ABSTRACT

Howida Abdallah Mohammad. Application of Molecular Biology for Production of Dairy Products of Medical Importance. Unpublished Ph.D. Dissertation, Ain Shams University, Faculty of Agriculture, Department of Food Science, 2006.

Classical phenylketonuria is the most common inborn error of amino acid metabolism. It is caused by deficiency of the phenylalanine hydroxylase enzyme. Accumulation of phenylalanine in brain leads to severe mental retardation. A low phenylalanine diet can significantly prevent the occurrence of mental retardation, and is so essential for the hyperphenylalaninemic mothers.

This study was planned to achieve a low phenylalanine formula prepared from cow skim milk with adding amino acids and vitamins to supply the needed recommended intakes of the essential amino acids, carbohydrates, fat, minerals and vitamins that are usually supplied by natural milk protein intake.

Skim milk hydrolysate was obtained using two genetically prepared and SDS/PAGE purified, immobilized proteolytic enzymes (papain and protease XXIII from *A.oryzae).* The debittering was accomplished by two methods based on activated carbon and barium sulphate adsorption in order to achieve the most balanced amino acids pattern with high biological value and palatable nonbitter taste to accomplish the feeding treatment aims.

BALB/c mice were used as a model for introduction of hyperphenylalaninemia with 3% L-phenylalanine and 0.3% L-ethionine. The histopathological, immunological, lipids, protein and blood profiles before and after mutagenesis were compared in blood, brain, kidneys and liver of the experimental mice model as well as feeding on the two achieved skim milk hydrolysates. Also, a comparison was done by feeding with one of the available commercial low-phenylalanine formula.

To conclude, one of the achieved skim milk hydrolysate treated with barium sulphate proved to be effective in preventing and reversing most of the pathological and abnormal phenylketonuria syndromes. It was nutritionally safe, microbiologically free, with high biological value, protein efficiency ratio, net protein ratio, and food efficiency. In the same time, it was characterized by being cheap and easy to obtain for straightaway use.

Key words: Phenylketonuria - mutagenic mice - low phenylalanine - activated carbon - barium sulphate - skim milk hydrolysate - protein efficiency ratio - net protein ratio - mental retardation.

CONTENTS

LIST OF ABBREVIATIONS

%	:Percentage
α	:Alpha
β	:Beta
κ	:Kaba
μ	:Micro
5-HTP	:5-hydroxytryptophan
AAM	:Amino acid mixture
AAP	:Acid amino peptidase
AAV	:Adeno-associated virus vector
ACE	:Angiotensin converting enzyme
AGE	:Agrose gel electropheresis
AOAC	:Association of official analytical chemist
ATP	:Adenosine triphosphate
BBB	:Blood brain barrier
BF	:Breastfed
BH^4	:Tetrahydrobiopterin
BMD	:Bone mineral density
BV	:Biological value
Ca	:Calcium
CBC	:Cell blood count
cDNA	:complementary DNA
CHD	:Congenital heart disease
DC	:Digestibility coefficient
DH	:Degree of hydrolysis
dl	:Deciliters
DNA	:Deoxynucleic acid
DPP	:Dipeptidyl peptidase
EDTA	:Ethylene diamine tetera acetic acid
ER	:Endoplamic reticulum

ESADD : Estimated safe and adequate daily dietary
Fe : Iron
FE : Feed efficiency
FF : Formula fed
g : Gram
GMP : Glycomacropeptide
H_2O_2 : Hydrogen peroxide
Hb : Hemoglobin
HCL : Hydrochloric acid
HDL : High density lipoproteins cholesterol
HMGR :3-hydroxy 3-methyl gltaryl coenzyme A reductase
HPA : Hyperphenylalaninaemia
HPLC :High performance liquid chromatography
Ig : Immunoglobulin
IQ : Intelligence quotient
IU : International units
kDa : Kilodalton
Kg : Kilogram
Km : Michaelis constant
LAP : Leucine amino peptidase
LBN : Lean body mass
LDL : Low density lipoprotein
Lf : Lactoferrin
LNAA : Large nutral amino acid
LP : Lactoperoxidase
LPP : Low phenylalanine peptides
LUPUFA : Long-chain polysaturated fatty acid
MHP : Mild hyperphenylalaninaemia
MPH : Milk protein hydrolyzates
MPKUCS : Maternal pku collaborative atudy
MRC : Medical Research Counical

mRNA : Messenger ribonucleic acid
MWD : Molecular weight distribution
N : Nitrogen
NaCl : Sodium chloride
NPN : Non protein nitrogen
NPR : Net protein ratio
NPU : Net protein utilization
PAH : Phenylalanine hydroxylase
PER : Protein efficiency ratio
pH : Negative log of hydrogen ion concentration
Phe : Phenylalanine
PHF : Partially hydrolyzed formula
PKU : Phenylketonuria
QDDPR : Dihydropteridine reductase
RBC : Red blood cells
RDA : Recommended daily allowance
REE : Resting energy expenditure
SBA : Secondary butyl alcohol
SDS/PAGE :Sodium dodecyl sulphate/Purified agrose gel electrophoresis
Se : Selenium
SMA : Synthetic milk adapted
TBN : Total body nitrogen
TCA : Trichloroacetic acid
TNBS : Trinitro-benzene-sulfonic acid
Tyr : Tyrosine
U : Unit
USA : United States of America
VLDL : Very low density lipoprotein
WHO : World Health Organization
WP :Whey proteins

WPC	:Whey protein concentrate
Zn	:Zinic