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## Development of bioanalytical parameters for the standardization of *Zingiber officinale*

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### ABSTRACT

**Objective:** To develop novel bioanalytical parameters for standardization of *Zingiber officinale* (*Z. officinale*) extract. **Methods:** Phytochemical analysis, solubility test, heavy metal analysis, antimicrobial study and fingerprint analysis through HPTLC method were carried out in the present investigation. Moreover quantitative analysis of rutin and quercetin in *Z. officinale* extract through HPTLC techniques were also performed in the present investigation. **Results:** Phytochemical analysis showed the presence of alkaloid, carbohydrate, tannin, steroid, triterpenoid, glycoside, triterpenoid, saponin, flavonoid, amino acid and protein. The total phenol and flavonoid content were found to be 0.59% and 4.50%. For quantitative analysis through HPTLC techniques, optimization of solvent system was done and the content of quercetin and rutin in the extract was found to be 3.22% (w/w) and 0.65% (w/w) respectively. **Conclusion:** These above mentioned parameters can be used as an important tool for the standardization of *Z. officinale* extract.

## 1. Introduction

*Zingiber officinale* (*Z. officinale*) Roscoe (family, Zingiberaceae), commonly known as ginger, is consumed worldwide as spice and flavoring agent. It is commercially cultivated in India, China, South East Asia, West Indies, Mexico, Africa, Fiji and Australia<sup>[1]</sup>. Biochemically, the main active components of *Z. officinale* is gingerol and shogaol, while zingiberene is obtained from ginger oil<sup>[2]</sup>.

In Ayurveda, *Z. officinale* is considered as valuable medicine due to rubefacient, antiasthmatic and stimulant to the gastrointestinal tract. It possesses antioxidant, hypoglycaemic, hypolipidaemic effects<sup>[1]</sup>. *Z. officinale* has a long history of use in ailments such as nausea, respiratory disorders, cardiovascular and rheumatic disorders. *Z. officinale* also has immunomodulatory properties and

is reported to inhibit various inflammatory mediators such as prostaglandins and proinflammatory cytokines<sup>[3]</sup>. Ginger extract has antibacterial, anticonvulsion, analgesic, antiulcer, antitumour, antifungal, anti-allergen, antiplatelet, antioxidant, antirhinoviral, antihepatotoxicity and antiarthritic activity<sup>[2,4]</sup>.

## 2. Material and methods

### 2.1. Development of bioanalytical parameters

Crude plant extract of *Z. officinale* was procured from Garlico Herbal Concentrate (M.P.), India. Phytochemical screening was conducted on the *Z. officinale* extract to confirm the presence of different phytoconstituents<sup>[5]</sup>. The presence of phytoconstituents in the extract was also analysed through thin layer chromatography (TLC) analysis<sup>[6]</sup>.

Parameters such as solubility in water, pH, moisture content, heavy metal analysis and microbiological analysis were also performed in the present investigation as per

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standard official methods[7,8]. Total phenol and flavonoid content were also determined according to the standard methods[9,10]. The quantification of rutin and quercetin in *Z. officinale* were determined through high performance thin layer chromatography (HPTLC) techniques (Table 1).

**Table 1.**  
Bioanalytical parameters of analysis.

Analysis	Estimation of rutin and quercetin in <i>Z. officinale</i> extract
Plate material	HPTLC Precoated plates Silica Gel Merck 60F <sub>254</sub>
Syringe	100 $\mu$ L Hamilton (Bonadzu, Switzerland)
Application mode	CAMAG Automatic TLC Sampler III
Development mode	Ascending
Scanning	CAMAG TLC scanner 3 with Cats software
Experimental conditions	Temperature (25 $\pm$ 2) $^{\circ}$ C, relative humidity 40%

### 3. Results

Phytochemical analysis showed the presence of alkaloid, carbohydrate, tannin, steroid, triterpenoid, glycoside, triterpenoid, saponin, flavonoid, amino acid and protein. TLC analysis showed two prominent spots with  $R_f$  0.20 and 0.90 value in *n*-hexane: diethyl amine (40:60) solvent system. However in the fingerprint analysis through HPTLC showed seven prominent spots in ethyl acetate: formic acid: glacial acetic acid: H<sub>2</sub>O (100:11:11:26) solvent system (Table 2). Moisture content and solubility in alcohol was found to be 6.94% and 32.10%. The total phenol and flavonoid content were found to be 0.59% and 4.50%. Further heavy metal level (lead, arsenic, mercury, cadmium) were found to be under the limit. Microbiological assay showed that *E. coli* and *Salmonella* was found to be absent whereas total bacterial count and yeast & moulds contents were found to be below the limit. For quantitative analysis through HPTLC techniques, optimization of solvent system was done and ethyl acetate: formic acid: glacial acetic acid: water (100:11:11:26) was found to be most suitable solvent system. The content of quercetin and rutin in *Z. officinale* was found to be 3.22% (w/w) and 0.65% (w/w) respectively.

**Table 2.**  
HPTLC fingerprint analysis of *Z. officinale* extract.

No of spot	Solvent system	$R_f$ value	Maximum peak height	Peak area (%)
7	Ethyl acetate:	0.07	140.6	10.87
	formic acid:	0.15	82.6	5.24
	glacial acetic	0.24	69.2	6.15
	a c i d : H <sub>2</sub> O	0.35	22.2	1.33
	(100:11:11:26)	0.42	341.4	36.52
		0.59	42.9	4.49
		0.96	538.8	35.41

### 4. Discussion

Physicochemical and phytochemical analysis was generally performed to ensure the identity, purity and quality of the drug. These parameters are also used to check the genuine nature of the crude drug, thus it plays an important role in preventing the possible steps of adulteration. Phytoconstituents obtained from natural sources have been gaining importance in the day by day due to the health promoting activity. So it is necessary to check the quality safety and efficacy of herbal drugs before its consumption. In the last few decades, an HPTLC technique has gained much popularity for standardization of the herbal drugs and formulations due to analysis of several samples simultaneously with small quantity of marker compound and mobile phase with very less time[11].

TLC and HPTLC techniques can be used to solve many qualitative and quantitative analytical problems in a wide range of fields, including medicine, pharmaceuticals, chemistry, biochemistry, food analysis, toxicology and environmental analysis[12,13]. Quality evaluation and standardization of the herbal preparation is a fundamental requirement of industry and other organization dealing with ayurvedic and herbal products. According to WHO guidelines, an herbal product needs to be standardized with respect to safety before releasing it into the market[14]. Heavy metals are important environmental pollutants and many of them are toxic even at very low concentrations. Heavy metal accumulations in edible plants can directly affect plant growth and an excess dietary intake of contaminated plants could also be dangerous for the health of humans and animals[15].

These above mentioned bioanalytical parameters can be utilized for the simultaneous analysis of different phytoconstituents present in the *Z. officinale* plant material. In future, this information may be useful as a standard to identify the adulterants and other related species.

### Conflict of interest

The authors declare they have no conflict of interests.

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