Phytases from Bifidobacteria for Human Nutrition

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Valencia - SPAIN

Myo-Inositol Hexakisphosphate or Phytic Acid and Phytates

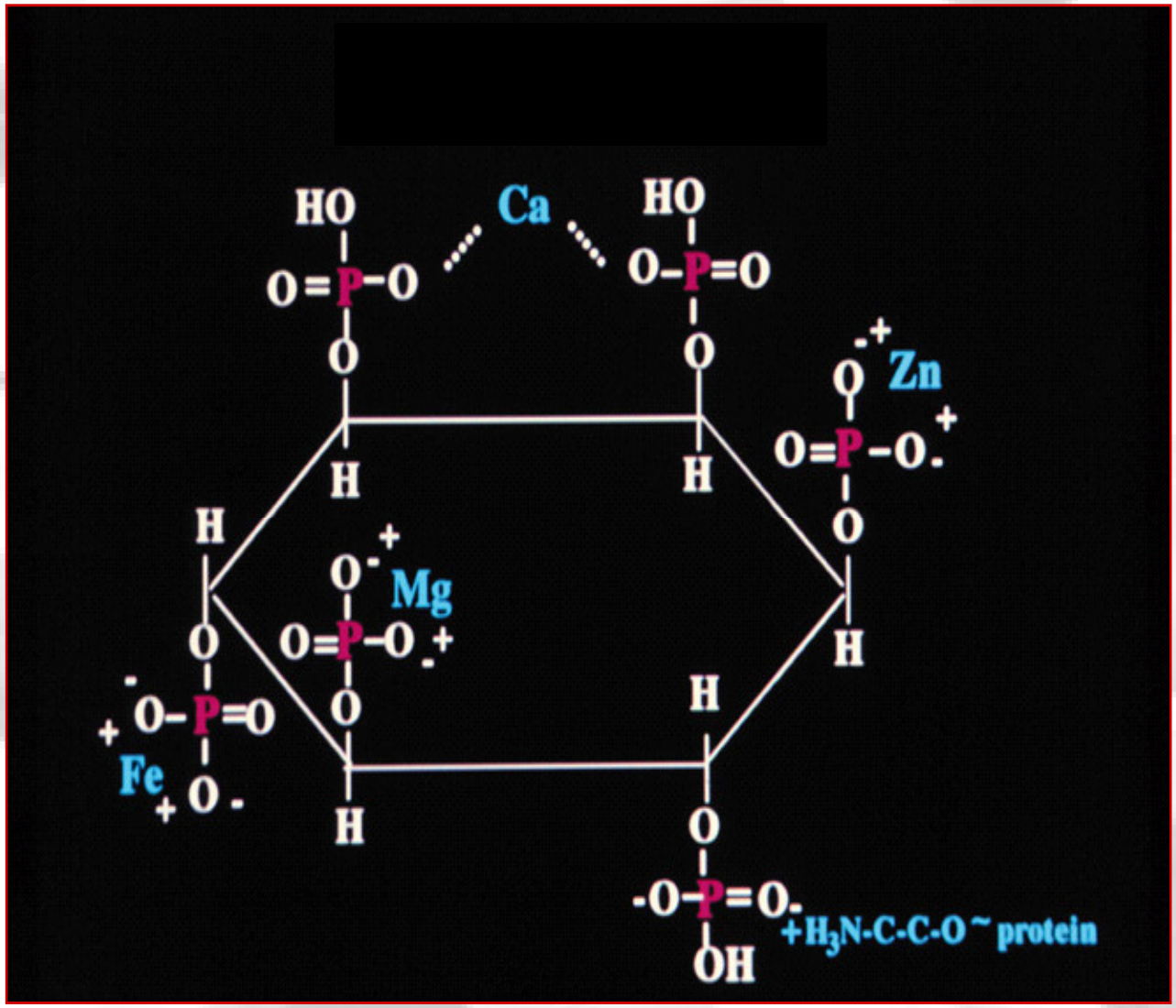
Are

- The primary source of inositol
- The major storage form of phosphorus in plants



Present in

- Seeds
- Cereal grains
- Legumes



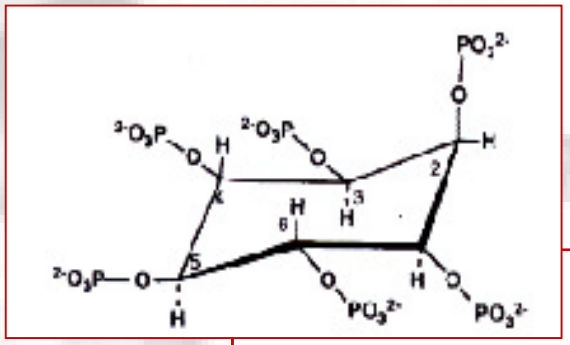
Sandberg et al-*J Nut* 48:185, 1982; Sandström and Sandberg-*J Tr El Elec Heal Dis* 6:99, 1992; Sandberg et al-*Am J Clin Nut* 70:240, 1999; Kennefick and Cashman-*Int J Food Sci Nut* 51:45, 2000; Lopez et al-*J Agric Food Chem* 49:2657, 2001; Haros et al-*J Agric Food Chem* 49:5450, 2001; Haros et *al-Eur Food Res Tech* 213:317, 2001; Hurrell et al-*Am J Clin Nut* 77:1213, 2003; Hurrell *Int J Vit Nut Res* 74:445, 2004; Sanz Penella et al.-*J Agric Food Chem* 57:10239, 2009

STRATEGIES FOR REDUCING OR ELIMINATING PHYTATES FROM FOOD AND/OR FEED INCLUDE:

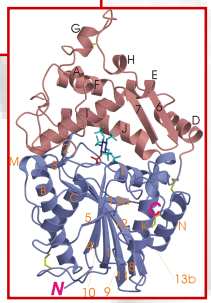
- Changes in food production processes (time, temperature, pH, etc)
- Selection of varieties with high phytase activity or low phytate content
- Genetic engineering (overexpression of plant phytase)
- Changes in agricultural conditions (optimization of fertilization)
- Addition of exogenous phytases (bacterial or fungal)
- Inclusion of phytate-degrading microorganisms

Faridi et al., 1983; Türk et al., 1996; Lopez et al., 2000; Sanz Penella et al., 2007; Sanz Penella et al., 2008; Palacios et al., 2008

PHYTIC ACID AND PHYTASE



PHYTASE
(phosphomonoesterase)
capable of hydrolyzing phytate (InsP_6) to free inorganic phophate (Pi), and lower *myo*-inositol phosphate esters (InsP_5-InsP_1)



InsP_6 —Pi→ InsP_5 —Pi→ InsP_4 —Pi→ InsP_3 —Pi→ InsP_2 —Pi→ InsP_1 —Pi→ *Myo*-Inositol

As the numbers of phosphate groups are progressively removed from phytate, te mineral binding strength decreases and solublity increases, forming weaker mineral complexes

Shears-*Biochim Biophys Acta* 1436: 49, 1998; Sandberg et al-*Am J Clin Nut* 70:240, 1999; Kennefick and Cashman-*Int J Food Sci Nut* 51:45, 2000; Hurrell et al-*Am J Clin Nut* 77:1213, 2003; Hurrell *Int J Vit Nut Res* 74:445, 2004; Shi et al-*Subcel Biochem* 39:265, 2006

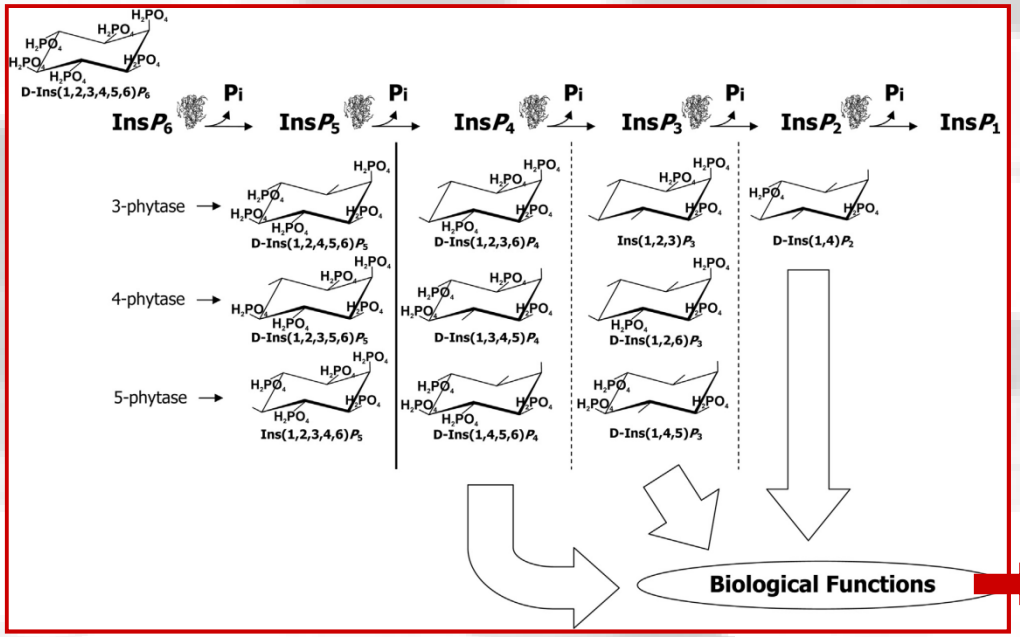


Figure. Sequence of phytic acid hydrolysis and generation of lower *myo*-inositol phosphate esters
Sanz-Penella and Haros, 2014

Anti-inflammatory functions
Secondary messengers

Three types of phytase are known, depending on the position on the inositol ring at which the initial attack of a phosphoester bond takes place:

❖ **3-phytase** (EC 3.1.3.8) (phytate hydrolysis starts at the D3 position of the *myo*-inositol ring) or **1-phytase** (name based on 1L-numbering system rather than 1D-numbering);

❖ **4-phytase** (EC 3.1.3.26) (phytate hydrolysis starts at D4 position of the *myo*-inositol ring) or **6-phytase** (name based on 1L-numbering system rather than 1D-numbering);

❖ **5-phytase** (EC 3.1.3.72) (phytate hydrolysis starts at D5 position of the *myo*-inositol ring).

In general, depending on the type of phytase, the first hydrolyzed phosphate group of the *myo*-inositol ring can vary in position, which could generate different positional isomers of InsP_5. Subsequently, the hydrolysis will continue on another specific phosphate group of each enzyme, and so on, generating a particular hydrolytic profile for each enzyme.

Sanz-Penella and Haros, *Whole grain and phytate-degrading human bifidobacteria.* In: Wheat and rice in disease prevention and health. Eds. Ronalds R. Watson, Victor R. Preedy, Sherma Zibadi. ISBN: 9780124017160, Elsevier **2014**

Microbiology Laboratory

Institute of Animal Reproduction and Food Research of the Polish Academy of Sciences

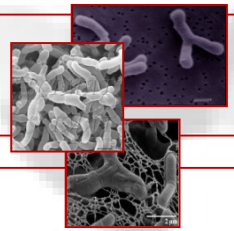
Olsztyn - Poland



CENEXFOOD

EU Centre of Excellence for Knowledge Transfer and Education in Food and Health

Beneficial effects of *Bifidobacteria*

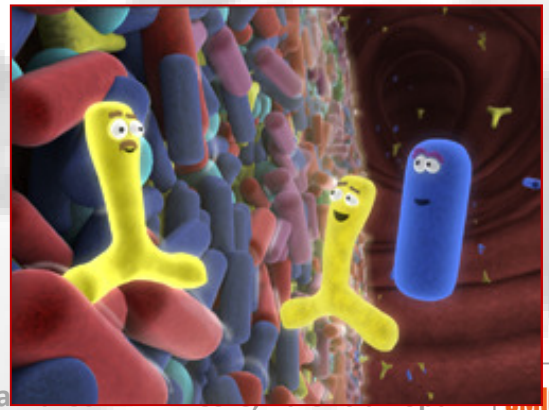


- Improvement of intestinal microbiota
- Protection against intestinal infections by different mechanisms:
  - Production of short chain fatty acids (specially lactic and acetic acids in the relations 2:3), succinic and formic acids.
  - Reduction of colonic pH
  - Production of antimicrobial substances
  - Reduction of the formation of putrefactive compounds like ammonia, phenols, indole, secondary bile acids.
  - Prevent the colonisation of the intestine by invading pathogens by competition for nutrients and attachment sites to the epithelial surfaces
- Prevention of constipation by stimulating intestinal motility for producing SCFA
- Reduction of serum cholesterol
- Increase the mineral absorption, specially a significant increase of Calcium and Magnesium absorption. The reduction of the colon pH explains in part the mechanism of absorption, but it is not clear yet.
- Anti colon cancer effect by reduction of intestinal transit time could contribute to cancer prevention and due to immunity enhancements
- Production of B-vitamins, B1, B2, B6, B12, nicotinic acid and folic acid

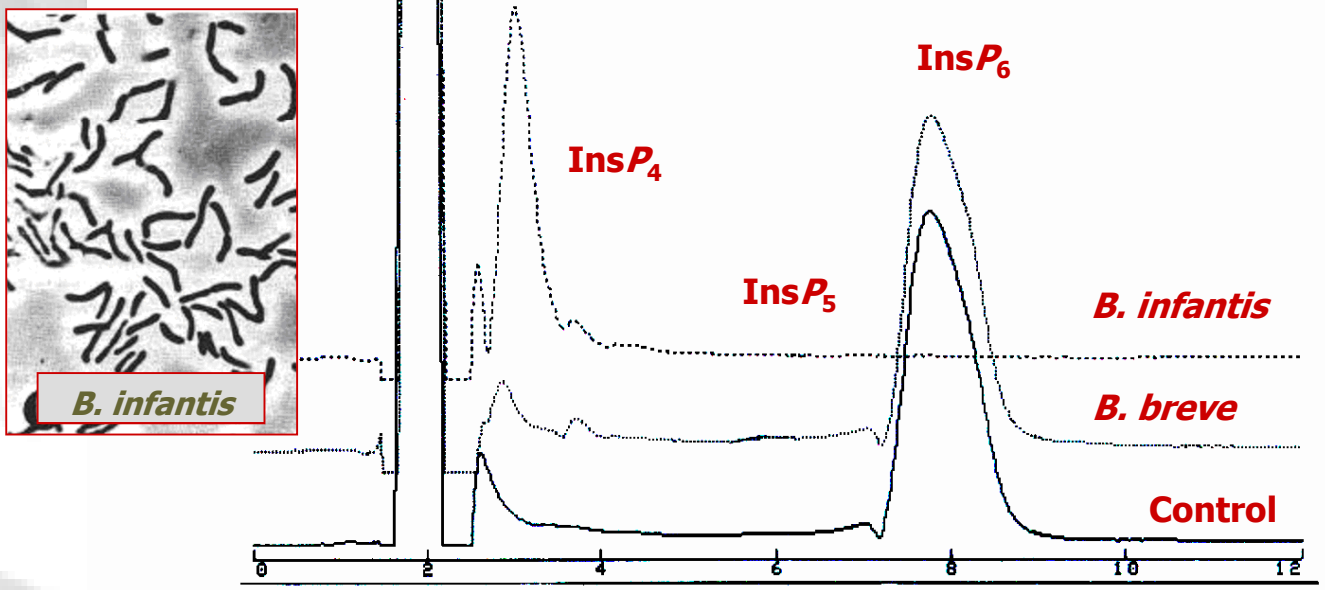
GRAS/QPS (Generally Regarder as Safe/Qualified Presumption of Safety)







Phytate degradation by *Bifidobacterium infantis*

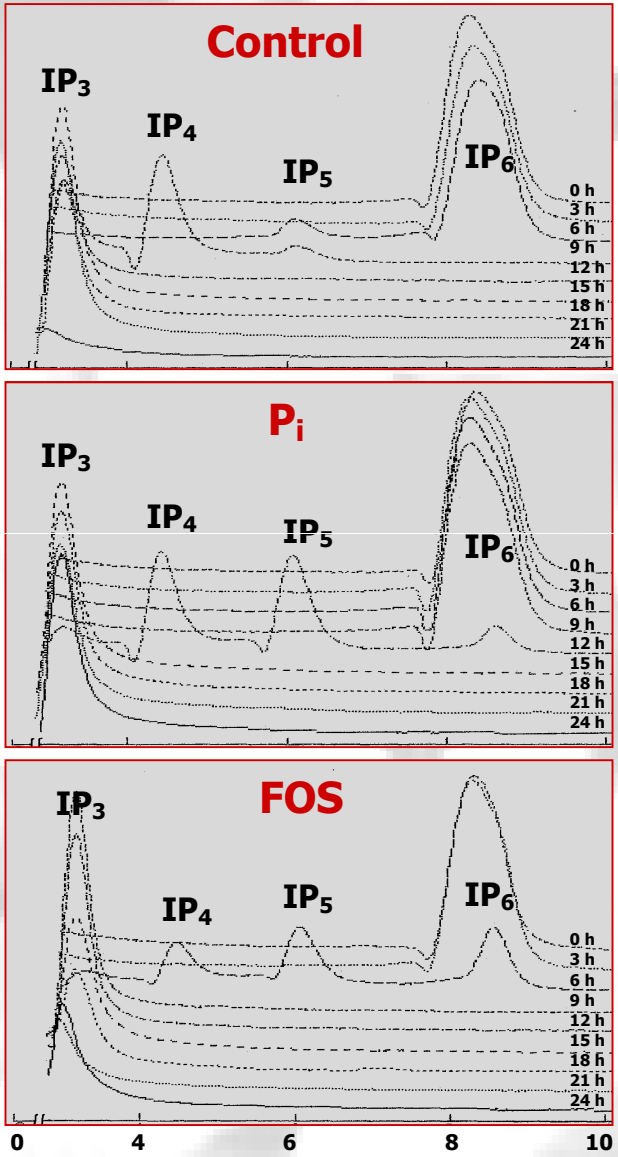


InsP_6: *myo*-inositol hexakisphosphate, **InsP_5**: *myo*-inositol pentakisphosphate, **InsP_4**: *myo*-inositol tetrakisphosphate, **InsP_3**: *myo*-inositol triphosphate

HPLC Method: mobile fase methanol/formic acid/tetrabutylamonium hydroxide; C-18 Chromasil column and refractometric detector

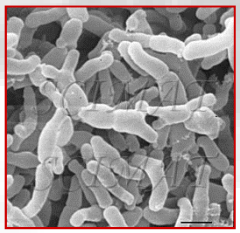
Haros et al-*FEMS Microbiol Lett* 247:231, 2005; Haros et al.-*Int J Food Microbiol* 117:76, 2007

Phytate degradation by *Bifidobacterium pseudocatenulatum*



Haros et al.-*Int J Food Microbiol* 117:76, 2007

FIGURE. Phytic acid degradation during *B. pseudocatenulatum* growth



Control: standard culture medium

P_i: standard culture medium with inorganic phosphate

FOS: standard culture medium replacing lactose by fructo-oligosaccharides (FOS)

$InsP_6$: phytic acid or hexakisphosphate of *myo*-inositol
$InsP_5$: pentakisphosphate of *myo*-inositol
$InsP_4$: tetrakisphosphate of *myo*-inositol
$InsP_3$: triphosphate of *myo*-inositol

Mobile phase: methanol/formic acid/tetrabutylamonium hydroxide; C-18 Chromasil column and refractometric detector

Food Science Division

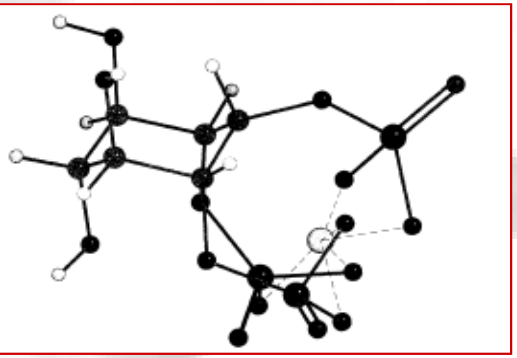
Department of Chemistry and Bioscience

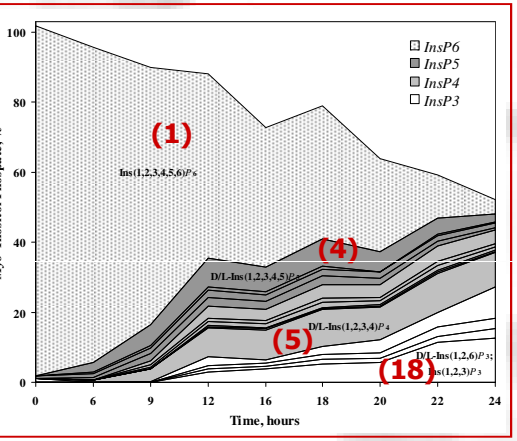


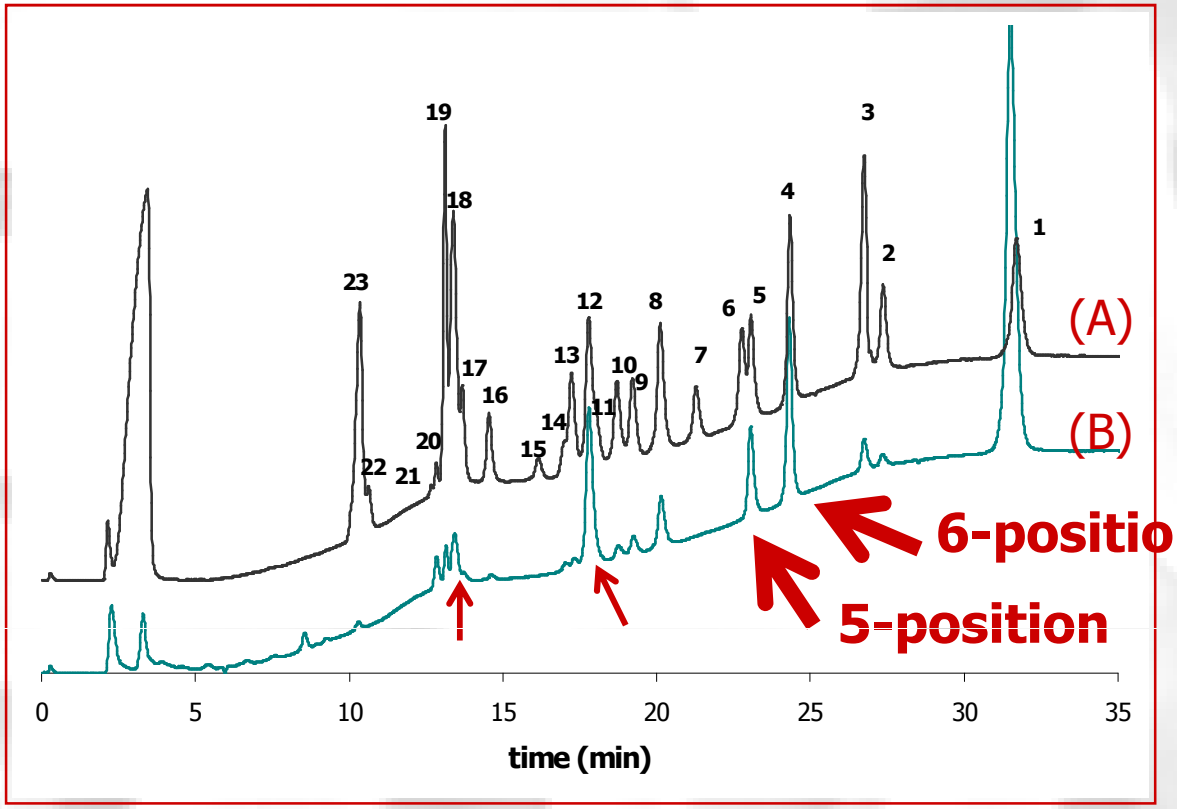
Chalmers University

Gothenburg - Sweden

InsP_6 hydrolysis by *B. pseudocatenulatum* phytase



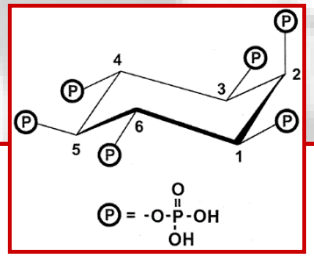


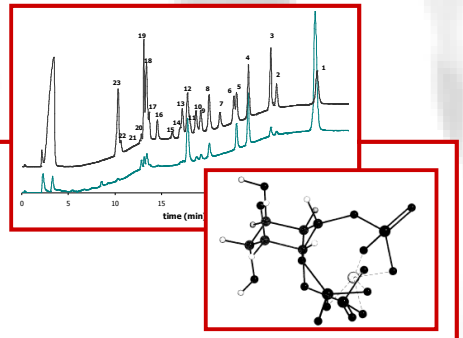


HPIC analysis of hydrolysis products of InsP_6 **(A)** InsP_6 hydrolysed by HCl, reference sample; **(B)** InsP_6 hydrolysis by *B. pseudocatenulatum* phytase, incubation time: 24 h. Unidentified peaks are assigned a star (*). Peaks Peaks: (1) Ins(1,2,3,4,5,6)P_6; (2) Ins(1,3,4,5,6)P_5; (3) D/L-Ins(1,2,4,5,6)P_5; (4) D/L-Ins(1,2,3,4,5)P_5; (5) Ins(1,2,3,4,6)P_5; (6) D/L-Ins(1,4,5,6)P_4; (7) Ins(2,4,5,6)P_4; (8) D/L-Ins(1,2,5,6)P_4; (9) D/L-Ins(1,3,4,5)P_4; (10) D/L-Ins(1,2,4,5)P_4; (11) Ins(1,3,4,6)P_4; (12) D/L-Ins(1,2,3,4)P_4; (13) D/L-Ins(1,2,4,6)P_4; (14) Ins(1,2,3,5)P_4; (15) Ins(4,5,6)P_3; (16) D/L-Ins(1,5,6)P_3; (17) D/L-Ins(1,4,5)P_3; (18) D/L-Ins(1,2,6)P_3, Ins(1,2,3)P_3; (19) D/L-Ins(1,2,4)P_3, Ins(1,3,4)P_3; (20)*; (21)*; (22) D/L-Ins(2,4)P_2; (23) D/L-Ins(1,2)P_2, D/L-Ins(4,5)P_2; Ins(2,5)P_2; D/L-Ins(1,4)P_2; D/L-Ins(1,6)P_2.
Mobile phase 5-95% HCl 1.0 M, HPIC PA-100 analytical column-Dionex, UV detection 290 nm

Haros et al-*Int J Food Microbiol* 135:7, 2009

Possible Pathway of InsP_6 hydrolysis





Ins (1,2,3,4,5,6)P_6

Ins(1,2,3,4,6)P_5 DL-Ins(1,2,3,4,5)P_5

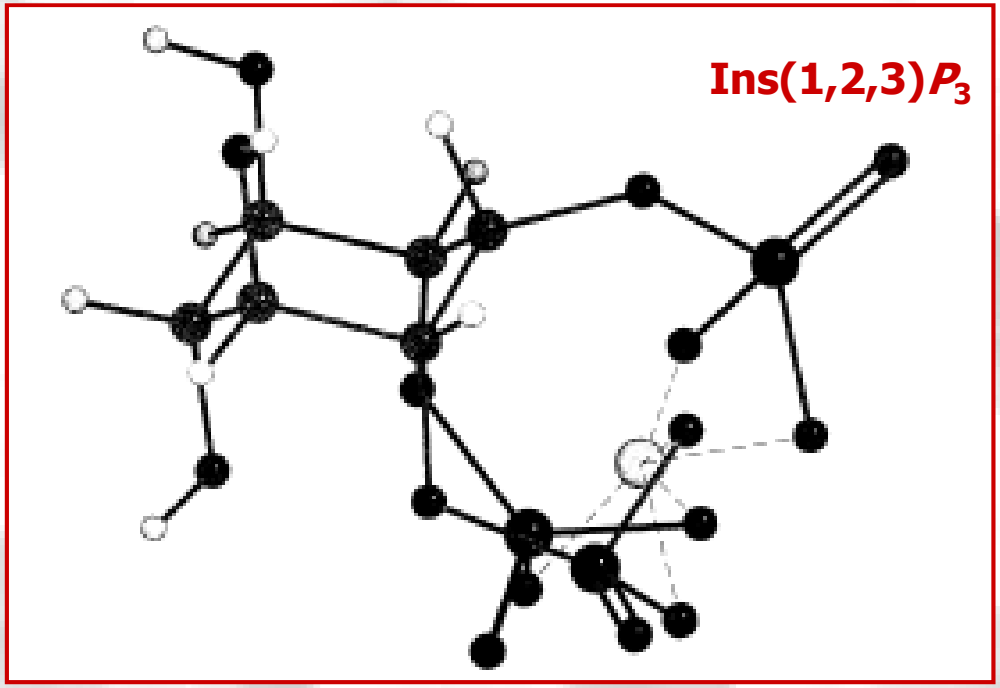
DL-Ins(1,2,3,4)P_4 DL-Ins(1,2,5,6)P_4

DL-Ins(1,2,4)P_3 Ins(1,2,3)P_3 Ins(1,3,4)P_3 DL-Ins(1,2,6)P_3

FIGURE. Possible pathway of phytate hydrolysis by the phytase from *B. pseudocatenulatum*

Haros et al-*Int J Food Microbiol* 135:7, 2009

Myo-inositol 1,2,3-triphosphate has been reported to be present in normal mammalian cells at concentrations of 1-10 mM, which it was suggested that this compound may have a role in iron chelation in the cell



- Has a configuration that uniquely provides a specific interaction with iron
- Increases its solubility
- Inhibits its ability to catalyse hydroxyl radical formation
- Has a role in iron chelation as an intracellular iron transport agent and/or cellular antioxidant
- Is potential candidate for enhancing mineral absorption

Hawkins et al-*Biochem J* 294:929, 1993; Barker et al-*Biochem J* 306:557, 1995; Phillippy and Graf-*Free Radical Biol Med* 22:939, 1997; Shen et al-*J Nut Biochem* 9:298, 1998; Dozol et al-*J Mol Struct* 643:171, 2002; Veiga et al-*J Biol Inorg Chem* 14:51, 2009

Cereal Group

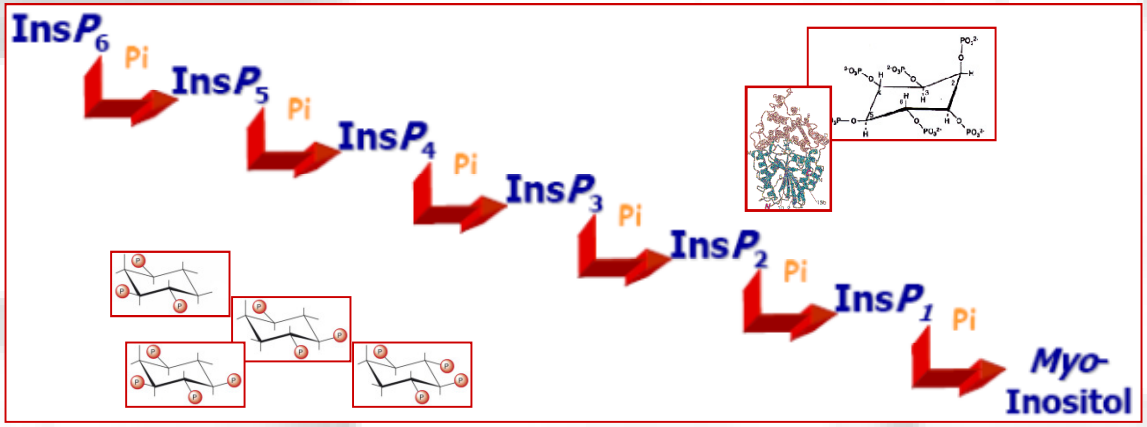
Institute of Agrochemistry and Food Technology

Valencia - SPAIN



Enzymatic phytate degradation in doughs depends on many factors:

- Fermentation time
- Temperature
- pH
- Water activity
- Flour extraction rate
- Starter culture
- Mineral content
- Aditives
- Bread-making process



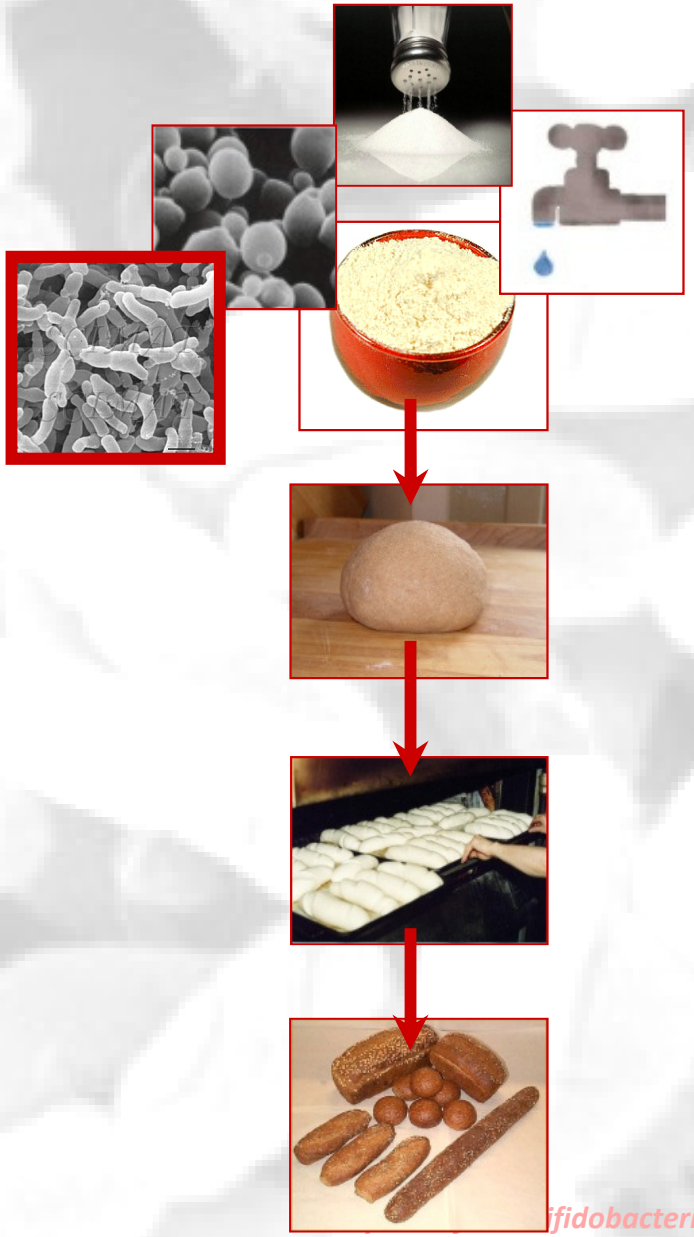
Türk and Sandberg-*J Cereal Sci* 15:281, 1992

The strategies to reduce or eliminate the phytate from food and/or feed include:

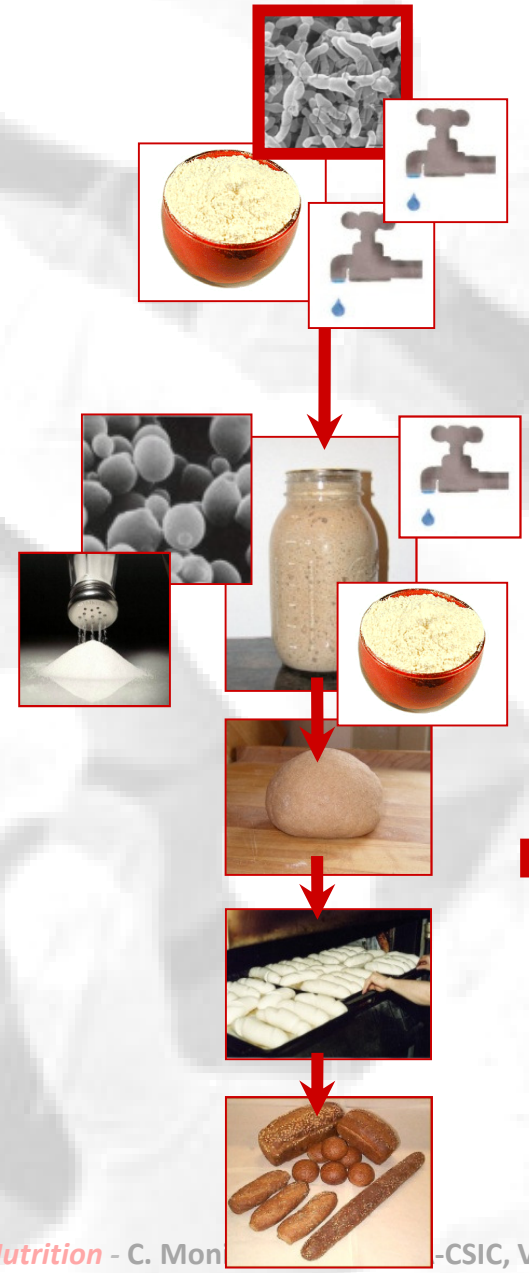
→ Addition of exogenous phytate-degrading enzymes
→ Genetic engineering
→ Agronomic conditions
→ Change the pH of the product
→ Food processes

Haros et al-*J Agric Food Chem* 49:5450, 2001; Haros et al-*Eur Food Res Tech* 213:317, 2001;
Sanz Penella et al-*J Cereal Sci* 48:715, 2008; Sanz Penella et al-*J Agric Food Chem* 57:10239, 2009

DIRECT PROCESS



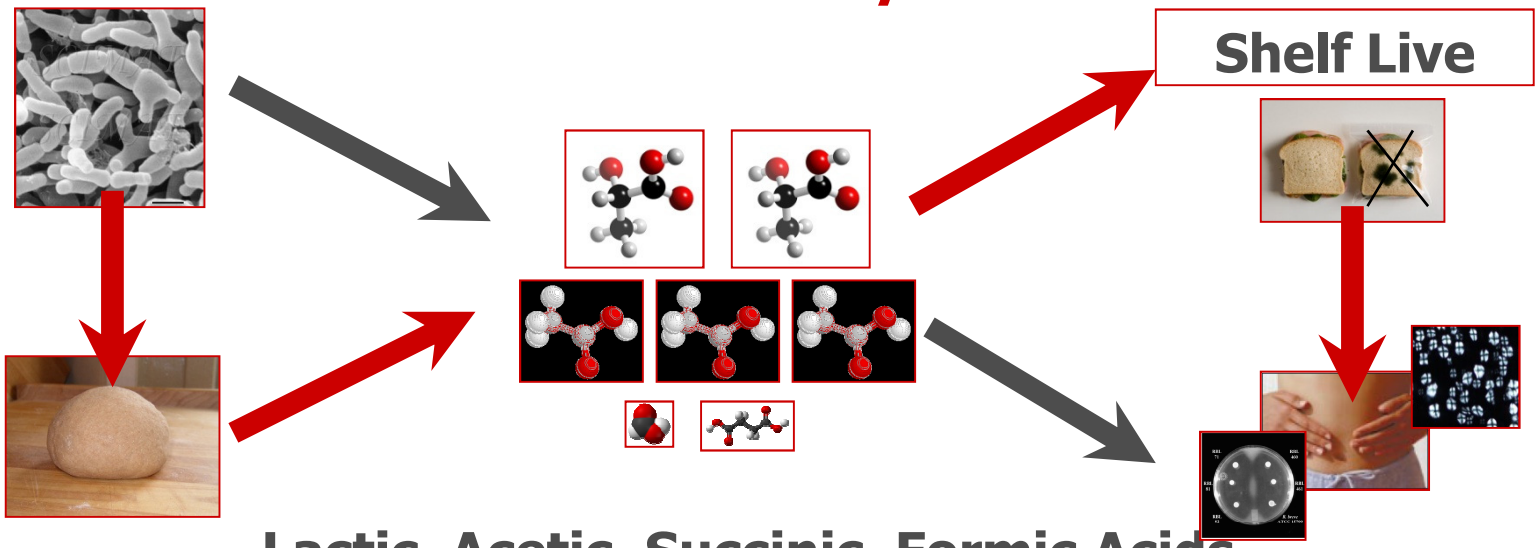
INDIRECT PROCESS



Sour Dough

Bread Making Process

Short Chain Fatty Acids



The whole wheat sourdough has great potential to modify the digestibility of starch by raising the lactic and acetic acid levels and increasing the mineral bioavailability. During sourdough fermentation, lactic acid bacteria produce metabolites and/or enzymes, which have been shown to have a positive effect on the texture, flavour and staling of bread

Östman et al-*J Cereal Sci* 42:300, 2005; Sanz Penella et al-*Proceeding of the International Conference on Cereals-their products and processing,* University of Debrecen, Hungary, 2008; Sanz Penella et *al-Food BioprocessTechnol*, 5:2370, 2012

Whole wheat sourdough bread

- **Control**
- **Sour Dough 5%**
- **Sour Dough 10%**
- **Sour Dough 15%**
- **Sour Dough 20%**

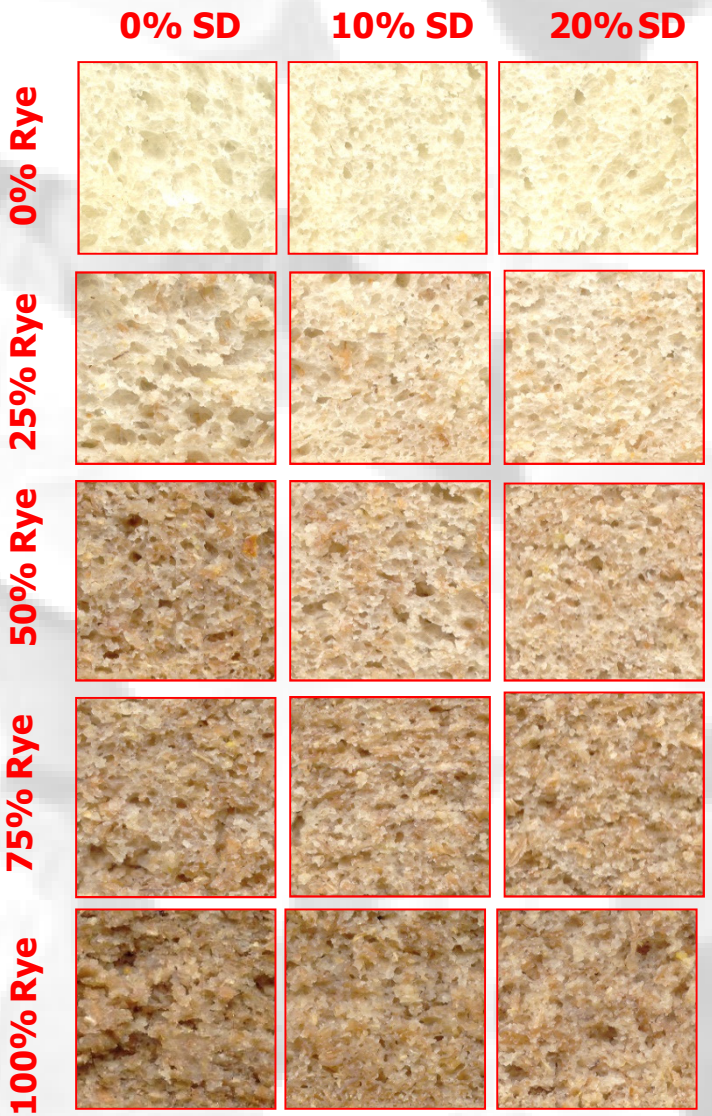
Sanz Penella et *al-Food Bioprocess Technol*, 5:2370, 2012



Bread inoculated with bifidobacteria had similar technological quality as the control sample in terms of: crumb and crust colour, crumb texture, crumb structure, specific bread volume, slice shape, and sensory analysis

SOUR DOUGH RYE BREAD





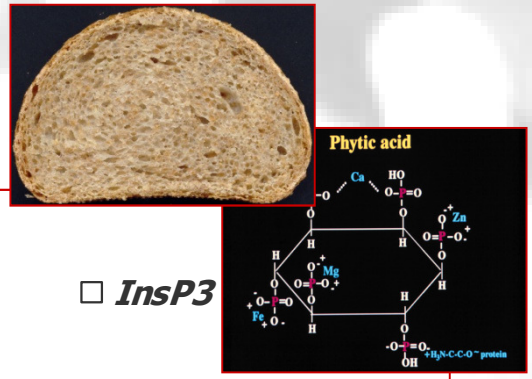
Sancho Robles et al. *Effect of sourdough fermentation with bifidobacteria on rye bread-making.* In: Proceedings of International Conference on Food Innovation FoodInnova, Nº13, pp 1-4 ISBN978-84-693-5010-2, 2010

SOUR DOUGH WHOLE RYE BREAD



Ruiz Arranz et al. *Effect of bifidobacterium starter on whole rye dough and bread quality.* In: Proceedings of international Conference on Food Innovation FoodInnova, Nº10, pp 1-4. ISBN978-84-693-5010-2, 2010

Myo-Inositol Phosphates - Sourdough bread



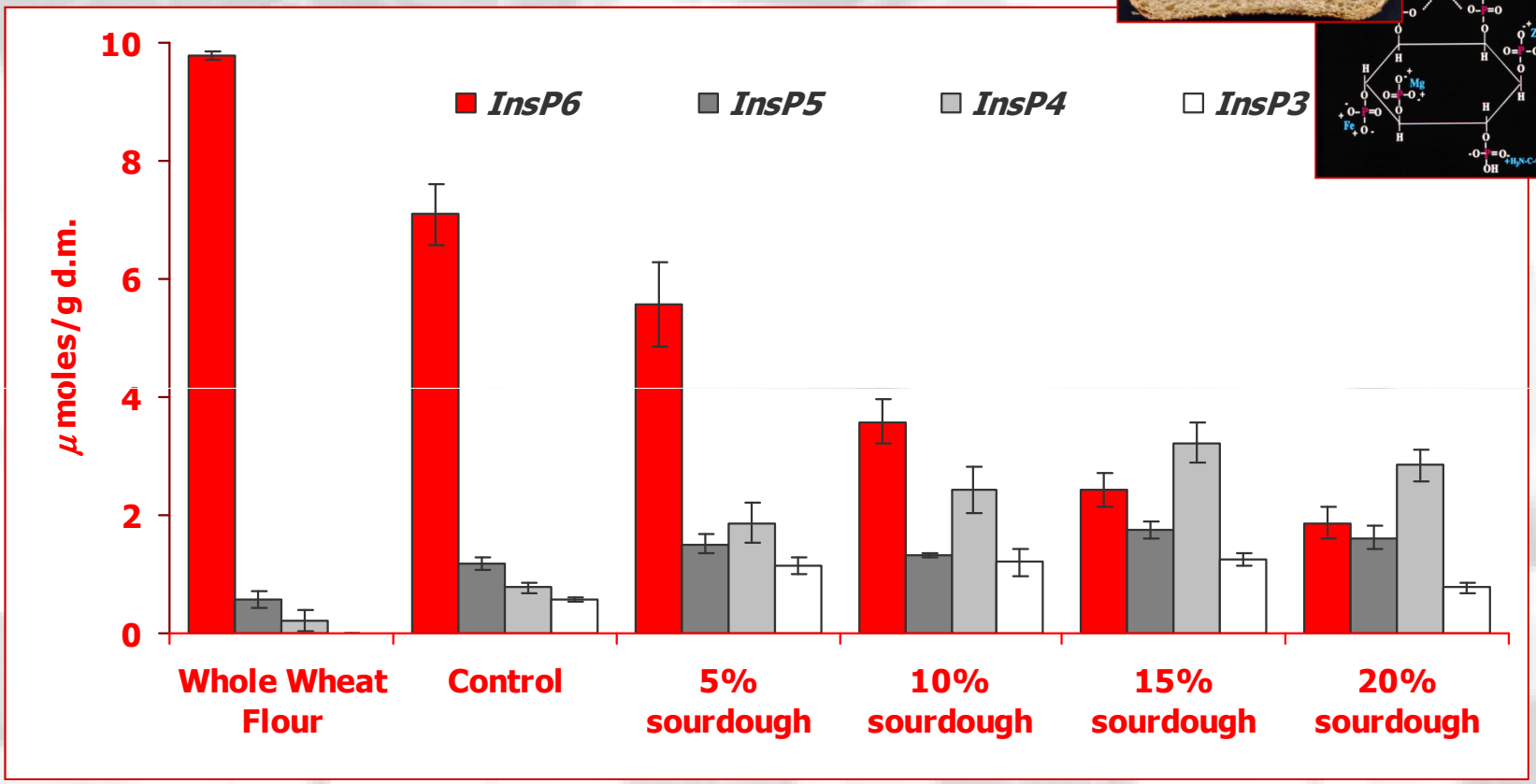


FIGURE. Effect of the sourdough concentration on *myo*-inositol phosphate levels in whole wheat sourbread

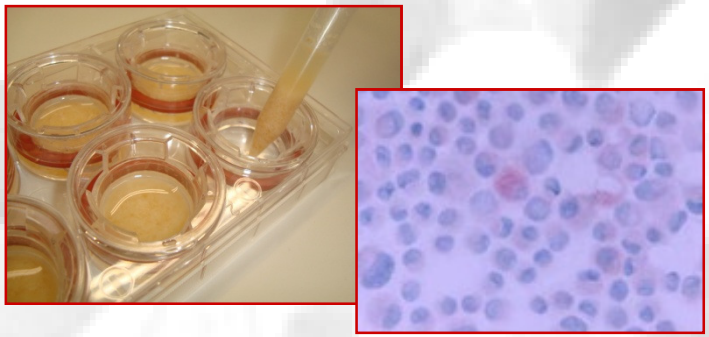
Sanz Penella et *al-Food BioprocessTechnol*, 5:2370, 2012

Bifidobacterium phytase-producer as starter in whole wheat sourdough bread

Table 1. Iron Dializability

Formulation	Iron dialyzable, μg/mL	Iron Dialyzability, %
Control	0.104±0.009a	2.2±0.2a
Fitasa	0.151±0.012a	4.0±0.3b
MM 10%	0.294±0.047b	6.2±0.9c
MM 20%	0.578±0.033d	10.8±0.6d
MM 20% - CA	0.398±0.064c	7.4±1.2c

Bread formulations: MM 10% y MM 20% are bread with 10 and 20% of sourdough inoculated with bifidobacteria, respectively. MM 20%-CA: control chemically acidified. Mean, n=3. Values followed by the same letter in the same column are not significant different ($p<0.05$).



Sanz-Penella et al., J Agric Food Chem, 60:3190, 2012

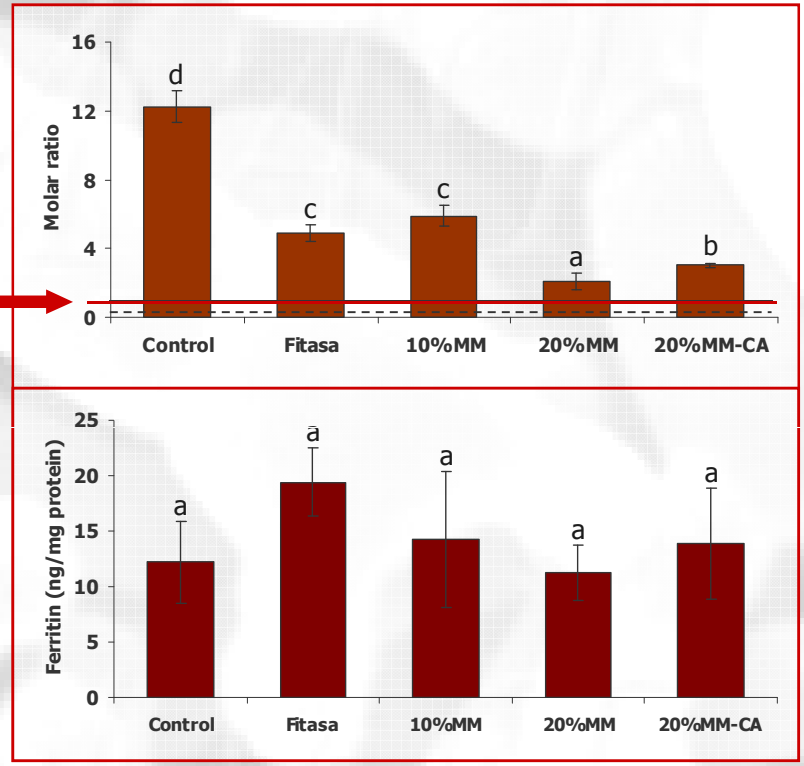
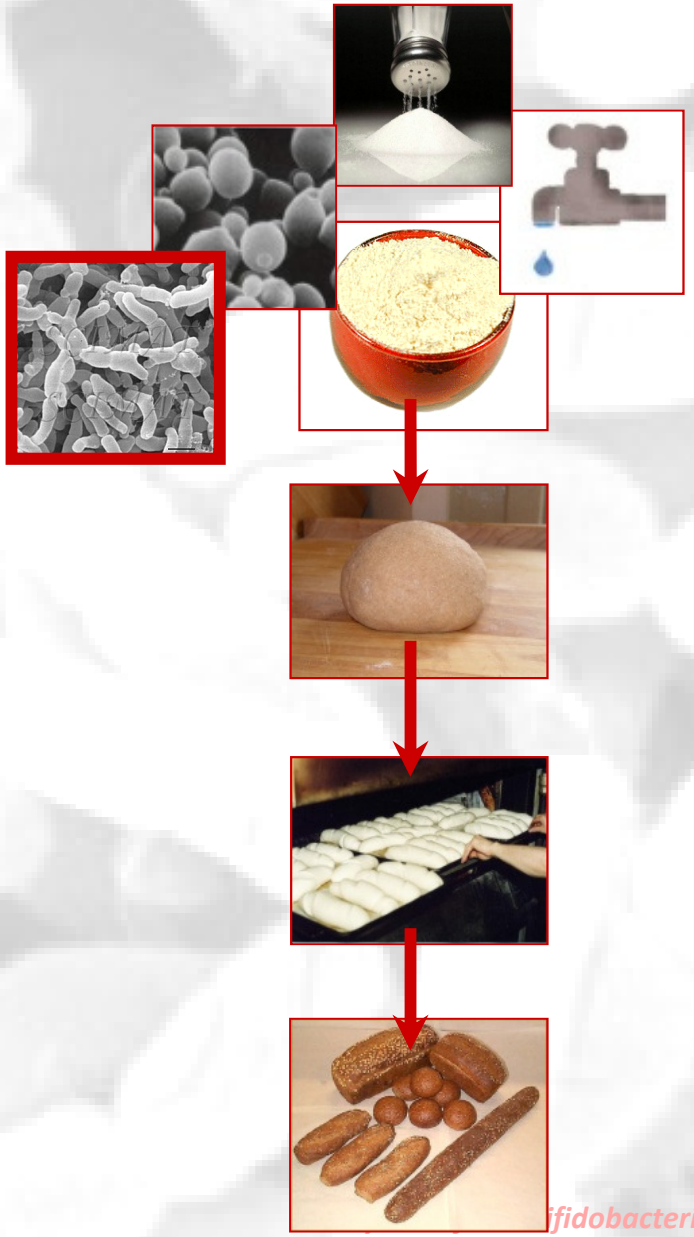
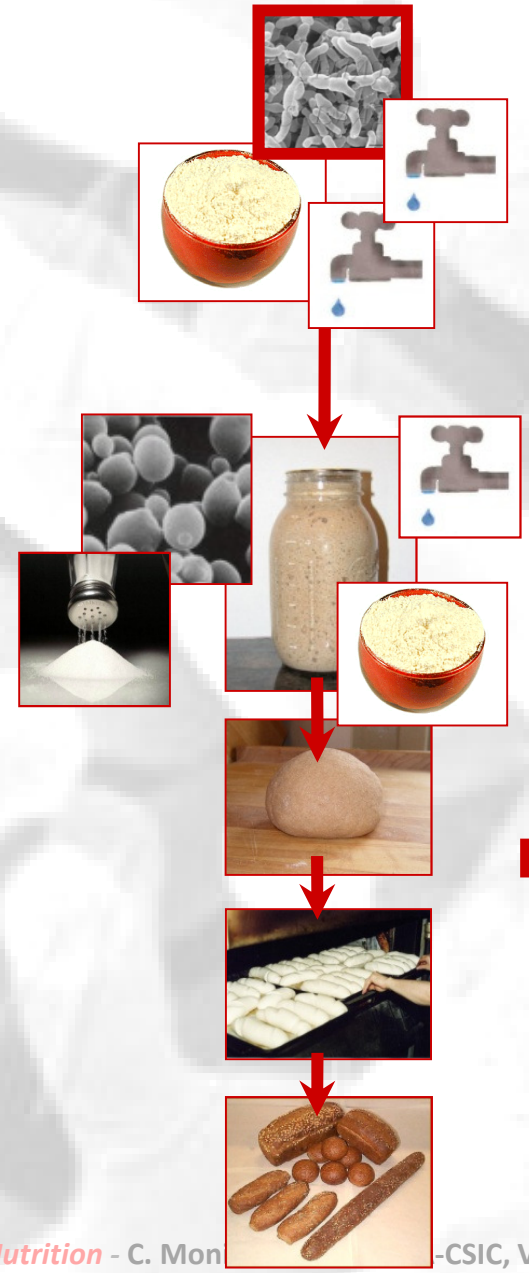


Figure 1. Effect of treatment on molar ratio InsP_6/Fe and ferritin formation in Caco-2 cells exposed to gastrointestinal digestion of loaves. Bars with the same letter are not significantly different ($p<0.05$).

DIRECT PROCESS



INDIRECT PROCESS



Sour Dough

Bread Making Process

100% Whole Wheat Bread – Direct Process

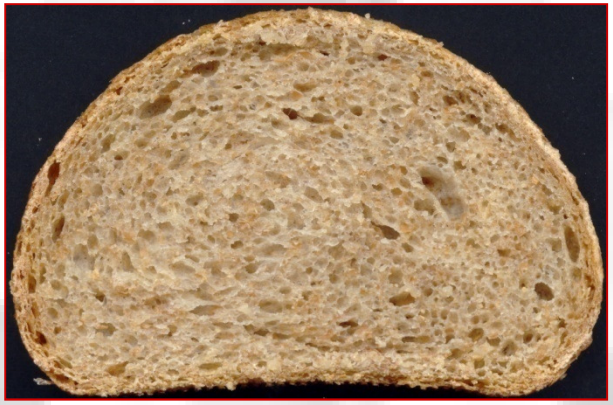
- Control





- Fungal Phytase





- Bifidobacteria





Sanz Penella et al -*J Agric Food Chem* 57:10239, 2009

Bread inoculated with bifidobacteria had similar technological quality as the control sample or bread with fungal phytase in terms of: crumb and crust colour, crumb texture, crumb structure, specific bread volume, slice shape, and sensory analysis, flavour

Phytates - InsP_6

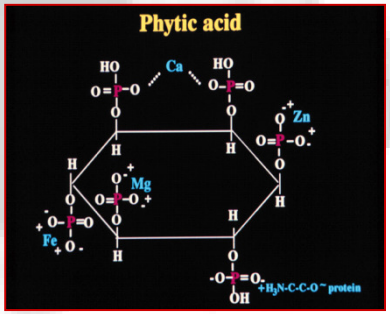
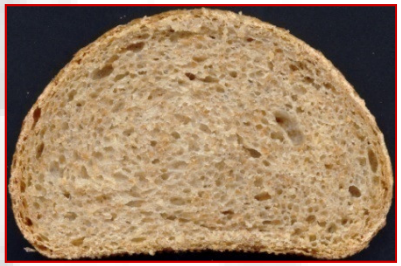
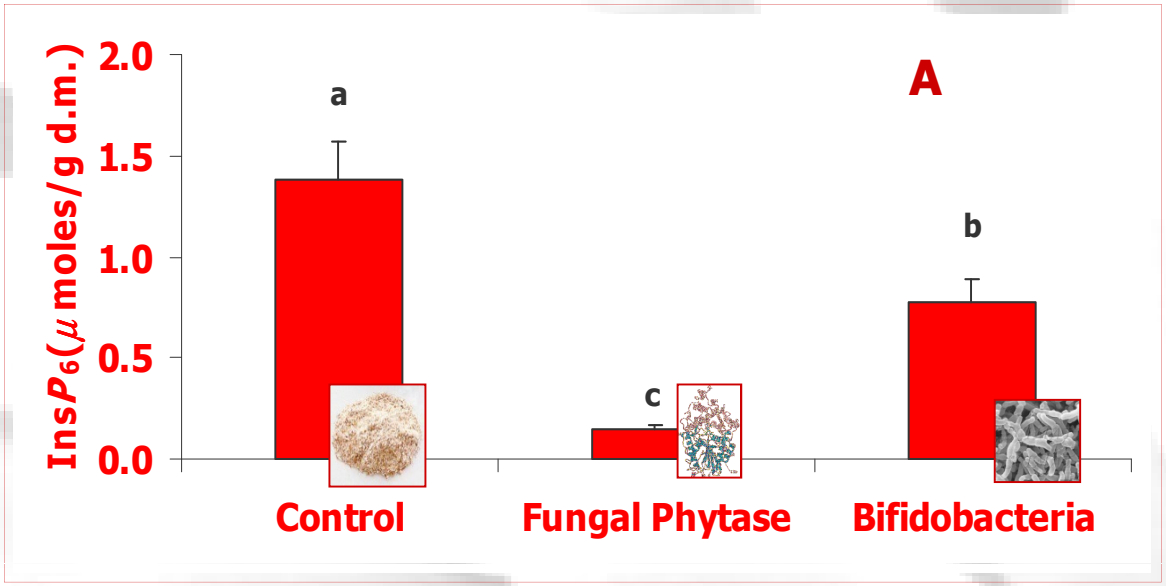


FIGURE. Residual level of phytate in bread. **A.** Whole wheat bread, **B.** Bran-Enriched Bread



Sanz Penella et al -*J Agric Food Chem* 57:10239, 2009



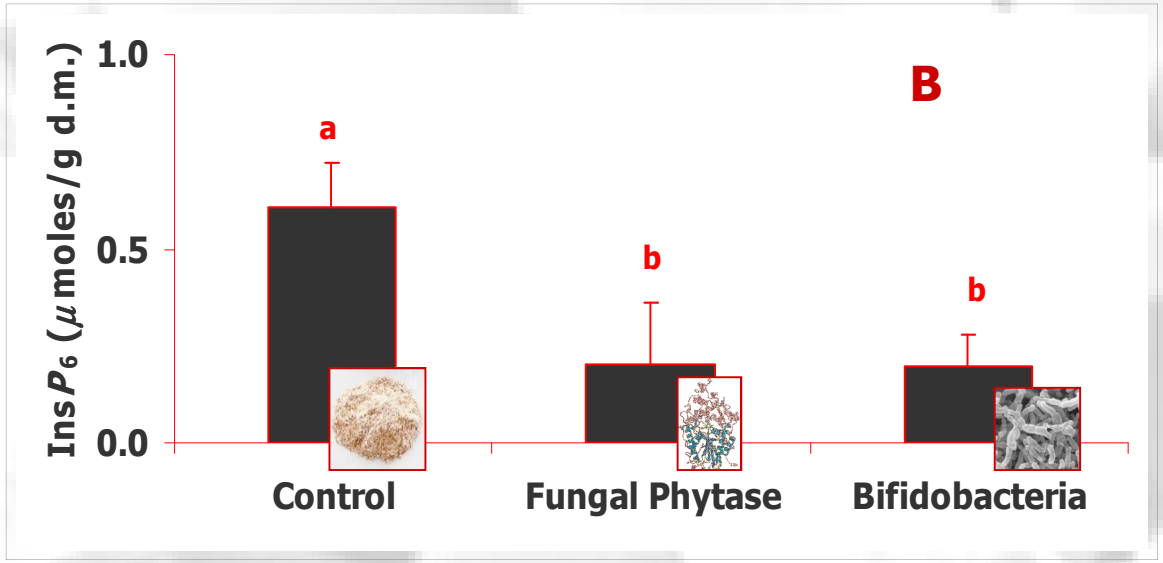


Table. *Myo*-inositol phosphate concentration in final product[a,b]

% Whole wheat flour	Treatment	μmoles/ g bread (d.m.)[c]				
		InsP_6	InsP_5	InsP_4	InsP_3	InsP_6+ InsP_5
100	C	1.383 a	0.797 a	2.255 a	1.472 a	2.180 a
	Phy	0.150 c	0.024 b	0.009 b	0.056 b	0.173 c
	Bif	0.770 b	0.712 a	2.384 a	1.594 a	1.482 b
50	C	0.607 a	0.260 a	0.845 a	1.269 a	0.867 a
	Phy	0.203 b	0.040 c	0.023 b	0.050 b	0.242 b
	Bif	0.199 b	0.145 b	0.850 a	1.310 a	0.345 b

[a]Control (C), fungal phytase addition (Phy) and *Bifidobacterium* addition (Bif).
[b]Mean, n=3. Values followed by the same letter in the same column are not statistically different at 95% confidence level (p<0.05). The statistical analysis of the different flours was made separately.
[c]InsP_3 to InsP_6: *myo*-inositol phosphate containing 3-6 phosphates per inositol residue

Sanz Penella et al-*J Agric Food Chem* 57:10239, 2009

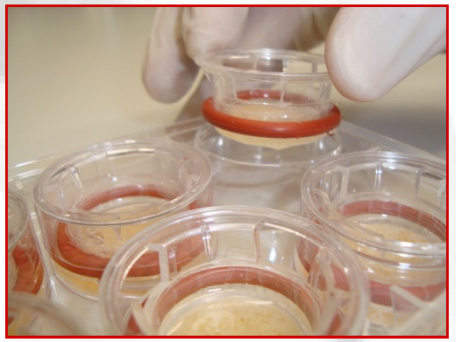
The inclusion of commercial fungal phytase in bread formulation significantly reduced the amount of phytate after one hour of fermentation. However it also hydrolyzed efficiently the lower *myo*-inositol phosphates.

Bifidobacterium phytase-producer as starter in whole wheat bread-making

Table. Effect of treatment in the iron dializability

% whole wheat flour	Treatment	Iron dialyzable, μg/mL	Iron Dialyzability , %
100	Control	0,107a	2,2a
	Fungal Phytase	0,125a	2.0a
	Bifidobacterum Phytase	0,243b	4.8b

Mean, n=3. Values followed by the same letter in the same column are not significant different ($p<0.05$).



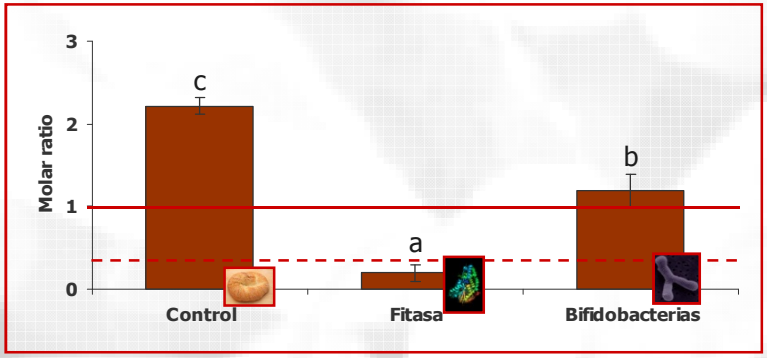


Figure 1. Effect of treatment on iron molar ratio (InsP_6/Fe). Bars with the same letter are not significantly different ($p<0.05$).

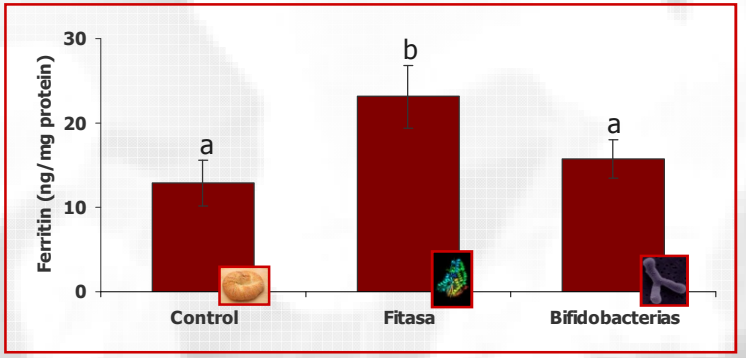


Figure 2. Ferritin in Caco-2 cells exposed to gastrointestinal digestion of loaves. Bars with the same letter are not significantly different ($p<0.05$).

Sanz-Penella et al., J Agric Food Chem, 60:3190, 2012

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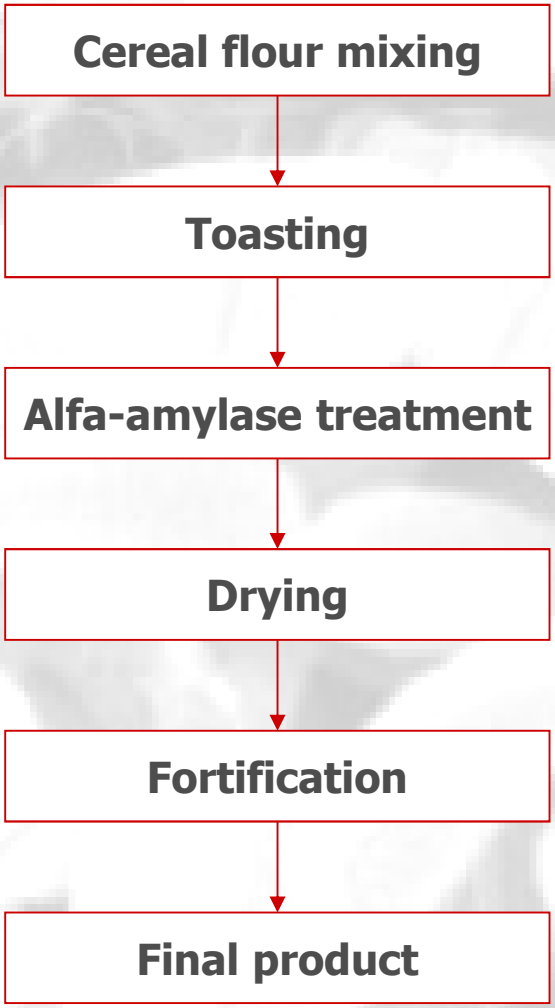
Valencia - SPAIN



Bifidobacterium Phytases Application in human nutrition

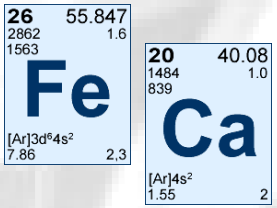
- **Infant cereals for porridge**
- **Breadmaking process**
- **Fermented soy milk with recombinant *lactobacilli***

Infant Cereal Process



Phytase addition

- 55ºC
- 20 minutes
- pH 5.5



Infant flours are commonly fortified with vitamins and minerals according to European legislation (Official Journal of Europen Union, Directive 2006/125/EEC; p. 16)

Sanz-Penella et *al.* - *J Agric Food Chem*, 60: 11787, 2012

Infant Cereal for Porridge

Table 3. Effect of phytases from different source on the phytate hydrolysis after treatment on cereal products

Sample of cereals	Phytate % Hydrolysis	InsP_6 μmoles/g d.b.
MC-Control (without phytase)	50.0	1.16c
MC-Commercial PhytaseA	83.6	0.38b
MC-*Bifidobacterium longum* phytase	90.1	0.23ab
MC-*Bifidobacterium pseudocatenulatum* phytase	93.6	0.15a
GF-Control (without phytase)	57.0	1.10c
GF-Commercial PhytaseA	68.4	0.61b
GF-*Bifidobacterium longum* phytase	86.0	0.27a
GF-*Bibidobacterium pseudocatenulatum* phytase	91.7	0.16a

Mean, n=3; values followed by the same letter in the same column are not significantly different at 95% confidence level. MC: multicereal sample; GF: gluten free sample; d.b.: dry basis.

Sanz-Penella et *al.* - *J Agric Food Chem*, 60: 11787, 2012

Bread with whole amaranth flour and phytases



		Fungal commercial Phytase	*Bifidobacterium pseudocatenulatum* Phytase	*Bifidobacterium longum* sp. *infantis* Phytase
Control Bread	HBC			
Whole Wheat Bread	HIC	HI FC	HIPS	HI IN
25% whole amaranth flour	HA 25C	HA25FC	HA 25 PS	HA 25 IN
50% whole amaranth flour	HA 50C	HA50FC	HA50PS	HA 50 IN

García-Mantrana et al., EFRT 238:853 2014

Bread with whole amaranth flour and phytases



		Fungal commercial Phytase	*Bifidobacterium pseudocatenulatum* Phytase	*Bifidobacterium longum* sp. *infantis* Phytase
Control Bread	control HB			
Whole Wheat Bread	control HI	HI FCOM	HI PS	HI IN
25% whole amaranth flour	control HA25	HA25 FCOM	HA25 PS	HA25 IN
50% whole amaranth flour	control HA50	HA50 FCOM	HA50 PS	HA50 IN

García-Mantrana et al., EFRT 238:853 2014

Effect of formutation on *myo*-inositol phosphates levels in breads

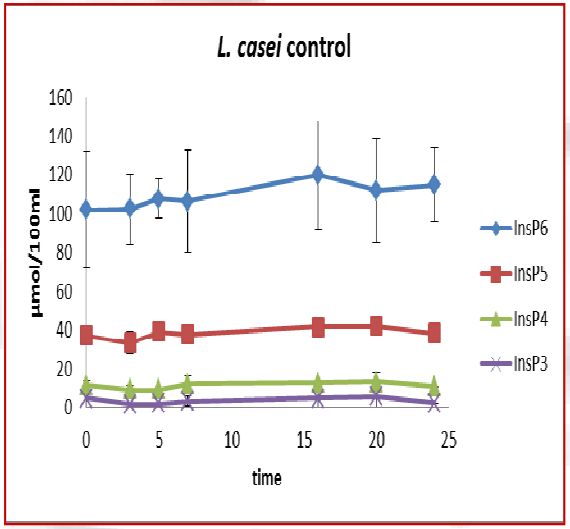
Sample		μmoles/g de pan d.m.				
	Phytase	InsP_6	InsP_5	InsP_4	InsP_3	InsP_6+InsP_5
HBC	---	n.d.	n.d.	n.d.	n.d.	n.d.
HIC	Control	2.3±0.5e	0.6±0.1e	0.3±0.1d	0.5±0.1f	2.9±0.7c
	Commercial	0.6±0.4ab	0.1±0.1ab	0.1±0.1a	0.1±0.1a	0.7±0.7a
	B. pseudocatenulatum	0.3±0.1a	0.1±0.1ab	0.1±0.1b	0.3±0.1e	0.5±0.1a
	B. longum	0.3±0.2a	0.1±0.1a	0.1±0.1ab	0.1±0.1a	0.4±0.3a
HA25	Control	1.8±0.3cd	0.3±0.1bcd	0.1±0.1b	0.2±0.1de	2.1±0.4b
	Commercial	1.3±0.1bc	0.2±0.1abc	0.1±0.1ab	0.1±0.1abc	1.5±0.2b
	B. pseudocatenulatum	0.2±0.1a	0.1±0.1a	0.1±0.0a	0.1±0.0ab	0.2±0.1a
	B. longum	0.2±0.1a	0.1±0.0a	0.1±0.0a	0.1±0.1abc	0.3±0.1a
HA50	Control	6.8±0.2g	1.0±0.2f	0.2±0.1c	0.2±0.1bcde	7.8±0.5e
	Commercial	3.4±0.6f	0.4±0.1cde	0.1±0.0ab	0.1±0.1abc	3.7±0.6d
	B. pseudocatenulatum	2.0±0.1de	0.3±0.1bcd	0.1±0.1ab	0.2±0.1abcd	2.4±0.0bc
	B. longum	3.5±0.3f	0.5±0.1de	0.1±0.0ab	0.2±0.1cde	3.9±0.4d

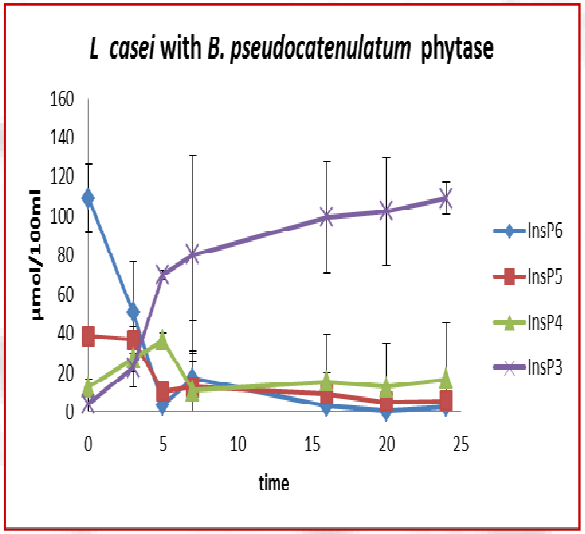
[a-g]Mean± SD, n=3. Values followed by the same letter in the same column are not significantly different at 95% confidence level. HBC: 100% wheat flour HIC: 100% whole wheat flour HA25: 25% whole amaranth flour/75% wheat flour HA50: 50% whole amaranth flour/50% wheat flour; n.d.: not detected; d.m.: dry matter; Ins$P_{6\text{-}3}$: hexakis, pentakis, tetrakis and tri phosphate of *myo*-inositol, respectively. García-Mantrana et *al.*, *Eur Food Res Technol.*, 238:853, 2014

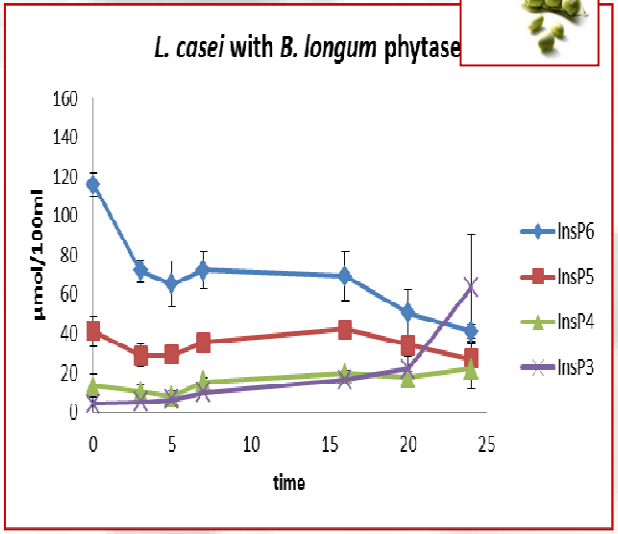
Effect recombinant strains of *L. casei* expressing bifidobacterial phytases on *myo*-inositol phosphates contents during soya-milk fermentation

Cloning and expression of bifidobacterial phytase genes in *Lactobacillus casei*. The phytase genes from *Bifidobacterium pseudocatenulatum* and *Bifidobacterium longum* spp *infantis* were cloned in the vector pNG8048e and transformed into *L. casei.* In this vector, expression of the cloned genes is under the control of a nisin-inducible promoter (NICE system).









Fermentation by recombinant strains of *L. casei* expressing bifidobacterial phytases removed a substantial amount of InsP_6 and led to accumulation of lower *myo*-inositol phosphates (InsP_3), particularly in the strain expressing the *B. pseudocatenulatum* phytase. This investigation showed the ability of modified *L. casei* to produce enzymes with technological relevance in functional foods application. The ability of other species, as recombinant *Lactobacillus plantarum* strains, to reduce anti-nutrients in fermented plant-based food is also under evaluation.

García-Mantrana et *al.*, Nutritional improvement of plant-based foods by fermentation with modified *Lactobacillus casei* producing phytase, Barcelona - BIET13

CONCLUSIONS

- Phytases from *B. longum* subsp. *infantis* ATCC15697 and *B. pseudocatenulatum* ATCC27919 can be categorized into the class of histidine acid phosphatases, which are active at acidic pH and have a histidine on the active site

- The enzymes belong to a new subclass within the histidine acid phytases, are highly specific for the hydrolysis of phytate, and render *myo*-inositol triphosphate (InsP_3) as the final hydrolysis product

- They represent the first phytases characterized from this group of probiotic microorganisms (GRAS/QPS), opening the possibilities for their use in the processing of high-phytate-content products for human or animal nutrition

- The phytate-degrading activity constitutes a novel metabolic trait which could contribute to the improvement of mineral absorption in the intestine as a nutritional probiotic feature with potential trophic effect in human gut



Computer-assisted three-dimensional structure of *Bifidobacterium pseudocatenulatum* phytase

Thank for your attention