Supplementary materials

Sourdough Fermentation Degrades Wheat Alpha-Amylase/Trypsin Inhibitor (ATI) and Reduces Pro-Inflammatory Activity

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Band 1

>sp|P17314|IAAC3_WHEAT Alpha-amylase/trypsin inhibitor CM3 OS=Triticum aestivum OX=4565 PE=1 SV=1

1 MACKSSCSLLLLAAVLLSVLAAASASGSCVPGVAFRTNLLPHCRDYVLQQ

51 TCGTFTPGSKLPEWMTSASIYSPGKPYLAKLYCCQELAEISQQCRCEALR

101 YFIALPVPSQPVDPR SGNVGESGLIDLPGCPR EMQWDFVR LLVAPGQCNL

151 ATIHNVR YCPAVEQPLWI

Band 2

>sp|P16851|IAAC2_WHEAT Alpha-amylase/trypsin inhibitor CM2 OS=Triticum aestivum OX=4565 PE=1 SV=2

1 MASKSSITHLLLAAVLVSVFAAAAATGPYCYPGMGLPSNPLEGCR EYVAQ

51 QTCGVGIVGSPVSTEPGNTPR DRCCKELYDASQHCRCEAVRYFIGRTSDP

101 NSGVLKDLPGCPREPQRDFAKVLVTPGHCNVMTVHNTPYCLGLDI

Figure S1. Protein sequences of identified ATI tetramers. Unique peptides underlined were identified by MALDI-TOF/TOF.

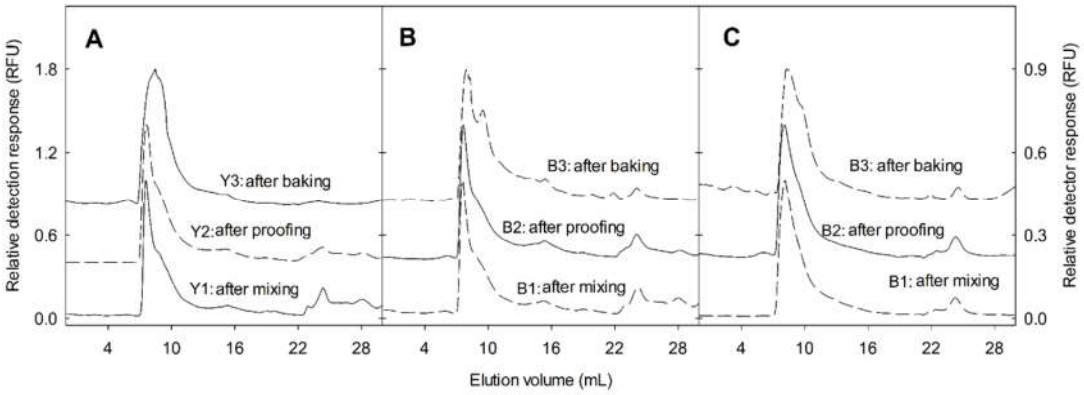

Figure S2. Size-exclusion chromatogram of fluorescence-labelled ATI in bread-making, **Panel A**: Yeast-fermented bread, **Panel B**: (III) chemically acidified dough at pH 5.0, **Panel C**: (V) Rye starter-wheat sourdough. FITC-labelled ATI was added to bread dough. ATI was extracted after mixing of sourdough, after sourdough fermentation, or after mixing of bread dough, after proofing, and after baking. FITC-ATI was separated by size exclusion chromatography coupled to a fluorescent detector set at excitation 488 nm and emission 530 nm. Chromatograms were normalized to the highest peak intensity in each chromatogram, and were offset by 0.4 RFU. The elution volume of the molecular marker bovine serum albumin (66 kDa) was 7.25 mL, of lysozyme (14 kDa) was 9.86 mL, and of glutathione (307 Da) was 17.35 mL. The Roman number and the sample letters are the same as those indicated in Table 1.