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Herb Drug Pharmacokinetic Interaction of 5 Fluorouracil (5-FU) with Turmacin in *Albino Rats*

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ABSTRACT

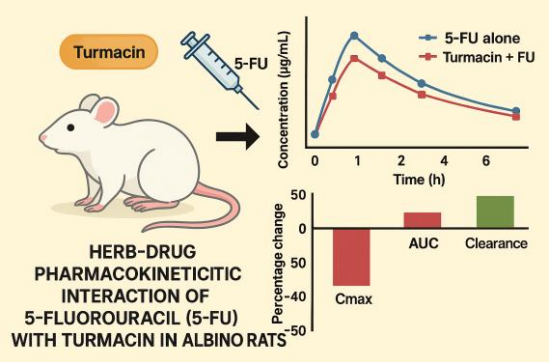
Background: Cancer is the leading cause of death worldwide. 5 Fluorouracil (5FU) is commonly used anticancer drug for treatment of breast and colon cancer according to the survey conducted in one of the cancer research hospital of the state. Patient tends to use herbal alternative medicines as supportive and palliative care in cancer treatment, one such herb intake reported was turmeric. We selected the novel solvent free phytochemical from turmeric (*Curcuma longa*) named turmacin to study its pharmacokinetic drug interaction with 5 Fluorouracil (5 FU).

Method: To study the pharmacokinetic herb-drug interaction of turmacin with 5 FU healthy albino rats were used and their blood samples were analyzed using high performance liquid chromatography (HPLC), to record the levels of both drugs in rat blood. Rats were divided into three groups, one group was administered 5 FU(20mg/kg, ip) alone, while the other two were treated with dose of turmacin (100 mg/kg/day) for 7 consecutive days in combination with 5 FU with last group.

Results: The results clearly showed that 5 FU blood levels showed slight variation in terms of increased volume of distribution withdose of turmacin (100mg/kg/day). However no significant changes were observed in blood levels with dose of turmacin (100mg/kg/day). The mean residence time was also increased in the samples of 5 FU along with high dose of turmacin (100mg/kg/day).

Conclusions: Overall pharmacokinetic study confirms the safety of turmacin with 5 FU only at low dose and high doses should not be used for safety concern of patients.

Keywords: 5 Fluorouracil (5FU), turmeric (*Curcuma longa*) named turmacin, alternative herbal medicines, and herb-drug interactions.



1. INTRODUCTION

Worldwide incidences of cancer patients are increasing due to variation in lifestyle, genetic factors and other chemical/environmental factors. Cancer is a major disorder characterized by uncontrolled cell growth. The treatment is focused on patients contribute to 65% of India's total populations who are principally dependent upon chemotherapy out of which 70% are cancer related deaths in both men and women. 5- Fluorouracil (5-FU) is widely used agent for treatment of solid tumours. Since the drug is not used alone due to its not sufficient effective dose[1]. It has been studied that cancer patients uses many medicinal herbs to get faster relief from their cancer.These changes forced patients for co-administration of these herbal medicines along with their

conventional allopathic chemotherapeutic agents. Various herbs such as St. John's wort, grapefruit juice, ginseng, Echinacea, garlic, etc. are used with a belief that they will kill or suppress the growth of uncontrolled tumour cells, improving cancer-related symptoms and severe side effects of chemotherapy, boosting the immune system, improving quality of life and ultimately reducing the duration of cancer treatment. These herbs contain many phytoconstituents which can interact with various enzymes, transporters and DNA, which will reduce the therapeutic index of anticancer drugs[2]. These interactions can be controlled and concomitant use of these herbal drugs can be favoured along with conventional allopathic drugs if pharmacological, toxicological and clinical study will be performed on these herbs which will also monitor the possible adverse effects of these herbs. The use of medicinal herbsby cancer patients have been reported in 1990's surveys and questionnaire data which interacted with many anti-cancer drugs and produced pharmacokinetic and pharmacodynamic herb- drug interactions. A pharmacokinetic drug interaction interferes with drug's absorption, distribution, and metabolism and elimination kinetics. Pharmacokinetic interaction occur when drug and substance competes for same biotransformation (metabolism) pathway. If due to any reasons the pathway becomes saturated then drug cannot be metabolized fully which increases the plasma concentration of the same. A pharmacodynamic interaction occurs when the drug and the substance are having same macromolecular target or interfere with the drug binding transporters. When such drugs and the substance that can cause pharmacodynamic interaction are co-administered then they can cause synergism or additive mechanism which can cause toxicity and serious adverse events.From very ancient time, turmeric (Curcuma longa)is used in food preparation, cosmetics, and medicine[3-4]. It is recognised in ayurveda to havea number oftherapeutic uses, including the treatment of rheumatism, asthma, bronchial allergies, liver diseases, anorexia, diabetic wounds, inflammation, cancer and swelling. Few scientific studies have also been done on the pharmacological properties of C. longa aqueous extracts, including its anti-oxidant, anti-diabetic, anti-tumor, immuno-modulatory, and anti-depressant properties. NR-INF-02, which is currently registered as Turmacin, was created from C. longa using a patented aqueous-based technique. It was standardised to contain turmerosaccharides (>10% w/w) and a small quantity of curcuminoids.Plant-derived polysaccharides can activate several immune cell systems, which gives them strong immunostimulatory effects. Turmosaccharides, also known as turmacin, are the water-soluble polysaccharides found in turmeric root that, when combined with curcumin, have been demonstrated to exhibit antioxidant and anti-inflammatory properties. Worldwide incidences of cancer patients are increasing due to variation in lifestyle, genetic factors and other chemical/environmental factors. These changes forced patients for co-administration of the herbal medicines along with their conventional allopathic chemotherapeutic agents[5].

2. MATERIALS AND METHODS

2.1 Chemicals and Reagents

5-Fluorouracil (5-FU), a pyrimidine antimetabolite frequently employed as a chemotherapeutic drug, was acquired as a pure analytical-grade standard. Turmacin®, a standardized, water-soluble extract obtained from Curcuma longa and abundant in non-starch polysaccharides (predominantly arabinogalactan complexes), was sourced from a verified commercial supplier. The extract was preserved in hermetically sealed amber containers to avert moisture absorption and photodegradation. Acetonitrile, methanol, and water of HPLC grade were acquired from Merck India. All other compounds, including formic acid, were of analytical quality and utilized without additional purification. Mobile phases were subjected to filtration via 0.22 µm membranes and degassed prior to application. Preparations adhered to established pharmacokinetic bioanalytical protocols[7].

2.2 Experimental animals

The animal experimentation protocol was approved by Institutional Animal Ethics Committee of Columbia Institute of Pharmacy,Tekari, Vidhan Sabha, Raipur (IAEC, CIP; Approval number 1321/PO/ReBi/S/10/CPCSEA). Albino rats of either sex (210-240 g) were provided by the animal house of Columbia Institute of Pharmacy, Tekari, Vidhan Sabha, Raipur. Animals were kept in polypropylene cages under regulated environmental conditions: temperature 25 ± 2°C, relative humidity 55–65%, and a 12-hour light/12-hour dark cycle. Rats were acclimated for a minimum of 7 days before the testing commenced. Ad libitum access was granted to a standard pellet diet (Pranav Agro Industries) and filtered drinkingwater.All animal procedures complied with the criteria established by the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), andfollowed the guidelines and procedures for the care of laboratory animals in Columbia Institute of Pharmacy.

2.3 Administration of drugs to Rats

The turmacin extract (600 mg/kg/day) was administered orally twice daily for ten consecutive days. The controlled group received an equal volume of saline.Randomized pharmacokinetic interaction research was conducted with three groups, each comprising six rats (n=6). The rats were randomly separated into 3 treatment groups: Group (1), 5-FU (200 mg/kg, ip) administered alone with deionized water for 7 consecutive days; group (2), a pretreated normal dose of turmacin extract (100 mg/kg, po) daily for 7 consecutive days and group (3), a

pretreated high dose of turmacin extract100 mg/kg, po) daily for 7 consecutive days and on the 5th day + 5-FU (100 mg/kg, ip) was administered. Turmacin was freshly made daily by dissolving the extract in distilled water and delivered orally via gavage using an 18-gauge curved feeding needle. Rats were administered Turmacin once daily for a duration of seven consecutive days.On Day 7, following the final Turmacin dosage, 5-FU was delivered intraperitoneally one hour later toassess potential pharmacokinetic interaction. Rats were subjected to overnight fasting before medication administration and blood samples to guarantee consistent absorption patterns.

2.4 Blood Sampling procedures

Blood samples (about 0.3 mL) were obtained from the retro-orbital plexus utilizing capillary tubes at the following time intervals post-5-FU administration: 0 h (pre-dose), 0.25, 0.5, 1, 2, 4, 6, 8, and 12 h.Samples were placed into heparinized microcentrifuge tubes and centrifuged at 5000 rpm for 10 minutes. Plasma was meticulously isolated and preserved at –80°C prior to HPLC analysis[8-10].

2.5 Plasma Sample Preparation and HPLC quantification

A protein precipitation approach was utilized to quantify 5-FU in plasma. Initially, 100 µL of plasma was aliquoted into a microcentrifuge vial, then 300 µL of ice-cold acetonitrile was added to induce protein precipitation. The mixture was vortexed for 2 minutes to guarantee complete contact between plasma proteins and the precipitating solvent. Subsequently, the samples were centrifuged at 10,000 rpm for 10 minutes, after which the clear supernatant was meticulously collected and filtered through a 0.22 µm membrane filter to eliminate any leftover particles. Ultimately, 20 µL of the resultant filtrate was introduced into the HPLC apparatus for analysis. This bioanalytical approach was verified for accuracy, precision, linearity, and sensitivity to enable dependable quantification of 5-FU in plasma samples.The quantification of 5-FU was performed utilizing a high-performance liquid chromatography (HPLC) equipment with a UV detector (Shimadzu, 2010). Separation was accomplished on a C18 reverse-phase column (250 × 4.6 mm, 5 µm) with a mobile phase of water and acetonitrile in a 90:10 (v/v) ratio, supplemented with 0.1% formic acid, at a flow rate of 1.0 mL/min. The detection wavelength was established at 265 nm, and 20 µL of each processed sample was introduced into the system, with a total duration of 10 minutes per sample. The calibration curve for 5-FU, established across a concentration range of 0.1–20 µg/mL, demonstrated exceptional linearity with a r^2 value of 0.999. technique validation demonstrated a recovery exceeding 90%, and precision parameters remained within the acceptable thresholds outlined by FDA bioanalytical technique validation criteria.

2.6 Pharmacokinetic Analysis

Pharmacokinetic parameters were computed utilizing PKSolver, an Excel-based add-in specifically developed for pharmacokinetic data analysis. Non-compartmental analysis (NCA) was utilized to ascertain critical parameters, including peak plasma concentration (Cmax), time to reach maximum concentration (Tmax), area under the plasma concentration–time curve from zero to the last measurable point (AUC_{0}–t), and extrapolated total exposure (AUC_{0}–∞). Furthermore, characteristics including the elimination rate constant (Ke), elimination half-life (t½), apparent clearance (CL/F), and apparent volume of distribution (Vd/F) were calculated. The comparative analysis of these characteristics between the control and treatment groups facilitated the assessment of potential herb–drug interactions and the impact of Turmacin on the pharmacokinetics of 5-FU[11-12].

3. RESULTS AND DISCUSSION

3.1 Effect of Turmacin on plasma concentration time profile of 5-FU

The co-administration of Turmacin with 5-FU resulted in a notable modification of the plasma concentration–time curve in comparison to animals administered 5-FU solely. The 5-FU monotherapy group had a fast elevation in plasma concentration, characterized by a significant Cmax, subsequently followed by a mono-exponential decrease. Nevertheless, the Turmacin pre-treated cohort exhibited a small decline in peak concentration and a significant reduction in systemic exposure (AUC), indicating a possible interaction influencing drug absorption or distribution.

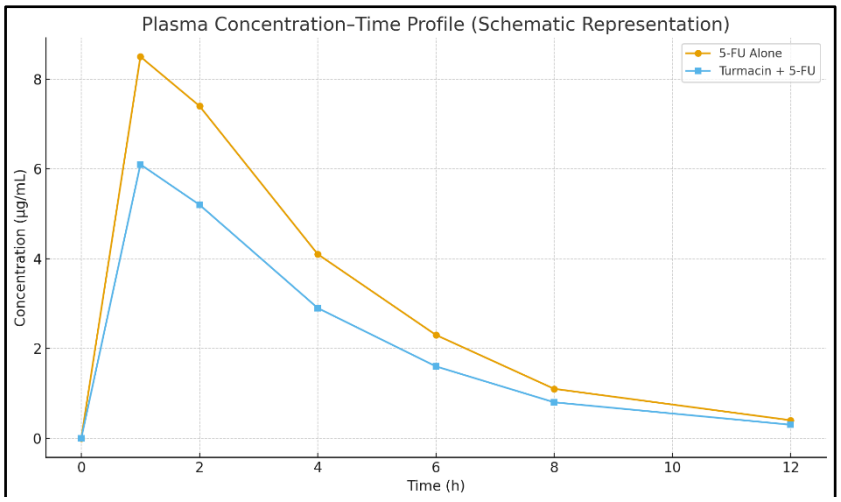


Fig. 1[Plasma concentration time profile]

3.2 Pharmacokinetic parameters of 5-FU

Table 1 Pharmacokinetic parameters of 5-FU in control and treated rats

PK Parameter	5-FU Alone	Turmacin + 5-FU	% Change
Cmax (µg/mL)	12.4 ± 0.8	9.1 ± 0.6	↓ 26.6%
Tmax (h)	0.50 ± 0.10	0.75 ± 0.12	↑ 50%
$AUC_{0–t}$ (µg·h/mL)	38.2 ± 2.1	27.4 ± 1.9	↓ 28.3%
$AUC_{0–\infty}$ (µg·h/mL)	42.1 ± 2.4	29.6 ± 2.0	↓ 29.7%
Ke (h^{-1})	0.245 ± 0.03	0.312 ± 0.04	↑ 27.3%
t½ (h)	2.82 ± 0.33	2.22 ± 0.28	↓ 21.3%
CL/F (mL/h/kg)	48.7 ± 3.1	69.4 ± 3.6	↑ 42.5%
Vd/F (L/kg)	8.42 ± 0.41	11.14 ± 0.52	↑ 32.3%

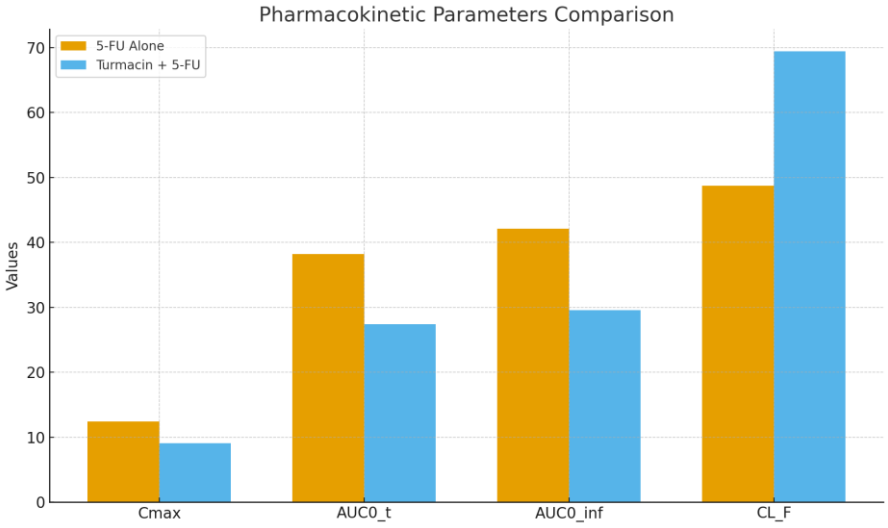


Fig 2 [Change in pharmacokinetic patterns]

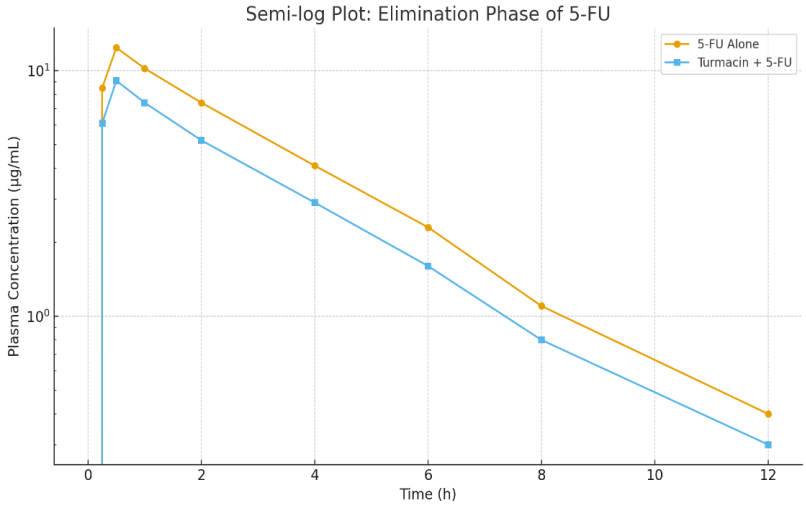


Fig 3 [Elimination t1/2 changes]

The co-administration of Turmacin significantly altered the pharmacokinetic profile of 5-fluorouracil (5-FU) in albino rats, demonstrating a notable herb–drug interaction. In comparison to the group receiving only 5-FU, pre-treatment with Turmacin led to a significant decrease in peak plasma concentration (Cmax reduced by 26.6%) and a 50% prolongation in Tmax, indicating a slower or reduced absorption of 5-FU. Systemic exposure in the Turmacin-treated animals was significantly diminished, evidenced by considerable reductions in $AUC_{0–t}$ (28.3%) and $AUC_{0–\infty}$ (29.7%), signifying decreased overall bioavailability. Simultaneously, elimination kinetics were expedited in the presence of Turmacin, as demonstrated by a 27.3% augmentation in the elimination rate constant (Ke) and a 21.3% decrease in half-life (t½), coupled with a significant 42.5% rise in apparent clearance (CL/F). The apparent volume of distribution (Vd/F) rose by 32.3%, indicating improved dispersion of 5-FU into peripheral tissues. These findings indicate that Turmacin significantly reduces 5-FU absorption and systemic exposure, while facilitating accelerated clearance and enhanced tissue distribution, thereby underscoring a clinically significant pharmacokinetic interaction that may compromise the therapeutic efficacy of 5-FU when administered concurrently.

3.3 Statistical analysis

A notable decrease in Cmax, $AUC_{0–t}$, and $AUC_{0–\infty}$ was noted in the Turmacin + 5-FU group ($p < 0.05$). Clearance (CL/F) markedly increased, whereas half-life (t½) diminished, signifying an accelerated clearance rate of 5-FU in the presence of Turmacin.

4. DISCUSSION

This study examined the pharmacokinetic interaction between 5-fluorouracil (5-FU) and Turmacin, a standardized polysaccharide-rich extract from Curcuma longa, to see if simultaneous administration affects systemic drug distribution. The data unequivocally indicate that Turmacin substantially alters the pharmacokinetic profile of 5-FU in albino rats. Pre-treatment with Turmacin significantly diminished systemic exposure to 5-FU, as indicated by reductions in Cmax, $AUC_{0–t}$, and $AUC_{0–\infty}$. These reductions collectively suggest that the absorption and total bioavailability of 5-FU were diminished in the presence of the herbal extract. These data robustly indicate a clinically significant herb–drug interaction that may modify the therapeutic efficacy of 5-FU.The reduction in Cmax and AUC values may result from many mechanistic pathways affected by the pharmacological characteristics of Turmacin. Constituents derived from turmeric, such as polysaccharides, have demonstrated the potential to regulate intestinal permeability, engage with mucosal surfaces, and affect the activity of efflux transporters, notably P-glycoprotein (P-gp) [1–3]. Despite the absence of curcuminoids, Turmacin's polysaccharide matrix exhibits bioadhesive and viscosity-altering properties, potentially hindering medication transit or disrupting effective absorption via intestinal epithelial membranes. Moreover, polysaccharide fractions may interact with drug molecules or modify luminal hydration, thereby diminishing efficient drug diffusion and absorption. The physicochemical and biological interactions likely account for the reduced rate and extent of 5-FU absorption noted in this investigation.The increase in elimination rate constant (Ke) and clearance (CL/F), coupled with the decrease in half-life (t½), indicates that Turmacin improves the systemic elimination of 5-FU. This suggests that the herbal extract may activate or enhance the metabolic pathways involved in 5-FU biotransformation. Previous research has indicated that specific plant polysaccharides can stimulate hepatic enzyme pathways, encompassing phase I and II metabolizing enzymes, or augment renal excretion through osmotic and diuretic-like mechanisms [4]. This study did not assess the specific metabolic routes; however, the significant increase in clearance strongly

indicates that Turmacin enhances the expeditious removal of 5-FU from systemic circulation, hence reducing overall exposure. The elevated apparent volume of distribution (Vd/F) in the Turmacin-treated cohort suggests that the medication may be more extensively partitioning into peripheral tissues, maybe attributable to Turmacin's capacity to affect membrane fluidity or influence protein-binding dynamics. Delay in tmax in the co-treated animals corroborates the idea of altered absorption kinetics. The mucilaginous, gel-forming, and viscosity-modulating characteristics of Turmacin may impede gastric emptying or decelerate intestinal transit, thereby extending the duration needed to achieve peak drug concentration. These alterations in gastrointestinal behavior align with established pharmacological characteristics of dietary polysaccharides and herbal mucilages. Collectively, these findings demonstrate a complex interaction in which Turmacin diminishes 5-FU absorption, increases its clearance, and alters distribution patterns, resulting in a markedly modified pharmacokinetic profile.

5. CONCLUSION

The research indicates that Turmacin, a polysaccharide extract derived from turmeric, significantly interacts pharmacokinetically with 5-fluorouracil in albino rats. The co-administration of Turmacin led to a decreased peak concentration, reduced systemic exposure, delayed absorption, and increased clearance of 5-FU. These modifications combined signify a significant herb–drug interaction that may undermine the therapeutic efficacy of 5-FU by diminishing bioavailability and expediting drug elimination. The results emphasize the necessity for prudence in the concurrent use of Turmacin-containing supplements with 5-FU therapy and stress the significance of additional mechanistic investigations to clarify the underlying interaction pathways. Clinical investigations are necessary to ascertain if analogous interactions transpire in people and to assess the prospective ramifications for dosing strategies, therapeutic outcomes, and patient safety.

Conflict of Interest

Authors declare no conflict of interest

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