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RESEARCH ARTICLE

Degradation of Erythromycin in Seafood Products by Different Thermal Treatments

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Abstract

Prawn (*Macrobrachium rosenbergii*) and tilapia (*Oreochromis niloticus*) are two important seafood farming species cultivated in Mekong river delta, Vietnam. Intensive aquaculture of these species has high risk of disease outbreaks caused by virus, bacteria, fungi, and other pathogens. Erythromycin is the most widely used antibiotic in the macrolide. Erythromycin has a low cytotoxicity for aquaculture. Prawn and tilapia intended for export have to meet the antibiotic residue standards of the importing countries. In the present research, different factors affecting erythromycin residue on prawn and tilapia muscles after thermal treatments were carefully investigated. The present results proved that among frozen, cooked, oil-fried and baked methods; the oil-frying treatment strongly degraded erythromycin at 86.88% in prawn and 87.02% in tilapia samples. Thermal processing by oil heating medium at high temperature caused a significant degradation in the concentration of erythromycin residue.

Keywords: *Prawn, Tilapia, Erythromycin, Thermal, Degradation, Antibiotic residue.*

Introduction

Prawn (*Macrobrachium rosenbergii*) and Nile tilapia (*Oreochromis niloticus)* have been considered two of the most important species of freshwater aquaculture in Viet Nam, especially in the Mekong River Delta. During their cultivation, bacterial necrosis is a common disease observed in adult prawn. Bacterial flora like *Pseudomonas fluorescens, Aeromonas* sp., *Lactococcus garvieae* and *Edwardsiella tarda* are responsible for necrosis syndrome. Meanwhile, the most significant common diseases in Nile tilapia (*Oreochromis niloticus)* culture are caused by *Streptococcus iniae, Aeromonas hydrophila, Trichodina* sp., *Flexibacter, Edwardsiella* spp [1].

Streptococcosis can cause mass death in even healthy tilapia fishes [2]. Erythromycin is an antibiotic efficient for the curement of different bacterial infections [3]. It is broad spectrum antibiotic that exhibits high activity against nearly all Grampositive and Gram-negative bacteria [1]. It's considered as a common alternative for treatments with penicillin allergies [4]. Oral erythromycin is an effective drug for alleviating feeding intolerance in preterms [5].

Erythromycin has limited effectiveness and bioavailability owing to its instability and conversion under acidic conditions via an intramolecular dehydration reaction [6].The macrolide antibiotic erythromycin has long been the chemotherapeutant of choice to effectively prevent and control *Streptococcus* spp on tilapia and necrosis on prawn [7]. There is an increasing demand of global fishery trade. Intensive culture system having high stocking density frequently lead to disease outbreak causing tremendous economic losses to the aquaculture industry. Antibiotics are commonly utilized to cure disease outbreaks in fish and prawn farming [8].

The over-dose usage of antibiotics in general and erythromycin in particular in aquaculture may cause development of antibiotic resistance among pathogens infecting cultured fishes and humans [9, 10]. Erythromycin is among antibiotics utilized in aquaculture apart from chloramphenicol, streptomycin, prefuran, and neomycin [11]. The degradation of veterinary drugs approved for use in aquaculture is very important in the evaluation of the impact of these drugs on the environment and to ensure safe food production [12]. The half-life of erythromycin appeared to be longer in seawater than in freshwater [13]. The existence of antibiotic residues in foodstuffs is of concern due to possible direct allergic or toxicity reactions as well as the appearances of resistant strains of bacteria in animals with pathways to humans [14].

Thermal degradation of tetracyclines has been studied at some depth in pork [15], chicken [16], sheep [17] and fish [18]. Zorraquino et al [19]. Examined the degradation of ß-lactams by time-temperature combinations. Hsieh et al [20]. Evaluated the heat stability of 14 veterinary antibiotics under a short-term heating scenario by characterization of their structural degradation and their relationship to resultant changes in antimicrobial activity. Ola Svahn and Erland Björklund [21].

Studied the thermal stability assessment of antibiotics in moderate temperature and subcritical water using a pressurized dynamic flow-through system. The thermal degradation of ß-lactams, quinolones, sulfonamides, and tetracyclines can be described using a first-order kinetic model [22]. Bacitracin and neomycin sulphate were heat-labile at 100°C and 121°C, whereas amoxicillin and amoxicillin/clavulanate seemed to be heat-stable at 100°C but were heat-labile at 121°C.

Enrofloxacin, gentamicin, kanamycin, tiamulin, tilmicosin, tylvalosin and colistin were quite stable to heat at 121°C [23]. Objective of the present study investigated the destruction of thermal treatments like frozen, cooked, oil-fried, baking to the erythromycin residue on prawn and tilapia muscles.

Material and Method

Material

Prawn and tilapia were collected from Dong Thap province, Vietnam. After collecting, they must quickly conveyed to laboratory for experiments. They were washed, eviscerated, beheaded, deboned, skinned, filleted and their muscles were collected separately. These muscles must be ground into homogenized form. Erythromycin in white powder and purity (96.5%) was purchased from DHG Pharma, Vietnam.

Researching Method

The homogenized samples were fortified with erythromycin at suitable amount to obtain different antibiotic residue levels (50, 75, 100, 125, 150 ng/g). The incurred samples were then coming to frozen (-18°C)/ cooked (100°C, 1.5 minutes)/ oil-frying (130°C, 1.5 minutes)/ baking (130°C, 1.5 minutes). These thermal-treated samples were then set at ambient temperature before analysis.

Antibiotic Residue Determination and Statistical Analysis

Erythromycin residue (ng/g) was quantified by stripping square wave voltammetry [1]. The experiments were run in triplicate with three different lots of samples. Statistical analysis was performed by the Stat graphics Centurion version XVI. The data were presented as mean± standard deviation. Probability value of less than 0.05 was considered statistically significant.

Result & Discussion

Cooking time and temperature are two key parameters affecting to antibiotic residues [24]. Reduction percentage is in significant and positive correlation with cooking time, sample weight percent and center temperature. In the present research, the incurred prawn and tilapia samples were frozen (-18°C)/ cooked (100°C, 1.5 minutes)/ oil-fried (130°C, 1.5 minutes)/ baked (130°C, 1.5 minutes).

Our results revealed that the oil-frying strongly degraded erythromycin compared to other thermal treatments (table 1 and 2). Our findings were similar to other reports. Ola Svahn and Erland Björklund proved that ß-lactams cefadroxil, cefuroxime, amoxicillin, penicillin V, and penicillin G showed a high degree of stability with a maximum degradation of less than 30 % at 150 °C.

Ciprofloxacin and norfloxacin showed a very high thermal stability up to 200 °C, as did trimethoprim and sulfamethoxazole. At 250 °C all antibiotics were either partly of fully removed. The tetracycline doxycycline showed a specific removal pattern probably involving both binding to metal surfaces at lower temperatures as well as degradation at increased temperatures. Gratacós-Cubarsí and Fernandez-García [16] revealed that boiling for 14 min and microwave heating for

6 min reduced the amount of tetracycline in chicken and pig meat to between 56 and 82 %.

Boiling in lamp meat for 30 min was necessary to decrease oxy tetracycline levels by 95 % [17]. Boiling avoids the lengthy lag phase of heating commonly encountered in the cooking of solid tissue and can thus easily assess the actual exposures of antibiotics to heat [25]. Zorraquino et al [26].Demonstrated that sterilization at 120 °C for 20 min showed a considerable inactivation of penicillins (65%) and cephalosporins (90%).

Roca et al [14].Showed that ciprofloxacin and norfloxacin in milk were the most sensitive with 12 % degradation at 120 °C for 20 min, while enrofloxacin, flumequine and oxolinic acid were more recalcitrant, being degraded to only 5 % or less. Ultra high temperature heating to 140 °C for 4 s, had basically no effects on degradation.

In another report, ranking of heat stability by antibiotic classes at 121 °C was highest for sulfonamides, followed by lincomycin, colistin, tetracyclines and ß-lactams while at 100 °C sulfonamides equaled lincomycin and and was greater than colistin but variability was observed within different tetracyclines and ß-lactams [20]. Erythromycin was shown to be heat-labile [25]. Beta-lactam was found to degrade quite rapidly at 37°C [27].Significant reduction in stability and antibacterial activity of azithromycin solution when subjected to 70 and 100˚C for 24 h.

While stability of tetracycline was significantly reduced when subjected to 70 and 100˚C for 24 h [28]. Sterilization at 120 ºC for 20 min inactivated 93% of erythromycin. Treatment at 140 ºC for 10 s resulted in 30% reduction of erythromycin. Pasteurization (60 ºC for 30 min) reduced 21% erythromycin [26].

Table 1: Degradation (%) of erythromycin from raw and thermal treated samples analyzed in fortified prawn and tilapia muscles at level of50 ng/g

Sample	Prawn		Tilapia	
Fortification at 50 ng/g	Mean±SD (ng/g)	Reduction (%)	Mean±SD (ng/g)	Reduction (%)
Raw	49.13±0.02[a]	1.74±0.02[d]	49.34±0.03[a]	1.32±0.02[d]
Frozen (-18°C)	49.05±0.00[a]	1.90±0.01[d]	49.27±0.02[a]	1.46±0.01[d]
Cooked (100°C, 1.5 min)	34.61±0.01[b]	30.78±0.00[c]	33.09±0.01[b]	33.82±0.00[c]
Oil frying (130°C, 1.5 min)	6.41±0.03[d]	87.18±0.02[a]	5.87±0.03[d]	88.26±0.02[a]
Baking (130°C, 1.5 min)	12.85±0.00[c]	74.30±0.01[b]	11.45±0.02[c]	77.10±0.01[b]

Note: the values were expressed as the mean of three repetitions; the same characters (denoted above), the difference between them was not significant (α = 5%).

Table 2: Degradation (%) of erythromycin from oil-fried samples analyzed in fortified prawn and tilapia muscles at different fortification levels (50, 75, 100, 125, 150 ng/g)

Oil frying (130°C, 1.5 min)	Prawn		Tilapia	
Fortification levels (ng/g)	Mean±SD (ng/g)	Reduction (%)	Mean±SD (ng/g)	Reduction (%)
50	6.41±0.03[e]	87.18±0.02[b]	5.87±0.03[e]	88.26±0.02[ab]
75	8.56±0.00[d]	88.59±0.00[a]	8.45±0.03[d]	88.73±0.02[a]
100	13.79±0.01[c]	86.21±0.01[cd]	14.02±0.02[c]	85.98±0.01[bc]
125	17.54±0.02[b]	85.97±0.00[d]	18.13±0.00[b]	85.50±0.00[c]
150	20.31±0.03[a]	86.46±0.02[c]	20.09±0.01[a]	86.61±0.00[b]
Average reduction (%)		**86.88±0.01**	Average reduction (%)	**87.02±0.01**

Note: the values were expressed as the mean of three repetitions; the same characters (denoted above), the difference between them was not significant (α = 5%)

Conclusion

Prawn and fish are becoming an increasingly important source of food for human consumption. There are the increasing concerns of environmental sustainability and consumer preference for safe food. Erythromycin is widely used in both the aquaculture and livestock industries. Erythromycin varies in its susceptibility to degradation by heating processes. Differential heat stability of erythromycin after heating was conclusively demonstrated. Stability of erythromycin residue under cooking conditions is very important to provide valuable information related to health safety aspects from a safety and toxicological point of view.

References

1. Nguyen Phuoc Minh, Tran Bich Lam, Nguyen Trong Giao (2011) Simultaneous determination of erythromycin A in giant prawn and tilapia in Mekong region by stripping square wave voltammetry. European Journal of Food Research and Review, 1: 01-14.

2. Minh NP, Lam TB, Giao NT, Quan NC (2011) Accumulation and clearance of orally administered erythromycin in adult Nile tilapia (Oreochromis niloticus). International Food Research Journal, 18: 95-100.
3. Shafia S, Praneeth Chandluri, Ramesh Ganpisetti, Lakshmi BVS, Aravinda Swami P (2016) Erythromycin use as broad spectrum anti biotic. World Journal of Pharmaceutical and Medical Research, 2: 1-5.
4. Alvarez-Elcoro S, Enzler MJ (1999) The macrolides: Erythromycin, clarithromycin, and azithromycin. Mayo Clin. Proc., 74: 613-634.
5. Madani A, Pishva N, Pourarian Sh, Zarkesh M (2004) The efficacy of oral erythromycin in enhancement of milk tolerance in premature infants: A randomized clinical trial. IJMS, 29: 1-4.
6. Erika Cyphert L, Jaqueline Wallat D, Jonathan Pokorski K, Horst von Recum A (2017) Erythromycin modification that improves its acidic stability while optimizing it for local drug delivery. Antibiotics, 6: 11.
7. Minh NP, Lam TB, Giao NT and Quan NC (2010) Tissue distribution and elimination of erythromycin in giant freshwater prawn (Macrobrachium rosenbergii) depletion. African Journal of Food Science, 4: 578-584.
8. Holmström K, Gräslund S, Wahlström A, Poungshompoo S, Bengtsson BE, Kautsky N (2003) Antibiotic use in shrimp farming and implications for environmental impacts and human health. International Journal of Food Science and Technology, 38: 255-266.
9. Katrin Holmström, Sara Gräslund, Ann Wahlström, Somlak Poungshompoo (2004) Antibiotic use in shrimp farming and implications for environmental impacts and human health. International Journal of Food Science & Technology, 38: 255-266.
10. Sanjoy Banerjee, Mei Chen Ooi, Mohamed Shariff, Helena Khatoon (2012) Antibiotic resistant Salmonella and Vibrio associated with farmed Litopenaeus vannamei. Scientific World Journal, 2012: 130136.
11. Heny Suseno, Sumi Hudiyono, Muslim Muslim (2016) Elimination of chloramphenicol by tiger shrimp (Penaeus monodon) and white shrimp (Litopenaeus vannamei). HAYATI Journal of Biosciences, 23: 117-120.
12. Jin-Wook Kwon (2016) Environmental impact assessment of veterinary drug on fish aquaculture for food safety. Drug Test Analysis, 8: 556-564.
13. Jin-Wook Kwon (2011) Guidance on estimating soil persistence and degradation kinetics from environmental fate studies on veterinary pharmaceuticals for environmental risk assessment. Korean J. Environ. Agric., 30: 68.
14. Roca M, Castillo M, Marti P, Althaus RL (2010) Effect of heating on the stability of quinolones in milk. Journal of Agriculture and Food Chemistry, 58: 5427-5431.
15. Fedeniuk RW, Shand PJ (1997) Effect of thermal processing and additives on the kinetics of oxytetracycline degradation in pork muscle. Journal of Agriculture and Food Chemistry, 45: 2252-2257.
16. Gratacós-Cubarsí M, Fernandez-García A (2007) Formation of tetracycline degradation products in chicken and pig meat under different thermal processing conditions. Journal of Agriculture and Food Chemistry, 55: 4610-4616.
17. Ibrahim A, Moats WA (1994) Effect of cooking procedures on oxytetracycline residues in lamb muscle. Journal of Agriculture and Food Chemistry, 42: 2561-2563.
18. Kitts DD, Yu C, Aoyama RG, Burt HM (1992) Oxytetracycline degradation in thermally processed farmed salmon. Journal of Agriculture and Food Chemistry, 40: 1977-1981.
19. Zorraquino MA, Roca M, Fernandez N, Molina MP, Althaus R (2008) Heat inactivation of ß-lactam antibiotics in milk. Journal of Food Protection, 71: 1193-1198.
20. Hsieh MK, Shyu CL, Liao JW, Franje CA, Huang YJ, Chang SK, Shih PY, Chou CC (2011) Correlation analysis of heat stability of veterinary antibiotics by structural degradation, changes in antimicrobial activity and genotoxicity. Veterinarni Medicina, 56: 274-285.
21. Ola Svahn, Erland Björklund (2015) Thermal stability assessment of antibiotics

in moderate temperature and subcritical water using a pressurized dynamic flow-through system. International Journal of Innovation and Applied Studies, 11: 872-880.

22. Lei Tian, Salma Khalil, Stéphane Bayen (2017) Effect of thermal treatments on the degradation of antibiotic residues in food. Critical Reviews in Food Science and Nutrition, 57: 3760-3770.
23. Thamthaweechok N, Tiengrim S, Thamlikitkul V (2018) Heat stability of antibiotics commonly used in food animals and agriculture in Thailand. J. Med. Assoc. Thai., 101: 863-867.
24. Heshmati A (2015) Impact of cooking procedures on antibacterial drug residues in foods: A review. Journal of Food Quality and Hazards Control, 2: 33-37.
25. Hassani M, Lazaro R, Perez C, Condon S, Pagan R (2008) Thermostability of oxytetracycline, tetracyclines, and doxycycline at ultrahigh temperatures. Journal of Agricultural and Food Chemistry, 56: 2676-2680.
26. Zorraquino MA, Althaus RL, Roca M, Molina MP (2011) Heat treatment effects on the antimicrobial activity of macrolide and lincosamide antibiotics in milk. Journal of Food Protection, 74: 311-315.
27. Samara E, Moriarty TF, Decosterd LA, Richards RG, Gautier E, Wahl P (2017) Antibiotic stability over six weeks in aqueous solution at body temperature with and without heat treatment that mimics the curing of bone cement. Bone Joint Res, 6: 296-306.
28. Kurjogi M, Issa Mohammad YH, Alghamdi S, Abdelrahman M, Satapute P, Jogaiah S (2019) Detection and determination of stability of the antibiotic residues in cow's milk. PLoS ONE, 14: 0223475.

