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Genetic Effects of Ultra-High-Diluted-Biomedicines Gall 30C, Gall 200C, and Gall 1000C may be Vaccines against Plant and COVID-19 Diseases: Improved Agriculture-Health-Medical-Pharmaceutical-Science-Technology-Communication-Issues!

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Abstract

The current coronavirus 2 (SARS-CoV-2) outbreaks of Coronavirus disease 2019 (COVID-19) pandemic have emphasized the vulnerability of human populations to novel viral pressures, causing an emergent global pandemic and badly impacts on agriculture-environment-health-socio-economy medical-pharmaceutical-science-technology-communication-issues. And India emphasis on okra, *Abelmoschus esculentus* (L.) Moench Cv. Ankur-40, the 'Nature's-Gift to Human-Disease-Free-Healthy-Life' and the most 'Economically-Important Number-One Consumption-Vegetable-Crops,' is damaged by different pathogens like nematodes, causing the root-knot (RK) disease which is easily controlled by different chemical-pesticides, and so it is an urgent need to develop potential epidemiological and biomedical vaccines. The Ultra-High-Diluted Biomedicines (UHDBM) Gall 30C, Gall 200C, and Gall 100C, prepared from Okra Root-Galls (ORG) or Gall MT, applied by foliar spray @ 20 mL/plant each group respectively, are highly effective against the root-knot disease of okra, with increasing fresh-plant growth and fruit production. The UHDBM; Gall 30C, Gall 200C, and Gall 1000-pretreatments are more effective than the untreated ones, and Gall 200C shows the most potential result in all respects. The genetic-effects of UHDBM thought to induce systemic acquired resistance response of the treated plants through the expression of pathogenesis-related (PR)-proteins-genes (22 to 4 numbers), which are more or less similar molecular range (295 kD to 11 kD) of many coronaviruses, and it will responsible for preventing RK and COVID-19 like variant-virus diseases by inducing defense-resistance or increasing innate-immunity, and enriching agriculture-environment-health-socio-economy-medical-pharmaceutical-science-technology-communication-issues (AEHSEMPSTCI) with the toxic-free world, and it may help to develop best potential new preventive treatments methods or drug or vaccines, in the field of '21st Century COVID-19 like Pandemic in the new normal situation in future.

Keywords: Genetic effects; Ultra-high-diluted-biomedicines; Vaccines; Plant-and-COVID-19-diseases; Agriculture-health-medical-pharmaceutical-science

Introduction

Pandemic problems

The recent coronavirus 2 (SARS-CoV-2), is just one of the nonillions of viruses on our planet, and scientists are rapidly identifying legions of the most recent species that confronting our next National Health Disaster - long-haul Covid-19, outbreaks of Coronavirus disease 2019 (COVID-19) pandemic have emphasized the vulnerability of human populations to novel viral pressures and genome variation of worldwide, causing an emergent global pandemic and badly impacts on agriculture-environment-health-socio-economy medical-pharmaceutical-science-technology-communication-issues, etc [1-20].

India emphasis on okra (lady finger) for the 'food security' which is the most important and crucial aspect of sustainable development in the agricultural sector and the backbone of the economy, but badly impact of the COVID-19 pandemic on food security, agriculture, massive consequences on health and livelihoods of developing countries [21], and it not only reduced incomes, but also disrupted supply chains, chronic and acute hunger was on the rise due to various factors including conflict, socio-economic conditions, natural hazards, climate change, and pests like the locust outbreak compounding this crisis across 23 countries with the other zoonotic diseases remain a recurrent threat. And it is reported that the okra, *Abelmoschus esculentus* (L.) Moench, is one the most economically important, commercially exploited, number one consumption in a

variety of ways vegetable crops, which is the oldest widely cultivated oligo purpose, significantly contribute for nutritional, medicinal and industrial application , and used as traditional medicine, achieving the 'Nature's Gift to human disease-free healthy life' multipurpose crop, achieving India first in the world for the fruits rich in vitamins, calcium, folic acid, carbohydrates, phosphorus, magnesium and potassium, iodine, mineral matters, and a good source of superior nutritional quality for human nutrition for preventing different diseases; like cardiovascular disease, type 2 diabetes, kidney diseases, skin infection, digestive diseases, some cancers, antioxidant, nootropic, eye, body immunity, blood pressure, obesity, asthma, constipation, heart disease, sexual health, and neurological disorders, etc., and mature fruit and stems contain crude fiber, used in the paper industry and sugarcane industry [22-25]. The enrich source of nutrients, minerals, and fibers of okra has invited to many pathogens, pests, and diseases infestation, and only the nematode pathogens causing root-knot, damage 10-40% of the total crop production annually, causing serious problems in our country and this indirectly affects our advanced agronomy-plant-breeding-horticulture-environment socio-economy green-science-technology-communication-issues, and agricultural-economy also. Though the pesticides are very much effective but it creates several problems in toxicity, pollution, cost-effectiveness, environment-friendliness, and biodiversity conservation. On the other hand the pesticides block functional first known gene transfer between plants to insects or animals, which is used in the host's defenses mechanism for new pest-control strategies also, and recently in the 31countries of the globe that higher airborne pollen concentrations correlated with increased SARS-CoV-2 infection rates [1-30].

Recently after long lockdowns in Purba Bardhaman, West Bengal, India (Figure 1), only two day shows that the total COVID-19 positive cases are 37494-37524, the total number of discharge cases are 36685-36718, the total number of COVID-19 death is 436-436, rate of recovery is 97.84%-97.85% and rate of mortality is 1.16%-1.16% respectively. So it is an urgent need to find out by developing policy-initiative, cheap, non-phytotoxic, and non-pollutant potential high-diluted-biomedicines (UHDBM) for preventing both the pandemic crisis by improving the agriculture system with the findings and other new research to develop future support and treatments.

Work done

It is already shown that the various; pure compounds, homeopathy, allelopathy, plant extracts, phytomedicine, bio-agents, intercropped-/multi cropped-biomedicines, bio-medicinal-meals, biomedicine-vaccine, social-vaccine, policy-developed global-vaccine, many models, etc. are applied to regulate against different diseases causing pathogens, control the plants- and animal- diseases causing pathogen [3-19,23-30]. But it's not achieving potential success all told the cases because of different causes.

It has also been observed recently that the animals- and plants-biomedicines; Nematode Extract (NE) or nematode MT (NMT) and Root Galls (RG) extract or Gall MT is the safe alternative method to control root-knot diseases caused by nematode pathogens, by inducing their natural defense response of the host plants [14-19,24-26,28-30,44].

Aims and Objectives

The purpose of the current study is to use and confirm the systematic signaling and induced natural defense or immunity in the host plants, *Abelmoschus esculentus* (L.) Moench Cv. Ankur-40, by applying UHDBM; Gall 30C, Gall 200C, and Gall 100C, prepared from Gall MT (GMT) or Gall Roots (GR) as a preventive measure against root-knot, and COVID-19 diseases ideas, and also to find out the actual reasons of the genetic effects on the UHDBM for preventing diseases.

The current paper contributes the development of the most policy-initiative, cheap, non-phytotoxic, non-pollutant, and side-effect free potential UHDBM global vaccines; Gall 30C, Gall 200C, and Gall 1000C, for controlling both the pandemic crisis; root-knot diseases, and the future-pandemic COVID-19 like virus diseases, by enriching agriculture-environment-health-socio-economy-medical-pharmaceutical-science-technology-communication-issues (AEHSEMPSTCI) with the findings and other new research to drug development for the future support and the best treatments or potential vaccine for all.

Materials and Methods

Location and preparation of High-Diluted Biomedicines (HDBM) Gall MT (GMT)

The UHDBM Gall MT (GMT), prepared from the root galls (RG) of Okra Plants (OP) which were collected from roots of the OP plants, *Abelmoschus esculentus* (L.) Moench Cv. Ankur-40, grown in the experimental garden of the Department of Zoology, VisvaBharati University, Santiniketan - 731235, and they were washed with sterile tap water, homogenizer and extracted with 90% ethanol at room temperature (25 ± 2°C) for 15 days and centrifuged at @ 3500 rpm for 5 minutes, and then the Gall Extract (GE) supernatant was collected and allowed to evaporate at room temperature (25 ± 2°C) and the biomedicines residues were kept over anhydrous calcium chloride ($CaCl_2$) for dehydration and stored at 4°C. The crud GE-biomedicines residue was mixed with sterile distilled water just before application on the test plants. The crude residue was diluted in 90% ethanol at 1mg/ml concentration and was prepared UHDBM, Gall MT (Original Solution or Crude Extract i.e. Mother Tincture) [3-9,12-19,26-30].

UHDBM Gall 30, Gall 200C and Gall 1000C preparation

For the preparation of UHDBM liquid drugs, the high-diluted GMT was diluted with 90% ethanol (1:100) proportionate in a round vial which was filled up to two-thirds of its space, tightly crocked, and the vials were given 10 powerful downward strokes of the arm for mechanical agitation (succession), forming the 1st centesimal potency named Gall 1C. All the subsequent potencies were prepared by further diluting each potency with 90% ethanol in the same proportion (1:100) and the mixture was given 10 powerful downward strokes. In this way, different potencies of both the drugs; Gall 30C, Gall 200C, and Gall 1000C, were prepared respectively [3-9,12-19,26-30].

UHDBM Gall 30, Gall 200C, and Gall 1000C test solutions preparation

For the preparation of ultra-high-diluted test-solution of the biomedicines; Gall 30C, Gall 200C, and Gall 1000C, were diluted (v/v) @ 1mL drug/20ml sterile distilled water (in the proportion of drug: water=1:20, containing 0.2mg drug) respectively, and the UHDBM-liquid control-solution of both the drugs were diluted (v/v) @ 1mL 90% ethanol/20ml sterile distilled water (in the proportion of drug: water=1:20) respectively, and the control solution was prepared for comparison to the preparation of test solutions, and stored at 4°C for treatments media [3-9,12-19,26-30].

Pot test and inoculation

Aseptically germinated seeds of *Abelmoschus esculentus* (L.) Moench Cv. Ankur-40 was sown at the rate of one seed/pot (32 cm diam.) containing a mixture of clay soil and composted manure (2:1

Daily Press Briefing
PurbaBardhaman
Date:26/06/2021 (Up to 5.00 P.M.)
(Report to by send by 7.00 P.M Daily)

Part – I: Related to COVID-19

i)	**Total No. of COVID Positive Patients found on the day of reporting ***	:	**31**
ii)	Total no of COVID positive patients**	:	**37393**
iii)	Total no of active patients as on today***	:	**382**
iv)	Total no of discharged cases	:	**36577**
v)	Total no of COVID death recorded	:	**434**
•	**Quarantine status :**		
vi)	Total no of persons under institutional quarantine	:	**0**
vii)	Total no of persons under house quarantine	:	**0**
viii)	Total no of persons under quarantine from Maharashtra, Delhi, Gujarat, Tamil Nadu & M.P.	:	**0**
ix)	Total no of persons under quarantine from other state of India	:	**0**
x)	Total no of persons relaxed from institutional quarantine	:	**0**
•	**Testing status :**		
xi)	Total no of Sample collected up to 25.06.2021	:	**276158**
xii)	Total no of Sample tested	:	**275622**
xiii)	Total no of Positive cases	:	**14682 (+53 repeat +ve)**
xiv)	Total no of negative Cases	:	**259695 (+1192 spillage/rejected)**
•	**Containment Zone status :**		
xv)	Total no of Containment Zone as on today	:	
xvi)	Total no of containment withdrawn	:	
•	**Analysis of Positive Persons Details : On date – Positive-**		
xvii)	Total No. of Migrant (Other State + Other Dist. of WB):	:	**00**
xviii)	No. of Persons in Safe House:	:	**07**
xix)	No. of Person in Covid Hospital:	:	**08**
xx)	No. of Persons in Home Isolations:	:	**28**
Analysis on COVID-19 +VeCases :			
xxi)	Rate of Recovery# (Percentage)	:	**97.82**
xxii)	Rate of Mortality# (Percentage)	:	**1.16**
Report on Sample Testing:			
xxiii)	Antigen Test	:	**1969**
xxiv)	RT-PCR Test	:	**706**
xiv)	Test Result within 24 Hrs.	:	**2046(RAT-1969+ RTPCR-87)**

#On Total Positive Case

Part- II:

Distribution of COVID Positive Cases found on 26/06/2021

Aushgram-I	**0**	**Galsi-II**	**0**	**Ketugram-I**	**1**	**Mongolkote**	**0**	**Burdwan Municipality**	**6**
Aushgram-II	**0**	**Jamalpur**	**3**	**Ketugram-II**	**0**	**Purbasthali-I**	**2**	**Dainhat Municipality**	**0**
Bhatar	**0**	**Kalna-I**	**5**	**Khandoghosh**	**2**	**Purbasthali-II**	**1**	**Guskara Municipality**	**0**
Burdwan-I	**0**	**Kalna-II**	**0**	**Manteswar**	**3**	**Raina-I**	**1**	**Kalna Municipality**	**2**
Burdwan-II	**2**	**Katwa-I**	**0**	**Memari-I**	**0**	**Raina-II**	**0**	**Katwa Municipality**	**0**
Galsi-I	**1**	**Katwa-II**	**0**	**Memari-II**	**1**	**Other District**	**1**	**Memari Municipality**	**0**

Analysis on COVID +Ve Cases on 26/06/2021		***COVID Positive as on today**	****Total Positive Cases**
Type	Symptomatic	03	2656
	Asymptomatic	28	34737
Total		**31**	**37393**
Contact Analysis	Primary Contact	01	1500
	Travel from High Burden Dist. of W.B.	00	217
	Travel from Other State	00	435
	No Travel History	30	35241
Total		**31**	**37393**

Daily Press Briefing
PurbaBardhaman
Date:29/06/2021 (Up to 5.00 P.M.)
(Report to by send by 7.00 P.M Daily)

Part – I: Related to COVID-19

i)	**Total No. of COVID Positive Patients found on the day of reporting ***	:	**30**
ii)	Total no of COVID positive patients**	:	**37524**
iii)	Total no of active patients as on today***	:	**370**
iv)	Total no of discharged cases	:	**36718**
v)	Total no of COVID death recorded	:	**436**
•	**Quarantine status :**		
vi)	Total no of persons under institutional quarantine	:	**0**
vii)	Total no of persons under house quarantine	:	**0**
viii)	Total no of persons under quarantine from Maharashtra, Delhi, Gujarat, Tamil Nadu & M.P.	:	**0**
ix)	Total no of persons under quarantine from other state of India	:	**0**
x)	Total no of persons relaxed from institutional quarantine	:	**0**
•	**Testing status :**		
xi)	Total no of Sample collected up to 28.06.2021	:	**277692**
xii)	Total no of Sample tested	:	**277318**
xiii)	Total no of Positive cases	:	**14751 (+53 repeat +ve)**
xiv)	Total no of negative Cases	:	**261322(+1192 spillage/rejected)**
•	**Containment Zone status :**		
xv)	Total no of Containment Zone as on today	:	
xvi)	Total no of containment withdrawn	:	
•	**Analysis of Positive Persons Details : On date – Positive-**		
xvii)	Total No. of Migrant (Other State + Other Dist. of WB):	:	**00**
xviii)	No. of Persons in Safe House:	:	**05**
xix)	No. of Person in Covid Hospital:	:	**03**
xx)	No. of Persons in Home Isolations:	:	**27**
Analysis on COVID-19 +VeCases :			
xxi)	Rate of Recovery# (Percentage)	:	**97.85**
xxii)	Rate of Mortality# (Percentage)	:	**1.16**
Report on Sample Testing:			
xxiii)	Antigen Test	:	**1514**
xxiv)	RT-PCR Test	:	**460**
xiv)	Test Result within 24 Hrs.	:	**1617 (RAT-1514+ RTPCR-103)**

#On Total Positive Case

Part- II:

Distribution of COVID Positive Cases found on 29/06/2021

Aushgram-I	0	Galsi-II	0	Ketugram-I	1	Mongolkote	0	Burdwan Municipality	4
Aushgram-II	0	Jamalpur	1	Ketugram-II	0	Purbasthali-I	0	Dainhat Municipality	2
Bhatar	1	Kalna-I	2	Khandoghosh	1	Purbasthali-II	1	Guskara Municipality	0
Burdwan-I	3	Kalna-II	0	Manteswar	0	Raina-I	3	Kalna Municipality	0
Burdwan-II	3	Katwa-I	1	Memari-I	0	Raina-II	0	Katwa Municipality	0
Galsi-I	2	Katwa-II	0	Memari-II	2	Other District	0	Memari Municipality	3

Analysis on COVID +Ve Cases on 29/06/2021		***COVID Positive as on today**	****Total Positive Cases**
Type	Symptomatic	03	2664
	Asymptomatic	27	34860
Total		**30**	**37524**
Contact Analysis	Primary Contact	00	1501
	Travel from High Burden Dist. of W.B.	00	217
	Travel from Other State	00	435
	No Travel History	30	35371
Total		**30**	**37524**

Figure 1: (Part-I & II**):** Daily Press Briefing of the Purba Bardhaman District (Date: 28-29/06/2021, Upto 5.00 P.M. Daily).

v/v). The soil-filled pots were treated with boiling water 5 (five) times. The pots were divided into four batches/groups; each numbering 10: (i) uninoculated untreated, (ii) inoculated untreated, (iii) Gall 30C-pretreated inoculated, and (iv) Gall 200C-pretreated inoculated, and (v) Gall 1000C-pretreated inoculated. All the pretreatments were done by foliar spray. The experiment was conducted outdoors at an ambient atmospheric temperature (27 ± 2°C) and relative humidity (75 ± 5%). Plants were inoculated at the 12-leaf stage (Day-25) with *M. incognita* (J2) @ 3425 ± 75 J_2-larvae/plant [3-9,12-19,26-31].

Treatments with UHDBM Gall 30, Gall 200C, and Gall 1000C test solutions

The UHDBM; Gall 30C, Gall 200C, and Gall 1000C -test solutions, were applied into the okra plants and sprayed on plants @20 mL/ treated plants three days before inoculation for pretreatments with nematodes-J_2 respectively. Control okra plants were treated with an equal amount (20 ml) of control solutions prepared with sterile distilled water, and the plants were regularly watered in the morning and evening. During spraying, the soil surface underneath each plant was covered with a polythene sheet. Plants in both uninoculated untreated and inoculated untreated groups received a spray of an equal amount of control solutions. All treatments were done in hygienic conditions [3-9,12-19,26-31]. All the data were analyzed by ANOVA (Analysis of Variance). The experiment was repeated thrice. Data from the last experiment are reported here.

Densitometer scanning of Okra Root (OR), Okra Root Galls (ORG), and Nematode Proteins (NP): The Okra Root Galls (ORG), OR, and Nematode Female (NF)-proteins separation was carried out by the method of Laemmli (1970) with the modifications as suggested by the LKB Instructional Manual (1986). A 10% separating gel and 5% stacking gel were used. The bands were scanned with a recording electrophoretic scanner (Biomidi, 96-300 densitometers). In figure 2 and table 1, the observation was recorded from the densitometer curve [4,7,14-19,27-32].

Tear of toxicity: Biomedicines; Gall 30C, Gall 200C, and Gall 1000C, has exposed directly for the study of toxic effect on nematode juveniles after 2 hours exposure periods at room temperature (20 ± 2°C) [4,13-18,23-35].

Harvesting: All the okra plants were uprooted 53 days after the sowing of seeds. The following measure measurement was taken: biomass of shoot, root, and fruits, root gall number, nematode population in roots (2 g) and soil (200 g), the protein content of root and fruits. Proteins were estimated by the Folin-phenol method [4,14-19,28-30,30-34]. All the data were analyzed by Analysis of Variance (ANOVA). The experiment was repeated five times with similar results and the data from the third experiment were represented in table 2.

In health-medical-pharmaceutical-science-technology: The different scientists, academicians, clinicians, scholars, researchers, students, farmers, administrators, institutions, communities, associations, teachers, staff, regulators, photographers, visitors, healthcare, media personnel, Burdwan Green Haunter and Students' Goal-NGO, and different club and social organizations, and organize street cornering, workshops, seminars, agriculture fair, health camp, campaign, aware, make the news, and publish in different journals emphasis on "Prevent okra and COVID-19 diseases for the genetic basis of the UHDBM"; Gall 30C, Gall 200C, and Gall 1000C enriching AEHSEMPSTCI [3-19,26-30].

Results

Effect on RK with UHDBM Gall 30, Gall 200C, and Gall 1000C

The pretreated with UHDBM; Gall 30, Gall 200C, and Gall 1000C significantly ($P \leq 0.05$ by ANOVA) increased plant growth in terms of the fresh biomass of shoot and fruits compared to the inoculated and untreated plants (Table 2). Root galls, nematode population in the root and soil, and root protein content were significantly ($P \leq 0.05$ by ANOVA) reduced in pretreated plants as compared to the untreated ones (Table 2). The number of fruits and protein content in green fruit was significantly reduced in inoculated untreated plants as compared to the uninoculated ones, and all the pretreatments with biomedicines; Gall 30, Gall 200C, and Gall 1000C showed better plant growth and lesser intensity of the root-knot disease, and Gall 200C showed the best results in all respect (Table 2).

Effect on toxicity: The UHDBM; Gall 30, Gall 200C, and Gall 1000C had not produced any direct toxic effect on nematode because no mortality occurs after 2 hours exposure period.

Genetic effects of OR, OGR and NF proteins: The table 1 and figure 2; show the genetic effects on molecular weight (kD) of Okra Root (OR), and Okra Gall Root (OGR) proteins for all pretreatments effects of UHDBM; Gall 30, Gall 200C, and Gall 1000C on *M. incognita* pathogens causing RK-disease of the OP. An analysis of root proteins of all 5-groups and nematodes female's (NF) proteins by electrophoresis and densitometer scanning of all the test OR and OGR-proteins show that all the UHDBM; Gall 30, Gall 200C, and Gall 1000C-pretreatments resulted in an increased number of proteins in the roots than uninoculated untreated and inoculated untreated groups; the highest number of root proteins in the UHDBM Gall 200C-pretreated inoculated group is 23, and next highest number of the root protein is 19 in the Gall 30C-pretreatment inoculated group, 16 in the Gall 1000C, and 15 in the inoculated untreated group and the lowest number of protein is 11 in the uninoculated untreated group respectively (Table 1 and Figure 2). The highest molecular weight of the OR-protein gene is 295 kD and the lowest molecular weight of the OR-protein genes is 11 kD. The lowest number of the new pathogenesis-related protein genes (PR-proteins) is 4 in the uninoculated untreated okra roots, and the highest number of the new PR-proteins-genes is 22 in the HDBM- Gall 200C-pretreated okra-pretreatment-group, 18 in the Gall 30C, and the same number i.e. 14 number of PR-proteins-genes is in both the ultra-high-diluted biomedicines Gall 1000C-pretreatment- and inoculated untreated -treatment-group respectively (Table 1 and Figure 2).

And the NF contained 18-proteins-genes and the molecular weight of the NF proteins-genes ranging from the lowest 12 kD to the highest 280 kD, and the total number of the PR-proteins-genes of NF is 16 in comparison to the uninoculated untreated group (Table 1 and Figure 2).

Discussion

On growth

The present study clearly showed that the OP-growth in terms of fresh biomass of shoot and fruits, and the number of fruits was higher than inoculated untreated groups in all the pretreatment groups of OP treated with the UHDBM; Gall 30, Gall 200C, and Gall 1000C, but reverse in the fresh biomass of roots of inoculated untreated one [4,14-19,28-30].

Table 1: Genetic effects of root proteins of the Gall 30C, Gall 200C, and Gall 1000C-pretreatmed okra and nematode female proteins.

Treatments Groups (Except NP)	Total number of proteins	Molecular weight (kD) of pretreatment groups																						Total number of PR-proteins
		Serial number of proteins																						
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	
I. Uninoculated Untreated	11	280	260	240	210	155	155	105	85	70	45	13	-	-	-	-	-	-	-	-	-	-	-	-	4
II. Inoculated Untreated	15	270	240	200	170	145	188	98	90	77	65	58	46	37	2	1	-	-	-	-	-	-	-		14
III. Gall 30C- Pretreated Inoculated	19	240	197	175	160	145	120	92	72	65	52	35	31	28	26	23	22	17	12.5	11	-	-	-	-	18
IV. Gall 200C-PretreatedInoculated	23	295	235	220	180	160	140	135	120	115	98	88	82	72	60	50	42	38	30	25	21	19	16.5	12.5	22
V. Gall 1000C- Pretreated Inoculated	16	260	210	150	135	120	110	95	80	72	60	50	37.5	28	23.5	16.5	11	-	-	-	-	-	-	-	14
Nematode Female Proteins (NFP)	18	280	260	250	200	190	165	150	135	85	55	40	38	27	23	22	18	15	12	-	-	-	-	-	16

'-' indicate no band and 'nematode female protein' not included in the treatments group.

Table 2: Effect of the biomedicines Gall 30C, Gall 200C and Gall 1000C-pretreated on okra plants[x] inoculated with root knot nematodes.

Treatments[y]	Fresh Biomass (g)[x]			Number of Fruits[x]	Number of Root Galls[x]	Nematode Population[x]		Protein[x] Content (%)		Number of Total Protein
	Shoot	Root	Fruits			Root (2 g)	Soil (200 g)	Root	Fruits	
I. Uninoculated Untreated	196.28a[z] ± 10.02	26.58c ± 1.22	32.16a ± 10.02	22.12b ± 1.02	00			1.24d ± 0.22	2.26a ± 0.02	11
II. Inoculated Untreated	75.01c ± 10.01	68.49a ± 6.01	13.27c ± 4.63	07.31c ± 2.01	424.13a ± 87.11	718a ± 22	31a ± 43	2.63a ± 0.01	1.12c ± 0.02	15
III. Gall 30C-Pretreated Inoculated	184.04b ± 1.02	30.31b ± 0.09	35.99c ± 0.01	24b ± 0.02	69b ± 1.11	53b ± 1.93	54b ± 2.02	1.38b ± 0.02	1.92c ± 0.02	19
IV. Gall 200C-Pretreated Inoculated	194.92a ± 2.02	27.10c ± 1.02	31.53a ± 4.23	25.68a ± 2.02	13.07d ± 2.31	29d ± 13	48c ± 24	1.29c ± 0.21	2.19b ± 0.03	23
V. Gall 1000C-Pretreated Inoculated	179.96b ± 2.42	29.22b ± 2.04	26.02b ± 7.08	24.24b ± 1.04	32.01c ± 1.03	42c ± 12	46c ± 24	1.37b ± 0.11	1.97b ± 0.03	16

'x'-Mean of 10 replicates with S.E.

'y'-Okra plants inoculated at 12-leaf stage (Day-25) with *M. incognita* juveniles (3425 ± 75J_2/pot), pretreated with GMT at 8-leaf stage (Day-22), and harvested 53 days after sowing of germinated seeds.

'z'-Means carrying same letters in a column are not significantly different (P ≤ 0.05) by analysis of variance.

On RK diseases

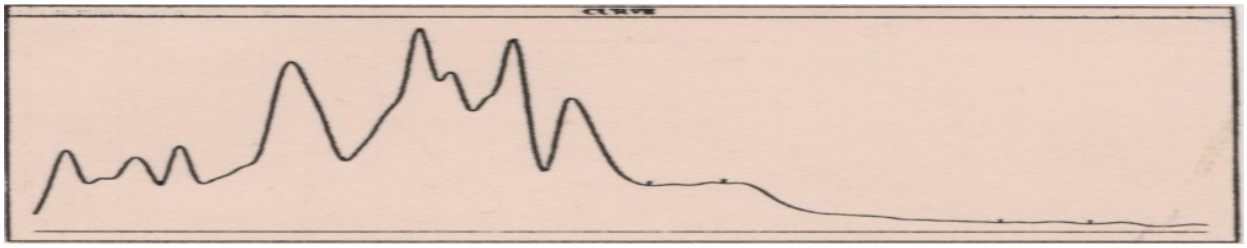
All the pretreatment groups of OP treated with UHDBM; Gall 30, Gall 200C, and Gall 1000C, decreased RK-diseases in terms of nematode infection in root gall number, and nematode population in root in comparison to inoculated untreated groups, and the population of nematode in the rhizospheric soil was the maximum with the group treated with Gall 30, Gall 200C, and Gall 1000C-biomedicines, and minimum with the inoculated untreated group, due to potential UHDBM, and it is also showed that the UHDBM; Gall 30, Gall 200C, and Gall 1000C, might induce synthesis of some antagonistic substance in the treated-OP, which is proved from the inoculated untreated ORG contained the highest protein-content due to presence of a large number of nematodes [4,14-19,28-30].

On toxicity

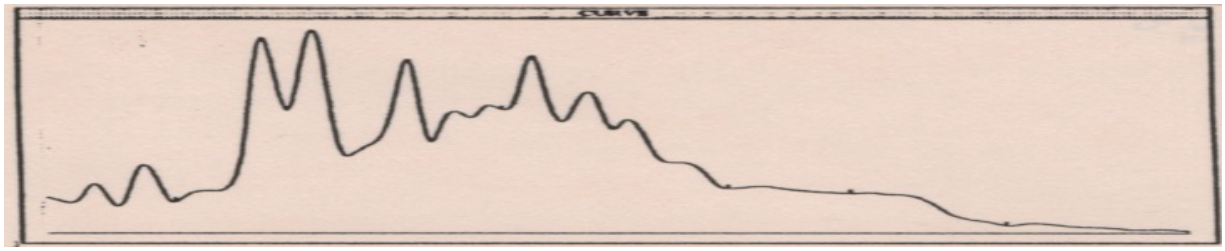
In the recent study is also showed that the UHDBM; Gall 30, Gall 200C, and Gall 1000C, had no direct toxic effects on nematodes juveniles, and OP, but it induced synthesis of some resistance substances in okra plants to *M. incognita* infection for preventing RK-diseases in the OP, and for these reasons, all the pretreated group had significantly greater fresh biomass of shoot-and fruits- plants than inoculated untreated one [4,14-19,28-30].

On defense response

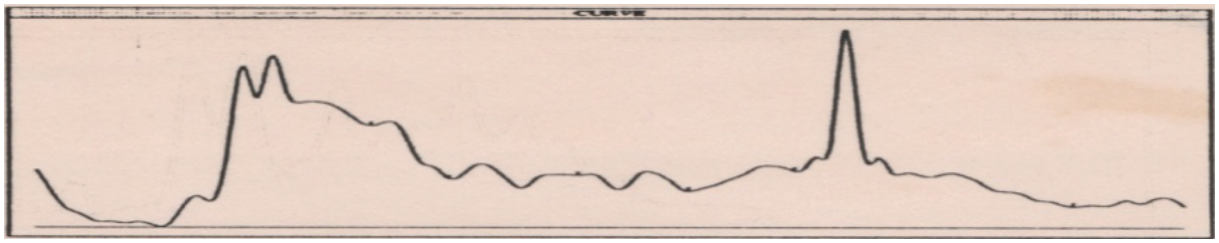
The present experiment showed that the UHDBM; Gall 30, Gall 200C, and Gall 1000C, act as really effective preventive biomedicines natural vaccines against plant diseases because of their defense



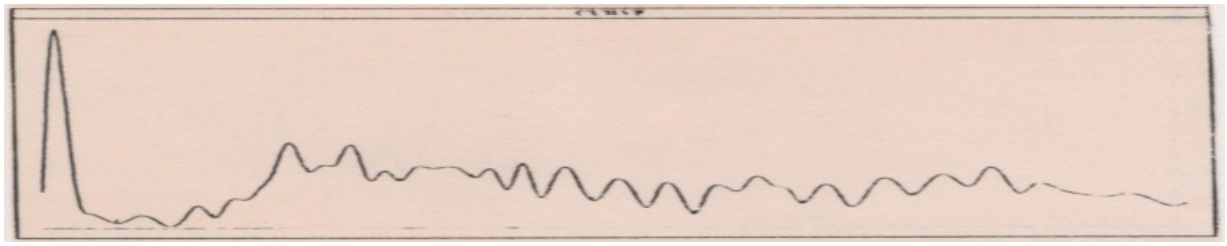
Uninoculated Untreated



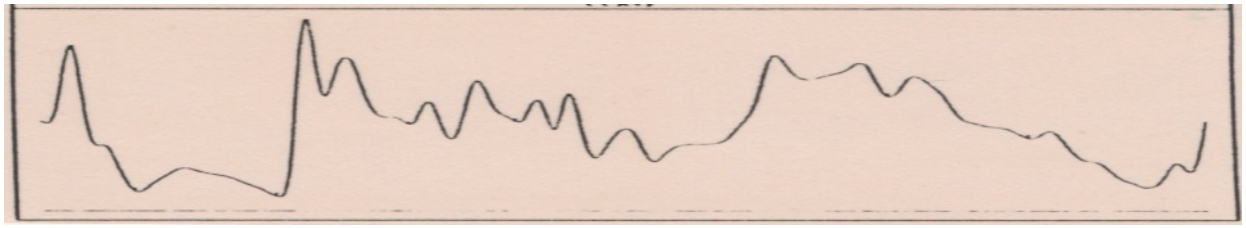
Inoculated Untreated



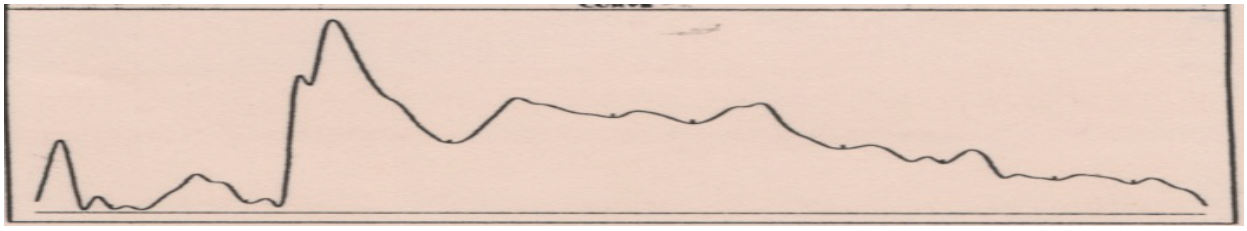
Gall 30C Inoculated



Gall 200C-Pretreated Inoculated



Gall 1000C-Prettreated Inoculated



Nematode Female Proteins (NFP)

Figure 2: Densitometry tracings of root proteins of the Gall 30C, Gall 200C and Gall 1000C-treated on okra with nematode proteins resolved on Poly Acrylamide Gel Electrophoresis (SDS-PAGE).

resistance, and it's known that the lectins accumulated in galled regions of the OR- infected with the RK-disease [38]. It's already known that many crop plants will be induced by acquiring systemic resistance for the localized virus- infection or non-pathogenic, and pathogenic-microorganisms or their culture-filtrates or gas or salicylic-acid, etc. protects plants from the numerous pathogens attack, by working systemically [4,14-19,28-47].

On pathogenesis-related (PR) of root proteins-genes

It is known that the *M. incognita* is known to share common antigens with its host plants [48], and Iqbal S, et al. (2020) informed that the attempt to 'Silence Genes' of the root-knot nematode, *M. incognita* results in diverse responses including an increase and no change in expression of some genes. So in the all the treated plant's roots show that the UHDBM; Gall 30, Gall 200C, and Gall 1000C-pretreatments resulted in an increased number of proteins-genes in the root than inoculated untreated-okra plants-group; the highest number of PR-proteins-genes in the UHDBM; Gall 30, Gall 200C, and Gall 1000C-pretreated group is 23, and the next highest number of the PR-protein-gene is 19 in the pretreated-Gall 30C -inoculated group, 16 in the Gall 1000C group, and 15 in the inoculated untreated group, and the lowest number of protein is 11 in the uninoculated untreated group respectively, which proved and confirmed that during infection with the nematode, host plants showed minimal defense responses to the nematode because of this antigenic similarity, and the different PR-proteins-genes of the ORG proteins ranging from 295 kD (the highest molecular weight protein) to 11 kD (the lowest molecular weight protein) of the OR-protein. And all the pretreated UHDBM; Gall 30, Gall 200C, and Gall 1000C-stimulate the synthesis of the number of different PR-proteins-antigens-genes that must induce defense responses in which the nematodes fail to survive [4,14-19,28-30], and it is also proved from the plant-nematode interaction, newly synthesized PR-proteins genes have been found in potato plants infected with the potato-cyst-nematodes *Globodera pallida* and *G. rostochiensis* [4,7,18,19,24,25,28,30,46,47]. It is also reported that Salicylic Acid (SA) increases resistance in plants against RK-diseases by inducing expression and accumulation of pathogenesis-related-I protein (14 kD, PR-I) in the sprayed plant-root and leaves, and it sprays enhances-PAL higher activity in infected-roots [4,7,18,19,24,25,28,39-47].

On PR-proteins-genes of NF

The sixteen PR-proteins-genes out of total eighteen proteins-genes of NF of *M. incognita* in comparison to the uninoculated untreated group, and the molecular weight of the nematode-proteins-genes ranging from 12 kD to 280 kD, already confirmed the potential efficacy of the UHDBM-Gall MT or GMT use as an effective stimulus for the expression of these various new 16 defense-related PR-proteins-genes might be provided resistance to nematode-infection in okra plants due to nematodes present in the gall roots also, and it can be preventing pathogenesis in patients with COVID-19 due to more or less proteins-genes range (240 kD to 26 kD) in different bovine- and human- coronavirus-structural-proteins which send genetic-information, and the SARS-CoV-2 genes may integrate with human-DNA to code the essential nonstructural-proteins like an RNA-polymerase also, and the nematode-proteins-genes slip into human-chromosomes and the diverse immunological-factors on viral-dissemination, immunotherapeutic-options, and inflammatory-responses, and need molecular-characterization and understanding of the human-coronavirus-life-cycle, structural and functional properties of SARS-CoV-2 spike-protein for potential antivirus-drug development, and analysis of therapeutic-targets for SARS-CoV-2 and discovery of potential-drugs by computational-methods, and genomic-characterization and epidemiology of 2019 or genomic-epidemiology has come of age during this pandemic affording to track SARS-CoV-2 sequences helped identify worrying-variants, with implications for virus-origins and receptor-binding also, for preventing the COVID-19, but researchers are blind to emerging-mutations in some-regions, and so it is thought, "the next pandemic by transforming food-systems for affordable healthy-diets" [1-33,39-44]. Recently it may help from the report that cellular senescence (SnC) contributes to inflammation, multiple-chronic-diseases, and age-related dysfunction, and the SnC become hyper-inflammatory in response to pathogen-associated-molecular-patterns (PAMPs), including SARS-CoV-2 Spike-protein-1, increasing expression of viral entry proteins and reducing anti-viral gene-expression in non-SnCs through a paracrine-mechanism [1,2,14-21].

On genetic effects of ORG-pathogenesis-related (PR)-proteins-genes

In the UHDBM-Gall 200C -pretreated Okra Root Galls (ORG), the 22 PR-proteins-genes out of total 23 proteins-genes in comparison to the uninoculated untreated okra plant group, and the molecular weight of the ORGs-proteins-genes ranging from lowest 12.5 kD to highest 295 kD, proved the potential efficacy of the pretreatment-UHDBM -Gall 200C act as the most effective stimulus for the expression of these many new 22 defense-related PR-proteins-genes which might be provided resistance to nematode-infection in okra plant. All the UHDBM; Gall 30, Gall 200C, and Gall 1000C-pretreatments collectively resulted in an increased number of PR-proteins-genes. And the genetic effects of UHDBM; Gall 30, Gall 200C, and Gall 1000C-pretreatments resulted in an increased number of proteins-genes, are thought to induce systemic acquired resistance response of all the pretreated plants through the expression of Pathogenesis-Related (PR)-proteins-genes (22 to 14 numbers), which are more or less similar molecular range (295 kD to 11 kD) of various coronavirus, and it will responsible for preventing RK and COVID-19 like virus diseases by inducing resistance of plants or increasing immunity of animals respectively, and enriched AEHSEMPSTCI [14-21].

Idea for Future Research

The UHDBM; Gall30C, Gall 200C, and Gall 1000C-pretreated ORG-proteins could be induced the production of new defense-related PR-genes in the test plants and might be confirmed, and in near future, synthetic production of the RG-proteins would be the most potential cost-effective personalized-biomedicine OR social vaccine OR vaccine against coronavirus-2 like pandemic diseases by increasing immunity, and helping policy initiative clinical research in all areas in the field of advanced agronomy-plant-breeding-horticulture- agricultural, aquatic sciences, environment, socio-economy, and green-science-technology-communication issues by preventing okra root-knot and COVID-19 also. Thus, RG-proteins will serve as very effective biomedicines that would be the most effective cheapest, non-phytotoxic, non-pollutant, conserve our biodiversity, and this vaccine might be the most effective against the delta variant of coronavirus, and scholars that give good scope for new development and future research in various fields of pathology-medical-pharmacology preventing fungal infections by increasing the post-COVID weakened lungs as well as the immune also [14-21]. And in the near future UHDBM; Gall30C, Gall 200C, and Gall 1000C pretreatment GR may be used as mixing vaccines to provoke potent immune responses for the real-world efficacy and rare side effects. And also help for the future mitigation strategies, and COVID appropriate and risk behavior at slums different areas also [3-19,24,23-30].

Conclusions

The UHDBM Gall 30C, Gall 200C, and Gall 1000C are again highly effective against the root-knot disease of okra, with increasing plant growth and fruit production. The UHDBM; Gall 30C, Gall 200C, and Gall 1000-pretreatments are more effective than the untreated ones, and Gall 200C shows the most potential result in all respects. The genetic-effects of UHDBM thought to induce systemic acquired resistance response of the treated plants through the expression of PR-proteins-genes (22 to 4 numbers), which are more or less similar molecular range (295 kD to 11 kD) of many coronaviruses, and it will be responsible for preventing plant and COVID-19 like variant-virus diseases by inducing defense-resistance or increasing innate-immunity, and enriching green-AEHSEMPSTCI with the toxic-free world, and it may help to develop best potential new preventive treatments methods or drug or vaccines, in the field of '21st-Century COVID-19 like a pandemic in the new normal situation in future. And in the near future pretreatment of gall root may be used as mixing vaccines to provoke potent immune responses for the real-world efficacy and rare side effects, and it also may help for the future mitigation strategies, and COVID appropriate and risk behavior at slums different areas.

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