *et al.* (2020) and Ghorai *et al.* (2021). Previous studies (Imam & Garba, 2013; Bulus *et al.*, 2017; Abraham *et al.*, 2018) have reported the occurrence of *Xiphinema* spp. in irrigated vegetable fields of Gombe, Kano and Kaduna states in Northern Nigeria. The incidence of *Xiphinema* spp. (dagger nematodes) as the principal vector transmitting ToRSV in the soil to both cultivated and uncultivated plant species (Tomlinson, 2014; Tuffen, 2018) in the study locations is an important factor contributing to the spread of the virus. Moreover, weed species have been reported to

serve as alternative hosts of ToRSV (OEPP/EPPO, 1991; Tuffen, 2018) in the field. Therefore, poor weed management within and around majority (70%) of the surveyed tomato fields is likewise an important factor influencing the incidence of ToRSV. Abraham *et al.* (2021b) recently documented nineteen weed species from 11 families to be naturally infected with ToRSV in Northern Nigeria among which *Gomphrena* and *Chenopodium* species have also been reported to spread the virus through its seed as reviewed by Sastry (2013). A common practice amongst most farmers (63%) is the continuous use of tomato untreated seeds collected from previous planting season. This practise by farmers could be another significant factor for ToRSV spread in the surveyed areas. Similarly, some farmers (18%) do purchase seedlings from uncertified local market vendors who raised seedlings in untreated soils that could be infested with ToRSV vector, hence, increasing the chances of seedling infection. The transmission of ToRSV through infected tomato seeds has been well documented (Sastry, 2013; OEPP/EPPO, 2016). Furthermore, some farming practices in the study area including the continuous and all-year-round cultivation of tomato, intercropping

and/or planting around tomato fields other vegetable crops susceptible to ToRSV without any deliberate control measures against both the virus and its vector allows for their uninterrupted thrive and spread in the region. Tan *et al.* (2022) have observed that *Xiphinema* spp., after they acquire ToRSV within 1 hour feeding on infected plant, they are capable of transmitting the virus for up to 2 years. This agrees with the report of Bernardo *et al.* (2018) that agricultural crop cultivation systems/pattern contributes to the incidence and spread of plant viruses in space and time.

This study reported the natural occurrence of ToRSV in field-grown tomato plants for the first time in Nigeria. This is the first report of ToRSV infecting tomato plants in Nigeria. Gombe and Kano states had the highest incidence of the virus. Farmer's unawareness about ToRSV and its effective management measures were key factors influencing the occurrence and distribution of the virus, hence, the need to educate farmers on the integrated management of the ToRSV in the region. In addition, Molecular characterization of the virus to determine its possible strain (s), relationship with other ToRSV species/strains reported elsewhere is also recommended.

**Table 1.** Virus disease symptoms and cropping history of the surveyed locations in Gombe, Jigawa and Kano States during the 2017 and 2018 dry seasons.

State	LGA	Farm size (Ha)	Variety grown	Cultivation		Surrounding crops	Crop growth Stage***	Cropping pattern	
				History (Years)	Seed source*				
<b>Gombe Akko</b>									
	Gadawo	0.5	UTC/Syria	20	PS	C, LC, M, CS, S	Pepper, Tomato, Okra	V	Mixed cropping with okra, pepper
	Kembu-Gingin Gada	1.4	Syria	25	LMV	C, S, LC, CS N, M	Okra, tomato	FW	Mixed cropping with okra
	Kembu	0.4	Syria/Tandino	>60	PS	C, S, CS LC, M	Tomato, pepper, watermelon	V	Sole cropping: rotate with water melon and pepper
<b>Kaltungo</b>									
	Gujuba	0.1	Syria	4	LMV	N, C, LC, M, CS	Pepper, maize, Corchorus	FW	Sole cropping: rotate with pepper and maize
	Awak	1.2	Roma VF	8	PS	C, LC, M, S, N	Tomato, sugarcane	V	Mixed cropping with cucumber
	Dogon Ruwa	1.5	Tandino	7	PS	N, C, LC, M, T, CS	Tomato, Okra, Onion, Maize	V	Mixed cropping with okra, pepper
<b>Yamaltu-Deba</b>									
	Dadin Kowa	0.4	Syria	5	PS	C, T, S, LC, M, CS	Sweet melon, maize	V	Mixed cropping with maize and sweet melon
	FCHTRF	0.3	Syria	15	PS	C, LC, M, S, CS	Okra, pepper	FW	Mixed cropping with okra
	Kwadon	1.1	Syria	30	PS	C, LC, M, S, T, N	Tomato, onions, maize	FW	Mixed cropping with maize
<b>Jigawa Hadejia</b>									
	Mai Alkama	1.1	Tandino	25	PS	C, N, LC, S, CS	Onion, tomato	V	Mixed cropping with pepper
	Hadejia	0.6	UTC	6	PS	LC, M, CS, N	Tomato, pepper	V	Sole cropping: rotate with pepper and onion

State	LGA	Farm size (Ha)	Variety grown	Cultivation			Surrounding crops	Crop growth Stage***	Cropping pattern
				History (Years)	Seed source*	Symptoms**			
Kano	Yayari	3.5	UTC	30	PS	C, LC, CS, M,	Tomato, pepper	F	Mixed cropping with okra and pepper
	<b>Kazaure</b>								
	Dabaza	1.4	UTC	8	CS	S, C, N, M, CS	Tomato, pepper, cassava	F	Sole cropping: rotate with pepper
	Dan Dutsi-Sadua	1.6	UTC (Graptor)	25	CS	C, S, LC, M, N	Tomato and pepper	F	Mixed cropping with okra, maize and cucumber
	Kurfi	0.6	Roma VF	10	PS	C, LC, M, N, CS	Tomato, Maize	F	Sole cropping: rotate with pepper
	<b>Kirikasama</b>								
	Tarabu	1.7	UTC	25	PS	N, C, LC, S, T	Pepper, tomato	F	Sole cropping: rotate with pepper
	Tarabu-Kumoyo	0.8	UTC	30	PS	C, S, M, T, LC, N	Maize	FW	Mixed cropping with rice and maize
	Marma Giryo	0.2	Roma VF	>30	PS	C, LC, M, S, N	Tomato, rice, maize	F	Mixed cropping with maize
	<b>Bagwai</b>								
	Dabino-Center 5	0.1	Roma VF	15	SC	C, S, LC, M, CS	Onion, Tomato, Maize	F	Mixed cropping with peas, onions and groundnut
	Dabino-Center 4	2	UTC	17	SC	C, LC, M, LC	Tomato, Maize, Groundnut	V	Mixed cropping with maize and peas
	Dabino-Center 3	1.1	Dan Jos	7	SC	S, LC, T, M, CS	Tomato, Cowpea, Maize	V	Mixed cropping with peas and groundnut
	<b>Garun Mallam</b>								
	Chiromawa	2.1	Roma VF	15	LMV	C, N, LC, M,	Tomato, maize,	V	Mixed cropping with peas
	Yantomu	0.8	UTC	>15	PS	C, M, LC, S, CS	Tomato, maize, peas and cucumber	FW	Mixed cropping with radish, pumpkin and cucumber
	Kadawa	2.1	Roma VF	7	LMV	M, CS, N, CL	Tomato, water melon, Maize, peas	V	Mixed cropping with peas and maize
	<b>Kura</b>								
Butalawa-fadama 1	1.4	UTC	27	PS	C, LC, N, S, M	Maize, Tomato, rice	F	Mixed cropping with maize, pepper and cabbage	
Butalawa-fadama 2	0.5	UTC	10	LMV	N, C, CS, T, M	Tomato, pepper	F	Mixed cropping with pepper and maize	
Butalawa-fadama 3	0.8	UTC	15	PS	S, CS, M, C	Tomato, maize, Cassava	V	Mixed cropping with maize and cassava,	

\*SC= Seed company; PS = Previous season; LMV = Purchased from local market vendors. \*\*C= Chlorosis; CS= Chlorotic spots; LC= Leaf curl; M= Mosaic; N= Necrosis; S= Stunting; T= Twisting; \*\*\*V= Vegetative; FW= Flowering.

## المخلص

ابراهيم، ب. و. بانوو، ب.د. كاشينا، م.د. أليجيبيجو وم.ب. ابراهيم. 2024. ظهور فيروس التبغ الحلقي للبندورة/الطماطم الذي يصيب نباتات البندورة/الطماطم في شمال نيجيريا. مجلة وقاية النبات العربية، 42(1): 43-48. <https://doi.org/10.22268/AJPP-001214>

أجري مسح حقل للكشف عن فيروس التبغ الحلقي في البندورة/الطماطم (ToRSV) في منطقة السافانا السودانية (جيجاوا، كانو، غومبي) من نيجيريا خلال موسمي الجفاف في 2017 و2018. ولهذه الغاية، تم اختيار ثلاث مناطق حكومية محلية لكل ولاية مما سبق، وذلك وفقاً لتاريخ زراعة محصول البندورة/الطماطم، وبواقع 3 حقول لكلٍ منها. بلغ عدد عينات أوراق البندورة التي جمعت عشوائياً من الحقول كافة (9 حقول) 2160 عينة، والتي تم اختبارها للكشف عن الفيروس باستخدام اختبار إلزا بالاحتواء الثلاثي للفيروس (TAS-ELISA). بينت النتائج انتشار فيروس ToRSV في الولايات الثلاث المدروسة، وسجلت أعلى نسب إصابة بالفيروس (29%) في منطقة Kaltungo وبفروق معنوية عن باقي المناطق ( $P \leq 0.05$ ) في ولاية غومبي، وتلتها منطقة Akko بنسبة إصابة بلغت 23.5%، في حين سجلت أقل نسبة إصابة (17.5%) في منطقة Yamaltu-Deba. أما في ولاية جيجاوا، فكانت أعلى نسبة إصابة (19.5%) في منطقة Kazaure، وتليها منطقة Kirikasama (16.6%) وكانت أدها في منطقة Hadejia (10.4%). وفي ولاية كانو، وسجلت أعلى نسب إصابة بالفيروس (31.1%) في منطقة Kura، وتلتها منطقة Bagwai (21.1%)، في حين كانت أقل نسبة إصابة (16%) في منطقة Garun Mallam. يعد هذا التسجيل الأول لفيروس التبغ الحلقي في البندورة/الطماطم (ToRSV) في نيجيريا؛ وإثمه من الضروري إعلام مزارعي البندورة/الطماطم بإجراءات الإدارة الفعالة لهذا الفيروس الذي يمكن أن يحد من إنتاجية المحصول في المنطقة.

كلمات مفتاحية: كشف، انتشار، نيجيريا، فيروس التبغ الحلقي للبندورة/الطماطم، TAS-ELISA.

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