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Research Article

NANOENCAPSULATION OF FOLIC ACID THROUGH ELECTROSPRAYING AND ITS FORTIFICATION IN YOGHURT

HINGU M.N. AND RAMANI V.M.*
Department of Dairy Microbiology, College of Dairy Science, Amreli, 365601, Kamdhenu University, Gandhinagar, 382010, India
*Corresponding Author: Email - vimalmramani@gmail.com

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Abstract- Folic acid is an essential component required for body functions and it's deficiency may lead to various disorders and diseases. Humans and animals, cannot synthesize folate and therefore it must be obtained through their diet from various vegetables and fruits. Naturally occurring folates lose more than half of its bioactivity due to harvesting and subsequent processing. Synthetic folic acid is degraded upon exposure to heat, UV, and sunlight resulting in lowering its bioavailability. Folic acid is a water soluble, so it is not stored in the body so the, leftover amounts of folate will leave the body Fortification of encapsulated folic acid can increase the bioavailability as it will reach intact in intestine. Nanoencapsulation protects the bioactive compound from adverse environmental conditions as well as nanoencapsulation improves the bioavailability, stability and controlled release at targeted site. So, in the present study, the nanoencapsulation of folic acid was carried out using the electrospraying for its fortification in yoghurt. This nanoencapsulated folic acid was fortified in the yoghurt and showed higher growth of yoghurt culture as compared to control yoghurt. The growth of *L. bulgaricus* and *S. thermophilus* was 7.11 log cfu/g and 7.35 log cfu/g in nanoencapsulated folic acid fortified yoghurt, respectively and in case of control yoghurt there was a lower growth of *L. bulgaricus* and *S. thermophilus i.e.,* 6.99 log cfu/g and 7.25 log cfu/g, respectively. As well as the net folic acid content was also found very high in nanoencapsulated folic acid fortified yoghurt as compared to control yoghurt. After 15 days of storage, the concentration of folic acid was 158.87 µg in the nanoencapsulated folic acid fortified yoghurt, whereas very low concentration of folic acid was found in control yoghurt *i.e.,* 19.32 µg.

Keywords- *Folic acid, Electrospraying, Nanoencapsulation, Whey protein concentrate, Bioavailability*

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Introduction

Folic acid (FA) is a water soluble vitamin and which is also known as vitamin B9. It is an essential component required for body functions and it's deficiency may lead to various disorders and diseases like atherosclerotic cardiovascular disease, shortness of breath, neurological and neuropsychiatric disorders and also congenital defects, anemia, homocysteinemia, mental confusion, irritability, depression and carcinogenesis, dementia and Alzheimer's disease [1-6]. It takes participation in several functions like biosynthesis of amino acids, nucleotides, neurotransmitters, and certain vitamins. So, it is required at all age health group [7]. Rogers *et al*., (2018) [8] analysed the total 11 surveys, which were reported earlier, and they mentioned that the prevalence of folate deficiency was more than 40% in most countries. Venkatesh *et al*., (2021) [9] analysed the data from the database like global index medicus, PubMed etc. for the folate deficiency in India and they reported 37% overall prevalence of folic acid deficiency.

Recommended dietary allowances (RDA) mentioned by the Food Safety and Standards Authority of India [10] for the folic acid is ranging from 25 to 500 µg/day for different group of people. This folic acid is consumed either in its natural form or mostly as a supplement to overcome the deficiency [11] but, the folic acid is removed from the body as it is water soluble and body cannot store it. So, there is a less bioavailability of folic acid and leads to its deficiency.

Nanoencapsulated particles as reduced to nanoscale, results in effective delivery system and it provides greater bioavailability of bioactive compounds due to their faster rate of digestion [12]. The nanoencapsulated folic acid increases the bioavailability [13,14]. Electrospraying is a versatile method for the nanoencapsulation which allows the direct incorporation of the bioactive compound in a polymeric solution. The electrospraying method is one of the modern techniques among different available techniques for the nanoencapsulation, which aids in the sustained and controlled release, higher surface to volume ratio, enhanced stability of bioactive, extend the long shelf life of coated nutraceuticals components [15, 16].

Yoghurt is an ancient food and has been a part of the human diet since time immemorial and has been promoted as a healthy food [17]. Yoghurt being a functional food, it renders several health benefits. Yoghurt is used as a tool for fortification of various components like peanut skin extract powder [18], modified starch, whey protein concentrate spirulina powder [19], seed flour of fenugreek and Moringa oleifera [20], turmeric powder [21], date palm [22], selenium [23]. Yoghurt is also used for the fortification of various nutraceutical components like chromium [24], barley β-glucan [25,26]. The present study has been undertaken for nanoencapsulation of folic acid through electrospraying and its fortification in yoghurt. As it render long-shelf life of coated bioactive components without varying organoleptic properties. As there is no application of high temperature, the nutritive value of product is being conserved.

Materials and Methods

Preparation of solution and optimization study of folic acid-WPC-proline-lactate for electrospraying

The solution of folic acid (Sigma Aldrich) was prepared as per the product information sheet of Sigma Aldrich *i.e.*, in 1 M NaOH (40mg/ml) (SD Fine chemicals) in amber coloured bottle and the WPC (AS-IT-IS brand) was prepared in different concentration *i.e.*, 10%, 20%, 30% and 40%. The different combination ratios of proline (P) (Sigma Aldrich) and lactate (LA) (Sigma Aldrich) with WPC and folic acid were used for preparation of solution for nanoencapsulation through electrospraying as indicated in [Table-1].

Table-1 Preparation of electrospraying solution with different combination

Sample Code	WPC %	P:LA ratio (w/v)	FA mcg/ml	Sample Code	WPC %	P:LA ratio (w/v)	FA mcg/ml
A	10	1:1 1:2 1:3 3:1 2:1	100	O	30	1:1 1:2 1:3 3:1 2:1	100
B			150	P			150
C			200	Q			200
D			400	R			400
E			600	S			600
F			800	T			800
G			1000	U			1000
H	20	1:1 1:2 1:3 3:1 2:1	100	V	40	1:1 1:2 1:3 3:1 2:1	100
I			150	W			150
J			200	X			200
K			400	Y			400
L			600	Z			600
M			800	AA			800
N			1000	AB			1000

Electrospraying process

Electrospraying instrument (Model FLUIDNATEK LE-10, Make - Bioincia S. L., Valencia, Spain) was used for electrospraying. The electrospraying system, equipped with a variable high voltage power supply. The prepared solutions [Table-1] were subjected to electrospraying at different flow rate, voltage and distance as indicated in [Table-2].

Table-2 Electrospraying of solutions at different flow rate, voltage and distance

Trial	Flow rate (µl/hr)	Voltage (kv)	Distance (cm)
1	200	13	10
2			12
3			14
4	200	15	10
5			12
6			14
7	200	17	10
8			12
9			14
10	300	13	10
11			12
12			14
13	300	15	10
14			12
15			14
16	300	17	10
17			12
18			14
19	400	13	10
20			12
21			14
22	400	15	10
23			12
24			14
25	400	17	10
26			12
27			14

After obtaining optimum tailor cone in the electrospraying at the flow rate, voltage and distance of 300 (µl/hr), 15 (kv) and 12 (cm), respectively, the following combinations as per [Table-3] were further carried out.

Table-3 Electrospraying of solutions at fixed flow rate and voltage but at different distance

Trial	Flow rate (µl/hr)	Voltage (kv)	Distance (cm)
28	300	15	11
29			12
30			13
31			14
32			15

Measurement of particle size and zeta potential

The nanoparticles produced through the electrospraying were dispersed in the distilled water for the particle size measurement and zeta potential in the particle size analyzer (zetasizer). Nanosight (Model - NS300, Make - Malvern Panalytical Ltd. UK) equipment was used for size distribution and concentration (number of particle) analysis.

Topographical Characteristics of Nanoencapsulated folic acid

Scanning electron microscope (SEM) (Model - ZEISS EVO-18, Make - Carl Zeiss, Germany; analysis carried out at Food Testing Laboratory, Junagadh Agricultural University, Junagadh, Gujarat) was used to study the size and topographical characteristics of nanoencapsulated folic acid powder samples.

Study on growth of yoghurt culture

For the *L. bulgaricus* and *S. thermophilus* count in yoghurt samples, the MRS agar (HiMedia) and M17 agar (HiMedia) has been used, respectively. The petri plates containing the appropriate dilutions were poured with melted and cooled the medium at around 45 °C, mixed properly and allowed to solidify. In case of MRS agar, a second layer of same agar (4-5 ml) was overlaid in each plate, after solidification of first layer. The Petri plates were then incubated at temperature of 37 ±1 °C for *Lactobacillus* count and 43 ±1 °C for *Streptococcus* count and typical colonies formed on the agar medium were counted after 48 h and the count is expressed as log cfu/g. The growth of yoghurt culture was recorded on the 0 day, 7th day and 15th day.

Standard curve preparation and quantification of folic acid

The quantitative analysis of folic acid was carried out by using the method developed by Kshirsagar *et al*., (2017) [27] with slight modification at λmax (281 nm) in double beam spectrophotometer (Model - UV- 1900, Make - Shimadzu, Japan). The method of Olmo *et al*. (2022) [28] was used with slight modification for the preparation of the standard curve of the folic acid and the different concentrations in the range of 100 µg/ml to 500 µg/ml were prepared in 1 M NaOH solution. Measurement for the calibration was done using the 1 M NaOH solution as a blank. The stock solution was prepared by weighing 100 mg folic acid and transferred it to the 100 ml volumetric flask. The working standard solution was prepared by diluting the 0.1 ml of stock solution in 10 ml of distilled water to have concentration of 100 µg/ml.

Statistical Analysis

The collected data were subjected to statistical analysis. Data were analysed by analysis of variance (ANOVA) and critical difference test at 5% level of significance ($P \leq 0.05$) to compare the different treatments means, with 3 replications with the help of WASP (Web Agri Stat Package) developed by the Indian Council of Agricultural Research (ICAR), New Delhi (https://ccari.icar.gov.in/wasp/index.php).

Results and Discussion

Preparation of solution and optimization study of folic acid-WPC-proline-lactate for electrospraying

The folic acid 150 µg, 10% WPC and proline to lactate ratio of 1:1 has been optimized for the nanoencapsulation process through electrospraying. The optimized flow rate, voltage and a tip-to-collector distance in the electrospraying process is mentioned in [Table-4]. The encapsulated powder was collected on flat collector which was wrapped with parchment paper in aluminum foil and packed in sterile sealed polythene bags and stored at 4°C and 37°C for analysis. The obtained powder was used for further analysis.

Table-4 Optimized process of electrospraying for nanoencapsulation of folic acid

Folic acid	WPC	P:LA ratio	Flow rate (µl/hr)	Voltage (kv)	Distance (cm)
150 µg	10%	1:1	300	15	13

Measurement of particle size and zeta potential

The particle size was analyzed through particle size analyzer, NanoSight and scanning electron microscope. The average size of the nanoparticles obtained was around 58 nm with average zeta potential of -17.25. The average size, zeta potential and concentration of nanoparticles of folic acid is given in [Table-5]. Average size and concentration of nanoencapsulated folic acid in NanoSight (NS300) is represented in [Fig-1]. Intensity of size of nanoencapsulated folic acid in the solution in NanoSight (NS300) is given in [Fig-2].

Table-5 Average size, zeta potential and concentration of nanoparticles of folic acid

Average Size (d.nm)		Average number of particle/ml
PSA	57.67 nm	3.85e+08 +/- 3.35e+06
NanoSight	59.24 nm	Average Zeta potential (mV)
SEM	56.79 nm	-17.25

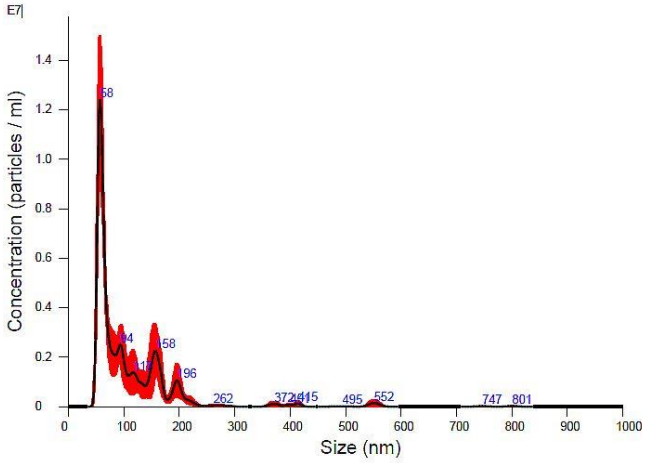

Fig-1 Average size and concentration of nanoencapsulated folic acid in NanoSight

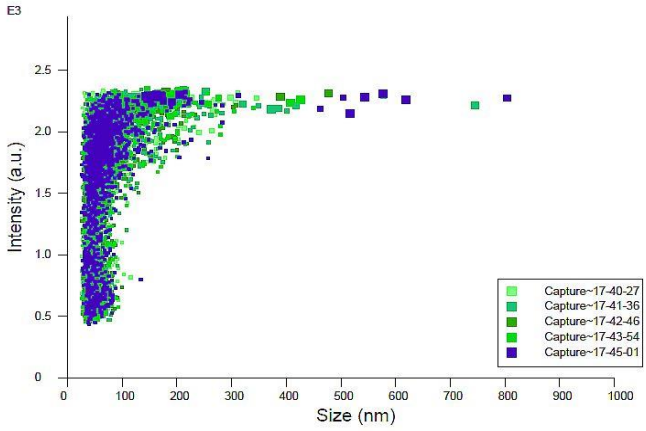

Fig-2 Intensity of size of nanoencapsulated folic acid in the solution in NanoSight

Pradeepkumar *et al*., (2019) [29] used deep eutectic solvents (DESs) *i.e.,* amino acid salt of proline and lactic acid to prepare DESs in different ratios of 1:1, 1:2, 1:3, 2:1, and 3.1 respectively for the nanoencapsulation. They observed nanoencapsulation efficiency of micelles up to 79±2% with the ratio of 1:2 and 1:3. The size of the micelles were 193 nm. Assadpour *et al*., (2016) [30] did the nanoencapsulation of folic acid by double emulsion method containing WPC/ pectin/ maltodextrin and they also reported the size of nanoparticle below 100nm. In the study of Parın *et al*., (2022) [31], the folic acid was sprayed using electrohydrodynamic system into the Polyvinyl alcohol (PVA) nanofiber and the average diameter of the fibre was ranging from 74.5 ± 15.4 to 291 ± 66.9 nm. They also mentioned that, the smooth structure of PVA/FA (folic acid) was found in the fibres, but folic acid cluster were not seen much on the PVA/FA fibre surface. The reason behind this mentioned was it may be due to the use of same solvent in both electrohydrodynamic process and thus folic acid clusters had been incorporated into the fibre structure. Evangelho *et al*., (2019) [32] encapsulated the folic acid with zein fibres. The mean diameter of zein and 1.5% folic acid nanocapsules were ranging from 369 nm to 702 nm. The lowest mean diameter found was of 268 nm for the combination of zein and 0.5% folic acid, whereas the largest mean diameter of 407 nm was found in the combination of zein with 1.0% of folic acid. Darwish *et al*., (2021) [33] studied the characteristics of nanoencapsulated iron (Fe) and folic acid (FA) fortified functional yogurt along with bovine serum albumin-nanoparticles (BSA-NPs). On the basis of results, they obtained for the zeta potential −24.6, and −21.20 mV for the different combinations of FA@ BSA-NPs, and Fe + FA@ BSA-NPs, respectively, they mentioned that the strong repulsive forces and electrostatic stabilization between them. Karami *et al*., (2020) [34] mentioned maximum zeta potential of −30 mV for electrostatically stabilized nanosuspensions. In one of the study conducted by Fathima *et al*., (2022) [35], the folic acid loaded chitosan nanoparticle (FA-Chi-NP) was formulated and had the zeta potential of +52 mV and 180 nm particle size. Further, they also mentioned about the controlled release of nanoparticles of folic acid in in vitro conditions. Pradeepkumar *et al*., (2019) [29], used the molar ratio of deep eutectic solvents (DES)-1:3 (Proline: Lactic acid) for the nanoencapsulation of folic acid using nanomicelles method and they found the zeta potential around -4.26±1 mV for micelles.

Topographical characteristics of nanoencapsulated folic acid

The topographical study of nanoencapsulated folic acid powders was carried out by using scanning electron microscopy (SEM). [Fig-3] shows Scanning Electron Microscopy (SEM) images of nanoencapsulated folic acid powders which have spherical shape without any fracture or crack on the surface of the nanocapsules. The size of nanoencapsulated folic acid was found in range between 53-90 nm.

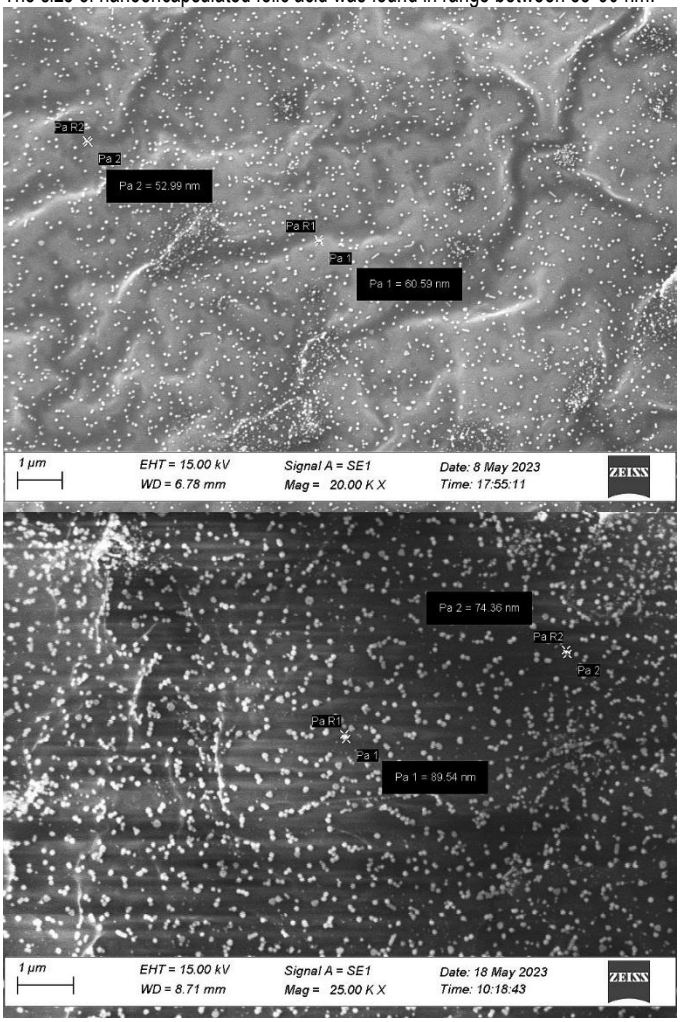

Fig-3 SEM images of nanoencapsulated curcumin powders with different combination (A) 1:10 FA: WPC with 1:1 ratio of P:LA (B) 1:10 FA: WPC with 1:2 ratio of P:LA

Pradeepkumar *et al*., (2019) [29] also reported the nanomicelles of DES-g-PGA-FA were spherical and average size of the particles was around 200 nm. Chowdhuri *et al*. (2015) [36] prepared the folic acid conjugated silver nanoparticles (Ag NPs). The folic acid was conjugated through electrostatic interaction on the surface of functionalized silver nanoparticles. To identify the size they used transmission electron microscope (TEM) and for the morphology they used field emission scanning electron microscopy (FESEM). The similar results were also found in the study of Gunduz *et al*., (2014) [37], where they prepared folic acid solution with dicyclohexyl carbodiimide (DCC) with 1:1 ratio and was added in dimethyl sulfoxide (DMSO) and was subjected to stirring for 2 h. The shape and morphology of folic acid- polyethylene glycol-magnetic nanoparticles (FA–PEG–MNP) were seen regular and spherical and the size was between 10–40 nm. Teng *et al*., (2013) [38] prepared the nanoparticles of soy protein isolate (SPI) that was conjugated with folic acid (FA) using ethanol desolvation method. In the SEM analysis, they also mentioned that the nanoparticles were spherical with smooth surfaces and the size of it varied from 150 to 250 nm.

Study on growth of yoghurt culture

The yoghurt (purchased from the local supermarket) was fortified with the nanoencapsulated folic acid in aseptic condition and was studied for the growth of *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus*. The growth of *L. bulgaricus* and *S. thermophilus* is given in [Table-6] and [Table-7], respectively and graphically represented in [Fig-4]. The control yoghurt sample and the yoghurt fortified with nanoencapsulated folic acid was stored in the refrigerated conditions and the growth of starters were recorded on 0 day, 7th day and 15th day. The growth of *L. bulgaricus* and *S. thermophilus* was comparatively higher in the yoghurt containing nanoencapsulated folic acid than control sample. In fortified yoghurt, the highest growth of *L. bulgaricus* was found on zero day *i.e.,* 7.28 log cfu/g which was followed by 7.18 log cfu/g and 7.11 log cfu/g on day 7 and 15, respectively in refrigerated storage. The growth of *L. bulgaricus* in control sample was 7.21 log cfu/g on 0 day, which was followed by 7.15 log cfu/g and 6.99 log cfu/g on 7th and 15th day, respectively.

In case of fortified yoghurt, the highest growth of *S. thermophilus* was found on zero day *i.e.,* 7.46 log cfu/g which was followed by 7.42 log cfu/g and 7.35 log cfu/g on day 7 and 15, respectively in refrigerated storage. The growth of *S. thermophilus* in control sample was 7.42 log cfu/g on 0 day, which was followed by 7.38 log cfu/g and 7.25 log cfu/g on 7th and 15th day, respectively.

Table-6 Survival growth of L. bulgaricus during refrigerated storage

Yoghurt	Growth (log cfu/g)		
	0 day	7 days	15 days
Control (LC)	7.21	7.15	6.99
Yoghurt with nanoencapsulated folic acid (LT1)	7.28	7.18	7.11

Table-7 Survival growth of S. thermophilus during refrigerated storage

Yoghurt	Growth (log cfu/g)		
	0 day	7 days	15 days
Control (LS)	7.42	7.38	7.25
Yoghurt with nanoencapsulated folic acid (ST1)	7.46	7.42	7.35

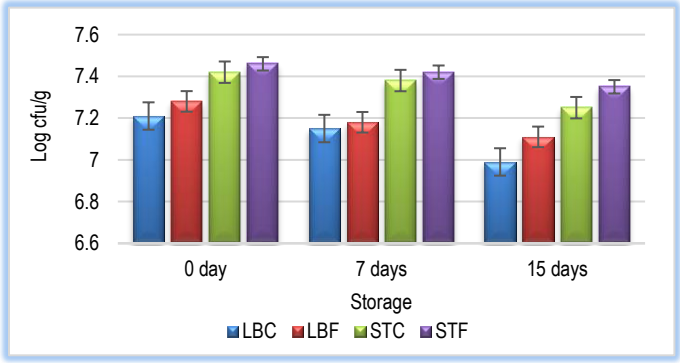

Fig-4 Growth of *L. bulgaricus* and *S. thermophilus* during refrigerated storage (LBC= Growth of *L. bulgaricus* in control yoghurt; LBF= Growth *of L. bulgaricus* in nanoencapsulated folic acid fortified yoghurt; STC= Growth of *S. thermophilus* in control yoghurt; STF= Growth of *S. thermophilus* in nanoencapsulated folic acid fortified yoghurt)

Darwish *et al*., (2021) [33] prepared the stirred fortified yoghurt (SFY). That was fortified with nanocapsules of folic acid (FA) and iron (Fe) with bovine serum albumin-nanoparticles (Fe + FA@BSA-NPs). The mean value of the growth of yoghurt culture was studied for 21 days of storage. They reported the mean growth of 7.30 log cfu/g and 7.10 log cfu/g for *S. thermophilus* and *L. bulgaricus*, respectively. Wu *et al*., (2017) [39] enriched the yoghurt with folic acid. Yoghurt was prepared using the *L. bulgaricus* and *S. thermophilus* along with *Lactobacillus plantarum*. They reported the growth around 9.12 log cfu/g after 14 days. Khalili *et al*., (2020) [40] studied the growth survival of yoghurt culture in which all the yoghurts were fortified with folic acid. They prepared the yoghurt with native culture of *L. bulgaricus* and *S. thermophilus* (Y1), native yoghurt culture along with Lactobacillus acidophilus and *Bifidobacterium animalis* subsp. lactis (Y2), native yoghurt culture along with *Lactobacillus plantarum* (Y3), native yoghurt culture along with *Lactococcus lactis* subsp. lactis (Y4) and native yoghurt culture along with other strain of *Lactobacillus plantarum*. They reported the minimum growth in Y2 *i.e.,* 5.86 ± 0.75 log cfu/ml, whereas the highest growth was in Y3 *i.e.,* 9.53 ± 0.50 log cfu/ml on the 14th day of refrigerated storage. Birollo *et al*., (2000) [41], used *L. delbrueckii* subsp. *bulgaricus* and *S. thermophilus* for the yoghurt manufacture at industrial level. They studied the viability of yoghurt cultures stored in cold storage of 6 °C up to 60 days. They mentioned that the initial growth of yoghurt culture in the appropriate media was higher. Subsequently, the growth of the yoghurt culture *i.e., L. delbrueckii* subsp. *bulgaricus* and *S. thermophilus* was reduced to 5 x 107 to 5 x 105 log cfu/g, respectively. Perez-Esteve *et al*., (2016) [42] studied the viability of yoghurt cultures in folic acid fortified yoghurt and yoghurt without any fortification. They reported the reduction in growth of yoghurt culture in both type of yoghurt, however the fortified yoghurt was having lower reduction of growth as compare to control sample.

Standard curve preparation and quantification of folic acid

The absorbance's of folic acid is shown in [Table-8] and [Fig-5]. Standard curve of folic acid was prepared by using different concentration of water soluble folic acid in distilled water. The optimum wavelength for maximum absorption of water soluble folic acid (λmax) is 281 nm [27]. The absorbance of standard folic acid solution was measured at 281 nm against distilled water as a blank. Standard curve was plotted with absorbance against concentration.

Table-8 Absorbance of folic acid at its different concentration in distilled water

No.	Concentration of folic acid (µg/ml)	Absorbance (WL- 281)
1	2	0.11
2	4	0.21
3	6	0.32
4	8	0.43
5	10	0.53

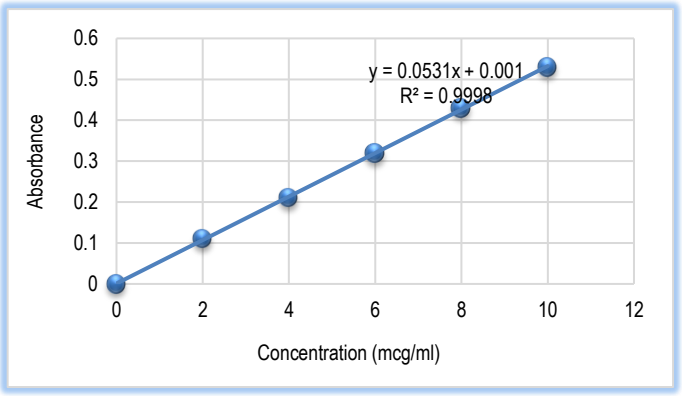

Fig-5 Standard curve of folic acid

Quantification of folic acid in the yoghurt during refrigerated storage

The changes in the folate concentration in the yoghurt during the 15 days of refrigerated storage study is given [Table-9] and graphically represented in [Fig-6]. The maximum concentration of 158.87 µg of folate was recorded after 15 days. The concentration of folic acid in yoghurt was continuously in increasing order *i.e.,* 74.34 µg, 95.71 µg and 158.87 µg on zero day, seven days, and fifteen days.

Table-9 Changes in the folic acid concentration in the yoghurt

Yoghurt	Folic acid concentration (µg)		
	0 day	7 days	15 days
Control (C)	39.37	28.72	19.32
Yoghurt with nanoencapsulated folic acid (TS1) (Total folic acid produced: nanoencapsulated folic acid + Folic acid produced by yoghurt cultures)	74.34	95.71	158.87
Net folic acid: Nanoencapsulated folic acid- NF (TS1-C)	34.97	66.99	139.55

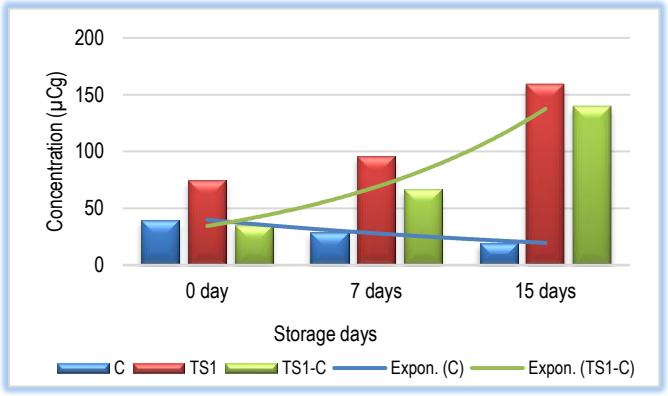

Fig-6 Changes in the folic acid concentration in the yoghurt
(C= Folic acid concentration in control yoghurt; TS1= Folic acid concentration in the yoghurt fortified with nanoencapsulated folic acid; NF= Net folic acid concentration calculated by difference *i.e.,* TS1-C)

The net folic acid increased was 44.66% after 7 days and 93.03% after 15 days of refrigerated storage. As such milk food is not a good source of dietary folate but the fermented milk food is producing folate content as a part of fermentation through lactic acid bacteria [43, 44, 45]. Even certain lactic cultures tend to increase the folate in the fermented milks including the *Lactobacillus* spp. and *Streptococcus* spp. [46, 47, 48, 49, 50]. Folic acid production by some organisms: *S. thermophilus* CSCC2000 produces 40-50 ng [51], *S. thermophilus* 908 produces ≈76 ng [52], L. lactis subsp. cremoris produces ≈60 ng [53]. But the increase of folic acid by this lactic acid bacteria is not fulfilling the requirement of daily dose of dietary folate.
Ruiz-Rico *et al*., (2017) [54] encapsulated the folic acid in the mesoporous silica for the food fortification purpose. In their fortification study, they reported the 77% of folic acid after 28 days of refrigerated storage. Rao and Shahani (1987) [55] reported the reduction of free folic acid from 9.8 µg to 1.6 µg in the yoghurt on 16th day of refrigerated storage.
Madziva *et al*., (2006) [56] encapsulated folic acid and used in the cheddar cheese- a ripened milk product. They studied the retention of folic acid during the cheddar cheese ripening up to 3 months. After 3 months of period, the concentration of folic acid was 360 µg in the cheese fortified with encapsulated folic acid whereas the concentration of folic acid was 109 µg in the cheese fortified with free folic acid. They concluded that the concentration of folic acid was continuously increasing in the cheese fortified with the encapsulated folic acid.

Conclusion

From the present study it is recommended to use electrospraying process for nanoencapsulation of folic acid. With the optimized combination *i.e.,* folic acid (150 µg), WPC (10%), ratio of proline and lactate (1:1) along with electrospraying conditions can be effectively used to get better nanoencapsulated folic acid. This nanoencapsulated folic acid can be fortified in the yoghurt, which supports the growth of yoghurt culture as well as it significantly increases the net folic acid that may fulfill the requirement of daily dosage as per the guidelines of the FSSAI.

Application of research: This nanoencapsulated folic acid produced through the electrospraying techniques can be used for the fortification in yoghurt and other milk based fermented foods.

Research Category: Nanoencapsulation, Electrospraying

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Research project name or number: Research station study

Author Contributions: All authors equally contributed

Author statement: All authors read, reviewed, agreed and approved the final manuscript. Note-All authors agreed that- Written informed consent was obtained from all participants prior to publish / enrolment

Study area / Sample Collection: College of Dairy Science, Amreli, 365601
Cultivar / Variety / Breed name: Dairy Animal

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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