

### REPUBLIC OF SOUTH AFRICA

#### REPUBLIEK VAN SUID AFRIKA

PATENTS ACT, 1978

## **CERTIFICATE**

In accordance with section 44 (1) of the Patents Act, No. 57 of 1978, it is hereby certified that:

#### ZHEJIANG UNIVERSITY

Has been granted a patent in respect of an invention described and claimed in complete specification deposited at the Patent Office under the number

#### 2021/10714

A copy of the complete specification is annexed, together with the relevant Form P2.

In testimony thereof, the seal of the Patent Office has been affixed at Pretoria with effect from the 28th day of April 2022

Registrar of Patents

## REPUBLIC OF SOUTH AFRICA PATENTS ACT, 1978 REGISTER OF PATENTS

Official application No.	Lodging date: Prov	isional Acceptance date			
21 01 2021/10714	22		47 28 March 2022		
International classification	Lodging date: Nat	ional phase	Granted date		
51 A23B	23 21 December	2021	28 April 2022		
71 Full name(s) of applicant(s)/P	atentee(s):				
Zhejiang University					
71 Applicant(s) substituted:			Date registrered		
71 Assignee(s):			Date registrered		
72 Full name(s) of inventor(s):					
(1) SUN, Chongde; (2) CAO, Jinpir	ng; (3) WU, Yu; (4) WA	NG, Yue; (5) KANG	G, Chen		
Priority claimed: Country	y   N	lumber	Date		
54 Title of invention					
METHOD OF MONITORING ACID F	ROT DURING CITRUS S	TORAGE AND TRAI	NSPORTATION		
Address of applicant(s)/patentee					
866 Yuhangtang Road, Xihu Disti	rict, Hangzhou , 31005	8, China			
74 Address for service					
Sibanda and Zantwijk, Oaktree C		, Oaklands (PO Bo	x 1615 Houghton 2041),		
Johannesburg, 2192, SOUTH AFR Reference no.: PT CP ZA000024.					
Neterence no.: 11_cr_2A000024	25 ([11315. ])				
61 Patent of addition No.		Date of any chang	Date of any change		
or in decire of addition no.		Date of any chang	<i>j</i> ~		
Fresh application based on.		Date of any change			
- Son approach subsection					

#### FORM P7

## REPUBLIC OF SOUTH AFRICA PATENTS ACT, 1978 COMPLETE SPECIFICATION

[Section 30(1) - Regulation 28]

OFFICIAL APPLICATION NO.		LODGING DATE				
21	01 2021/10714	22 21 December 2021				
51	INTERNATIONAL CLASSIFICATION A23B					
FULL NAME(S) OF APPLICANT(S)						
71	Zhejiang University					
FULL NAME(S) OF INVENTORS(S)						
72	SUN, Chongde CAO, Jinping WU, Yu WANG, Yue KANG, Chen					
TITLE OF INVENTION						
54	4 METHOD OF MONITORING ACID ROT DURING CITRUS STORAGE AND TRANSPORTATION					

# METHOD OF MONITORING ACID ROT DURING CITRUS STORAGE AND TRANSPORTATION

#### **TECHNICAL FIELD**

[01] The present disclosure belongs to a field of post-harvest preservation of fruits, and relates to a method of monitoring acid rot during citrus storage and transportation.

#### BACKGROUND ART

- [02] Acid rot is also known as white mildew and wet rot and is one of the main diseases in the process of storage and transportation of citrus fruits after harvest, which is caused by citrus Geotrichum candidum. Once the acid rot develops, the fruit quickly rots into a sticky mass like a rotten persimmon within a few days, and the juice flows out and emits a strong sour smell. The juice contains a large number of pathogenic bacteria, and the fruits near diseased fruits are very susceptible to infection, resulting in large-scale decay of citrus. At present, types of preservatives that can be used for acid rot prevention and treatment are limited, and the disease course develops too fast to make the prevention and treatment difficult. Therefore, in an actual storage and logistics process, once acid rot occurs, the diseased fruits should be picked out in time, and the storage, transportation and sales strategy should be adjusted in time according to the development of the disease to sell as soon as possible.
- [03] During storage or long-distance transportation of citruses, a large number of fruits are stacked in a relatively confined space, which is more likely to cause a large area of acid rot. At present, the actual production is mainly through regular inspections by personnel to monitor the occurrence of diseases. The acid rot fruits will emit a pungent sour smell, which is easy to identify. However, the human sense of smell is easy to adapt to environment and sensitivity decreases rapidly. In order to find diseased fruits from densely stacked fruits, a small number of samples need to be sampled and observed one by one. For the fruits located under the stack, they need to be removed one by one in order to find the existence of diseased fruits. Therefore, it is difficult to find the fruits with acid rot during storage and transportation. Once it appears, a large area of rot has often appeared, and it is difficult to recover the economic loss. Utilizing its characteristic smell to develop more sensitive and objective detection techniques is an effective means to detect citrus acid rot in time.

#### **SUMMARY**

- **[04]** The present disclosure utilizes a characteristic of citrus acid rot fruits with easy-to-identify sour smell to propose a sensitive and objective disease monitoring method, and aims to provide a technology solution for early detection of acid rot fruits during mass storage and transportation of citrus.
- [05] The technical solution of the present disclosure includes the following steps:
- [06] a, dividing citrus acid rot fruits into disease courses according to a size of a disease spot;

- [07] b, placing diseased fruits of different disease courses in a confined space to simulate a storage and transportation environment, and recording a ratio of a diseased area of the fruits placed in the confined space to a total area of the fruits as  $Y_i$ ;
- [08] c, using an electronic nose collector to insert into an airtight container with fruits to extract gas components for electronic nose detection, wherein a total response time of a single electronic nose is t, and a data collection interval is once per second, the electronic nose includes n sensors generating corresponding sensor data for a to-be-monitored gas respectively;
- [09] d, collecting electronic nose data of fruits of each disease course, wherein for a j-th course,  $0 \le j \le 5$ , for an i-th sensor signal of the electronic nose,  $1 \le i \le n$ ; extracting sensor feature values, wherein the sensor feature values comprise a maximum sensor data value  $F_{max,i,j}$ , a sum of sensor data values per second  $F_{sum,i,j}$ , the sensor data at 0.5 t  $F_{0.5t,i,j}$ , the sensor data at t  $F_{t,i,j}$ ;
- [10] e, taking each of the sensor feature values of the electronic nose as an independent variable, the ratio of the diseased area of the fruits placed in the confined space to the total area of the fruits as a dependent variable of a model to establish a prediction model by using partial least squares regression, equations are as follows:

```
[11] Y = a_{max,1}F_{max,1} + a_{sum,1}F_{sum,1} + a_{0.5t,1}F_{0.5t,1} + a_{t,1}F_{t,1}
```

[12] 
$$+ a_{\text{max},2}F_{\text{max},2} + a_{\text{sum},2}F_{\text{sum},2} + a_{0.5t,2}F_{0.5t,2} + a_{t,2}F_{t,2}$$

[13] .....

[14] 
$$+ a_{\text{max},n}F_{\text{max},n} + a_{\text{sum},n}F_{\text{sum},n} + a_{0.5t,n}F_{0.5t,n} + a_{t,n}F_{t,n}$$

[15] + b

- [16] wherein,  $a_{\text{max,i}}$ ,  $a_{\text{sum,i}}$ ,  $a_{0.5t,i}$  and  $a_{t,i}$  are coefficients corresponding to the sensor feature values, and b is an intercept.
- [17] As a preference, the sensors are 12 sensors in built-in sensors of the electronic nose (whose model is Alpha-Mos FOX4000), including TA/2, T40/1, T40/2, P30/2, P40/2, P30/1, PA/2, T70/2, P40/1, P10/2, P10/1 and T30/1.
- [18] As a preference, the total response time t of a single electronic nose is 120s in step c.
- [19] The present disclosure establishes a model for the acid rot in storage and transportation process of citrus fruits. At each node in the storage and transportation process of the citrus, the occurrence of diseases in the storage and transportation space is evaluated according to the established model, so as to adjust the storage, transportation and sales strategy in time.

#### BRIEF DESCRIPTION OF THE DRAWINGS

- [20] FIG. 1 is a table of various sensors and corresponding performance of an electronic nose used in the present disclosure;
- [21] FIG. 2 is a typical response curve of 18 sensors of an electronic nose to citrus samples used in the present disclosure;
- [22] FIG. 3 is a radar chart of multi-sensor response of citrus acid rot fruits and normal fruits;
- [23] FIG. 4 is correlation coefficients between sensor response values of an electronic nose and acid rot disease course development of citrus fruits.

#### DETAILED DESCRIPTION OF THE EMBODIMENTS

- [24] The present disclosure will be further described with reference to the drawings and embodiments. It should be understood that these embodiments are only for illustrative purposes, and are not used to limit the scope of application of the present disclosure.
- [25] Embodiments:
- [26] Fruit material: citrus ripe fruits of Citrus unshiu Marc. cv. Nobis from Taizhou, Zhejiang (harvested on October 12, 2018), Citrus reticulata Blanco cv. Ponkan from Quzhou, Zhejiang (harvested on December 21, 2018), "Newhall" navel orange (Citrus sinensis Osbeck cv. Newhall) from Yichang, Hubei (harvested on December 19, 2018), and "Citrus Hybrida" from Ningbo, Zhejiang (December 20, 2018) are used as test materials.
- [27] Test methods: after four kinds of citrus fruits are inoculated with acid rot, 9 fruits of uniform size are selected from each group and placed in a tasteless closed lock box with a breathing valve to simulate a storage and transportation environment, 3 per box, 3 boxes per group. According to a size of a disease spot, disease courses are divided into: normal fruit, grade I diseased fruit (with disease spot diameter 10 mm), grade II diseased fruit (with disease spot diameter 30 mm), grade IV diseased fruit (with disease spot diameter 50 mm), grade IV diseased fruit (with disease spot diameter 70 mm, half-fruit rot) and grade V diseased fruit (whole fruit rot). At different stages of the development of the disease courses, the gas in the environment inside the boxes is collected and quickly injected into an electronic nose (Alpha-Mos FOX4000, France) for detection. A total time of data collection is 120s, and a reading interval is 1 time per second.
- [28] Test results:
- [29] 18 sensors in the electronic nose Alpha-Mos FOX4000 have different sensitivity to citrus acid rot. Some sensors are not sensitive to volatile smell of acid rot, and are slightly redundant in electronic nose detection and data analysis. In order to better screen out sensors that are sensitive to citrus acid rot, the four types of citrus are inoculated with pathogenic bacteria, and signal values of the electronic nose are detected every 24 hours, and the signal values are used for the electronic nose radar analysis for more intuitively exploring response degree of each sensor to the smell of acid rot. The results show that 12 sensors TA/2, T40/1, T40/2, P30/2, P40/2, PA/2, P30/1, T70/2, P10/2, P40/1, P10/1, T30/1 responds to the smell of acid rot fruits (FIG. 1 ~ FIG. 3).
- [30] Further response signal feature value data of 12 sensors with response values are extracted, and performed linear fitting and correlation analysis. The results show that correlation coefficients between response values of the 12 sensors and spot size of acid rot all exceed 0.7, indicating that response intensity of the electronic nose sensor has a high correlation with the disease course of acid rot (FIG. 4).
- [31] Then electronic nose data of fruits of each disease course are collected, wherein for the j-th course,  $0 \le j \le 5$ , for the i-th sensor signal of the electronic nose,  $1 \le i \le n$ ; sensor feature values are extracted, wherein the sensor feature values include a maximum sensor data value  $F_{max,i,j}$ , a sum of sensor data values per second  $F_{sum,i,j}$ , the

sensor data at  $0.5 t F_{0.5t,i,j}$ , the sensor data at  $t F_{t,i,j}$ ;

[32] each of the sensor feature values of the electronic nose is taken as an independent variable, a ratio of a diseased area of the fruits placed in a confined space to a total area of the fruits is taken as a dependent variable of a model to establish a prediction model by using partial least squares regression, equations are as follows:

```
 \begin{array}{lllll} \textbf{[33]} & Y = a_{max,1}F_{max,1} + a_{sum,1}F_{sum,1} + a_{0.5t,1}F_{0.5t,1} + a_{t,1}F_{t,1} \\ \textbf{[34]} & + a_{max,2}F_{max,2} + a_{sum,2}F_{sum,2} + a_{0.5t,2}F_{0.5t,2} + a_{t,2}F_{t,2} \\ \textbf{[35]} & \dots \\ \textbf{[36]} & + a_{max,n}F_{max,n} + a_{sum,n}F_{sum,n} + a_{0.5t,n}F_{0.5t,n} + a_{t,n}F_{t,n} \\ \textbf{[37]} & + b \end{array}
```

[38] wherein,  $a_{\text{max,i}}$ ,  $a_{\text{sum,i}}$ ,  $a_{0.5t,i}$  and  $a_{t,i}$  are coefficients corresponding to the sensor feature values, and b is an intercept.

#### WHAT IS CLAIMED IS:

- 1. A method of monitoring acid rot during citrus storage and transportation, comprising following steps:
- a, dividing citrus acid rot fruits into disease courses according to a size of a disease spot;
- b, placing diseased fruits of different disease courses in a confined space to simulate a storage and transportation environment, and recording a ratio of a diseased area of the fruits placed in the confined space to a total area of the fruits as  $Y_i$ ;
- c, using an electronic nose collector to insert into an airtight container with fruits to extract gas components for electronic nose detection, wherein a total response time of a single electronic nose is t, and a data collection interval is once per second, the electronic nose comprises n sensors generating corresponding sensor data for a to-be-monitored gas respectively;
- d, collecting electronic nose data of fruits of each disease course, wherein for a j-th course,  $0 \le j \le 5$ , for an i-th sensor signal of the electronic nose,  $1 \le i \le n$ ; extracting sensor feature values, wherein the sensor feature values comprise a maximum sensor data value  $F_{\text{max},i,j}$ , a sum of sensor data values per second  $F_{\text{sum},i,j}$ , the sensor data at 0.5 t  $F_{0.5t,i,j}$ , the sensor data at 0.5 t
- e, taking each of the sensor feature values of the electronic nose as an independent variable, the ratio of the diseased area of the fruits placed in the confined space to the total area of the fruits as a dependent variable of a model to establish a prediction model by using partial least squares regression, equations are as follows:

$$\begin{split} Y &= a_{\text{max},1} F_{\text{max},1} + a_{\text{sum},1} F_{\text{sum},1} + a_{0.5t,1} F_{0.5t,1} + a_{t,1} F_{t,1} \\ &+ a_{\text{max},2} F_{\text{max},2} + a_{\text{sum},2} F_{\text{sum},2} + a_{0.5t,2} F_{0.5t,2} + a_{t,2} F_{t,2} \\ &\cdots \\ &+ a_{\text{max},n} F_{\text{max},n} + a_{\text{sum},n} F_{\text{sum},n} + a_{0.5t,n} F_{0.5t,n} + a_{t,n} F_{t,n} \\ &+ b \end{split}$$

wherein,  $a_{max,i}$ ,  $a_{sum,i}$ ,  $a_{0.5t,i}$  and  $a_{t,i}$  are coefficients corresponding to the sensor feature values, and b is an intercept.

- 2. The method of monitoring acid rot during citrus storage and transportation according to claim 1, wherein the sensors are 12 sensors in built-in sensors of the electronic nose (whose model is Alpha-Mos FOX4000), comprising TA/2, T40/1, T40/2, P30/2, P40/2, P30/1, PA/2, T70/2, P40/1, P10/2, P10/1 and T30/1.
- 3. The method of monitoring acid rot during citrus storage and transportation according to claim 1, wherein, the total response time t of a single electronic nose is 120 s in step c.

DA van Zantwijk

Sibanda & Zantwijk Patent Attorneys

## **DRAWINGS**

Sensor name	Performance description		
T30/1	Sensitive to organic compounds		
P10/1	Sensitive to combustible gases (hydrocarbons)		
P10/2	Sensitive to flammable gas (methane)		
P40/1	Sensitive to gases with strong oxidation ability (fluorine)		
T70/2	Sensitive to aromatic compounds		
PA/2	Sensitive to organic compounds (ethanol and amine compounds, etc.)		
P30/1	Sensitive to organic compounds		
P40/2	Sensitive to gases with strong oxidation ability (hydrogen sulfide,		
	ammonia)		
P30/2	Sensitive to organic compounds (ethanol)		
T40/2	Sensitive to gases with strong oxidation ability		
T40/1	Sensitive to gases with strong oxidation ability		
TA/2	Sensitive to organic compounds		

FIG. 1

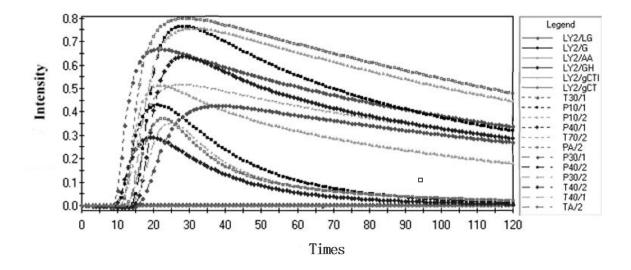


FIG. 2

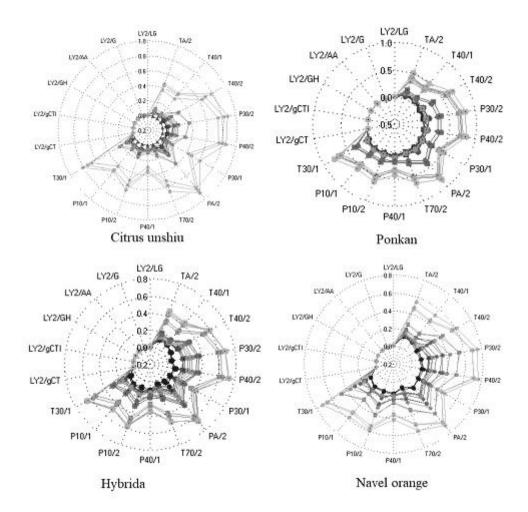


FIG. 3

Sensor	Citrus unshiu	Ponkan	Hybrida	Navel orange
T30/1	0.8708	0.878	0.9294	0.9051
P10/1	0.7661	0.865	0.8743	0.8227
P10/2	0.8558	0.865	0.9153	0.9051
P40/1	0.7659	0.8849	0.8666	0.7835
T70/2	0.8542	0.8731	0.9064	0.8665
PA/2	0.9036	0.902	0.9709	0.958
P30/1	0.8708	0.9013	0.972	0.9674
P40/2	0.8946	0.8806	0.9439	0.913
P30/2	0.9053	0.8993	0.9652	0.9471
T40/2	0.8928	0.9041	0.9592	0.9236
T40/1	0.7193	0.835	0.8267	0.7349
TA/2	0.7281	0.8439	0.8404	0.7609

FIG. 4

#### ABSTRACT OF THE DISCLOSURE

The present disclosure relates to a method of monitoring acid rot during citrus storage and transportation, and provides a technical solution for early detection and timely treatment of acid rot fruits in citrus mass storage and transportation. The present solution uses the acid rot fruits to emit a special sour smell that is easy to distinguish, and has a characteristic response signal in the electronic nose, so that it can specifically identify and monitor the occurrence of diseases during storage and transportation. Utilizing that the acid rot fruits will emit a special sour smell that is easy to distinguish, and has a characteristic response signal in an electronic nose, the present solution can specifically identify and monitor the occurrence of diseases during storage and transportation. This present solution collects citrus acid rot fruits of different disease courses, collects signals of the electronic nose, and establishes a prediction model.

#### ABSTRACT DRAWING

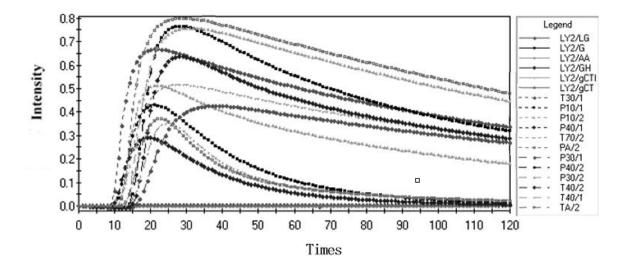


FIG. 2