

Fluorescence method for the assessment of homogeneity of granular mixtures

Metoda fluorescencyjna do oceny stanu jednorodności mieszanin ziarnistych

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Abstract

This paper presents a consecutive stage of the research aimed at developing a methodology for the application of fluorescence to evaluate the homogeneity of granular and loose mixtures. At this stage it was examined which of the selected components of feeds are suitable to be used in the proposed method. The experiments were conducted with the use of three components: wheat, kardi, barley. Each of the materials was wet dressed with a solution of a fluorescent material. In the study the substances having the characteristics of fluorescence under UV light such as: tinopal, rhodamine B, uranine, eosin, were applied. The assessment of the homogeneity of a ternary granular system was performed based on the share of a tracer, which was a component coated with a fluorescence substance. In the method a computer image analysis was used. Based on the conducted observations and statistical calculations it was demonstrated that only kardi meets the requirements necessary for the application in the tested method. The remaining components: wheat and barley were excluded. Moreover, types and concentration of fluorescent substances were determined and the methodology was clarified.

Keywords: fluorescence, homogeneity of mixtures, mixing, tracer

Abstrakt

Artykuł przedstawia kolejny etap badań mających na celu opracowanie metodyki zastosowania fluorescencji do oceny homogeniczności mieszanin ziarnistych i sypkich. Na tym etapie badano, które z wybranych komponentów mieszanek paszowych nadają się do zastosowania proponowanej metody. Doświadczenia przeprowadzono z wykorzystaniem trzech składników: pszenicy, kardii, jęczmienia. Każdy z elementów zaprawiano na mokro roztworem substancji fluorescencyjnej. Do badań wykorzystano wybrane substancje posiadające cechy fluorescencji w świetle UV: tinopal, rodamina b, uranina, eozyna. Ocenę stanu jednorodności trójskładnikowego układu ziarnistego określano na podstawie udziału traseru, czyli

danego składnika pokrytego substancją fluorescencyjną. W metodzie wykorzystano komputerową analizę obrazu. Na podstawie dokonanych obserwacji oraz obliczeń statystycznych, wykazano, iż tylko kardi spełnia wymagania niezbędne do zastosowania w testowanej metodzie. Pozostałe składniki: pszenica i jęczmień zostały odrzucone. Dodatkowo określono rodzaje i stężenie substancji fluorescencyjnych oraz doprecyzowano metodykę.

Słowa kluczowe: fluorescencja, jednorodność mieszanin, mieszanie, traser

Introduction

For thousands of years people have been using the mixing of solid particles for different purposes. Mixing of loose and granular components is a key element in the production of ceramic and pharmaceutical materials, plastics processing, production of food and feed, production of mineral fertilizers, mining or metallurgical industry (Alonso and Alguacil, 1999). This paper presents the results of experiments using granular components applied in the production of feeds.

A correctly performed mixing process is essential in the industrial production of feed. However, the appropriate quality is characterized by a random distribution of the components throughout the feed. These mixtures must guarantee an adequate content of the individual components in each dose of the feed (Beumer, 1991). Homogeneity of the compound feed should constitute a key element of quality control in feed manufacturing plants. Currently, feeds contain many substances such as: vitamins, minerals, amino acids added in small amounts. This favors the process of segregation and causes that the efficiency of the mixing process becomes even more important (Groesbeck et al., 2007; Rocha, 2015).

The assessment of homogeneity of granular mixtures, necessary mixing time and distribution of the individual components in the mixture, requires collecting numerous samples. Sampling irreversibly affects the mixture at the point of collection, and at the same time in the entire volume. However, systematic sampling at specified points are necessary to evaluate the process of mixing and compare mixtures under different conditions (Holdrich, 2002; Daumann and Nirschl, 2008). Depending on the statistical requirements, the size of an individual sample should range from 4 g to 50 g (Daumann and Nirschl, 2008). Most of experiments in this field are based exactly on invasive sampling (Sudah et al., 2002; Clark et al., 2007; Lemieux et al., 2007; Królczyk et al., 2010; Matuszek, 2013; Rocha et al., 2015). However, it is possible to find publications describing noninvasive tools (Lai et al., 2001, 2004; Lai and Cooney, 2004; Djuragic et al., 2009). Regardless of the method of sampling the next important step is the assessment of a condition of the mixture. In this area numerous studies are still conducted, and new tools and methods are developed and implemented by, among others, the author of the present study (Eisenberg, 1992; Realpe and Velazquez, 2003; Djuragic et al., 2009; Siiriä and Yliruusi, 2009; Obregón et al., 2010; Matuszek and Szwedziak, 2013; Matuszek, 2015; Realpe et al., 2015).

The results of an experiment conducted by the author of this studies are aimed to develop a new method for the assessment of homogeneity of granular mixtures and

constitute a consecutive stage of the research. The proposed method is based on an assessment of the content of a key ingredient (tracer) coated with a fluorescent substance. In the first phase of observation a description of the proposed method using maize as a key ingredient was performed (publication in press). The current stage includes the results of using other ingredients: wheat, kardi, barley, as a tracer.

Materials and methods

The experiments were performed in a laboratory equipped with a station for mixing granular materials (flow mixer), a chamber for acquisition of an image in the UV light, an analytical balance accurate to 0.01 g and a computer with self-developed software for BMP recorded image processing.

The mixing system consisted of three components at different ratio: green peas (60%), sorghum (30%) and tracer (10%). Components characteristic for feeds including: wheat, barley, kardi were used as tracers. The tracer was treated with solutions of selected fluorescent substances: tinopal (0.30%, 0.03% solutions), uranine (0.50%, 0.30%, 0.03% solutions), rhodamine b (0.01%, 0.001% solutions) and eosin (0.30%, 0.50% solutions). The selection of an individual fluorescent substance and its concentration for a given tracer was performed based on the results obtained in the earlier stages of the research (Matuszek and Szwedziak, 2013), as well as current observations, conducted during the experiments. As a consequence, fluorescent substances presented in Tables 1-3 were used for the selected components (wheat, barley and kardi). A tracer (at an amount of 100 g) and the remaining components were placed always in the same manner in the mixing chamber before the start of the mixing process. The volume of the mixed material was 1,000 g. The mixing process was carried out by consecutive ten flows. After mixing, 10 samples were collected from the entire volume of the tank. The volume of an individual sample was 40 g. For each system a certain number of test series (Tables 1-3) was performed in triplicate. The obtained samples were placed on a glass Petri dish measuring 120*20 mm (Ø * height). Then, the dish was placed in a horizontal position in a rectangular, sealed chamber equipped with fluorescent UV lamps. The material of the chamber assured no access of external light. The sample was placed in a horizontal position on a movable shelf. After closing the chamber, the sample was in its central part. The control of lighting in the chamber (switching on/off UV fluorescent lamps or a normal bulb) was performed from the outside. The objective of the camera was situated directly above the sample. The acquired images in the BMP format (1,600*1,200 pixels, Figure 1) were then analyzed using the PATAN application developed by M. Krótkiewicz. In the study circular area three following classes were indicated: 1 class – tracer, 2 class – green peas, 3 class – sorghum. As a result of the analysis, information about the content of the tracer in the study sample was obtained. Repeatability and reliability of the acquired images require maintaining clearly defined conditions. These conditions were met by the use of an appropriate test station. After the acquisition of the image, the tracer was manually separated in order to determine its mass, to verify the reliability of the obtained result.

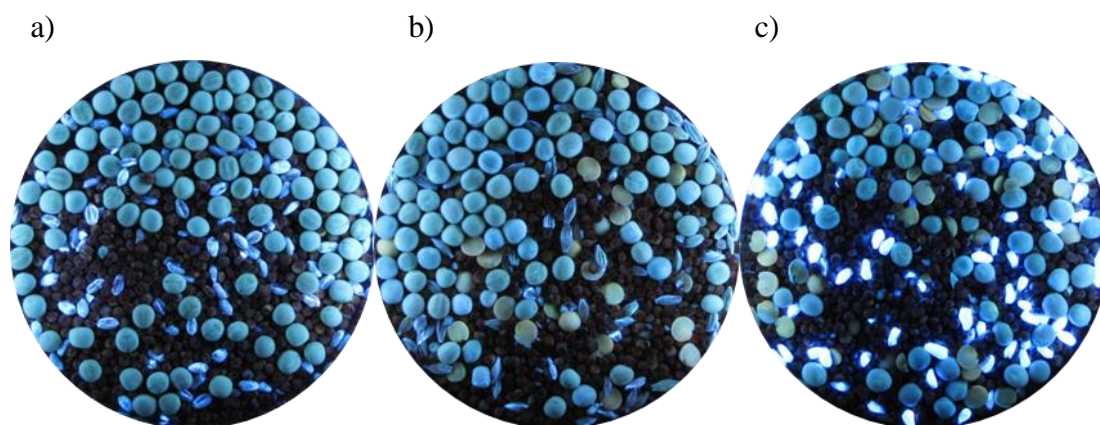


Figure 1. A sample of the mixture under UV light with a tracer coated with a 0.30% solution of Tinopal, a) wheat, b) barley, c) kardi

Rysunek 1. Próbkę mieszaniny w świetle UV z traserem pokrytym 0,30% roztworem Tinopalu, a) pszenica, b) jęczmień, c) kardi

Verification of the proposed method consisted in comparing the obtained result with a percentage of occupancy of the tracer. This parameter was determined based on a comparison of mass (measurement using an electronic balance, g) and content of the tracer (measurement using a computer image analysis) for a particular quantity of grains from 5 to 100 items. In this way an algorithm was obtained to determine the percentage of a tracer based on the information on its weight. A 5.00% error was assumed as a verification threshold.

Results and discussion

The obtained results (average content of the tracer, standard deviation, absolute error and percentage error) based on the proposed method and verification method, were presented in Tables 1-3.

Table 1. Results of the content of wheat in the compound feed
 Tabela 1. Wyniki oceny udziału pszenicy w mieszance paszowej

Fluorescent solution (%)	Computer image analysis ^a		Gravimetric method ^a		Absolute error	Percentage error (%)
	Mean (%)	SD	Mean, (%)	SD		
Tinopal						
0.30	4.58	0.95	7.36	1.48	2.79	35.33
0.03	7.93	1.01	6.95	1.57	1.92	30.87
Uranine						
0.30	3.87	1.09	7.13	1.31	3.26	46.44
Rhodamine B						
0.01	5.95	1.07	7.38	1.13	1.43	19.45
0.001	5.45	0.98	7.48	1.08	2.03	27.23

^a Mean of three analyses and then samples

^a Średnia dla trzech serii badań i 10 próbek

Table 2. Results of the content of barley in the compound feed
 Table 2. Wyniki oceny udziału jęczmienia w mieszance paszowej

Fluorescent solution (%)	Computer image analysis ^a		Gravimetric method ^a		Absolute error	Percentage error (%)
	Mean (%)	SD	Mean (%)	SD		
Tinopal						
0.30	25.22	4.17	7.05	1.06	18.17	268.06
0.03	33.04	4.31	6.79	1.75	26.24	434.84
Uranine						
0.30	4.19	0.82	7.39	1.31	3.21	43.13
Rhodamine B						
0.01	4.30	0.81	7.58	1.01	3.28	42.96
0.001	11.08	1.00	7.27	1.31	3.81	55.63

^a Mean of three analyses and then samples

^a Średnia dla trzech serii badań i 10 próbek

Table 3. Results of the content of kardi in the compound feed

Tabela 3. Wyniki udziału kardi w mieszance paszowej

Fluorescent solution (%)	Computer-aided image analysis ^a		Gravimetric method ^a		Absolute error	Percentage error (%)
	Mean (%)	SD	Mean (%)	SD		
Tinopal						
0.30	6.76	1.51	6.83	1.50	0.22	3.17
0.03	7.38	0.99	6.78	1.40	0.67	11.82
Uranine						
0.50	6.58	1.09	6.47	1.08	0.28	4.28
0.30	7.10	1.21	6.42	1.43	0.68	12.51
0.03	9.06	0.86	6.27	0.77	2.79	47.23
Rhodamine B						
0.01	7.77	0.73	7.59	0.70	0.20	2.68
0.001	8.22	1.23	6.30	1.34	5.75	33.11
Eosin						
0.30	6.13	0.64	6.80	0.82	0.70	9.80
0.50	6.12	0.85	6.48	0.92	0.54	8.21

^a Mean of three analyses and then samples

^a Średnia dla trzech serii badań i 10 próbek

Based on the comparison of the proposed method with the verification method, significant differences not only between the two methods for assessment of the tracer content, but also depending on the applied fluorescent substance, can be observed (Tables 1-3).

The lowest percentage error, in the case of wheat, of 19.45% was recorded for rhodamine b at a concentration of 0.01%. In turn, the largest difference was observed for the use of 0.30% uranine ($\delta=46.44\%$). Based on the obtained results wheat was excluded as a carrier for a fluorescent substance (Table 1).

Verification of the method using barley as a tracer indicates a lack of relevance of application of this component in the proposed method. The range of error is

considerable and equals from 42.96% for Rhodamine B 0.01% up to the value of 434.84% for Tinopal 0.03% (Table 2). In therefore, it was decided to exclude barley as a carrier for a fluorescent substance.

In the case of kardi the authors decided to performed additionally tests with uranine (0.50%, 0.03%) and eosin (0.30%, 0.50%). It can be observed (Table 3) that the adopted assumption ($\delta \leq 5.00\%$) was obtained for the following fluorescence substances: Tinopal 0.30% ($\delta = 3.17\%$), Uranine 0.50% ($\delta = 4.28\%$) and Rhodamine B 0.01% ($\delta = 2.68\%$). For the remaining solutions a percentage error higher than 5.00% (Table 3) was obtained. It can also be noted that compared to the results obtained using wheat and barley, the margin of error is significantly smaller (δ from 8.21 to 47.23%).

Based on the above-mentioned conclusions, it was decided to conduct a comparative statistical analysis of the results obtained using kardi. For this purpose t-Student test was used ($r=2$, $k=19$). Statistical calculations were performed using *Statistica* software (Statsoft, 2015).

For calculations of t-Student statistics, the following assumptions were made:

1. Each of the two populations (population should be understood as the method for the assessment of the share of the tracer) have normal distribution (this assumption was verified using the Shapiro-Wilk test).
2. Both populations have equal variances (this assumption was verified by the Fisher test).

The null hypothesis is:

$$H_0: \mu_1 = \mu_2 \quad (1)$$

The mean of the tested populations are equal at the value of $r = 2$.

The alternative hypothesis is:

$$H_1: \mu_1 \neq \mu_2 \quad (2)$$

The mean of the tested populations are different at the value of $r = 2$.

The analysis of hypotheses was performed based on t-Student test for independent samples, assuming a significance level $\alpha = 0.05$ (Aczel, 2005; Stanis, 2006).

The results of the comparative statistical analysis are presented in Table 4.

Table 4. Results of the comparative statistical analysis of the content of kardi in the mixture

Tabela 4. Wyniki porównawczej analizy statystycznej udziału kardi w mieszance

Fluorescent solution (%)	t-Student test	
	t	p
	Tinopal	
0.30	-0.10	0.920
0.03	1.05	0.308
	Uranine	
0.50	0.19	0.845
0.30	1.09	0.289
0.03	7.25	0.000
	Rhodamine B	
0.01	0.55	0.590
0.001	3.17	0.005
	Eosin	
0.30	1.91	0.073
0.50	0.86	0.401

Coming to the analysis of the obtained results it can be observed that for the populations of Tinopal 0.30%, 0.03%, Uranine 0.50%, 0.30%, Rhodamine B 0.01%, Eosin 0.30% and 0.50% no differences were observed. In turn, in the case of Uranine 0.03% and Rhodamine B 0.001% there is no reason to accept the null hypothesis concerning the lack of differences in the results obtained using the test method and verification method (Table 4). Comparing the results to the value of the percentage error (Table 3) it was assumed to be reliable that kardi as a tracer coated with a fluorescence substance at a concentration: 0.30% for Tinopal, 0.50% for Uranine, 0.01% for Rhodamine B is useful for the assessment of homogeneity of a ternary granular mixture according to the proposed method.

Based on the obtained results and observations described in the present and earlier works of the author, a methodology for determining the homogeneity of multi-component granular mixtures using fluorescent substances was developed.

Among all tested components, only two are suitable for use in the test method, and they are maize and kardi. These components are wet dressed with solutions of suitable fluorescent substances. In the case of maize it is: 0.30% solution of Tinopal or 0.001% solution of Rhodamine B. In turn, in the case of kardi it is: 0.30% solution of Tinopal, 0.50% solution of Uranine or 0.01% solution of Rhodamine B. The method can be applied to assess the homogeneity of granular (not grinded or before grinded) mixtures under laboratory conditions. The amount of a tracer introduced into the mixer should constitute 10% of the total volume of the mixed material. The mass of a single sample of a granular mixture should equal 40 g. The number of samples should be consistent with the standards. A collected sample should be placed on a Petri dish having dimensions of 120*20 mm, covering its entire surface.

Subsequently, the sample should be placed in a sealed chamber equipped with UV lighting (15 W fluorescent lamps). Then, a photograph of the sample in the BMP format should be taken and the content of the tracer should be estimated using the computer image analysis. Based on this information it is possible to perform calculations of homogeneity of the feed using for this purpose known parameters (e.g. coefficient of variation, degrees of mixing).

The application of staining of granular components to evaluation the grain behavior, the blender performance, and estimation of mixture homogeneity was the subject of several studies (Sato and Miyanami, 1988; Weinekötter and Reh, 1994; Poux et al., 1995; Alonso and Alguacil, 1999; Boss et al., 2002; Realpe and Velazquez, 2003; Matuszek and Tukiendorf, 2007; Dauman and Nirschl, 2008; Obregón et al., 2010). These researches based commonly by the estimation of the proportion of the selected component from the binary system with a strongly varied coloration (e.g. red and blue or white and black). In addition, the study was conducted using materials made of plastics or other with certain shapes, such as steel balls. The obtained results indicated that the observation of behavior of colored particles can be useful to assessment the parameters of the mixing process. The X-ray fluorescence phenomenon for evaluating the content of sulfur and chlorine in feeds was reported by Necemer et al. (2003). Whereas the laser-induced fluorescence (LIF) was the subject of investigations to assess the homogeneity of powders used in the pharmaceutical industry, which were used by, inter alia, Lai et al. (2001) and Karumanchi et al. (2011).

The ability of Tinopal to shine in ultraviolet light was used to evaluate the work of sprayers (Hołownicki et al., 2012). This publication has inspired the author to start investigating the implementation of UV-fluorescence in the assessment of homogeneity of granular mixtures. The proposed method allows, inter alia, to determine the share of the actual ingredient constituting, for example, the component present in the feed.

Conclusions

The comparative statistical analysis demonstrated no statistically significant differences in the content of kardi obtained with the use of two methods for the following solutions of fluorescent substances: 0.30%, 0.03% Tinopal, 0.50%, 0.30% Uranine, 0.01% Rhodamine B, 0.30% and 0.50% Eosine. Based on the conducted tests it was observed that wheat and barley are not suitable for use in the fluorescent method. In the case of kardi the adopted assumptions were met for three solutions of the following fluorescent substances: 0.30% Tinopal, 0.50% Uranine and 0.01% Rhodamine B. Based on the presented results and previous observations, a methodology for the assessment of homogeneity of granular mixtures based on the content of a tracer coated with a fluorescence substance was developed.

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