

Research Article

EFFECT OF NATURAL ELICITORS ON PHYSICAL AND SENSORY QUALITIES OF FENUGREEK (*Trigonella foenum-graecum* L) SPROUTS

ANWAR HUSSAIN AND IMTIYAZ MURTAZA

Division of Post Harvest Technology, Sher e Kashmir University of Agricultural Science and Technology of Kashmir, 180001, India *Corresponding Author: Email- yokcan63101@gmail.com

Received: April 20, 2018; Revised: April 24, 2018; Accepted: April 26, 2018; Published: April 30, 2018

Abstract- Sprouts of fenugreek cultivars *viz.*, '*Methi Local*' and '*Methi Shalimar Improved*' were grown pretreated with natural elicitors such as vitamin C (100 µM and (500 µM)), folic acid ((50 µM) and 100 µM) and chitosan (1000 ppm and 1500 ppm). Physical parameters and sensory properties of these sprouts were studied at different stages (0, 2nd, 5th, 8th and 10th days) of sprouting. The results showed significant variations between the selected cultivars regarding the studied parameters. The pre-treatments increased both the parameters of the sprouts during the sprouting process and among these, chitosan (1500 ppm) was almost the favourable candidate in doing so. In most of the cases, the peak stage where the parameters were maximum was the 8th day of the sprouting. This study concluded that the physical and organoleptic properties of the fenugreek sprouts were enhanced during the process of sprouting and these parameters were further increased by the pre-treatment of selected natural food grade additives.

Keywords- Fenugreek sprouts, Seed treatment, Sprouting, Statistical analysis

Citation: Anwar Hussain and Imtiyaz Murtaza (2018) Effect of Natural Elicitors on Physical and Sensory Qualities of Fenugreek (*Trigonella foenum-graecum* L) Sprouts. International Journal of Microbiology Research, ISSN: 0975-5276 & E-ISSN: 0975-9174, Volume 10, Issue 4, pp.-1160-1163.

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DOI: http://dx.doi.org/10.9735/0975-5276.10.4.1160-1163

Introduction

Fenugreek (*Trigonella foenum-graecum* L.) is a leguminous herb cultivated in India and North African countries. It belongs to the family Fabaceae. India is the major producer of fenugreek and its main consumer for culinary and medicinal uses. The seeds are used as spices worldwide, whereas the leaves are used as green leafy vegetables in the diet. Fenugreek seeds are bitter to taste and are known for a long time for their medicinal qualities. The seeds of fenugreek are used as a spice for seasoning, a flavoring agent and in comparatively larger quantities in making soups and pan cakes. In the indigenous system of medicine in India, it is effective against anorexia and is a gastric stimulant [1, 2].

Sprouts are one of the most complete and nutritionally beneficial of all foods. Their nutritional value was discovered by the Chinese thousands of years ago. Recently, in USA, numerous scientific studies suggest the importance of sprouts in a healthy diet. Such type of functional food components has been associated with the prevention and/or treatment of at least four of the leading causes of death in USA, cancer, cardiovascular disease, diabetes and hypertension and with the prevention and/or treatment of other medical ailments such as osteoporosis, neural tube defects, arthritis abnormal and bowel function. Although the use of sprouts as a food source for man is old as man's use of seeds, it is only in recent times that science has begun to unravel the chemistry of sprouting seed and its potential significance in both human and animal nutrition.

The objective of the current research was to examine the effect of exogenously applied natural elicitors which would be food grade in quality on physical properties and quality of fenugreek sprouts. The elicitors used were vitamin C, folic acid and chitosan. Both vitamin C and folic acid are water soluble vitamins and chitosan is a natural polymer, appear to be promising candidates regarding the physical changes in fenugreek sprouts.

Materials and Methods

The experimental work was carried out in the Biochemistry laboratory of the Division of Post Harvest Technology, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir, J & K, India during the year 2016-17.

Seed treatment and sprouting

Seeds of the fenugreek cultivars *i.e.*, '*Methi Local*' and '*Methi Shalimar Improved*' were obtained from the Division of Vegetable Science, SKUAST-K. The elicitors used in the study were folic acid (Titan Biotech Ltd.), vitamin C (S.d. fine-chem Ltd.) and chitosan (HiMedia Laboratories Pvt. Ltd.) as pretreatments. Dry seeds of the selected cultivars were soaked in conical flasks containing different pre-treatments *viz.*, distilled water (control), vitamin C (100 μ M), vitamin C (500 μ M), folic acid (50 μ M), folic acid (100 μ M), chitosan (1000 ppm) and chitosan (1500 ppm). The flasks were then placed on a shaker at a speed of 120 rpm at room temperature for 18 hours. The pre-soaked seeds were washed in distilled water and germinated in glass jars. The germinating seeds were kept moist with distilled water and sprouts were raised for 10 days. Sprouts were collected on 0, 2nd, 5th, 8th and 10th day of sprouting for analysis.

Physical changes

Sprout length was recorded on representative sample of a sprout in triplicate using a measuring scale. A representative sample of a sprout in triplicate was weighed on an electronic balance and average sprout weight was calculated. For measuring physiological change (loss or gain) in weight from each treatment, 3 sprouts in 3 replications (1 sprout/replication) were taken. Per cent change in weight was calculated by subtracting the final weight of sprouts from initial weight of sprouts as per equation given below

 $PCW(\%) = \frac{\text{Original weight} - \text{subsequent weight at analysis}}{O(1) + 1} \times 100$

Original weight Moisture content in samples was determined by vacuum oven method [3]. 500 mg of sample in triplicate were weighed accurately and dried at 70 ± 2 °C for 12 hours to constant weight under vacuum of 750 mm Hg. The loss in weight was determined to calculate the moisture content.

Sensory evaluation

Treated samples were evaluated for quality by a panel of 10 judges for colour, taste, aroma, texture and overall acceptability using 4-point scale.

- 4. Excellent
- 3. Good
- 2. Fair
- 1. Poor

Statistical analysis of data

Data obtained were subjected to statistical analysis following the CRD model and the variation among the treatment means was tested for significance of analysis of variance technique described by Gomez and Gomez [4]. Levels of significance used for T-test was $p \le 0.05$ from the table given by Fisher [5]. The critical difference has also been worked out.

Results and discussion

Sprout length

Study with respect to sprout length indicated that individual length of fenugreek sprouts differed between the two varieties of fenugreek on all days except on 0 day [Table-1]. Sprouting resulted in a continuous increase in sprout length in both the varieties and on 8th day (peak day) the variety '*Methi Shalimar Improved*' had maximum length of 101.71 mm as compared to variety '*Methi Local*' having length of 99.43 mm. Increase in length has been reported by Mc Kinney, *et al.* [6] in soyabean, No, *et al.* [7] in soyabean and Cho, *et al.* [8] in sunflower. Various pre-treatments increase the pace of length during sprouting. Among all the treatments, Chitosan (1500 ppm) was able to maintain the maximum sprout length against control (water treated) samples with minimum sprout length on all days. The former resulted in 105.5 mm length as compared to the latter with 94.5 mm on 8th day of sprouting. Lee, *et al.* [9] and Choi, *et al.* [10] found that chitosan treatment increased growth rate of soyabean sprouts.

Table-1 Effect of chemical treatments and storage on length of fenugreek sp	routs

Treatment	Length (mm)						
	0 day	2 nd day	5 th day	8 th day	10 th day		
Variety							
V1(Methi Local)	4.21	41.42	67.57	99.43	98.86		
V2(Methi Shalimar Improved)	4.23	44.42	72.57	101.71	100.86		
CD (p≤0.05)	NS	1.04	0.62	1.13	0.96		
Chemical							
T ₀ (Control)	4.19	39.50	63.50	94.50	93.50		
T₁(Vitamin C (100 µM)	4.20	41.50	67.00	98.00	97.00		
T ₂ (Vitamin C (500 µM)	4.22	43.50	70.50	101.00	99.50		
T ₃ (Folic acid (50 µM)	4.23	42.50	69.50	99.50	99.00		
T ₄ (Folic acid (100 µM)	4.24	44.50	72.50	102.50	101.50		
T₅(Chitosan (1000 ppm)	4.24	43.50	72.50	103.00	103.00		
T₀(Chitosan (1500 ppm)	4.25	45.50	75.00	105.50	105.50		
CD (p≤0.05)	NS	1.96	1.16	2.12	1.80		

Sprout weight

In spite of minimum sprout length, the variety '*Methi Local*' possessed maximum sprout weight on 2nd and 5th day of sprouting than the variety '*Methi Shalimar Improved*' [Table-2]. The weight of both the varieties were found to be increased during sprouting process and maximum value was observed on 8th day with '*Methi Local*' having 116.29 mg as compared to that of '*Methi Shalimar Improved*' having 115.71 mg. The findings are in agreement with Cevallos-Casals and Cisneros-

Zevallos [11] in fava bean, soyabean, mungbean, fenugreek, lentil, alfalfa and sunflower, No, *et al.* [7] in soyabean and Cho, *et al.* [8] in sunflower. The various pre-treatments significantly influence the pace of increase in sprout length and Folic acid (100 μ M) resulted in maximum and Vitamin C (100 μ M) resulted in minimum length on all days. In previous studies, it has been shown that the treatment of pea seedlings with exogenous growth factors such as folic acid and vitamin C resulted in an increase in agronomic vigour properties [12].

Table-2 Effect of chemical treatments and storage on weight of fenugreek sprouts

Treatment	Weight (mg)					
	0 day	2 nd day	5 th day	8 th day	10 th day	
Variety						
V1(Methi Local)	46.14	69.85	93.42	116.29	115.00	
V ₂ (Methi Shalimar Improved)	46.08	68.28	90.42	115.71	114.52	
CD (p≤0.05)	NS	0.98	0.98	NS	NS	
Chemical						
T₀(Control)	46.00	69.00	91.00	115.00	113.00	
T₁(Vitamin C (100 μM)	46.00	67.50	89.00	112.50	111.00	
T ₂ (Vitamin C (500 µM)	46.53	68.50	91.00	116.00	114.00	
T ₃ (Folic acid (50 µM)	46.00	71.00	94.00	118.00	116.50	
T₄(Folic acid (100 μM)	46.26	73.50	97.00	121.00	119.50	
T₅(Chitosan (1000 ppm)	46.00	68.00	90.00	114.00	114.00	
T ₆ (Chitosan (1500 ppm)	46.00	68.00	91.50	115.50	115.50	
CD (p≤0.05)	NS	1.83	1.84	2.14	2.23	

Change in weight

The data in [Table-3] revealed that, there was a gain in weight during sprouting up to 8th day followed by weight loss on 10th day. But the weight gain continuously decreased from 2nd day up to the above-mentioned day. The gain in weight of *'Methi Local'* was significantly higher (33.87 %) as compared to a lower gain (32.61 %) in *'Methi Shalimar Improved'* on 2nd day as well as on all other days. These values reached up to 21.84 % in the former and 19.66 % in the latter variety. The loss in weight was 1.5 % in case of *'Methi Local'* and 1.39% in *'Methi Shalimar Improved'*. Samples treated with Folic acid (100 µM) resulted in highest gain in weight (37.18 %) on 2nd day against the lowest gain of 30.28 % in Chitosan (1500 ppm). However, control samples resulted in highest loss of 1.78 % against the minimum loss of 0.97 % on 10th day. Lee, *et al.* [9] and Choi, *et al.* [10] found that chitosan treatment increased weight of soyabean sprouts.

Table-3 Effect of chemical treatments and storage on change in weight of fenugreek sprouts

Treatment	Gain ir	Gain in weight (%)						
	0day	2 nd day	5 th day	8 th day	10 th day			
Variety								
V ₁ (Methi Local)	0.00	33.87	25.23	21.84	1.50			
V2(Methi Shalimar Improved)	0.00	32.61	24.49	19.66	1.39			
CD (p≤0.05)	NS	0.82	0.68	0.47	NS			
Chemical								
T₀(Control)	0.00	33.31	24.44	21.05	1.78			
T₁(Vitamin C (100 µM)	0.00	32.05	23.93	20.70	1.31			
T ₂ (Vitamin C (500 µM)	0.00	32.84	24.72	21.54	1.75			
T₃(Folic acid (50 µM)	0.00	35.19	24.07	19.83	1.44			
T ₄ (Folic acid (100 µM)	0.00	37.18	24.58	20.33	1.70			
T₅(Chitosan (1000 ppm)	0.00	30.28	25.67	20.49	1.20			
T ₆ (Chitosan (1500 ppm)	0.00	31.83	26.65	21.30	0.97			
CD (p≤0.05)	NS	1.54	1.28	0.89	NS			

Moisture content

It is evident from the data given in [Table-4] that, the moisture content in samples of variety '*Methi Local*' had maximum moisture content as compared to the variety '*Methi Shalimar Improved*' of on all days except on 0 day. There was a continuous increase in the moisture content of both the varieties up to 8th day on which moisture content was 85.78 % in '*Methi Local*' and 84.58 % in '*Methi Shalimar*

Improved'. Increase in moisture content during sprouting are in confirmity with that of Cevallos-Casals and Cisneros-Zevallos [11] in fava bean, soyabean, mungbean, fenugreek, lentil, alfalfa and sunflower. The treatment Folic acid (100 μ M) resulted in maximum influence on moisture content of sprouts and the minimum influence was shown by Chitosan (1000 ppm) on all days. The values were maximum on the peak day *i.e.*, 8th day (87.86 % and 82.78 % for the mentioned parameters respectively.

Table-4 Effect of chemical treatments and storage on moisture content of fenugreek sprouts

Treatment	Moisture (%)					
	0 day	2 nd day	5 th day	8 th day	10 th day	
Variety						
V1(Methi Local)	20.11	50.81	77.07	85.78	85.12	
V ₂ (Methi Shalimar Improved)	20.07	49.58	75.90	84.58	83.85	
CD (p≤0.05)	NS	0.16	0.23	0.16	0.16	
Chemical						
T ₀ (Control)	20.00	49.74	74.52	84.59	83.55	
T₁(Vitamin C (100 µM)	20.00	48.74	73.92	83.86	82.85	
T ₂ (Vitamin C (500 µM)	20.37	50.27	75.67	85.45	84.59	
T ₃ (Folic acid (50 µM)	20.00	51.53	79.29	86.37	85.56	
T₄(Folic acid (100 μM)	20.28	53.29	81.00	88.99	87.86	
T ₅ (Chitosan (1000 ppm)	20.00	47.99	73.85	82.78	82.78	
T ₆ (Chitosan (1500 ppm)	20.00	49.83	76.15	84.21	84.21	
CD (p≤0.05)	NS	0.31	0.43	0.31	0.31	

Sensory qualities

Colour

It can be speculated from the data given in [Table-5] that the sensory quality based on organoleptic scoring of colour of sprouts from fenugreek varieties differed non-significantly on all days of observation. However, colour of both the varieties got improved up to 8th day of sprouting then decrease sharply on last stage *i.e.*, on 10th day. The present findings are in agreement with Khalil *et al.* [12] in chickpea cultivars. Best quality in terms of colour was observed in both concentrations (1000 and 1500 ppm) of Chitosan on all days and also in both concentration of Vitamin C (100 and 500 μ M) and Folic acid (50 and 100 μ M) on 8th day of sprouting. The highest score of 4 was observed on 8th day by these elicitors. However, control samples resulted in poorest sensory quality on all days.

Table-5 Effect of chemical treatments and storage on colour of fenugreek sprouts

Treatment	Colour					
	0 day	2 nd day	5 th day	8 th day	10 th day	
Variety						
V ₁ (Methi Local)	1.00	2.55	3.50	3.97	3.77	
V ₂ (Methi Shalimar Improved)	1.00	2.61	3.52	3.98	3.82	
CD (p≤0.05)	NS	NS	NS	NS	NS	
Chemical						
T ₀ (Control)	1.00	2.00	3.05	3.85	3.57	
T₁(Vitamin C (100 µM)	1.00	2.45	3.20	4.00	3.67	
T ₂ (Vitamin C (500 µM)	1.00	2.55	3.40	4.00	3.72	
T ₃ (Folic acid (50 µM)	1.00	2.55	3.40	4.00	3.77	
T ₄ (Folic acid (100 µM)	1.00	2.75	3.50	4.00	3.82	
T₅(Chitosan (1000 ppm)	1.00	2.90	4.00	4.00	4.00	
T ₆ (Chitosan (1500 ppm)	1.00	2.90	4.00	4.00	4.00	
CD (p≤0.05)	NS	0.24	0.16	NS	0.11	

Taste

Non significant differences were observed in the interactions of varieties and treatments as per the data given in [Table-6]. As for the taste of sprouts is concerned, there was again an unremarkable difference between the two varieties of fenugreek *viz.*, '*Methi Local*' and '*Methi Shalimar Improved*' on all the stages. However same trend shown by colour was found by the sensory parameter, texture *i.e.*, continuous increase up to 8th day followed by a sharp decrease.

Increase in taste has been reported by Khalil, *et al.* [12] in chickpea cultivars. It was the treatment Chitosan (1500 ppm) which resulted in maximum scoring on all days of sprouting and the treatment control resulted in lowest scoring of taste. It was the 8^{th} day where the discussed treatments had maximum effect.

Table-6 Effect of chemical treatments and storage on taste of fenugreek sprouts

Treatment	Taste						
	0day	2 nd day	5 th day	8 th day	10 th day		
Variety							
V1(Methi Local)	1.00	2.14	2.86	3.27	2.50		
V ₂ (Methi Shalimar improved)	1.00	2.25	2.94	3.36	2.57		
CD (p≤0.05)	NS	NS	NS	NS	NS		
Chemical							
T ₀ (Control)	1.00	1.60	2.55	3.05	2.05		
T₁(Vitamin C (100 µM)	1.00	2.15	2.75	3.15	2.45		
T ₂ (Vitamin C (500 µM)	1.00	2.25	2.90	3.25	2.55		
T₃(Folic acid (50 µM)	1.00	2.25	2.85	3.35	2.55		
T ₄ (Folic acid (100 µM)	1.00	2.35	3.05	3.43	2.65		
T₅(Chitosan (1000 ppm)	1.00	2.35	3.07	3.43	2.70		
T ₆ (Chitosan (1500 ppm)	1.00	2.45	3.15	3.55	2.80		
CD (p≤0.05)	NS	0.23	0.17	0.21	0.23		

Aroma

A higher scoring of aroma in variety '*Methi Shalimar Improved*' as compared to the variety '*Methi Local*' was obtained in present study except on 0 day, as per data of [Table-7]. Also, like other parameters, 8th day was the peak stage of sprouting where '*Methi Local*' had aroma score of 3.42 and '*Methi Shalimar Improved*' had score of 3.50. Increase in colour during sprouting are in conformity with that of Khalil, *et al.* [12] in chickpea cultivars. It is evident from the results regarding the aroma of fenugreek sprouts that Chitosan (1500 ppm) was proved to be the best treatment on 5th, 8th and 10th days where as control was the poorest treatment on these very days.

Table-7 Effect of chemical treatments and storage on aroma of fenugreek sprouts

Treatment	Aroma						
	0day	2 nd day	5 th day	8 th day	10 th day		
Variety							
V1(Methi Local)	1.00	2.04	3.00	3.42	2.62		
V ₂ (Methi Shalimar Improved)	1.00	2.06	3.05	3.50	2.67		
CD (p≤0.05)	NS	0.03	0.04	0.06	0.02		
Chemical							
T ₀ (Control)	1.00	2.01	2.80	3.25	2.52		
T₁(Vitamin C (100 µM)	1.00	2.03	3.01	3.42	2.56		
T ₂ (Vitamin C (500 µM)	1.00	2.06	3.05	3.45	2.60		
T ₃ (Folic acid (50 µM)	1.00	2.05	3.05	3.48	2.61		
T ₄ (Folic acid (100 µM)	1.00	2.08	3.07	3.52	2.67		
T ₅ (Chitosan (1000 ppm)	1.00	2.06	3.09	3.54	2.75		
T ₆ (Chitosan (1500 ppm)	1.00	2.08	3.11	3.57	2.81		
CD (p≤0.05)	NS	NS	0.08	0.11	0.07		

Texture

Non-significant differences were observed in the interaction effects of varieties and treatments [Table-8]. The two varieties of fenugreek in terms of sprout texture were non-significantly different. But a continuous increase in the said parameter was observed and the peak stage was the 8th day. Khalil *et al.* [12] reported increase in texture score during sprouting in chickpea cultivars. Among the various pre-treatments Chitosan (1500 ppm) was the most effective elicitor for maintaining the sprout texture against the least effective treatment control.

Table-8 Effect of chemical treatments and storage on texture of fenugreek sprouts

Treatment	Texture					
	0 day	2 nd day	5 th day	8 th day	10 th day	
Variety						
V1(Methi Local)	1.00	2.58	3.45	3.85	3.02	
V ₂ (Methi Shalimar Improved)	1.00	2.67	3.53	3.90	3.11	
CD (p≤0.05)	NS	NS	NS	NS	NS	
Chemical						
T₀(Control)	1.00	2.05	3.10	3.85	2.10	
T₁(Vitamin C (100 µM)	1.00	2.62	3.47	3.92	2.85	
T ₂ (Vitamin C (500 µM)	1.00	2.72	3.57	3.97	2.95	
T₃(Folic acid (50 µM)	1.00	2.50	3.27	3.67	2.75	
T ₄ (Folic acid (100 µM)	1.00	2.57	3.32	3.72	2.85	
T₅(Chitosan (1000 ppm)	1.00	2.90	3.75	4.00	4.00	
T ₆ (Chitosan (1500 ppm)	1.00	3.05	3.95	4.00	4.00	
CD (p≤0.05)	NS	0.18	0.18	0.21	0.24	

Overall acceptability

Perusal of the data given in [Table-9] reported that, a higher value of overall acceptability was observed in '*Methi Shalimar Improved*' on 2^{nd} and 10^{th} day as compared to the variety '*Methi Local*' where these two varieties were non-significantly different on 0, 5th and 8th days.

Continuous increase up to 8th day was observed followed by a sharp decrease on the last day. The increase in overall acceptability is in consonance with findings of Khalil, *et al.* [12] in chickpea cultivars. Chitosan (1500 ppm) treated samples resulted in maximum score of overall acceptability on all days where as control treated samples resulted in minimum effect on overall acceptability of fenugreek sprouts.

Table-9 Effect of chemical treatments and storage on overall acceptability of fenugreek sprouts

Treatment	Overall acceptability					
	0 day	2 nd day	5 th day	8 th day	10 th day	
Variety						
V1(Methi Local)	1.00	2.33	3.21	3.63	2.98	
V ₂ (Methi Shalimar Improved)	1.00	2.40	3.26	3.67	3.04	
CD (p≤0.05)	NS	0.04	NS	NS	0.05	
Chemical						
T₀(Control)	1.00	1.91	2.87	3.46	2.56	
T₁(Vitamin C (100 µM)	1.00	2.31	3.12	3.62	2.88	
T ₂ (Vitamin C (500 µM)	1.00	2.39	3.23	3.67	2.95	
T ₃ (Folic acid (50 µM)	1.00	2.34	3.14	3.62	2.92	
T ₄ (Folic acid (100 µM)	1.00	2.43	3.23	3.67	3.00	
T₅(Chitosan (1000 ppm)	1.00	2.55	3.52	3.74	3.36	
T ₆ (Chitosan (1500 ppm)	1.00	2.61	3.55	3.78	3.40	
CD (p≤0.05)	NS	0.09	0.11	0.10	0.06	

Conclusion

There was increase in both physical and sensory properties of fenugreek during the process of sprouting. The elicitors used in the study also resulted into the increase in physical as well as sensory parameters of the fenugreek sprouts. The most effective day for eliciting the selected parameters in sprouts by using the pre-treatments was the 8th day in most of the cases.

Application of research: Sprouts can be used instead of vegetables owing to its freshness and organoleptically acceptable. Because of its, health benefits besides nutritional package as proven during extensive research previously done by several researchers, it can be a promising functional food.

Research category: Sprouts; chitosan; fenugreek; organoleptic properties; folic acid

Abbreviations: µM: Micro-molar, ppm: Parts Per Million, CRD: Completely

Randomized Design, Hg: Mercury

Acknowledgement/Funding: Authors are thankful to Sher-e Kashmir University of Agricultural Science and Technology of Kashmir, 180001, India for financial support to carry the present investigation.

Research Guide or Chairperson of research: Dr. Imtiyaz Murtaza

University: Sher-e Kashmir University of Agricultural Science and Technology of Kashmir, 180001, India. Research project name or number: Nil

Author Contributions: All authors equally contributed to the research

Author statement: All authors read, reviewed, agree and approved the final manuscript

Conflict of Interest: None declared

Ethical approval: This article does not contain any studies with human participants or animals performed by any of the authors.

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