



UNIVERSITI PUTRA MALAYSIA

***ANALYSIS OF AMINO ACIDS FOR DETECTION OF PORK IN
FRESH AND COOKED MEAT BY HIGH PERFORMANCE LIQUID
CHROMATOGRAPHY FOR HALAL AUTHENTICATION***

RAMIN JORFI

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AND COOKED MEAT BY HIGH PERFORMANCE LIQUID
CHROMATOGRAPHY FOR HALAL AUTHENTICATION**

By

RAMIN JORFI

**Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia,
in Fulfillment of the Requirement for the Degree of Master of Science**

April 2012

*Especially Dedicated to My Beloved
Parents, for Their Pray, Patience,
Patronage and Understanding*



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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of the requirement for the degree of Master of Science

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April 2012

Chairman: Associate Professor Shuhaimi bin Mustafa, PhD

Institute: Halal Products Research Institute

Many religions have some strict requirements about what should be used as food. In Islam, foods containing ingredients from pig sources are haram (unlawful) for the Muslims to consume. In the current global world, therefore, a reliable method to detect pork adulteration is needed to protect Muslim consumers from intentional or non-intentional fraud. Among the various analytical methods, high performance liquid chromatography (HPLC) has been widely used in many food authentication studies. In the current study, reverse phase high performance liquid chromatography (RP-HPLC) with ultraviolet (UV) detection system and pre-column derivatization with o-phthaldialdehyde (OPA) and 3-mercaptopropionic acid (3MPA) was applied to detect the presence of pork in beef, chevon, chicken and mutton at three different forms of raw, cooked and mixed meats. The identity and quantity of the amino acids were assessed by comparison of the retention times and peak areas of the standard amino acids. The results of this study showed that VAL, SER, ALA, HIS and ARG can be used to distinguish raw pork from the other meat types. However, analyzing

the cooked meats revealed that ASP, THR and TYR are potential markers for pork adulteration. In addition, it was observed that crude protein (CP) percentages of the raw meats were not changed significantly after cooking. Regarding the mixed meats, analysis of the results showed that 10-90% pork adulteration in beef can be identified using the CYS and CP as markers. Nonetheless, this was not the same in other types of meat in that VAL and CP for chicken and VAL and phenylalanine for mutton showed potential as markers of 10-90% pork adulteration. There were no amino acids identified as a marker of pork adulteration in chevon. However, PHE may consider as a relative indicator of pork adulteration in chevon in only percentages of more than 20%.

In conclusion, the results of the current study indicate that amino acid profile analysis using RP-HPLC can be employed as a marker for pork adulteration in beef, chevon, chicken and mutton in both forms of raw and cooked. With regards to the raw and mixed meat analysis, it can be concluded that VAL is a potential marker to distinguish pork adulteration in mutton and chicken. Further studies needed to find an appropriate marker in case of the beef and chevon. Moreover, ASP, THR and TYR appeared to be good markers of pork adulteration in cooked beef, mutton, chevon and chicken.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Master Sains

ANALISIS ASID AMINO DALAM MENGESAN DAGING KHINZIR YANG TERDAPAT DALAM DAGING MENTAH DAN DAGING YANG DIMASAK DENGAN MENGGUNAKAN CECAIR KROMATOGRAPHY YANG BERPRESTASI TINGGI UNTUK PENENTUAN HALAL SEBENAR

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Kebanyakan agama adalah sangat prihatin terhadap keperluan bahan untuk dijadikan sebagai sumber makanan. Dalam Islam, makanan yang mengandungi bahan dari sumber khinzir adalah haram (tidak dibenarkan) kepada orang islam untuk digunakan. Dalam dunia global kini, maka, cara yang mudah diperlukan dalam mengesan penggunaan daging khinzir dalam produk makanan dalam melindungi pengguna Muslim dari penipuan secara sengaja atau tidak disengajakan.

Di antara pelbagai cara analisis, penggunaan cecair kromatography (HPLC) yang berkuasa tinggi telah digunakan secara meluas dalam penggunaan kajian makanan. Dalam kajian terkini, RP-HPLC dengan pengesanan sistem UV dan pra-kolum yang diperolehi dengan o-phthaldialdehyde (OPA) dan asid 3-mercaptopropionik (3MPA) telah diaplikasikan untuk mengesan kehadiran daging khinzir yang terdapat dalam

daging lembu, daging kambing, daging ayam dan daging biri-biri dalam 3 keadaan iaitu mentah, yang dimasak dan campuran daging.

Dalam penentuan dan pengukuran asid amino telah dinilai dengan menggunakan perbandingan pengekalan masa dan kawasan tahap bagi asid amino. Keputusan menunjukkan bahawa VAL, SER, ALA, HIS dan ARG telah digunakan untuk membezakan daging khinzir berbanding daripada daging lain.

Walaupun, analisa terhadap daging yang dimasak telah menunjukkan bahawa ASP, THR dan TYR adalah petanda yang boleh digunakan untuk mengesan kehadiran daging khinzir. Juga, ia telah menunjukkan peratus protin dalam daging mentah adalah tidak berubah secara signifikan selepas dimasak. Berdasarkan dari campuran daging, analisa menunjukkan bahawa 10-90% kehadiran daging khinzir dalam daging lembu boleh dikesan dengan menggunakan CYS dan CP sebagai penanda aras. Ianya adalah tidak sama dalam jenis daging yang berlainan yang mana VAL dan CP untuk pengesanan daging ayam serta VAL dan phenylalanine untuk daging biri-biri menunjukkan penanda aras yang berpotensi sebanyak 10-90% dalam pengesanan daging khinzir. Ianya menunjukkan bahawa tiada asid amino yang boleh digunakan sebagai indikator penentuan daging khinzir di dalam daging kambing. Walaupun, PHE mungkin boleh dianggap sebagai penanda aras dalam penentuan daging khinzir di dalam daging kambing tidak lebih dari 20 peratus.

Kesimpulannya, keputusan menunjukkan bahawa analisis asid amino menggunakan RP-HPLC boleh dijadikan sebagai penanda aras dalam pengesanan daging khinzir dalam daging lembu, daging kambing, daging ayam dan daging biri-biri dalam

keadaan mentah dan yang telah dimasak. Bagi analisis daging mentah dan daging campuran, ianya boleh disimpulkan bahawa valine adalah penanda aras yang berpotensi dalam mengesan kehadiran daging khinzir dalam daging biri-biri dan daging ayam. Kajian yang lebih terperinci diperlukan untuk mendapatkan penanda aras yang berpotensi bagi kajian dalam daging lembu dan daging kambing. Tambahan pula, asparagine, threonine dan tyrosine menunjukkan boleh dijadikan penanda aras yang baik bagi pengesanan daging khinzir dalam daging yang dimasak, daging lembu, daging kambing dan daging ayam. Penemuan daripada kajian ini boleh dijadikan sebagai rujukan asas dalam kajian penentuan makanan halal.

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I certify that a Thesis Examination Committee has met on (27 April 2012) to conduct the final examination of Ramin Jorfi on his thesis entitled “Analysis of Amino Acids for Detection of Pork in Fresh and Cooked Meat by High Performance Liquid Chromatography for Halal Authentication” in accordance with the Universities and University Colleges Act 1971 and the Constitution of the Universiti Putra Malaysia [P.U. (A) 106] 15 March 1998. The Committee recommends that the student be awarded the Master of Science.

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DECLARATION

I declare that the thesis is my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously, and is not concurrently, submitted for any other degree at Universiti Putra Malaysia or at any other institutions.

RAMIN JORFI

Date: April 2012



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