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CLINICAL & BASIC RESEARCH

Effect of *Khat* (*Catha edulis*) Use on the Bioavailability, Plasma Levels and Antimalarial Activity of Chloroquine

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تأثير استخدام القات (كاثا أدليس) على التوافر الحيوي، مستويات البلازما ونشاط الكلوروكين المضاد للملاريا

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abstract: ***Objectives:*** This study aimed to evaluate the effect of *khat* (*Catha edulis*) on chloroquine (CQ) bioavailability in healthy Yemeni adults and its effect on CQ plasma levels and parasite clearance among malaria patients. ***Methods:*** This study took place between January and April 2007 in Bajil and Sana'a, Yemen. Two CQ doses (600 mg each) were given to 15 healthy males on separate occasions; the first dose was followed by a *khat*-chewing session (phase one) while controls abstained from *khat*-chewing for the second (phase two). Additionally, 103 patients with *Plasmodium falciparum*-induced malaria, including both regular *khat* chewers (n = 57) and non-*khat* chewers (n = 46), were treated with CQ (25 mg/kg) over three days. Pharmacokinetic parameters were analysed among both controls and malaria patients. Parasite clearance was also investigated for the latter group. ***Results:*** The mean area under the concentration-time curve (AUC) was 2,108.9 versus 2,797.4 ng/hour/mL, mean peak plasma concentration (C_{max}) was 415.6 versus 508.7 ng/mL and mean time to reach C_{max} was 3.8 versus 3.6 hours for controls in phase one versus phase two, respectively; both AUC and C_{max} levels were significantly reduced by *khat*-chewing (P <0.050). For *khat*- versus non-*khat*-chewing malaria patients, mean plasma CQ concentrations were 266.4 ng/mL versus 427.5 ng/mL (P <0.001). Furthermore, CQ was effective in 71.7% and 75.4% of non-*khat* and *khat*-chewing malaria patients, respectively (P = 0.823). ***Conclusion:*** *Khat*-chewing was found to significantly reduce plasma CQ levels among healthy volunteers and malaria patients. While receiving CQ treatment, patients should be advised not to chew *khat*.

Keywords: Khat; Catha edulis; Malaria; Plasmodium falciparum; Chloroquine; Yemen.

الملخص: أهداف: هدفت هذه الدراسة إلى تقييم تأثير القات على التوافر البيولوجي لعقار الكلوروكين (CQ) في البالغين الأصحاء اليمنيين وتأثيرها على مستويات البلازما للكلوروكين وقدرته على إزالة الطفيليات بين مرضى الملاريا. **منهجية:** أجريت هذه الدراسة في الفترة ما بين يناير وأبريل 2007 في باجل وصنعاء اليمن. أعطيت جرعتين من الكلوروكين 600 ملغ لكل منهما إلى 15 للذكور الأصحاء في عدة مناسبات منفصلة وأعقبت الجرعة الأولى جلسة لمضغ القات (المرحلة الأولى)، بينما امتنعت المجموعة الضابطة عن مضغ القات (المرحلة الثانية). بالإضافة إلى ذلك، تم علاج 103 مريضا بالملاريا التي يسببها البلازموديوم المنجلي وشملت المرضى الذين يمضغون القات بانتظام (ن = 57) والذين لايمضغون القات (ن = 46)، بواسطة جرعة 25 ملغ/كلغ من عقار الكلوروكين على مدى ثلاثة أيام. وقد تم تحليل حركية الدواء في كل من المجموعة الضابطة و مرضى الملاريا. وتم التحقق من إزالة الطفيليات أيضا للمجموعة الثانية. **نتائج:** كان متوسط المنطقة تحت منحنى التركيز الزمني هو 2,108.9 مقابل 2,797.4 نانوغرام/ساعة/مل، بينما كان متوسط ذروة تركيز البلازما (C_{max}) 415.6 مقابل 508.7 نانوغرام/مل ومتوسط وقت الوصول إلى C_{max} كان 3.8 مقابل 3.6 ساعة للمجموعة الضابطة في المرحلة الأولى مقابل المرحلة الثانية، على التوالي. انخفض متوسط المنطقة تحت منحنى التركيز الزمني و ال C_{max} بشكل ملحوظ احصائيا مع مضغ القات (P <0.050). كانت متوسط تركيزات البلازما للكلوروكين هي 427.5 نانوغرام/مل المرضى الذين لايمضغون القات مقابل 266.4 نانوغرام/مل في في المرضى ماضغي القات (P <0.001). وعلاوة على ذلك، كان عقار الكلوروكين فعالا في71.7% في المرضى الذين لايمضغون القات مقابل و 75.4% من مرضى الملاريا ماضغي القات على التوالي (P = 0.823). الخاتمة: لمضغ القات تاثير كبير على انحفاض تركيز عقار الكلوروكين في الذكور الأصحاء ومرضى الملاريا. لذلك يجب نصح هؤلاء المرضى بالتوقف عن مضغ القات اثناء تلقي العلاج.

كلمات مفتاحية: قات؛ كاثا أدليس؛ الملاريا؛ البلازموديوم المنجلي؛ الكلوروكين؛ اليمن.

Advances in Knowledge

- *This is the first study to reveal that the use of khat (Catha edulis) significantly reduces plasma chloroquine (CQ) levels among malaria patients receiving CQ treatment.*
- *The results of this study may be used as a reference for future research on possible interactions between khat and other drugs.*

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Application to Patient Care

- *Chewing khat was found to significantly reduce plasma CQ levels among malaria patients. Health workers and medication manufacturers should be educated regarding the potential risk of CQ and khat co-administration and patients should be advised to abstain from khat-chewing whilst undergoing CQ treatment.*
- *The rate of CQ resistance noted in this study was above the threshold recommended by the World Health Organization. This indicates that current antimalarial drug practices in Yemen need to be modified.*

Certain dietary substances can affect the absorption of medications, including antimalarial drugs.[1–6] Drug-food, drug-drug and drug-herb interactions are factors that can alter the pharmacokinetic activity of a drug; these interactions can occur during absorption, distribution, metabolism or excretion.[7] *Catha edulis* of the *Celastraceae* family, also known as *khat*, is a plant widely cultivated in East Africa (e.g. Kenya and Ethiopia) and the Arabian Peninsula.[8] The main active substances in fresh *khat* leaves are alkaloids with amphetamine-like properties which have euphoric and stimulatory effects.[9] In Yemen, *khat*-chewing is a widespread habit; approximately 80–85% of male and 50–60% of female adults in Northern Yemen chew *khat* at least once a week.[8] The concurrent use of *khat* with standard medications is anecdotally purported to be common practice in Yemen. Attef *et al*. found that *khat*-chewing among healthy Yemeni adults significantly reduced the bioavailability of the antibiotics ampicillin and amoxicillin.[10] Another study observed a significant reduction in the pharmacokinetic parameters of tetracycline-hydrochloride among healthy Yemeni adults after a *khat*-chewing trial.[11]

Malaria is a serious health concern in Yemen, with an estimated 800,000–900,000 malaria cases occurring each year.[12] The disease is responsible for significant child morbidity and mortality; 1% of deaths among Yemeni children under five years of age are attributed to malaria.[13] Recently, a study revealed that repeated dosing with extracts of *khat* reduced parasite loads among Swiss albino mice with malaria infections.[14] However, Ketema *et al*. found that *khat* exhibited only mild antimalarial activity among *khat* chewers in Ethiopia.[15] Chloroquine (CQ) is one of the most common antimalarial drugs prescribed by Yemeni physicians.[16] Consequently, determining whether *khat*-chewing has an effect on CQ is of great importance. This study was undertaken to investigate the effect of *khat* use on CQ bioavailability in both healthy volunteers and malaria patients. In addition, the effect of *khat* use on parasitaemia and parasite clearance among the latter group was also examined.

Methods

This study was carried out between January and April 2007 in Bajil and Sana'a, Yemen. Healthy volunteers and malaria patients made up the control and patient groups, respectively. A total of 15 healthy males >18 years old and between 45–64 kg were recruited from Sana'a, the capital city of Yemen, which is free of malaria.[16] The healthy volunteers were selected from a group of respondents to an advertisement requesting participants for an unpaid pharmacological study. Volunteers with blood film examinations negative for malaria parasites, normal standard laboratory test results (including haemoglobin [Hb] levels, a complete blood count and kidney and liver function tests) and who were otherwise healthy with no history of clinical illness were included in the control group. Individuals with a history of taking antimalarial or prophylactic drugs over the preceding 12 months and those who were unwilling to abstain from *khat*-chewing for a period of 72 hours were excluded. For the patient group, all patients >18 years old who presented to the National Malaria Control Centre in Bajil, Al-Hudaydah Governorate, West Yemen, with microscopy-confirmed *Plasmodium falciparum*-induced malaria were recruited (n = 103). This area was identified as a meso- to hyperendemic area for malaria between January–April 2007.[17] All patients had a *P. falciparum*-positive thick or thin blood smear and were seeking treatment for uncomplicated malaria with no signs of severe malaria, such as an inability to drink/eat or sit/stand upright, repeated vomiting, convulsions, lethargy or unconsciousness and Hb levels of <7 g/dL. The presence of a documented fever was not required for enrolment in the study.

The control group underwent two phases of CQ administration. In the first phase, a single dose of CQ (600 mg) was administered orally to the healthy volunteers immediately before a *khat*-chewing session which involved chewing ~250 g of fresh *khat* leaves for 4–5 hours. The second phase occurred four weeks later; participants received a second single dose of CQ (600 mg) after having been instructed to abstain from *khat*-chewing for the preceding 48 hours. In addition, they were also requested to refrain from *khat*-chewing for 24 hours on the day of dosing. All of the participants clearly understood the purpose of the study and had agreed to abstain from *khat*-chewing for the required period of time. The principle investigator contacted all of the volunteers two days before their second CQ dose to reiterate the necessity of abstaining from *khat*-chewing. For both stages, 3

mL of venous blood was collected from the subjects before and during CQ administration, at 0.5, one, two, three, four, six, eight, 12 and 24 hours after each dose. Samples were transferred to heparinised tubes and the plasma was separated and stored at -75 ˚C until analysis. Several studies have shown that CQ levels are negligible four weeks after a single dose.[18,19] Furthermore, active components of *khat*, cathine and cathinone are reportedly undetectable 24 hours after chewing.[20] As a result, residual levels of either the first dose of CQ or *khat* would not interact with the CQ dose administered during the second phase.

Malaria patients were divided into regular *khat* chewers and non-*khat* chewers. Patients were considered *khat* chewers if they reported chewing *khat* more than three days a week. For *khat*-chewing patients, the amount and type of *khat* used and the timing of the *khat*-chewing sessions was not controlled by the researchers. On average, *khat* chewers reported using 230–350 g of fresh *khat* leaves for 4–5 hours per session. All of the malaria patients received oral CQ tablets administered over three days, at 10 mg/kg for the first two days and 5 mg/kg on day three, resulting in a total dose of 25 mg/kg. The full dose was readministered if the patient vomited within 30 minutes of the initial dose. A total of 3 mL of venous blood was drawn and transferred into heparinised tubes the day before CQ dosing (day zero) and during CQ treatment (days 1–3), 24 hours after the dose had been administered. Body temperature was recorded during treatment as a marker of illness/clinical cure. In addition, parasitaemia was routinely assessed on each visit by counting the number of asexual parasites against at least 200 white blood cells (WBCs) in thick blood films, assuming a normal WBC count of 8,000/µL.[21] Patients were followed-up for three days after the first dose of CQ.

A validated high-performance liquid chromatography (HPLC) method was used to determine plasma CQ concentrations in the blood samples of both healthy controls and malaria patients using the LC-2010HT system (Shimadzu Corp., Kyoto, Japan) with an auto sampler.[22] Ultraviolet detection of eluted peaks was performed at 340 nm. Chromatographic separation was carried out using the reversed-phase 150 mm x 4.6 mm Hypersil™ BDS C18 HPLC Columns (Thermo Fisher Scientific Inc., Waltham, Massachusetts, USA) with a 5 µm particle size. The mobile phase contained a mixture of water and acetonitrile (ratio: 85:15%) as well as 1% triethylamine adjusted to a pH of 2.8 with concentrated phosphoric acid and a flow rate of 1 mL/minute. The pharmacokinetic parameters of CQ were determined by measuring the area under the concentration-time curve (AUC) from time zero (before the CQ dosing) to 24 hours after CQ dosing, maximal plasma drug concentration (C_{max}) and time taken to reach maximum plasma concentration (T_{max}). These parameters were calculated using the compartmental/trapezoidal method with Kinetica Software for PK/PD Data Analysis, Simulation and Reporting, Version 4.4 (Thermo Fisher Scientific Inc.). Values were reported as means ± standard deviation.

Among the malaria patients, parasitological and clinical responses to CQ treatment were classified according to different treatment protocols devised by the World Health Organization (WHO) and Osorio *et al*.[23–25] In terms of parasitological response, the 1973 WHO protocol was utilised—patients were deemed to exhibit sensitivity (S)/resistance level I (RI) if the *P. falciparum* parasites had cleared by day two of CQ treatment; RII if there was incomplete clearance of parasites by day two; and RIII if parasite density on day two was ≥25% that of the parasite density observed on day zero.[23] Clinical responses to CQ were classified according to definitions used within the 1996 WHO treatment protocols and methods of Osorio *et al*.[24,25] Early treatment failure (ETF) was defined as (1) the development of any danger signs, such as fever and vomiting, severe malaria or parasite positivity on days 1–3 of CQ administration; (2) higher parasitaemia levels on day two than day zero; or (3) parasite density on day three ≥25% that of day zero.[24] Treatment success (TS) was classified as negative parasitaemia on day three.[24] Late treatment failure (LTF) was defined as the development of any danger signs, severe malaria, parasitaemia or fever after day three or an unscheduled return to seek medical help because of clinical deterioration or parasitaemia on any scheduled follow-up day on or after day seven.[25] In this study, follow-up after three days was not possible. Patients who failed to respond to CQ treatment were prescribed sulfadoxine/pyrimethamine. During follow-up, patients received a single dose of primaquine (0.75 mg/kg) at the end of the follow-up period if they were found to have gametocytaemia.

The t-test for paired values was used to compare pharmacokinetic parameters of CQ obtained after the two phases of the control group trial and to compare results between *khat*-chewing and non-*khat*-chewing malaria patients. Geometric means were used to summarise parasite densities for malaria patients. Parasite clearance and the association with *khat* use were analysed with a two-sided Fisher's exact test. A *P* value of <0.050 was used as the level of significance.

This study was approved by the institutional review boards of the Faculty of Medicine & Health Sciences at Sana'a University and the National Malaria Control Programme (NMCP) in Sana'a. In addition, ethical

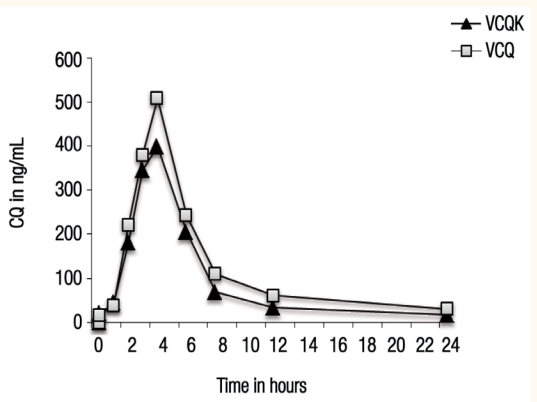

Figure 1: Mean plasma concentration-time curves of chloroquine* after a *khat*-chewing session† and after a 48-hour *khat*-chewing abstinence period among healthy adult males in Yemen (N = 15).

CQ = chloroquine; VCQK = after a khat-chewing session; VCQ = after a 48-hour khat-chewing abstinence period.

**Single dose of 600 mg. †Approximately 250 g of fresh khat leaves for 4–5 hours.*

permission was granted by the Liverpool School of Tropical Medicine in Liverpool, UK. Informed consent was obtained from all individuals before participation in the study.

Results

The control group ranged in age from 20–30 years old and had a mean Hb level of 16.2 g/dL and mean body weight of 62.86 kg. In phase two versus phase one, there was a significant reduction in mean C_{max} and AUC among the healthy controls (415.6 versus 508.7 ng/mL and 2,108.9 versus 2,797.4 ng/hour/mL; *P* <0.001 and 0.002, respectively) [Figure 1]. However, no significant difference was seen in mean T_{max} (3.8 versus 3.6 hours; *P* = 0.81) [Table 1]. The malaria patient group ranged in age from 20–45 years old and there were 57 *khat* chewers and 46 non-*khat* chewers. Of the *khat* and non-*khat* chewers, 25 (43.9%) and 27 (58.7%) were female and 32 (56.1%) and 19 (41.3%) were male, respectively. All patients had a temperature of >37 °C prior to treatment and had normal axillary temperatures by day three of CQ treatment. Plasma CQ levels among *khat*- and non-*khat*-chewing malaria patients according to day of treatment are presented in Table 2. Plasma CQ levels were significantly reduced among *khat*-chewing malaria patients compared to their non-*khat*-chewing counterparts (266.4 ± 164.3 and 427.5 ± 125.6, respectively; *P* <0.001).

Table 1: Effect of *khat*-chewing on the pharmacokinetic parameters of chloroquine* among healthy adult males in Yemen (N = 15)

Parameter	Mean ± SD		*P* value
	After 48-hour *khat*-chewing abstinence period	After *khat*-chewing session†	
AUC in ng/hour/mL	2,797.4 ± 845.9	2,108.9 ± 682.3	0.002
C_{max} in ng/mL	508.7 ± 106.4	415.6 ± 103.1	<0.001
T_{max} in hours	3.6 ± 0.5	3.8 ± 0.4	0.810

SD = standard deviation; AUC = area under the time-concentration curve; C_{max} = peak plasma concentration; T_{max} = time to reach peak concentration.

**Single dose of 600 mg. †Approximately 250 g of fresh khat leaves for 4–5 hours.*

Before CQ treatment, parasite density among the malaria patients ranged from 48–256,800 to 32–111,760/µL for non-*khat* and *khat* chewers, respectively. No danger signs or severe malaria symptoms were recorded for any of the patients. Geometric mean parasitaemia before treatment was 1,494/µL (95% confidence interval [CI]: 771–3,837/µL) and 1,861/µL (95% CI: 902–2,890/µL) for *khat* and non-*khat* chewers, respectively. On day three of CQ treatment, the geometric mean parasite density was 3/µL (95% CI: 1.7–5.2/µL) and 4/µL (95% CI: 1.9–7.9/µL) for *khat* and non-*khat* chewers, respectively. There was no significant difference in mean parasite density (*P* = 0.652 and 0.563, respectively) and axillary temperature (*P* = 0.140 and 0.145, respectively) between *khat* and non-*khat* chewers before treatment and on day three [Table 3].

Among the malaria patients, parasitaemia had cleared completely (S/RI) by day two of CQ treatment for 61 patients (59.2%) while the remaining 42 patients (40.8%) showed resistance at either the RII or RIII levels. Among non-*khat* chewers, 27 (58.7%) patients were completely clear of parasites by day two, while 19 (41.3%) showed parasitological resistance. However, CQ was effective for 34 *khat* chewers (59.6%) while 23 non-*khat* chewers (40.4%) were not clear of parasites by day two. There was no significant difference in overall resistance (RII and RIII) between the two groups (*P* = 0.394). Among the patient group, 76 (73.8%) had

Table 2: Effect of *khat*-chewing on chloroquine plasma levels among malaria patients in Yemen after chloroquine treatment* (N = 103)

Day of treatment	Mean plasma level ± SD		*P* value
	Non-*khat* chewers (n = 46)	*Khat* chewers (n = 57)	
Day one	216.5 ± 94.8	145.4 ± 73.8	<0.001
Day two	313.3 ± 123.4	202.7 ± 130.1	<0.001
Day three	427.5 ± 125.6	266.4 ± 164.3	<0.001

SD = standard deviation.

**Total dose of 25 mg/kg over three days.*

Table 3: Parasitaemia and axillary temperature before (day zero) and after (day three) chloroquine treatment* among *khat*- and non-*khat*-chewing malaria patients in Yemen (N = 103)

	Mean† parasitaemia per µL (95% CI)		Mean axillary temperature in °C ± SD	
	Day 0	Day 3	Day 0	Day 3
***Khat* chewers (n = 57)**	1,494 (771–3,837)	3 (1.7–5.2)	37.5 ± 0.54	36.8 ± 0.11
Non-*khat* chewers (n = 46)	1,861 (902–2,890)	4 (1.9–7.9)	37.4 ± 0.66	36.8 ± 0.14
***P* value**	0.653	0.563	0.140	0.145

CI = confidence interval; SD = standard deviation.
**Total dose of 25 mg/kg over three days. †Geometric mean.*

TS/LTF while 27 (26.2%) had ETF. Overall, rates of TS/LTF were 71.7% and 75.4% among non-*khat*- and *khat*-chewing malaria patients, respectively (P = 0.823). There was no significant difference between *khat* and non-*khat* chewers in terms of ETF (P = 0.823) [Table 4].

Discussion

The co-administration of CQ and *khat* was found to significantly affect the pharmacokinetics of CQ among both healthy controls and malaria patients in Yemen. This was comparable to reported reductions in the pharmacokinetic activity of tetracycline hydrochloride and the low bioavailability of ampicillin and amoxicillin when co-administered with *khat*.[10,11] A possible drug-plant CQ-*khat* interaction exists, as indicated by the significantly reduced CQ AUC and C_{max} values observed with *khat* use in the present study. The mechanisms underlying this interaction are unknown; however, one possible mechanism may relate to the interaction of the drug with some of *khat*'s components—tannic acid, cathinone and cathine—which are known to cause the formation of insoluble complexes and non-absorbable compounds.[10,11] In particular, it has been hypothesised that tannins in *khat* may interfere with absorption processes in the gastrointestinal tract.[10] Changes in gastric acidity and motility or the delay of gastric emptying may slow absorption of CQ, as these changes will prolong the duration of time that the drug remains in the stomach.[3,26] Finally, increased diuresis is often reported among *khat* chewers, possibly due to fluid intake during *khat*-chewing sessions;[26] this may lead to increased urinary excretion of CQ. Other possible mechanisms of pharmacokinetic interactions between *khat* and CQ require further investigation.

In the current study, CQ plasma levels were significantly reduced among *khat*-chewing malaria patients in comparison to non-*khat*-chewing patients. This may be associated with the general health of this patient group with regards to malnutrition, anaemia and parasitic infestation; Tulpule *et al*. found faster clearance of CQ in undernourished subjects.[5] In general, *khat*-chewing-induced symptoms include loss of appetite and malnutrition.[9] This may also affect digestion or absorption of CQ. In addition, differences in plasma CQ levels between healthy controls and malaria patients could be related to the age differences between the two groups. Edwards *et al*. reported that CQ blood concentrations and AUC were significantly higher in malaria patients receiving oral CQ treatment than in healthy subjects, suggesting that malaria induces absorption changes and increases systemic exposure to CQ.[27] In the current study, reduction in the plasma CQ levels among *khat*-chewing malaria patients could be related to impaired gastrointestinal absorption due to the effect of *khat* on CQ absorption.

In the current study, the rate of CQ resistance among the malaria patients was above the acceptable level of resistance determined by the WHO.[24] Several studies conducted in different governorates of Yemen have also demonstrated high to medium rates of CQ treatment failure, including rates of 61% in the

Table 4: *In vivo Plasmodium falciparum* response to chloroquine treatment* in *khat*- and non-*khat*-chewing malaria patients in Yemen (N = 103)

	n (%)					
	S/RI†	RII†	RIII†	Overall R†	TS‡/LTF§	ETF‡
***Khat* chewers (n = 57)**	34 (59.6)¶	17 (29.8)	6 (10.6)	23 (40.4)¶	43 (75.4)//	14 (24.6)//
Non-*khat* chewers (n = 46)	27 (58.7)	10 (21.7)	9 (19.6)	19 (41.3)	33 (71.7)	13 (28.3)
Total	61 (59.2)	27 (26.2)	15 (14.6)	42 (40.8)	76 (73.8)	27 (26.2)

S = sensitivity; R = resistance level; TS = treatment success; LTF = late treatment failure; ETF = early treatment failure.
**Total dose of 25 mg/kg over three days. †According to the 1973 treatment protocol of the World Health Organization.[23] ‡According to values obtained on day three of treatment and the 1996 treatment protocol of the World Health Organization.[24] §According to values obtained on day three of treatment and the methods of Osorio et al.[25] ¶P = 0.394. //P = 0.823.*

Al-Musaimeer district of Lahj Governorate and 16.1% in the Hethran and Al-Mafatch districts of Taiz Governorate.[28,29] In addition, Al-Mekhlafi *et al*. observed a high frequency of *P. falciparum* CQ resistance transporter T76 mutations, suggesting considerable CQ resistance among cases of *P. falciparum*-induced malaria in Yemen.[30] In the current study, parasite densities differed between *khat*- and non-*khat*-chewing malaria patients, although this finding was not significant. Ketema *et al*. similarly observed the mildly antiplasmodial characteristics of *khat* leaves.[15] Antimicrobial activity has also been reported.[31] Several studies have shown that plant alkaloids, flavonoids and tannins demonstrate *in vivo* and *in vitro* antimalarial activity against CQ-sensitive and -resistant strains of *P. falciparum*.[32,33] However, further studies are needed to investigate the antimalarial activity of *khat*. The limited reduction in parasite densities observed in the current study might be attributed to the antiplasmodial activity of certain compounds in *khat* leaves.

With regards to treatment outcomes, no significant differences were found between *khat*- and non-*khat*-chewing malaria patients in the current study. However, rates of successful treatment may have been overestimated, as certain patients might have been classified as cases of LTF instead of TS had they been followed-up for a longer period of time (e.g. 14–28 days). The lack of follow-up and utilisation of non-age-matched control and patient groups are limitations of the present study. Additionally, the difference between cases of S or RI could not be distinguished at day two; CQ resistance may therefore have been underestimated due to the short follow-up period. A further limitation was the use of CQ over artemisinin-based combination therapy (ACT). In 2005, the NMCP in Yemen replaced CQ with ACT as the first line of antimalarial therapy. Unfortunately, however, this policy was not implemented in practice until 2009.[16,17] Nevertheless, regardless of current antimalarial treatment guidelines, CQ remains the second most common antimalarial drug prescribed by physicians in public and private health facilities in Yemen.[16] As such, the results of this study should be utilised to educate health workers and medication manufacturers in Yemen as to the potential risk of CQ co-administration among *khat*-chewing patients. Current anti-malarial drug practices in Yemen should be modified accordingly and physicians should advise patients not to chew *khat* whilst undergoing CQ treatment.

Conclusion

The findings of this study revealed that CQ plasma levels were significantly reduced by *khat*-chewing; however, *khat*-chewing had no significant influence on rates of *P. falciparum* parasite clearance among malaria patients. The rate of CQ resistance among the patient group was above the acceptable levels determined by the WHO, indicating high CQ resistance in these areas of Yemen. Based on these preliminary findings, patients should be advised not to chew *khat* whilst undergoing CQ treatment.

CONFLICT OF INTEREST

The authors declare no conflicts of interest.

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