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# In vitro gastrointestinal digestion of a bisdemethoxycurcumin-rich *Curcuma longa* extract and its oral bioavailability in rats

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## Abstract

**Background:** Nonetheless curcumin has potential health benefits, its low bioavailability limits the application of conventional turmeric extract with curcumin as major curcuminoid. This is a comparative study to assess the stability, bioaccessibility and biological activity of BDMC in standardized *C. longa* extract (REVERC3) relative to curcumin in regular turmeric extract (RTE). Here we report the preparation of a standardized *Curcuma longa* extract (REVERC3[™]) standardized to contain $75 \pm 5$ w/w % bisdemethoxycurcumin (BDMC), $1.2 \pm 0.8$ w/w % curcumin and $10 \pm 5$ w/w % demethoxycurcumin (DMC). The turmeric extracts were subjected to in vitro gastrointestinal digestion and the curcuminoids in undigested and digested samples were analyzed using HPLC to determine the bioaccessibility. Further, the undigested and digested samples were evaluated for lipase inhibition and antioxidant activities. Male Wistar rats were administered with single dose (1000 mg/kg) of standardized *C. longa* extract and RTE to determine the plasma concentration of BDMC and curcumin respectively at different time points using LCMS/MS.

**Results:** The bioaccessibility of BDMC was significantly higher than curcumin ($p < 0.05$). BDMC was found superior to curcumin having significant lipase inhibitory effect ($p < 0.01$), ABTS radical scavenging ($p < 0.05$), and nitric oxide scavenging activities ($p < 0.01$). Interestingly, the relative bioavailability of BDMC in standardized *C. longa* extract was 18.76 compared to curcumin. The $C_{max}$ of BDMC was 4.4-fold higher than curcumin.

**Conclusion:** BDMC is reported to have higher bioaccessibility and bioavailability than curcumin. Our findings rationalize use of BDMC-enriched standardized *C. longa* extract for improved physiological benefits counteracting the regular turmeric extract with less bioavailable curcumin as major curcuminoid.

**Keywords:** Turmeric, Curcuminoids, Gastrointestinal, Rats

## Background

One of the extensively being used culinary spice in medicinal preparations is *Curcuma longa* L. (Turmeric). The rhizomes of the plant are a rich source of bioactive polyphenols called curcuminoids [curcumin, demethoxycurcumin (DMC) and bisdemethoxycurcumin (BDMC)] (Paramasivam et al. 2009). Turmeric extract conventionally contains higher content of curcumin (70–75%) which attributes majorly to its biological activities (Hewlings and Kalman 2017). Over the years, ample data through clinical and experimental studies have been documented of turmeric extract (Kalpravidh et al. 2010; Lim et al. 2011; Aditya et al. 2012; Panahi et al. 2014). The therapeutic benefits of curcumin include anti-inflammatory, antioxidant, anticancer actions and treatment of digestive disorders (Jurenka 2009; Anand et al. 2008; Thavorn et al. 2014). Nevertheless, curcumin is a therapeutically potent molecule, its poor bioavailability and chemical instability limits the effective use of it (Anand

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et al. 2007; Siviero et al. 2015). DMC (17%) and BDMC (3%) along with curcumin synergistically contribute to the pharmacological activities of turmeric extract (Sandur et al. 2007; Amalraj et al. 2017). These analogues of curcumin are reported to be effective individually with a better stability compared to curcumin (Yodkeeree et al. 2009).

It is well-known that bioavailability of dietary ingredients is a function of gastrointestinal transformation and bioaccessibility (Cuomo et al. 2018). These factors are not favorable in case of curcumin, thus restricting its bioavailability. More recently Wang et al. (2017) reported the absorption of the three curcuminoids through active transport mechanism. Here we propose that a standardized *C. longa* extract containing BDMC as the major curcuminoid (REVERC3™), comparatively more stable and active than regular turmeric extract (RTE) following the in vitro gastrointestinal digestion. The objective of this study was to compare the bioaccessibility and bioavailability of BDMC with curcumin. In this comparative study we have attempted to evaluate the stability of BDMC over curcumin in the intestinal phase alongside comparing the anti-lipase and antioxidant activities of turmeric extracts enriched with respective curcuminoids, following in vitro digestion. Further the oral bioavailability of BDMC in standardized *C. longa* extract was compared with curcumin in RTE.

## Methods
### Chemicals and reagents
Bisdemethoxycurcumin (HPLC grade, 98%), curcumin (HPLC grade, 98.6%), 2,2 diphenyl-1-picrylhydrazyl (DPPH), 2, 2′-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS), sodium nitroprusside, nitro blue tetrazolium (NBT), porcine pancreatic lipase and *p*-nitrophenylbutyrate (*p*-NPB) were purchased from Sigma Aldrich. All other chemicals used were of LR grade.

### Collection of plant material
The dried turmeric rhizomes were collected from parts of Salem district, Tamil Nadu, India during the month of March. The *C. longa* raw material was taxonomically authenticated at R&D Center, Viya Herbs Pvt Ltd., Bangalore, India (Voucher specimen no. VH/CUR/VS20-02).

### Preparation of BDMC-enriched standardized C. longa extract
Powdered turmeric rhizomes (100 g) were extracted using six bed volumes of ethanol (95–98% v/v) at 65 °C for 2 h in a solvent extractor. The extraction was repeated two times and the combined extract after filtration was evaporated to dryness to yield a curcuminoid-rich crude extract. 10.5 g of extract mixed with 25 g silica gel was
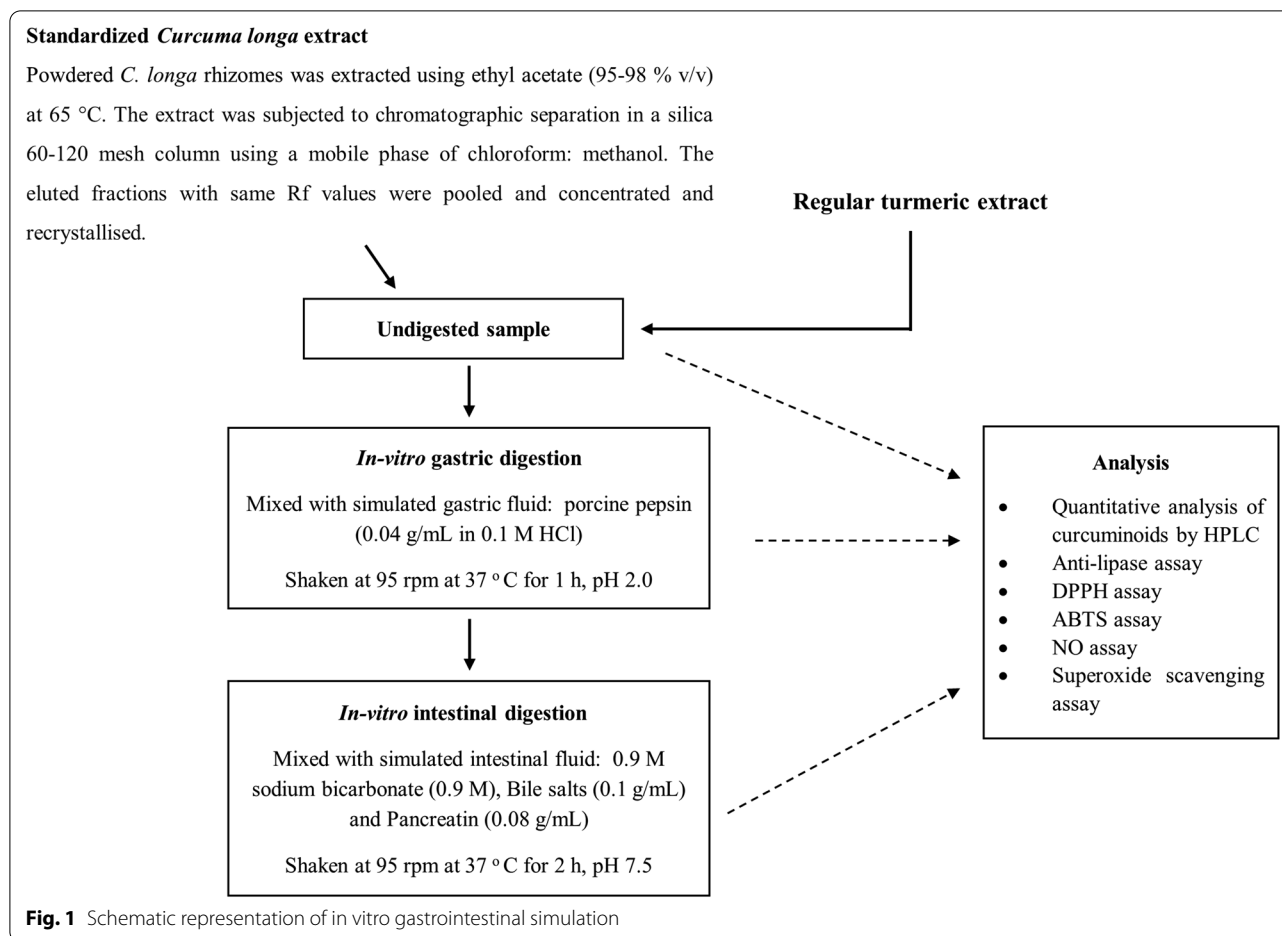
further subjected to chromatographic separation in a 60–120 mesh column (46 × 2 cm) containing 150 g of silica gel, using a mobile phase of ethyl acetate: ethanol. The separation was carried out with a beginning ratio of ethyl acetate (99.0%): ethanol (1.0%) and increasing the polarity thereafter with the increment of 1% each time to collect the fractions. Twenty eluted fractions (50 mL each) were analyzed for the presence of curcuminoids using thin layer chromatography (TLC) silica gel (Merk-60 F254, 0.25 mm thick) plate. Chloroform:ethanol:glacial acetic acid (94:5:1) was as the developing solvent system for TLC. Based on the Rf values, the fractions were pooled and evaporated to dryness. The individual curcuminoids isolated by column chromatography were further purified by recrystallization using methanol: ethyl acetate in the ratio 5:2 at 7 °C. The crystals obtained were separated by filtration. The recrystallized fraction containing > 70% BDMC by HPLC was considered as BDMC-enriched standardized *C. longa* extract. In-house prepared regular turmeric extract (RTE) (95% curcuminoids) was used for the comparison in this study. Preparation of regular turmeric extract is detailed in Additional file 1.

### In vitro gastrointestinal simulation
The turmeric extracts were subjected to simulated gastrointestinal digestion described elsewhere with slight modifications (Ryan et al. 2008). Briefly, 20 mL of the saline solution containing 1 mL plant extracts were acidified to pH 2.0 by adding porcine pepsin preparation (40 mg/mL in 0.1 M HCl) and allowed for gastric digestion at 37 °C in a shaker incubator. After 1 h, the pH of the solution was adjusted to 5.3 using 0.9 M NaHCO$_3$ solution. To this added 200 μL of bovine and porcine bile extract solution (100 mg/mL in saline), and 100 μL of pancreatin solution (80 mg/mL in saline). The pH was readjusted to 7.5 using 1 M NaOH followed by incubation at 37 °C for 2 h to accomplish the intestinal digestion phase. Aliquots of samples before and after digestion were centrifuged at 5000*g*; upper phase used for anti-lipase and antioxidant assays. The samples were further used for HPLC analysis to determine the bioaccessibility. Figure 1 shows the schematic overview of the in vitro simulated digestion.

### HPLC analysis
In this study, HPLC was used to quantify the content of BDMC in the standardized *C. longa* extract using a Shimadzu LC2030 C Prominence-i (Japan) system equipped with a high sensitivity LC2030 ultraviolet (UV) detector and LabSolutions software. Separation was achieved in Kinetex C-18 column (100 Å, 150 mm × 4.6 mm, 5 μm pore size), injecting 10 μL with an autoinjector, using an isocratic elution composed of 0.1% Formic acid: Acetonitrile (50:50) at a flowrate of 1 mL/min.

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**Standardized *Curcuma longa* extract**

Powdered *C. longa* rhizomes was extracted using ethyl acetate (95-98 % v/v) at 65 °C. The extract was subjected to chromatographic separation in a silica 60-120 mesh column using a mobile phase of chloroform: methanol. The eluted fractions with same Rf values were pooled and concentrated and recrystallised.

**Regular turmeric extract**

**Undigested sample**

***In-vitro* gastric digestion**

Mixed with simulated gastric fluid: porcine pepsin (0.04 g/mL in 0.1 M HCl)

Shaken at 95 rpm at 37 °C for 1 h, pH 2.0

***In-vitro* intestinal digestion**

Mixed with simulated intestinal fluid: 0.9 M sodium bicarbonate (0.9 M), Bile salts (0.1 g/mL) and Pancreatin (0.08 g/mL)

Shaken at 95 rpm at 37 °C for 2 h, pH 7.5

**Analysis**

- Quantitative analysis of curcuminoids by HPLC
- Anti-lipase assay
- DPPH assay
- ABTS assay
- NO assay
- Superoxide scavenging assay

**Fig. 1** Schematic representation of in vitro gastrointestinal simulation

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HPLC analysis of the samples before and after gastrointestinal digestion was performed following the in-house validated chromatographic conditions as above. Corresponding standards for BDMC and curcumin were injected to identify the compounds by retention time. The chromatograms of reference standards are provided in Additional file 2.

**Pancreatic lipase inhibition assay of the digested extracts**

The digested extracts were examined for lipase enzyme inhibition following the method of Kim et al. (2012). Briefly, the reaction mixture consisted of 6 μL of lipase solution (pH 6.8) from the enzyme stock solution of 2.5 mg/mL, 169 μL of Tris buffer (pH 7.0) and 20 μL of digested or undigested samples. The mixture was allowed to stand for 15 min at 37 °C with a subsequent addition of 5 μL *p*-nitrophenylbutyrate (*p*-NPB) substrate solution (10 mM in dimethyl formamide). The active enzyme hydrolyses *p*-NPB to nitrophenol which is measured at 405 nm using UV–visible spectrophotometer. The assay was performed in triplicate for each sample. Percentage

inhibition of enzyme activity was determined using the formula:

$$\text{Inhibition \%} = 100 - \{ B - b/A - a \times 100 \}$$

where '*A*' is the enzyme activity without sample, '*a*' is the negative control without sample, '*B*' is the activity with sample, and '*b*' is the negative control with sample.

**Determination of antioxidant activity**

The free radical scavenging ability of the extracts before and after gastrointestinal digestion was investigated using the following assays.

**DPPH free radical scavenging assay**

DPPH scavenging activity was done following the method of Soler-Rivas et al. (2000) with some modifications. Briefly, 10 μL of each extract at distinct phases of digestion were mixed with 100 μL of freshly prepared methanolic solution of DPPH (90 μM) and diluted with 190 μL of methanol in a clear 96-well microplate. Trolox was used as the standard and methanol as

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negative control. After 30 min of incubation in the dark at ambient temperature, the absorbance was measured at 515 nm in a microplate reader. The radical scavenging activity was expressed as Trolox equivalents per gram dry weight (mg/g TE dry weight). The percentage inhibition was calculated using the formula:

$$\text{DPPH scavenging activity} = (A_b - A_s / A_b) \times 100$$

where $A_b$ is the absorbance of blank; $A_s$ is the absorbance of sample.

### ABTS$^{.+}$ radical cation scavenging assay

The ABTS radical scavenging activity of the samples was determined following the method described by Re et al. (1999). The ABTS stock solution was prepared using 7 mM ABTS and 2.45 mM potassium persulfate, incubated in dark for 16 h. The resultant solution was diluted to an absorbance of 0.700 at 734 nm. 10 μL of the undigested and digested extract samples were mixed with 190 μL of ABTS reagent solution and the absorbance was determined at 734 nm. The results were expressed as percentage scavenging activity.

### Nitric oxide scavenging

Nitric oxide radical (NO) scavenging was measured with slight modification (Venkatachalam and Muthukrishnan 2012). Briefly, the reaction mixture (3.0 mL) containing sodium nitroprusside (5 mM) in phosphate-buffered saline (pH 7.3), with or without the extract at different phase of digestion, was incubated at 25 °C for 90 min. The nitric oxide radical thus generated interacted with oxygen to produce the nitrite ion, which was assayed at 30-min intervals by mixing 1.0 mL incubation mixture with an equal amount of Griess reagent. The absorbance of the chromophore (purple azo dye) formed during the diazotization of nitrite ions with sulfanilamide and subsequent coupling with NED was measured at 546 nm.

### Superoxide radical scavenging assay

This assay was based on the reduction of nitro blue tetrazolium (NBT) (Venkatachalam and Muthukrishnan 2012) in the presence of NADH and phenazine methosulfate under aerobic condition. The 3 mL reaction mixture in Tris buffer (0.02 M, pH 8.0) contained 50 μL of 1 M NBT, 150 μL of l M NADH with or without sample. The reaction was started by adding 15 μL of 1 M phenazine methosulfate to the mixture and the absorbance change was recorded at 560 nm after 2 min. Percent inhibition was calculated against a control without the extract.

### In vivo *bioavailability study*
### Animals

Twelve healthy male Wistar rats aged 10–12-week-old (250–300 g) were used for the bioavailability study. The animals were procured from Biogen Laboratory Animal Facility, Bangalore, India (Reg No. 971/PO/RcBiBt/S/06/CPCSEA). All the animals were housed in air-conditioned room under temperature (22 ± 3 °C) and humidity (30–70%) controlled environment. The rats were fed standard rodent diet and water ad libitum. The animal experimentation protocol was approved by the Institutional Animal Ethics Committee (IAEC) of Vidya Herbs Pvt Ltd., Bangalore, India (VHPL/PCL/IAEC/02/2021).

After 7-day acclimatization, the animals were fasted overnight prior to the experiment, with free access to water. Rats were randomized into two groups ($n = 6$): standardized *C. longa* extract and RTE treatment groups. The sample size was determined by power analysis (Festing and Altman 2002) using the formula,

$$\text{Sample size} = 2\,\text{SD}^2(1.96 + 0.842)^2 / d^2$$

where SD is standard deviation; $d$ is the effect size.

The extracts were formulated in a mixture of Cremaphor, Tween 80, ethanol, and water. The extracts were administered by oral gavage at a single dose of 1000 mg/kg. The animals were anesthetised in anaesthesia chamber supplied with 2% gaseous isoflurane. Blood samples were collected from tail vein at 0.25, 0.5, 1, 2, 4, 8, 12 and 24 h after oral administration of extracts. The blood samples collected in heparinized tubes were centrifuged at 3000 rpm for 10 min. The plasma samples were stored at − 20 °C for further analysis. All the animals were rehabilitated after experimentation in accordance with the guidelines laid down by CPCSEA (Committee for the Purpose of Control and Supervision of Experiments on Animals), Government of India. The animal experiment is reported in accordance with the ARRIVE checklist (Additional file 3).

### Sample preparation

The plasma samples were extracted using the method described elsewhere with slight modifications (Prasain et al. 2007). Briefly, 50 μL of plasma samples were mixed with 150 μL of methanol and centrifuged at 7000 rpm for 10 min to precipitate the proteins. 100 μL of the supernatant collected and injected 5 μL into the LCMS/MS system.

### Chromatographic conditions for quantification of BDMC and curcumin

LCMS/MS 8050 System (Shimadzu, Japan) equipped with an electrospray ionisation interface (ESI) was used to quantify BDMC and curcumin concentrations in

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plasma samples. The analysis was conducted on a Kinetex C18 column (150 mm × 2.5 mm, 2.6 μm). The mobile phase consisted of 0.2% formic acid and acetonitrile at a flow rate of 0.2 mL/min.

### Statistical analysis

The data were analyzed using GraphPad Prism 9.0. The statistical analysis was performed by ANOVA followed by post hoc test. The values were considered statistically significant at $p < 0.05$.
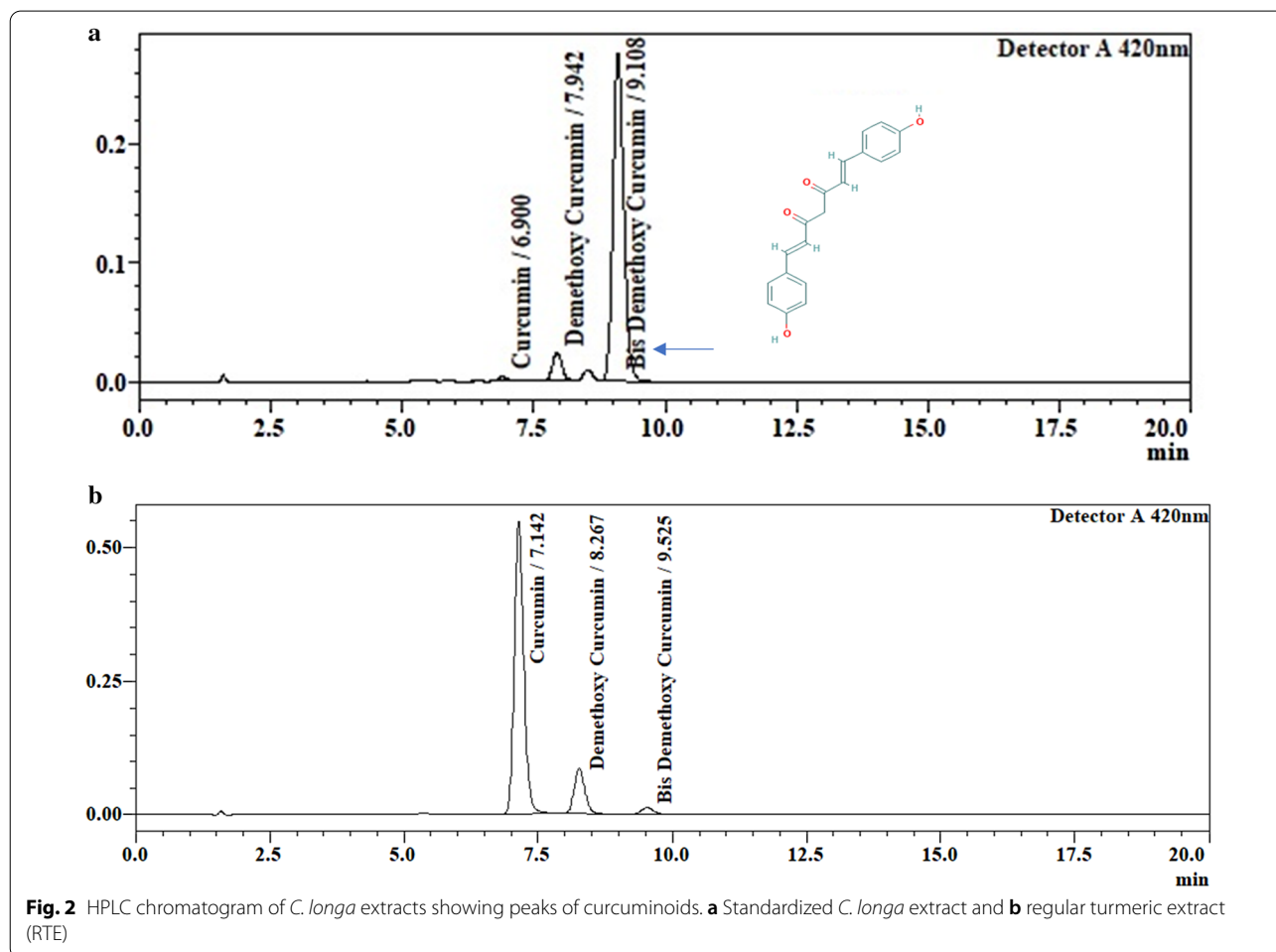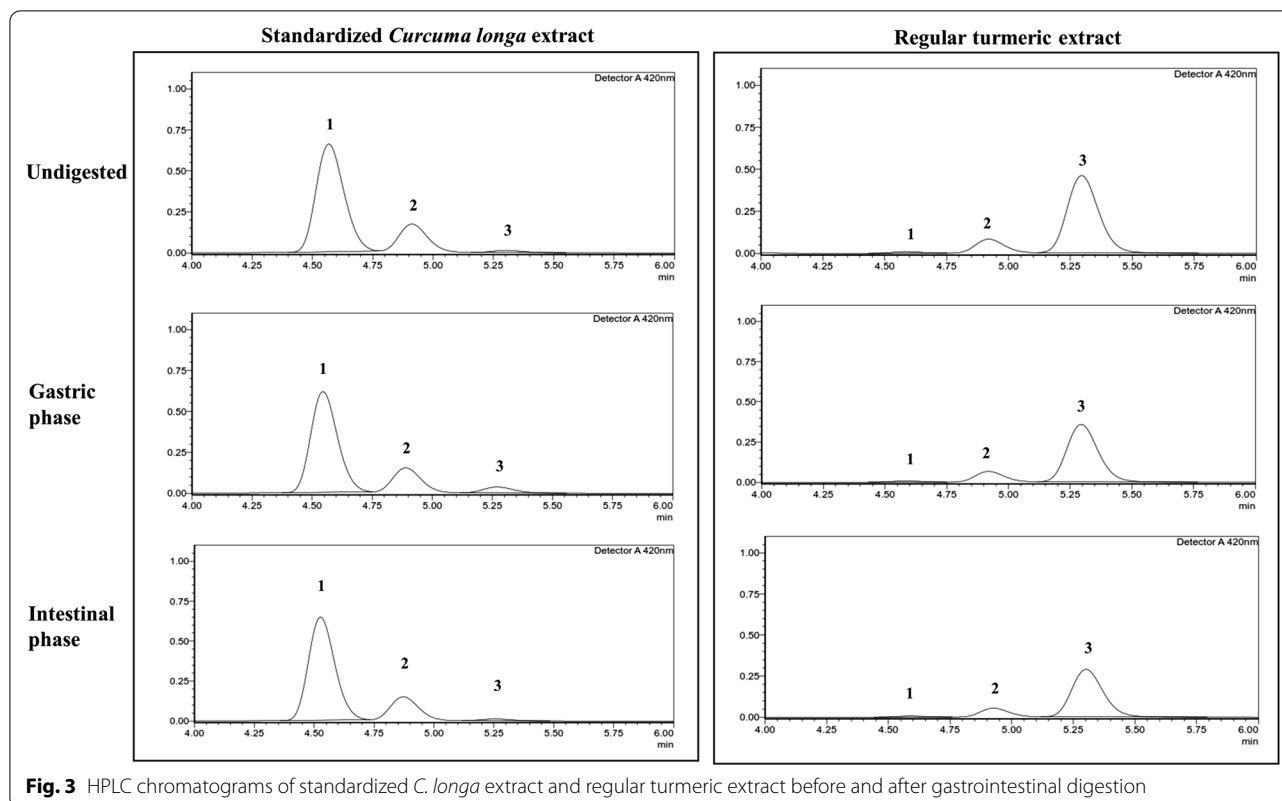
### Results

We have prepared the standardized and conventional turmeric extracts using solvent extraction and recrystallization method. The final yield of the standardized turmeric extract was 6.5% w/w while the yield of regular turmeric extract was 7.8% w/w. The chromatographic analysis using HPLC revealed that the

standardized *C. longa* extract contains 70–75% BDMC as the major curcuminoid (Fig. 2).

### Bioaccessibility after simulated digestion

In this study, we have performed the HPLC analysis to determine the stability and bioaccessibility of BDMC in standardized *C. longa* extract compared to RTE containing equivalent concentration of curcumin during in vitro gastrointestinal digestion. Figure 3 shows the comparative HPLC chromatograms of standardized *C. longa* extract and RTE samples after digestion. The bioaccessibility of BDMC (standardized *C. longa* extract) with respect to undigested sample, was considerably higher ($p < 0.05$) compared to curcumin (RTE). The concentration of BDMC in standardized *C. longa* extract samples at different digestive phases remained unchanged. However, the curcumin content in RTE was markedly reduced in the intestinal phase as compared to the undigested samples (Fig. 4).



**Fig. 2** HPLC chromatogram of *C. longa* extracts showing peaks of curcuminoids. **a** Standardized *C. longa* extract and **b** regular turmeric extract (RTE)

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**Fig. 3** HPLC chromatograms of standardized *C. longa* extract and regular turmeric extract before and after gastrointestinal digestion

***Anti-lipase and antioxidant activities of the extracts after* in vitro *digestion***

The digested turmeric extracts in simulated gastric and intestinal phases were evaluated for the inhibitory effects against pancreatic lipase, the enzyme responsible for digestion of dietary fat (Fig. 5). It was noted that the lipase inhibitory activity was markedly increased in both standardized *C. longa* extract (57.17%) and RTE (45.71) samples in the intestinal digestive phase compared to respective undigested samples. However, the lipase inhibitory effect of standardized *C. longa* extract was significantly higher than RTE ($p < 0.05$). The inhibitory activity of digested standardized *C. longa* extract and RTE samples were 1.65-fold and 2.05-fold higher than the undigested samples, respectively.
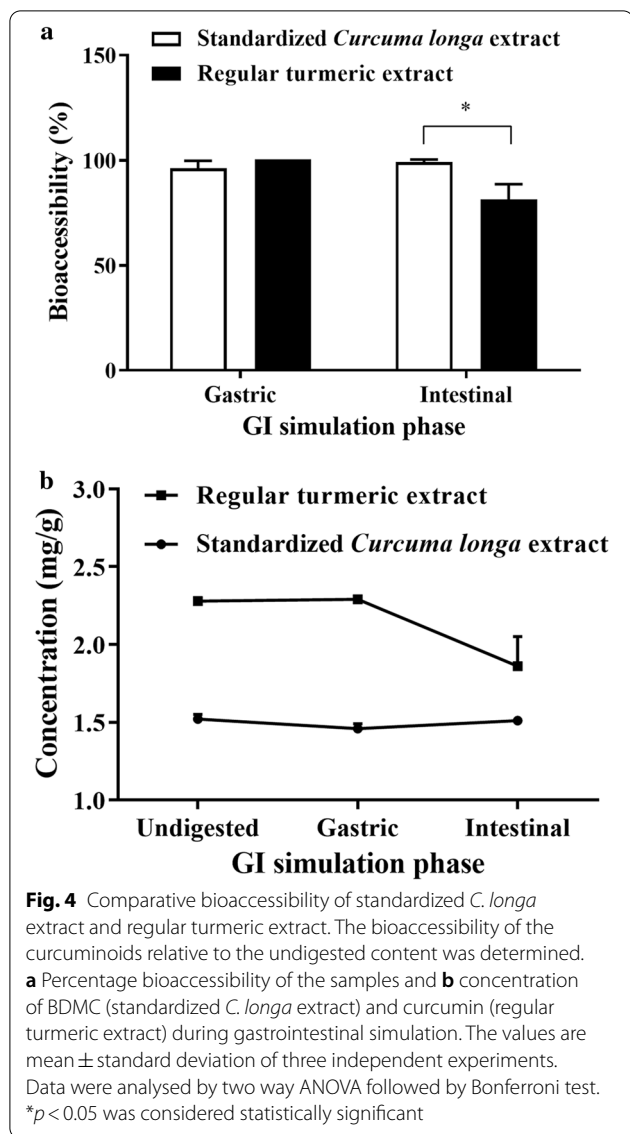
The digested samples were further investigated for free radical scavenging ability using different in vitro assays (Table 1). The DPPH radical scavenging activity of undigested standardized *C. longa* extract samples was 5.8%. The activity was markedly increased in the gastric phase (27.3%) following by a decline during intestinal digestion phase (9.3%). Similar trend was observed in RTE samples. The digested extracts exhibited considerable increase in ABTS cation scavenging activity from undigested to intestinal digestion phase.

The percentage inhibition of ABTS radical was significantly higher ($p < 0.01$) in standardized *C. longa* extract samples (67.36%) as compared to the RTE samples (61.58%) in the intestinal digestion phase.

The NO scavenging activity of the standardized *C. longa* extract was decreased from 62.2% in the undigested sample to 45.6% after intestinal digestion. The undigested RTE sample showed significantly lower NO scavenging activity (25.9%) as compared to the standardized *C. longa* extract ($p < 0.001$). However, the activity was increased to 64% in the gastric phase followed by a drastic decline (19%) during intestinal digestion. The standardized *C. longa* extract exhibited superior NO scavenging activity compared to RTE before and after digestion ($p < 0.001$). The superoxide radical scavenging activity of undigested turmeric extract samples were markedly increased in the gastric phase while the activity was reduced in the intestinal phase. After gastrointestinal digestion, the standardized *C. longa* extract showed significantly higher activity than RTE extract ($p < 0.05$).

**Oral bioavailability of BDMC and curcumin**

In the present study, blood levels of BDMC and curcumin after oral administration of standardized *C.*

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**Fig. 4** Comparative bioaccessibility of standardized *C. longa* extract and regular turmeric extract. The bioaccessibility of the curcuminoids relative to the undigested content was determined. **a** Percentage bioaccessibility of the samples and **b** concentration of BDMC (standardized *C. longa* extract) and curcumin (regular turmeric extract) during gastrointestinal simulation. The values are mean ± standard deviation of three independent experiments. Data were analysed by two way ANOVA followed by Bonferroni test. *$p < 0.05$ was considered statistically significant



**Fig. 5** Anti-lipase activity of standardized *C. longa* extract and regular turmeric extract after gastrointestinal digestion. The values are mean ± standard deviation of three independent experiments. Data were analysed by two way ANOVA followed by Bonferroni test. *$p < 0.05$; **$p < 0.01$ were considered statistically significant

*longa* extract and RTE respectively, were evaluated at different time points. The oral bioavailability of BDMC in standardized *C. longa* extract was compared with curcumin in RTE. Figure 6 shows the mean plasma concentration–time of BDMC (standardized *C. longa* extract) and curcumin (RTE). The pharmacokinetic measures including $C_{max}$, $T_{max}$ and AUC $_{0\text{-}Last}$ are provided in Fig. 6. Plasma levels of curcumin in RTE showed the $C_{max}$ at 0.5 h, rapidly decreasing thereafter. Interestingly, oral administration of standardized *C. longa* extract resulted in a sustained release of BDMC into systemic circulation over 24 h. However, BDMC showed $C_{max}$ at 0.5 h similar to curcumin. There was a 4.4-fold higher $C_{max}$ observed for BDMC
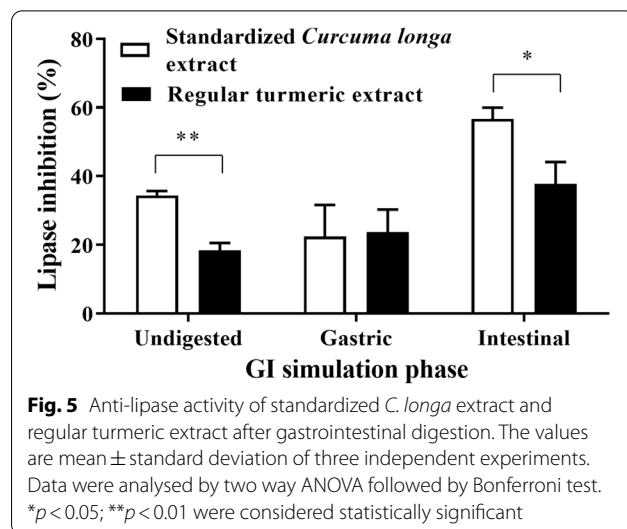
than curcumin. The AUC$_{0\text{-}Last}$ of BDMC was markedly increased in animals administered with the standardized *C. longa* extract compared to curcumin in RTE. The relative bioavailability of BDMC was 18.76 compared to curcumin.

## Discussion

One of the major setbacks towards the effective use of curcumin is its limited bioaccessibility and bioavailability. Attempts have been made to improve the solubility and stability of curcumin enabling the molecule to be more bioavailable (Sanidad et al. 2019; Mohammadian et al. 2019; Pan et al. 2018; Semalty et al. 2010; Das et al. 2010). Here we have reversed the curcuminoid composition conventionally found in regular turmeric extract to produce the BDMC enriched standardized *C. longa* extract. We have subjected the extracts to in vitro gastrointestinal digestion and compared the stability and bioaccessibility of BDMC and curcumin.

In vitro models of digestive simulation are used to quantify the compounds that remain bioaccessible (Versantvoort et al. 2005; Minekus 2014). In this study, we found that BDMC was more stable during the gastrointestinal digestion while the concentration of curcumin declined significantly in the intestinal digestive phase. Bioaccessibility is greatly influenced by the solubility of the molecule (Fu et al. 2016). Curcumin is sparingly soluble in water under acidic and neutral conditions while decompose at alkaline pH (Kharat et al. 2017). On the contrary, BDMC is more stable molecule due to the lack of methoxy groups present otherwise in curcumin (Gordon et al. 2015). In agreement to this, we found that BDMC in the standardized *C. longa* extract was more bioaccessible than curcumin in RTE.

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**Table 1** Antioxidant activities of the turmeric extracts before and after gastrointestinal digestion

| Digestive phase | Standardized *C. longa* extract | Regular turmeric extract | *p* value[#] |
|---|---|---|---|
| *DPPH radical scavenging (%)* | | | |
| Undigested | $5.77 \pm 1.21$ | $10.65 \pm 0.71$ | 0.004** |
| Gastric | $27.33 \pm 0.46$ | $28.43 \pm 1.55$ | 0.304 |
| Intestinal | $9.31 \pm 1.07$ | $11.96 \pm 1.91$ | 0.104 |
| **Digestive phase** | **Standardized *C. longa* extract** | **Regular turmeric extract** | ***p* value*** |
| *ABTS cation scavenging (%)* | | | |
| Undigested | $25.71 \pm 1.37$ | $23.69 \pm 2.47$ | 0.283 |
| Gastric | $15.29 \pm 1.87$ | $14.43 \pm 0.60$ | 0.492 |
| Intestinal | $67.36 \pm 2.20$ | $61.58 \pm 1.36$ | 0.018* |
| *Nitric oxide scavenging (%)* | | | |
| Undigested | $62.20 \pm 5.98$ | $25.89 \pm 3.4$ | < 0.001*** |
| Gastric | $49.11 \pm 3.82$ | $64.01 \pm 4.38$ | 0.011* |
| Intestinal | $45.55 \pm 3.86$ | $18.86 \pm 5.24$ | 0.002** |
| *Superoxide radical scavenging (%)* | | | |
| Undigested | $36.98 \pm 0.83$ | $24.35 \pm 0.62$ | < 0.001*** |
| Gastric | $86.47 \pm 2.14$ | $87.28 \pm 0.91$ | 0.576 |
| Intestinal | $72.58 \pm 0.85$ | $68.94 \pm 2.17$ | 0.054 |

The values are presented as mean $\pm$ standard deviation of three independent experiments. [#]Independent *t* test. *$p < 0.05$; **$p < 0.01$; ***$p < 0.001$

| | $C_{max}$ (ng/mL) | $T_{max}$ (h) | $AUC_{0\text{-Last}}$ |
|---|---|---|---|
| **BDMC (Standardized *Curcuma longa* extract)** | $5.91 \pm 0.43$ | 0.5 | $91.02 \pm 0.28$ |
| **Curcumin (Regular turmeric extract)** | $1.34 \pm 0.44$ | 0.5 | $4.85 \pm 0.77$ |
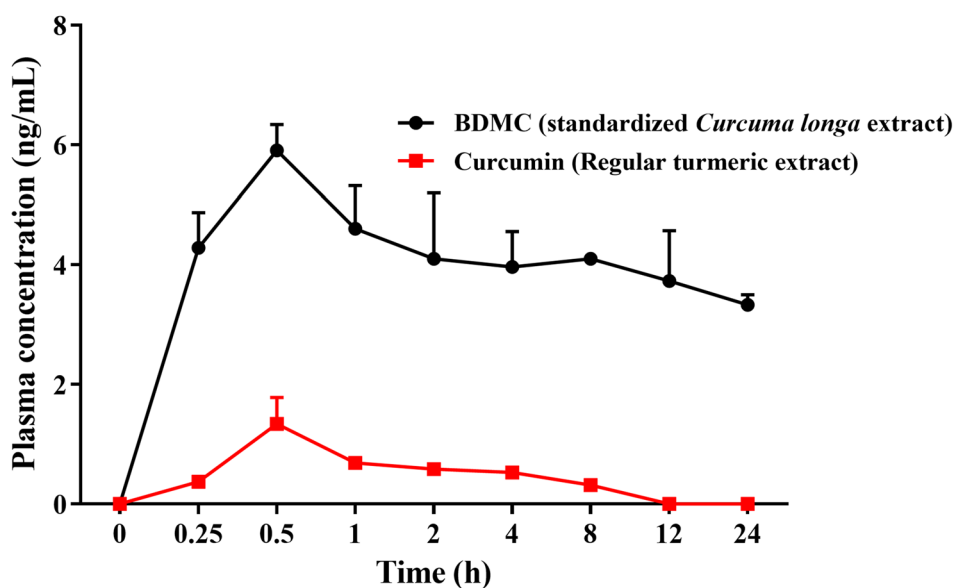


**Fig. 6** Oral bioavailability of BDMC and curcumin in rats

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BDMC is reported to be more biologically active among the curcuminoids (Cashman et al. 2008; Sandur et al. 2007). Here, we have performed several assays to confirm the radical scavenging ability of the extracts. Previously Sivabalan and Anuradha (2010) demonstrated that BDMC exerts profound antioxidant activity compared to curcumin. Similarly, in this study the standardized *C. longa* extract with higher BDMC content showed significant antioxidant activity at different digestive phases as compared to RTE. In addition, lipase inhibitory effect of standardized *C. longa* extract was significantly higher than RTE in the intestinal phase. Improved bioaccessibility of BDMC is suggestive of higher bioavailability compared to curcumin. In line with the results of in vitro study, BDMC was found have higher bioavailability relative to curcumin when administered to rats. The data on bioavailability of BDMC in rats could form the basis for further investigations in human subjects.

The present study has several limitations. Here we have evaluated the bioavailability of BDMC relative to that of curcumin in regular turmeric extract. The absolute bioavailability along with tissue distribution, metabolism and excretion will essentially add on to the potential of BDMC.

## Conclusion

The colon bioaccessibility and bioavailability of BDMC in a standardized *C. longa* extract compared with curcumin in the regular turmeric extract is reported. Findings from this study suggest the use of turmeric extract with higher content of BDMC over regular turmeric extract for improved health benefits. However, these claims must be validated further through human clinical studies.

### Abbreviations

ABTS: 2, 2'-Azino-bis (3-ethylbenzothiazoline-6-sulfonic acid); ANOVA: Analysis of variance; AUC: Area under the curve; BDMC: Bisdemethoxycurcumin; $C_{max}$: Peak plasma concentration; DPPH: 2,2 Diphenyl-1-picrylhydrazyl; ESI: Electrospray ionisation interface; HPLC: High performance liquid chromatography; IAEC: Institutional Animal Ethics Committee; NBT: Nitro blue tetrazolium; NO: Nitric oxide; *p*-NPB: *p*-Nitrophenylbutyrate; RTE: Regular turmeric extract; TLC: Thin layer chromatography; $T_{max}$: Time to achieve maximum plasma concentration.

## Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s42269-021-00544-8.

**Additional file 1.** Solvent extraction method of extraction of curcuminoids from turmeric rhizomes.

**Additional file 2.** Analytical method and chromatographic conditions for the HPLC analysis of reference standards along with the chromatograms.

**Additional file 3.** Checklist from the author in accordance with ARRIVE guideline.

### Availability of data and materials
The data sets used and/or analyzed during the current study available from the corresponding author on reasonable request.

### Declarations

### Ethics approval and consent to participate
The protocols involving the experimental laboratory animals were approved by the Institutional Animal Ethics Committee (IAEC) of Vidya Herbs Pvt Ltd., Bangalore, India (VHPL/PCL/IAEC/02/2021).

### Consent for publication
Not applicable.

### Competing interests
The authors declare no competing interests.

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