

Valorisation of food waste to chemicals: recovery of volatile fatty acids with functionalised magnetic nanoparticles

Authors: Angela Di Marcantonio, Lucia Placanica

Supervisor: Prof. Maurizio Masi

Co-Supervisor: Prof. Filippo Rossi, Ing. Elisa Lacroce

Abstract

Based on the current research interest in volatile fatty acids (VFAs) recovery and the wide applicability of nanoparticles, this work aims to propose a novel method for VFAs recovery from fermented food waste using iron oxide magnetic nanoparticles (MNPs). Synthesis of MNPs was performed via the chemical co-precipitation method and the obtained particles were characterised using XRD, FT-IR, CHNS, DLS, Z-potential, and SEM. A fit-for-purpose MNPs functionalisation was carried out using APTES; FT-IR, DLS, Z-potential, and CHNS were used to assess the successful bonding of the ligand to the surface. Adsorption tests were performed on a synthetic aqueous VFAs solution, both with naked and coated MNPs, with two different agitation methods and four distinct contact times. The adsorption was successful in all the conditions and nanoparticles didn't show strong selectivity for one analyte over the others. The two tests showing uniformly high uptakes for all analytes were replicated, but showed lower VFAs uptake values, indicating a high experimental variability. These replicates were used to perform desorption tests in deionised water and methanol. The desorption was verified with both solvents. A second adsorption test was conducted after desorption and resulted in similar capture values with respect to the first cycle despite the desorption was not complete. Finally, considering the vital role of the coating for the use of MNPs, the functionalisation reaction was optimized by maximising the number of amine groups on the nanoparticles' surface. The highest functionalisation percentage achieved is 10.7 %, using the 5-hours procedure with hydrolysed-APTES and MNPs/APTES molar ratio equal to 1:4.

Keywords: food waste, volatile fatty acids, iron oxides magnetic nanoparticles, APTES

1. Introduction

Food waste and losses are generated at each step of the food journey from farm to fork, ranking high on the list of global priorities. This major problem accounts for more than 1.3 billion tons of food wastage worldwide, corresponding to 1/3 of the overall food production [1].

Prevention and remediation methods are available and implemented to some extent. Attention from virtuous companies, consumer awareness, and new technological solutions that extend the life of the food products are some of the factors that prevent the transformation of food into waste; but when it is not possible to avoid food waste, nor to re-use it for human or animal consumption, the last valid option is to recover from it useful resources. The treatment methods currently implemented are focused on the production of biofertilizers, energy, and fuels (biogas and biomethane primarily) [2]. These processes degrade a complex and rich substrate yielding low value-added products. This thesis is part of the REVENUE project that proposes a 3-routes platform for REcovery of high Value products, ENergy, and bio-

fertilizer from Urban biowaste fully exploiting the potential of food waste. To see this process implemented in the future, it is necessary to overcome the main obstacle represented by the separation of VFAs from the fermentation broth. To face the challenge a novel method is proposed in this thesis work: the use of iron oxide magnetic nanoparticles (MNPs) to selectively recover VFAs and release them for further purification.

2. Materials

Ethanol (C_2H_6O , MW=46.07 g/mol, $\geq 99.8\%$), sodium acetate ($C_2H_3NaO_2$, MW=82.03 g/mol, $\geq 99.0\%$), sodium propionate ($C_3H_5NaO_2$, MW=96.06 g/mol, $\geq 99.0\%$), sodium butyrate ($C_4H_7NaO_2$, MW=110.09 g/mol, 98%), isobutyric acid ($C_4H_8O_2$, MW=88.11 g/mol, 99%), valeric acid ($C_5H_{10}O_2$, MW=102.13 g/mol, ≥ 99.0), isovaleric acid ($C_5H_{10}O_2$, MW=102.13 g/mol, 99%), sodium hexanoate ($C_6H_{11}NaO_2$, MW=138.14 g/mol, 99-100%) and (3-Aminopropyl)triethoxysilane (APTES, $C_9H_{23}NO_3Si$, MW=221.37 g/mol, 99%) were purchased from Sigma-Aldrich®, Germany. Colour-fix pH indicator

sticks (pH-Fix 0-14, No. 10642751), 25 mm syringe filter (nylon, 0.2 μm , No. 15121499) and heating plate equipped with a thermocouple were obtained by Fisherbrand[®]. Iron (II) chloride tetrahydrate ($\text{FeCl}_2 \cdot 4\text{H}_2\text{O}$), Iron (III) chloride hexahydrate ($\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$), sodium hydroxide (anhydrous pellets) and methanol were from Carlo Erba reagents[®]. Hydrochloric acid (1M, volumetric solution) by Fisher chemical, Fisher Scientific[®].

VirTis BenchTop Pro with Omnitronics by SP scientific was used for lyophilisation; LABOROTA 4000-efficient by Heidolph[®] was used to rotary evaporate F-MNPs and hydrolysed APTES; the centrifuge employed was the Sorvall ST 8 by ThermoFisher Scientific whereas the shaker was the Multi Reax by Heidolph[®]. Lastly, the neodymium fishing magnet (model: NJD90) by Wukong[®] was deployed for magnetic decantation. All reagents and solvents were used as received without further purifications.

3. Experimental methods

MNPs synthesis

The co-precipitation reaction was tested using ammonium hydroxide, sodium hydroxide and an excess of sodium hydroxide. This last was selected due to faster magnetic decantation, the procedure is described hereby.

In a three-necked round bottom flask, 1.057 g of $\text{FeCl}_2 \cdot 4\text{H}_2\text{O}$ and 2.523 g of $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ were dissolved in 106.6 mL of deionized water. The solution was stirred at 100° C and 400 rpm for 1 hour, under a nitrogen atmosphere [3][4]. After this time, 33.3 mL of NaOH (9.98M) were rapidly added to the reaction mixture, which was stirred for another hour. Then the mixture was cooled at room temperature. MNPs were magnetically collected with an external magnet and washed three times with deionised water.

MNPs functionalisation

APTES was hydrolysed according to the procedure described by [5]. The coating reaction was executed according to the procedure described by [3] and [5].

In a three-necked round bottom flask, 555 mg of MNPs were dissolved in 50 mL of deionized water and 22.5 mL of hydrolysed APTES (0.918 M) were added. The mixture was stirred for 5 hours at 60°C under a nitrogen atmosphere. After cooling at room temperature, the functionalized nanoparticles (F-MNPs) were washed once with deionised water, once with ethanol and rotary evaporated at 50°C for 5 minutes. The same procedure (i.e. 5-hour procedure) was followed using different molar ratios of MNPs to hydrolyzed-APTES (1:4 and 1:18). For comparison also the procedure described by [6], [7] (i.e. 1-hour

procedure) was conducted using a ratio of 1:4. Both procedures were tested using APTES and hydrolyzed-APTES.

Synthetic VFAs solution preparation

The synthetic VFAs solution was prepared according to the acids' concentration after fermentation obtained by IRSA-CNR researchers. Both VFAs retrieved as salts and VFAs retrieved as acids were added, and the final pH was adjusted to be equal to 7.

Adsorption experiments

For each experiment 10 mg of dry nanoparticles have been weighted and dissolved in 2 mL of VFAs solution. Tests were carried out under two different types of agitation: shaker and magnetic stirring. For the latter, a small stirring magnet has been added for the duration of the experiments and removed at the end. Both types of agitation experiments were conducted using four different times, namely 5 minutes, 30 minutes, 1 hour, and 2 hours, with MNPs and F-MNPs. The separation was performed through centrifugation, operating at 6000 rpm for 6 minutes. Finally, the supernatants have been filtered and analysed by gas chromatography (GC).

Desorption experiments

Water and methanol (MeOH) were used as solvents for desorption. F-MNPs were collected from the adsorption experiments and dissolved in 4 mL of solvent. All desorption tests were performed magnetically stirring for 30 minutes at 40°C. The separation was carried out through centrifugation, operating at 6000 rpm for 6 minutes. Then, the supernatants have been filtered and analysed with GC. The samples for which the desorption was performed in MeOH undergone an additional step: 4 mL of deionised water were added, and the samples were rotary evaporated to remove as much MeOH as possible.

Second-cycle adsorption experiments

Nanoparticles were collected from the desorption experiments, dissolved in 2 mL VFAs solution, and then the same procedure of the first set of adsorption experiments was followed.

4. Results and discussion

MNPs and F-MNPs characterization

The XRD diffractogram of MNPs is similar to what is reported in the literature [9]–[14]. It showed well-

defined peaks indicating good sample crystallinity and confirmed the presence of magnetite in the sample. The intermediate Akaganeite and Calcium carbonate residues from water were also detected. The average crystallite size estimated using the Scherrer equation **Error! Reference source not found.**), applied to the reflection peak at $\theta = 0.6186 \text{ rad}$, was 7.65 nm. The functionalization of the surface was verified by IR. The F-MNPs' spectra showed a strong band around 1500 cm^{-1} associated with the Si-O stretching vibration [13] [14] [17], this is absent in the MNPs sample. The Z-potential at pH values equal to 2, 4, 6, 7, 8, 10 and 12 was measured to further confirm functionalization and to observe the behaviour of surface groups. The isoelectric point of MNPs was at around pH=4.5 while the one of F-MNPs was near pH=9, indicating a change in surface groups. CHNS revealed an increase in nitrogen, carbon and hydrogen content after functionalization. The functionalization percentage was computed as $F = \frac{\omega_N \cdot MW_H}{\omega_H \cdot MW_N} \cdot 100$, where ω_N is the mass percentage of N of F-MNPs, ω_H is the mass percentage of H of MNPs, and MW_N and MW_H are the respective atomic weights, this resulted in $F = 7.7\%$. The DLS was employed to estimate the particle size for MNPs and F-MNPs, three consecutive measurements were performed for each sample. The mean size and the standard deviation increase in consecutive measurements both for MNPs and F-MNPs. This observation is explained by the formation of particle aggregates favoured by high surface energies and possibly magnetic forces [8], greater aggregation was observed for uncoated particles, consistent with literature findings [8] [14]. Aggregates were also observed in SEM micrograph.

Adsorption tests

From GC characterization of supernatants, the adsorption percentages were computed for each analyte i in each sample as $A\% = \frac{C_{i,0} - C_i}{C_{i,0}} \cdot 100$, where $C_{i,0}$ is the concentration in the starting VFA solution and C_i is the concentration in the supernatant. The resulting adsorption percentages ranged from 3 % to 41 %, the overall average adsorption percentage was 18 % and the standard deviation was 9 %. Replicating the tests for the two samples with the highest $A\%$ a high experimental variability was observed. The results obtained confirm the adsorption of VFAs by F-MNPs and MNPs under agitation. There was no evidence of a strong selectivity in the capture process. Among the possible causes for variability, the inhomogeneity in the aggregation extent in different adsorption samples was considered the most plausible.

Desorption tests

The desorption percentage for each analyte i , in each sample, was defined as the ratio between the milligrams of i in the supernatant after desorption ($mg_{i,des}$) and the milligrams of i adsorbed on the nanoparticles ($mg_{i,ads}$), estimated as:

$$D_i\% = \frac{mg_{i,des}}{mg_{i,ads}} \cdot 100 = \frac{C_{i,des} \cdot 4 \cdot 10^{-3}}{(C_{i,0} - C_i) \cdot 2 \cdot 10^{-3}} \cdot 100$$

Where $C_{i,0}$ is the concentration of i in the starting VFA solution, and C_i is the concentration of i in the adsorption supernatant expressed in [mg/L]; $C_{i,des}$ is the concentration in the desorption supernatant expressed in [mg/L]; $4 \cdot 10^{-3}$ is the desorption solvent's volume in [L] and $2 \cdot 10^{-3}$ is the liquid volume for adsorption in [L]. The highest desorption percentage obtained was 52 % and the lowest was 6 %, with an average of 21 % and a standard deviation of 14 %, indicating a highly varied sample of measurements. The results obtained confirm that short-chain carboxylates are present in the supernatants after the desorption step. The presence of methanol and unidentified peaks reduced the accuracy of the analysis. Desorption was not verified for all analytes in all samples, but the available data confirm the reversible nature of the capture process.

Second cycle adsorption

The developed nanoparticles are aimed to be employed multiple times for VFA extraction. This possibility was validated by performing a second adsorption test on nanoparticles separated after desorption. The adsorption percentages ranged between 2 % and 35 %. Nanoparticles captured on average 18 % of the carboxylates, with a standard deviation of 8 %. The quantity of carboxylates adsorbed on the surface of F-MNPs during this second adsorption step is relatively high considering that the previous desorption was not complete, and so part of the interaction sites on F-MNPs' surface was already occupied. These non-desorbed VFAs reduce the number of amines groups available for interaction, add steric hindrance but may also interact with carboxylates in solution due to their high affinity. Additionally, these nanoparticles might be more disaggregated and have more surface available as they have undergone adsorption, desorption, and adsorption again, so have been stirred for longer times and at higher temperatures (40° C during desorption) than the nanoparticles of the first adsorption.

Coating reaction optimization

Considering the importance of functionalisation, and the lack of studies investigating the effects of parameters involved, part of the work was devoted to optimising the functionalisation reaction. The optimization aims at maximizing the number of amine

groups on the surface of nanoparticles, as measured by the functionalization percentage, and the obtained results are summarised in Table 1.

Table 1: Functionalization percentages of tests using different coating agents, molar ratios and reaction procedures.

Coating agent	MNPs/ Coating agent	Reaction procedure	Fun. %
Hydrolysed-APTES	1:9	5-hours	7.7 %
Hydrolysed-APTES	1:18	5-hours	5.9 %
Hydrolysed-APTES	1:4	5-hours	10.7 %
Hydrolysed-APTES	1:4	1-hour	8.4 %
APTES	1:4	1-hour	2.6 %
APTES	1:4	5-hours	8 %

The F-MNPs obtained with hydrolysed APTES, 1:4 molar ratio and the 5-hour procedure have the highest functionalization percentage. The low functionalisation percentages obtained are satisfactory considering the steric hindrance of the ligand used.

5. Conclusions

Production of valuable products starting from organic food waste is a prolific field of research in this decade, it's an opportunity to exploit wastes that represent a burden and that otherwise would be disposed of.

This work is framed in this context and has proposed an innovative way to recover VFAs from diluted aqueous solutions using functionalized magnetic nanoparticles.

Synthesis and characterization of both MNPs and F-MNPs were performed.

The adsorption was successful, and nanoparticles didn't show strong selectivity for one analyte over the others; a high experimental variability was observed.

Desorption of VFAs in deionised water and methanol was verified even though residues of methanol reduced the number of accurate data available. For further analysis, the development of a new analytical method or new calibration lines is suggested. The desorption was not complete indicating a resilient interaction between carboxylates and amine groups, that may require peculiar conditions for desorption. The possibilities of using volatile alkali, acidic environments or high boiling organic solvents for desorption should be investigated in the future.

Second adsorption tests resulted in similar capture values with respect to the first cycle, despite the desorption was not complete.

The tests conducted verified the feasibility of each process step. From these experiments, it is not possible to conclude whether it is optimal to use MNPs or F-MNPs for adsorption, nor to identify which are the best conditions for each step (e.g. agitation method, contact time, temperature, pH) as only one replicate per experiment was performed. Optimization of each step

to improve performance should be the object of further studies.

This first experimental study of this novel methodology paves the way for future studies on many different aspects. A more in depth-characterization of the nanoparticles should be performed to better understand their aggregation, degradation under different conditions, magnetic response, type of interaction with carboxylates, and size distribution both before and after functionalization. The interaction of nanoparticles with fermentation culture could also be compelling to enable in-situ extraction of VFAs. Additionally, adsorption and desorption steps should be investigated to reduce the variability observed, identify the governing equations, and select the optimal conditions. These future research phases should use real fermentation broth to observe if capture is hindered by poisoning of the nanoparticles' surface with other compounds in solution. Finally, a detailed cost analysis should be conducted to support technological decisions in order to achieve a concretely feasible process.

Bibliography

- [1] J. Gustavsson, C. Cederberg, and U. Sonesson, "Global food losses and food waste - Extent, causes and prevention," 2011.
- [2] ISPRA, *Rapporto Rifiuti Urbani, Edizione 2020*, vol. 44, no. 8. 2020.
- [3] S. K. Mahmad Rozi, S. Bakhshaei, N. S. Abdul Manan, and S. Mohamad, "Superhydrophobic magnetic nanoparticle-free fatty acid regenerated from waste cooking oil for the enrichment of carcinogenic polycyclic aromatic hydrocarbons in sewage sludges and landfill leachates," *RSC Adv.*, vol. 6, no. 90, pp. 87719–87729, 2016.
- [4] C. C. Hua *et al.*, "Size-controlled synthesis and characterization of Fe₃O₄ nanoparticles by chemical coprecipitation method," *Sains Malaysiana*, vol. 37, no. 4, pp. 389–394, 2008.
- [5] B. Feng *et al.*, "Synthesis of Fe₃O₄/APTES/PEG diacid functionalized magnetic nanoparticles for MR imaging," *Colloids Surfaces A Physicochem. Eng. Asp.*, vol. 328, no. 1–3, pp. 52–59, 2008.
- [6] X. C. Shen, X. Z. Fang, Y. H. Zhou, and H. Liang, "Synthesis and characterization of 3-aminopropyltriethoxysilane-modified superparamagnetic magnetite nanoparticles," *Chem. Lett.*, vol. 33, no. 11, pp. 1468–1469, 2004.
- [7] K. Can, M. Ozmen, and M. Ersoz, "Immobilization of albumin on aminosilane modified superparamagnetic magnetite nanoparticles and its characterization,"

Colloids Surfaces B Biointerfaces, vol. 71, no. 1, pp. 154–159, 2009.

- [8] Y. Wei, B. Han, X. Hu, Y. Lin, X. Wang, and X. Deng, "Synthesis of Fe₃O₄ nanoparticles and their magnetic properties," *Procedia Eng.*, vol. 27, no. 2011, pp. 632–637, 2012.
- [9] H. El Ghandour, H. M. Zidan, M. M. H. Khalil, and M. I. M. Ismail, "Synthesis and some physical properties of magnetite (Fe₃O₄) nanoparticles," *Int. J. Electrochem. Sci.*, vol. 7, no. 6, pp. 5734–5745, 2012.
- [10] C. Xu, X. Lu, and H. Dai, "The Synthesis of Size-Adjustable Superparamagnetism Fe₃O₄ Hollow Microspheres," *Nanoscale Res. Lett.*, vol. 12, no. 1, pp. 4–13, 2017.
- [11] V. C. Karade *et al.*, "APTES monolayer coverage on self-assembled magnetic nanospheres for controlled release of anticancer drug Nintedanib," *Sci. Rep.*, vol. 11, no. 1, pp. 1–12, 2021.
- [12] T. Ahn, J. H. Kim, H. M. Yang, J. W. Lee, and J. D. Kim, "Formation pathways of magnetite nanoparticles by coprecipitation method," *J. Phys. Chem. C*, vol. 116, no. 10, pp. 6069–6076, 2012.
- [13] S. Villa, P. Riani, F. Locardi, and F. Canepa, "Functionalization of Fe₃O₄ NPs by silanization: Use of amine (APTES) and thiol (MPTMS) silanes and their physical characterization," *Materials (Basel)*, vol. 9, no. 10, 2016.
- [14] D. Maity and D. C. Agrawal, "Synthesis of iron oxide nanoparticles under oxidizing environment and their stabilization in aqueous and non-aqueous media," *J. Magn. Magn. Mater.*, vol. 308, no. 1, pp. 46–55, 2007.