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Mixtures of nanotracers based on manganese and iron oxides for quality control of liquid feeds

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Abstract

Modern strategies in terms of animal nutrition require increasingly sophisticated feed compositions, which, in turn, increase the importance of achieving homogeneous mixtures, especially for young animals. Nutritionally optimized rations play a critical role in supporting rapid development and maximizing nutrient utilization.

To control the uniformity of mixing of dry feeds, ferromagnetic microtracers based on iron oxides are actively used, which are effective due to their magnetic properties, allowing easy detection and control of the homogeneity of mixtures. The limited effectiveness of such tracers in liquid feed applications led to the development of innovative iron oxide-based magnetic nanomarkers specifically developed for use in liquid mixtures.

Ensuring homogeneous mixing of liquid feeds is especially crucial, as it can be influenced by the choice of surfactant. Various emulsifying agents, including dimethylamine oleate and ammonium oleate, influence the dimensions and clustering properties of nanoparticles in different manners. Compared to ammonium oleate, DMAOA proved to promote improved particle dispersion and greater suspension stability – factors essential for preserving feed quality. To measure the manganese content in the magnetic nanoparticles $Fe_xMn_yO_z$, researchers applied a modified spectrophotometric method.

This allows obtaining accurate data on the composition of nanoparticles, which is important for ensuring the effectiveness of quality control in feed production.

Keywords: manganese-iron oxide nanotracers; feed mixing uniformity; spectrophotometric determination of manganese; magnetic nanotracers; quality control; dimethylamine salt of oleic acid.

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1. Introduction

The production of liquid compound feeds, as well as dry ones, is an important component of the agricultural sector. A critical element in the manufacturing of mixed feed rations is the mixing procedure, which guarantees stable composition and nutritional balance. As feed composition grows increasingly complex, including a wider variety of ingredients, the demand for achieving thorough homogeneity in feed mixtures becomes even more pressing [1]. Uniform mixing of feed ingredients is essential to ensure that the nutritional requirements of various livestock and poultry species are fully met. Consistent composition of feeds promotes uniform distribution of nutrients and reduces the likelihood of component segregation, which could otherwise result in nutritional imbalances for animals [2]. To solve these problems, modern mixing technologies are used, such as ribbon mixers and vertical blenders, which are equipped with intelligent control systems. These systems allow prompt correction of the feed formulation based on real-time data, which increases the efficiency of the production process.

Ensuring a proper mixing process is essential for producing nutritionally complete and balanced feeds. Given the growing use of industrial amino acids and a wide range of additives, the importance of precise mixing continues to increase.

Despite being included in minimal amounts, these components play a vital role in determining the feed nutritional value. To ensure that all animals equally receive all essential nutrients, accurate and thorough mixing becomes indispensable.

Moreover, the effectiveness of the mixing process carries both economic and ethical consequences. If mixing is excessive, it may result in the degradation of sensitive compounds such as vitamins and medications, or lead to ingredient separation. Conversely, inadequate mixing can produce feed batches with imbalanced nutrient content, potentially causing financial losses for producers and increasing the risk of pharmaceutical residues entering the food chain.

Therefore, ensuring the correct mixing of ingredients is one of the main tasks in the production of compound feeds, which requires not only technical precision, but also constant monitoring of the process to ensure optimal results for both animal health and economic benefit for producers. Liquid compound feeds have the following advantages when used: they provide animals with the necessary amount of moisture, which can be especially important in hot climates or in conditions where access to water is limited; nutrients are absorbed faster and easier by the animal's body; they reduce the risk of loss of trace elements or vitamins during transportation or storage, as nutrients better "bind" in a liquid environment; they are more attractive to animals due to their consistency and more pleasant taste. Liquid compound feeds are best suited for certain types of animals that have special hydration needs, specific digestive systems or other nutritional requirements, their use is advisable for the following animals:

1. Cattle – dairy cows: liquid feeds can help increase feed intake and milk production. Cows need a lot of moisture, especially in high temperatures, so liquid feeds help meet their hydration needs [3]. In those cases when animals are fattened for meat production, liquid feeds can improve nutrient absorption and provide a uniform energy intake for body weight gain [4].

Molasses, in either dry or liquid form, is a good source of dietary sugar for dairy cows [5]. In particular, liquid molasses can be used to transport minerals and rumen-fermentable carbohydrates, but studies on its effectiveness compared to solid molasses are contradictory [6]. In addition, liquid molasses is inconvenient to store and transport. The main problem with using soybean molasses is storage and transportation, as it is usually stored in bulk containers, which can be inconvenient for small producers.

2. Young pigs have high hydration and nutrient absorption needs [7]. Liquid feeds are easier for young pigs to digest, help balance their diet, and improve growth and development [8]. Also for piglets, the use of liquid feeds helps to increase energy efficiency and ensure faster weight gain [9].

Ensuring complete uniformity in the mixture represents a crucial phase in the manufacture of solid and fluid feed formulations, as outlined earlier. To evaluate the mixing quality of multi-component dry compound feeds, a statistical approach based on particle distribution is commonly applied. This technique determines the consistency of ingredient dispersion, helping ensure that each feed portion maintains nutritional uniformity.

Initially outlined in the GMP+BA2 guideline "Residue Control," the protocol for evaluating consistency in dry feed and premix compositions – through techniques like microtracer analysis – has been progressively improved. Currently, this procedure is standardized as TS1.11: Control of Residues and Homogeneity [10].

Throughout the past three decades, ferromagnetic microtracers have been widely and effectively applied in feed production [11]. Unlike earlier tracer types, these particles can be quickly and easily extracted from bulk mixtures using magnetic separators, offering a more efficient solution for quality control.

Multiple studies conducted by Eisenberg (1979, 1980, 1987) [12–14], considered the use of ferromagnetic microtracers (MTs), which were patented and manufactured by the San Francisco-based company MicroTracers Inc. This technique involves incorporating MTs into the feed mixture during production as a trace additive, typically at a recommended rate of 50 grams per metric ton of ready-made feed.

Composed either of iron (Microtracer F) or stainless steel (Microtracer FS), these microtracers feature particle sizes ranging between 150 and 350 microns. The third type of MTs (Microtracer RF) consists of reduced iron powder, with most of the particles below 150 micron in size. All three types of MTs have various food dyes adsorbed on their surface (Table 1).

It is obvious that in several cases MTs are manufactured using a combination of two dyes (#6 and 7).

During the formulation of vitamin-mineral or medicinal premixes, Microtracers® are used to confirm the presence of the premix in the final feed product. These tracers also help identify patented feed additives and the feeds containing them. For quantitative analysis, Microtracers® provide a means to evaluate the effectiveness of the mixing process. Additionally, they can be employed to verify the thoroughness of equipment cleaning between the production of batches, ensuring minimal cross-contamination.

2. Methods

In accordance with established feed production standards, mixing equipment shall demonstrate high efficiency. For compound feed mixtures, where the dilution ratio is approximately 1:10.000, the acceptable coefficient of variation (CV) shall not exceed 15%. In contrast, premix production, which involves more concentrated mixtures (1:100.000), requires a CV of no more than 10%.

Table 1

Nature of various food dyes used in manufacturing MTs

# dye or combination of dyes	Name by FDA* registration	Trade or chemical name	Appearance of color developed from particles of MTs
1	FD&C Red #40	Allura Red AC	Red
2	FD&C Yellow #5	Tartrazine	Yellow
3	FD&C Yellow #6	Sunset Yellow	Orange
4	FD&C Blue #1	Brilliant blue FCF	Blue
5	FD&C Blue #2	Indigo Carmine	Dark Blue
6	FD&C Yellow #5 FD&C Blue #1	Tartrazine Brilliant blue FCF	Green
7	FD&C Red #40 FD&C Blue #1	Allura Red AC Brilliant blue FCF	Violet

* Food Drug Administration, USA

Table 2

Quantification of Microtracers® particles in sequentially collected feed samples during the discharge process from the compound feed mixer (measured in replicates)

Number of Samples Analyzed, 20				Tracer Recovery	91.36
114	120	118	130	Mean	127.40
123	114	126	150	Standard deviation	12.64
124	116	119	126	Coefficient of variation (%)	10.18
116	120	**140**	151	Coefficient of variation-Poisson (%)	8.86
132	120	134	155	Chi-Square (χ^2)	23.84
Probability (%)					16.04
Conclusion: A probability exceeding 5% indicates a satisfactory level of mixing for the red tracer.					

When assessing the mixture uniformity using direct analytical methods, and in line with regulatory standards [10], the level of probability (p%) is interpreted as follows:

$p \leqslant 1\%$: poor uniformity, indicating a likely significant inconsistency;

$1\% < p < 5\%$: possible deviation; the result is inconclusive and retesting is recommended;

$p \geqslant 5\%$: indicates satisfactory mixing uniformity.

To evaluate mixing quality, the results obtained from measurements were subjected to statistical analysis, with the coefficient of variation serving as a key indicator (see Table 2).

The overall number of tracer particles (e.g., 140 in Fig. 1) is obtained by counting the visible coloured marks on the filter paper – this can be done manually or through specialized software such as the TraCo system for image analysis and evaluation. For precision, the statistical analysis follows the principles of the Poisson distribution.

Iron-containing Microtracers® are generally ineffective for incorporation into liquid feed for-

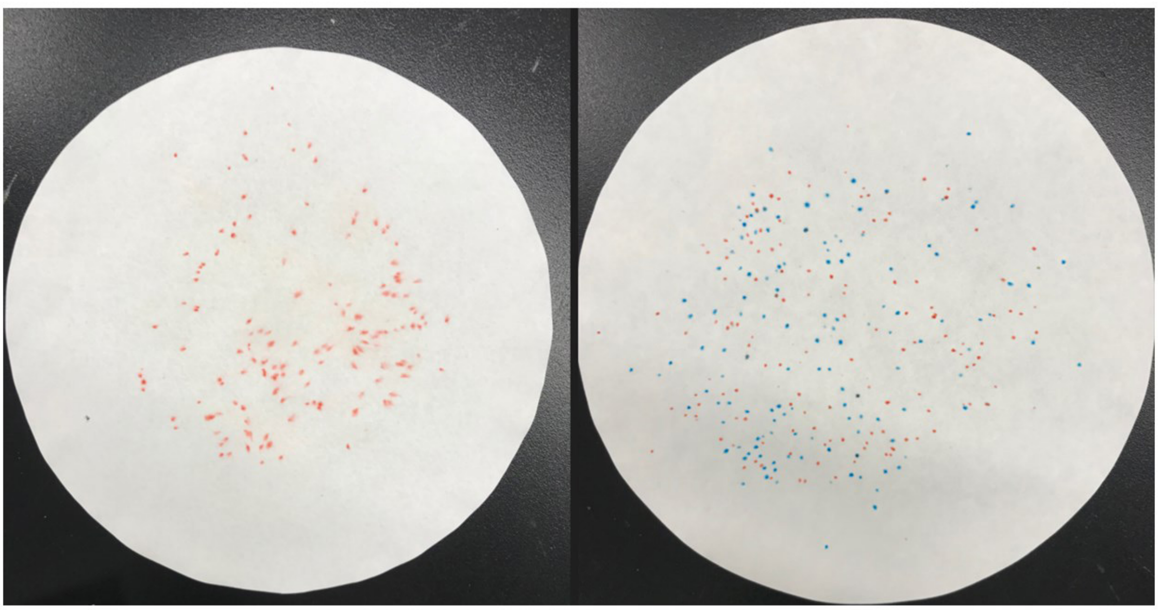


Fig. 1. Sample filter papers displaying Microtracers® particles utilized for quantitative analysis of microtracer

mulations, for tagging fluid supplements like enzymes, or for evaluating their dispersion in premixes and complete feed products. To address the rising need for such applications, a novel extractable magnetic nanotracer – comprising iron oxide particles dispersed in a magnetically responsive fluid – has been developed and tested [14].

Results and discussion

The process of producing a magnetic liquid generally consists of two key steps: first, the generation of magnetic particles of the required size, and second, their stabilization within a carrier fluid. Researchers have published numerous studies and reviews on the synthesis of magnetic nanoparticles, owing to their broad applications – not only in nanoparticle production but also in fields such as magnetic data storage, catalysis, electronics, and electrical engineering [15].

Analysis of the literature reveals that a two-step process is among the most promising methods for producing ferromagnetic materials, particularly magnetic nanoparticles (MNPs).

First, researchers synthesize ferromagnetic liquids, commonly referred to as ferrofluids [16].

Next, these ferrofluid droplets are dispersed into an aqueous environment with the help of a suitable surfactant.

This dispersion step plays a crucial role in stabilizing and evenly distributing the magnetic nanoparticles in liquid feeds, offering a more efficient alternative to traditional iron-based Microtracers® [17].

A major challenge in determining nanoparticle size is ensuring precise and accurate measurements that truly reflect their actual size, which presents significant metrological and mathematical difficulties, particularly in the nanoparticle tracking analysis (NTA) method [18].

Synthesis of Ferrofluid Using Fe and Mn Oxides

The following procedure was used to prepare ferrofluid based on iron oxide/manganese oxide with two amendments:

a) a mixture of $FeCl_2{\cdot}4H_2O$ and $MnCl_2{\cdot}4H_2O$ (8:2 wt/wt) was used instead of ferric chloride [19];

b) food grade heptane was used instead of kerosene.

To produce a ferrofluid based on iron and manganese oxides, a modified synthesis method was applied [17]. Initially, 258 g of $FeCl_3{\cdot}6H_2O$, 86.8 g of $FeCl_2{\cdot}4H_2O$, and 21.7 g of $MnCl_2{\cdot}4H_2O$ were dissolved by stirring with a non-magnetic glass rod. In a separate step, 350 ml of a 28% aqueous ammonia solution was diluted with an equal volume of water and gradually added to the iron salt solution over 75 seconds. The resulting colloidal magnetite mixture was continuously stirred and heated to a temperature of 90 °C. At the same time, 40 ml of oleic acid was combined with 460 ml of heptane and heated to 90 °C. Once both the ammonia-based magnetite and the organic solution reached 90 °C, they were mixed and stirred for 15 minutes.

After the reaction, the upper organic phase was separated, resulting in a stable ferromagnetic fluid with a density of 1.038 g/ml, a viscosity of 4.3 cP and a saturation magnetization of 252 gauss.

After synthesis, the ferrofluid was mixed into a 0.5% aqueous surfactant solution – either ammonium oleate or dimethylamine salt of oleic acid (DMAOA) – at a mass ratio between 1:100 and 1:400. The mixture was then filtered using Whatman filter paper with a 5-micron pore size. The resulting stable suspension was used for both nanoparticle size determination and colorimetric quantification of manganese content in the particles.

$Fe_xMn_yO_z$ Testing

A stable suspension of ferromagnetic iron/manganese oxide nanoparticles ($Fe_xMn_yO_z$) offers several potential applications, including:

- evaluating the mixing uniformity of liquid feed additives;
- liquid supplements such as enzymes can be encoded using this approach;
- the method enables effective monitoring of additive distribution in both premixes and ready-made feeds.

Tested both in laboratory and production environments, the $Fe_xMn_yO_z$ suspension – composed primarily of iron oxide (over 95%) with a minor fraction of manganese oxide (less than 5%) – proved effective for assessing liquid feed mixing and additive tracking.

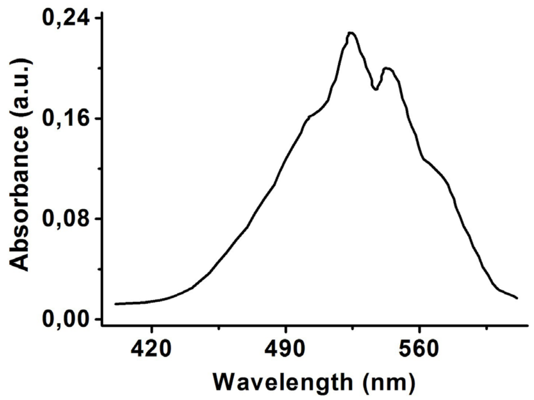
During laboratory trials, researchers introduced a 100 ppm suspension of the nanotracer, pre-mixed with a liquid enzyme (final enzyme concentration in feed: 110 ppm), into feed.

Serving as a magnetic marker, $Fe_xMn_yO_z$ nanoparticles were subsequently extracted and demonstrated a recovery rate of at least 75%, confirming their suitability for tracing in feed systems. Manganese concentration was determined using a modified spectrophotometric analysis, with previously reported sensitivity tests indicating a LOD of 0.45 mM and a LOQ of 1.51 mM [20].

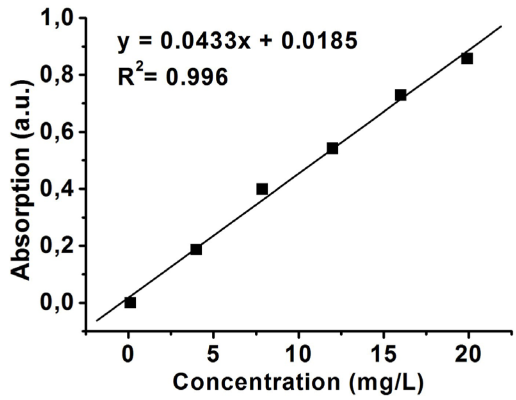
Determination of the Mn content in a suspension of nanoparticles

A four-step procedure was used:

1. Obtaining ferromagnetic nanoparticles from the suspension using a neodymium magnet with a plastic coating;
2. Dissolving nanoparticles in 20% aqueous HCl;
3. Adjusting the pH of the prepared solution and adding potassium periodate;



a



b

Fig. 2. Absorbance spectrum of permanganate anions – *a*;
calibration curve for determination of Mn content in iron oxide/manganese oxide nanoparticles at λ_{max} = 525 nm – *b*

4. Determination of the optical density of permanganate anions at 525 nm.

Fig. 2*a* shows the absorption spectrum of permanganate ions, which has three absorption peaks at wavelengths of 507, 525, and 545 nm. The absorption peaks at wavelengths of 525 and 545 nm are clearly pronounced, and the most pronounced absorption peak is at wavelength 525 nm [20].

Therefore, the wavelength of 525 nm was chosen for spectrophotometric study. According to the results of measuring standard samples, a calibration curve (Fig. 2*b*) with a linear regression coefficient (R^2>0.996) was obtained, which was used to determine the molar absorption coefficients.

Such a decrease in the variance of the linear regression at low concentrations indicates that this technique for spectrophotometric quantification of the permanganate ion concentration is not applicable to samples with a MnO_4- concentration lower than 6.3 mM (0.75 mg/L).

Conclusions

A method for synthesizing a ferromagnetic nanotracer $Fe_xMn_yO_z$ based on iron and manganese oxides has been developed, enabling the formation of a stable suspension in aqueous surfactant solutions.

The use of $Fe_xMn_yO_z$ as a nanotracer proved effective for evaluating the mixing quality of liquid feed under laboratory conditions.

Out of all the surfactants evaluated for dispersing ferromagnetic nanoparticles in the suspension, dimethylamine salt of oleic acid proved to be the most effective. Providing a more consistent particle size distribution along with improved stability, this surfactant plays a crucial role in ensuring accurate and reliable assessment of feed mixture quality.

Суміші нанотрейсерів оксиду мангану і заліза для контролю якості рідких кормів для тварин

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Анотація

Сучасні тенденції годівлі тварин (особливо молодих) вимагають дедалі складніших рецептур, що збільшує необхідність застосування однорідних сумішей. Це збалансований корм, який сприяє швидкому росту тварин та ефективному споживанню поживних речовин. Рівномірність змішування сухих кормів контролюють із використанням феромагнітних (на основі ферум оксидів) мікротрейсерів. Завдяки магнітним властивостям мікротрейсерам можна легко та ефективно виявляти та контролювати рівномірність змішування кормів. Однак використання таких індикаторів для кормів у рідкому стані є менш ефективним, що спонукало до розробки нових магнітних нанотрейсерів із ферум оксидом, які підходять саме для рідких сумішей. Наведено етапи та процеси перспективних методів отримання феромагнітних матеріалів – магнітних наночастинок, які забезпечують стабілізацію магнітних нанотрейсерів та їх рівномірний розподіл у рідких кормах. Описано методику приготування ферорідини на основі оксиду заліза/оксиду марганцю. Лабораторні дослідження суспензії нанотрейсерів, змішаних із рідким ферментом, доданим до корму з подальшою екстракцією феромагнітних наночастинок, показали не менше 75% відновлення. Особливо важливо, щоб при змішуванні рідкі корми залишались однорідними. На цей процес впливають різні ПАР. Наприклад, амоній олеат та диметиламін олеат (DMAOA) по-різному можуть впливати на агрегацію наночастинок та їх розмір. Встановлено, що кращі стабільність та дисперсію частинок забезпечує саме DMAOA, що є критичним для збереження якості корму. Для визначення концентрації марганцю у складі магнітних наночастинок $Fe_xMn_yO_z$ використано модифікований спектрофотометричний метод. Це дозволяє отримати точні дані про склад наночастинок, що важливо для забезпечення ефективності контролю якості при виробництві кормів.

Ключові слова: нанотрейсери оксиду марганцю та заліза; рівномірне змішування кормів; спектрофотометричне визначення марганцю; магнітні нанотрейсери; контроль якості; диметиламінна сіль олеїнової кислоти.

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