# The Amino Acid Sequence Determination of a New Variant of Kunitz Soybean Trypsin Inhibitor (SBTi-A2)

*Xin Hua, Xie Kefang, Dong Aiwu, Yan Qingyan, and Gu Qimin*
Biochemistry Department
Fudan University
Shanghai 20043
P.R. of China

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## Introduction

A substantial amount of Kunitz trypsin inhibitor (SBTi-A2) exists in soybean seeds. Three codominant alleles, Ti-a[Soybase], Ti-b[Soybase] and Ti-c[Soybase] were identified and their protein sequences have been determined. Ti-d was reported by Zhao Shuwen (1992), as a new variant of SBTi-A2 discovered in the Gansu province of China. The amino acid sequence of Ti protein was deduced from its DNA sequence which is obtained by PCR and determined by DNA sequence analysis in our laboratory.

## Materials and methods

DNA fragment encoding Ti-d protein was obtained from PCR method by using primers designed on the basis of Ti-a sequence. After that, the amplified DNA fragment was cloned into plasmid and subject to DNA sequencing by dideoxynucleoside chain termination method.

## Results and discussion

As shown in Figure 1, two amino acid residues in Ti-d are different from those in Ti-a, one is located in N-terminal signal peptide, the other is located in the mature protein, which is Glu(69) in Ti-a turned out to be Lys(69) in Ti-d . The result is consistent with the slower migration of the whole Ti-d protein and the CNBr degraded N-terminal fragment on native PAGE. The translated

amino acid sequence of Ti-d mature protein is also different from Ti-b and Ti-c (Table 1).

References

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