Analysis of the amino acid profile of milk, an important supplier of essential amino acids





#### Introduction

Lactoprotein is one of the main nutrients in milk, it is composed of 20 kinds of amino acids. Different amino acid ratios and arrangements form different types of proteins. Those proteins play an important role in maintaining the growth and renewal of body tissues, participating in various chemical reactions, and providing heat energy for physiological activities.

Lactoprotein is considered to be one of the proteins with the highest nutritional value. The main reason is that its amino acid content and composition ratio are basically close to the amount and ratio required by the human body for synthesis.

The nutritional value of protein is measured by whether its amino acid composition is reasonable and complete. Amino acids can be divided into two categories: essential amino acids and non-essential amino acids according to needs of human body. The socalled essential amino acids refer to amino acids that are needed by the human body but cannot be synthesized by itself or the synthesis speed cannot meet the needs and need to be obtained from food. There are nine kinds of essential amino acids in human body, they are Leu, Ile, Lys, Met, Phe, Thr, Try, Val and His. Lactoprotein is rich in the above nine kinds of amino acids, but there are certain differences between different protein types. The content of essential amino acids contained in casein is 45.1 g/100 g, while whey protein is 50.9 g/100 g. Therefore, whey protein is recognized as the best protein by nutritionists. According to the chemical scoring method, human milk and whole eggs score 100 points, milk scores 95 points, and whey protein scores as high as 104 points. The essential amino acid content of ideal protein and the recommended amount for 1year-old children (1991) have proposed by FAO and WHO. Compare which with the content of essential amino acids in lactoprotein in milk, they are very close.

Therefore, milk is considered to be one of the most nutritious foods. FAO and WHO have also proposed the daily requirement of essential amino acids for healthy adults. Suppose that an adult's weight is 60 kg, then drinking 500 mL milk a day can meet his daily needs of essential amino acids except for Met + Cys.

### Sample Preparation and Analysis

For the analysis of free, physiological amino acids in two types of milk a long-life low-fat country milk "Milk I" with a fat content of 1.5% and a long-life full cream "Milk II" with a fat content of 3.5% were selected for the studies. From both samples, previously homogenized

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by vigorous shaking, 1 ml each is added to a reaction vessel and centrifuged at 14500 rpm for 7 min. The fat is deposited on the surface of the samples. From the solutions below 400  $\mu$ L of the sample was mixed with 100  $\mu$ L of precipitation solution were added and deposited in the refrigerator for 20 min for the protein precipitation. The sample was centrifuged for 5 min at 14000 rpm. 100  $\mu$ L of the particle free solution were mixed with 100  $\mu$ L of the particle free solution were mixed with 100  $\mu$ L sample dilution buffer (including internal standard norleucin with a final concentration of 100 nmol/mL).

# Table 1: Determined amino acid concentrations in thetwo types of milk studied.

Amino Acid	"Milk I"	"Milk II"
	nmol/mL	nmol/mL
P-Ser	9.0	16.9
Tau	63.3	57.5
PEA	86.2	90.3
Urea	> 3.000.0	> 3.500.0
Asp	23.7	40.6
Thr	10.1	10.2
Ser	13.9	8.2
Asn	Not detected	Not detected
Glu	318.5	292.5
a-AAA	4.4	4.2
Gly	94.3	98.7
Ala	38.9	42.5
Cit	3.2	3.8
a-ABA	4.0	4.4
Val	16.0	12.9

Met	Not detected	Not detected
Leu	6.5	4.5
lle	5.8	3.6
Tyr	3.4	3.1
Phe	2.6	1.5
H-Cystine	Not detected	Not detected
b-AiBA	242.3	141.9
g-ABA	31.9	33.1
His	3.1	3.4
Orn	9.0	7.8
Lys	32.3	28.6
NH4	445.8	513.3
EOHNH2	112.5	193.2
Arg	21.4	18.0
Pro	22.7	23.3

The samples were analyzed by the Amino Acid Analyzer ARACUS, manufactured and distributed by membraPure GmbH worldwide. ARACUS is using the classic routine analysis of amino acids by post-column derivatization with ninhydrin and the detection at 440 nm and 570 nm.



Figure 1: Amino Acid Analyzer ARACUS





Figure 2: Comparison of a physiological amino acid standard (blue) with "Milk I" sample (black, top image) and "Milk II" sample (black, bottom image). The detection was performed at 440 nm and 570 nm. The concentrations of the individual amino acids were determined using a known concentration of the standard amino acid mixture.



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