



Received for publication, October, 10, 2019

Accepted, December, 27, 2019

Original paper

INULIN: unique among the polyglucides with significant functional properties and biotechnological perspectives

SILVANA MIHAELA DĂNĂILĂ-GUIDEA¹, RADIANA MARIA TAMBA-BEREHOIU²,
CIPRIAN-NICOLAE POPA^{2*1}, RADU TOMA¹, LUMINIȚA VIȘAN¹, VASILICA SIMION¹

¹University of Agricultural Sciences and Veterinary Medicine Bucharest, Faculty of Biotechnology, Bucharest, Romania

²Milling, Bakery, Research & Consulting, Bragadiru City, Diamantului street., 8B, Ilfov District, Romania

Abstract

Inulins are plants reserve polyglucides with functional properties that depend significantly on the degree of polymerization and their molecular mass (polydispersity index). Most nutritionists recommend increasing dietary inulin levels due to an impressive number of potential therapeutic effects on the health of the gastrointestinal tract, especially colon, but do not exclude a number of systemic effects related to the prevention of certain cancers and to the reducing of inflammatory processes in the body. However, there are medical studies that correlate fructolysis in the small intestine (possible for oligofructans of up to 10 carbon atoms) with negative effects, characteristic of irritable bowel syndrome. The most common method of obtaining inulins is the extraction from chicory in a manner similar to sugar obtaining. Fermentative or enzymatic synthesis of inulin are also being developed, but they are more expensive. With the exception of sugar beet, most transgenic plants accumulate significantly lower amounts of fructans (10 mg/g) compared to plants that naturally produce and accumulate fructans (60-150 mg/g). We can recall a significant number of industrial applications of inulin, from the formulation of drugs used in colon disorders and medical tests for kidney evaluation to various uses in the food industry (dairy, bakery) or water treatment.

Keywords

Inulins, oligofructoses, biotechnologies, therapeutics and industrial applications.

To cite this article: DĂNĂILĂ-GUIDEA SM, TAMBA-BEREHOIU RM, POPA C-N, RADU TOMA, VIȘAN L, SIMION V. *INULIN: unique among the polyglucides with significant functional properties and biotechnological perspectives. Rom Biotechnol Lett.* 2020; 25(2): 1386-1395. DOI: 10.25083/rbl/25.2/1386.1395

✉ *Corresponding author: CIPRIAN-NICOLAE POPA, Milling, Bakery, Research & Consulting, Bragadiru City, Diamantului street., 8B, Ilfov District, Romania
E-mail: cipnpopa@yahoo.com

Introduction

Inulin is a generic term for various mixtures of fructose polymers synthesized in nature, as reserve substances (those of plant origin). But fructose polymers also have a role in protecting the microbial cell against abiotic stress factors, as well as in its ability to adhere to structures in the living environment. Inulin was discovered in 1804 by the German scientist Rose in the hot water extract of *Inula helenium*, but its name was established by Thomas in 1811 (CANTOR, 2008 [1]); PHILLIPS & al, 2009 [2]).

Fructose polymers are characterized by polymerization rates, generally ranging from 2 to 60, and represent besides starch, the second reserve polyglucid as spreading, in the vegetable kingdom (over 36,000 plants produce inulin) (CANTOR, 2008 [1]). Inulin can be found in a large number of bacteria and in several fungal species (BANGUELA & al, 2006 [3]). Human nutrition provides an amount of inulin, estimated at 1-11 g/day, but inulin can be tolerated up to 20 g/day (BONNEMA & al, 2010 [4]). The interest for inulin has been determined by its unique complex of functional, technological and nutritional properties, which can be capitalized in: food and feed industry, chemical, pharmaceutical industries, medical field.

This paper aims to make a review of the literature on one of the most interesting agronomic resources available: inulin. Thus, a large number of papers have been studied, on inulin frequency in nature, its obtaining, perspectives in producing inulin transgenic plants, fermentative or enzymatic processes of obtaining, its structure, its physico-chemical properties, its therapeutic effect and the sphere of the industrial applications.

Polyglucides natural sources

Frequency in nature. There are significant differences between types of fructans, determined by their sources (vegetal, bacterial or fungal). These differences relate to: the degree of polymerization, the presence of ramifications, the type of linkage between fructose units and the position of the glucose residue (BANGUELA & al, 2006 [3]). Differentiation of fructans is based on the type of linkage between two adjacent fructose molecules. Inulins contain in a large proportion 2-1 fructosyl-fructose linkages, while in levans prevail the 2-6 fructosyl-fructose linkages, and occasional 1-2 linkages type (HIDAKA & al, 1988 [5]). In microorganisms, the inulin degree of polymerization (PD) varies depending on their source (Table 1).

In the vegetal kingdom, the inulin P.D. varies between 2-60. The main sources of vegetal origin inulin are: chicory root, garlic bulb rich in inulin and onion bulb (BEMILLER, 2018 [6]; CANTOR, 2008 [1]; MOSHFEGH & al, 1999 [7]). Another plant used for its medicinal properties and containing appreciable amounts of fructose polymers, called inulin, is *Helianthus tuberosus* known as Jerusalem artichoke (DĂNĂILĂ-GUIDEA & al, 2016 [8]).

Table 1. Degree of Polymerization (P.D.) of inulin in microorganisms

Polymerisation degree PD	Source
10 ⁴ -10 ⁶	Gram positive bacteria (<i>Streptococcus mutans</i> , <i>Lactobacillus reuteri</i> , <i>Leuconostoc citreum</i>)
2-10	Most fungi (<i>Aspergillus</i> genus -with few exceptions, <i>Aureobasidium</i> , <i>Penicillium</i> , <i>Fusarium</i> , <i>Pestalotiopsis</i> , <i>Myrothecium</i> , <i>Trichoderma</i> , and <i>Phytophthora</i>)
30	<i>Aspergillus sydawi</i>

The highest P.D. was observed in inulin from *Cynara scolymus* root, up to 250 monomers. Inulin is present in cereals, respectively in wheat, rye and barley, in relatively small quantities, being concentrated mainly in their pericarp (for example in wheat bran) (CANTOR, 2008 [1]). The P.D. and the percentages of vegetal inulins can be seen in Table 2.

Inulin from wheat and barley, also called graminans, are branched structures, where both types of β 2-1 and β 2-6 fructosyl-fructose linkages are present (SURIANO & al, 2018[9]; BANCAL & al, 1992[10]).

The inulin P.D. from vegetal sources is significantly influenced by harvest time and subsequent storage conditions (RONKART & al, 2006 [11]).

The average molecular weight of inulin is also important because the ratio between it and the average P.D. is a measure of the polydispersity of the sample (a value equal to 1 shows that the molecules have the same length, so they are monodisperse).

Table 2. Degree of Polymerization (P.D.) of inulin in vegetal resources

D	Source	%
2-60	Chicory root (<i>Chicorium intybus</i>)	35.7-47.6
2-50	Jerusalem artichokes (<i>Helianthus tuberosus</i> L)	16-20
2-50	Garlic bulb	9-16
2-12	Onion bulb	1.1-7.5
2-250	Artichoke (<i>Cynara scolymus</i>)	2-6.8
2-8	Wheat bran	1-4

The P.D. and the polydispersity index of inulin significantly influences the physicochemical properties and the potential for its use in various applications (BLECKER & al, 2003 [12]; STEPTO & al, 2009 [13]). In most living organisms, fructans are synthesized starting from sucrose, by a dual displacement mechanism which involves the formation and hydrolysis of an intermediate fructosyl-enzyme.

Microbiological source. Bacterial fructosyltransferases catalyze the transfer of fructosyl residue from sucrose to a wide range of acceptor substrates: water (sucrose hydrolysis), sucrose (kestosis synthesis), fructan (fructan polymerization), glucose (sucrose synthesis), or fructose (difructose synthesis). Most bacteria produce levans using enzymes (levansucrases; E.C.2.4.1.10.) that catalyse the formation of both types of fructosyl-fructose (β 2-1 and β 2-6). Enzymes, called inulosucrases, produce inulin type polymers (E.C.2.4.1.9). For plants, fructan synthesis always involves fructosyl transferases, which have specificity for the donor and acceptor substrate of the fructosyl residue. For example, the enzyme sucrose: sucrose 1 fructosyl transferase transfers a fructosyl residue from one sucrose molecule to another, with the formation of the triglycid intermediate 1 kestose and glucose. The same enzyme may, to a lesser extent, use 1 kestose as acceptor substrate to form β 2-1 binding and produce tetra or pentaglucides such as nistose or fructosylnistose (VAN der MEER & al, 1998 [14]). Other fructosyltransferases are involved in elongation and branching of fructans. β -2-6 linkages can occur in plants by fructan-fructan-6-fructose enzyme activity, which catalyzes the transfer of the terminal fructose from 1 kestose to the sucrose glucose residue, forming a neokestose (CHALMERS & al, 2003 [15]).

The process of inulin obtaining and biotechnological perspectives. Industrial procedures for the inulin production from sucrose, have been successfully developed. Ferments produced by *Aspergillus niger* or *Aureobasidium pullulans* are used to obtain fructo-oligosaccharides with a low P.D. (max. 5). *Aspergillus oryzae* produces oligo-fructoses such as nystose 1-kestose, nystose and fructosyl, from sucrose, but also possesses an enzyme that hydrolyses sucrose (HEYRAUD & al, 1984 [16]; KURAKAKE & al, 2007 [17]). Various studies have shown the ability of fructosyltransferases from *E. coli*, *Streptococcus mutans*, *Bacillus subtilis*, *Lactobacillus* sp. or *Leuconostoc citreum* to produce high molecular weight inulins from sucrose (HEYER & al, 1998 [18]; WADA & al, 2005 [19]). Conversion of sucrose to levan with bacterial levansucrases is extremely costly. The enzymes remain attached to the fructose polymer and the reaction mixture has high viscosity (BANGUELA & al, 2006 [3]). Linear inulin with P.D. 10-30 is obtained from the chicory root by extraction in hot water followed by filtration, elimination of colloidal materials and purification on ion exchange resins. The product is dried by spraying (BANGUELA & al, 2006 [3]).

Various process parameters can significantly influence the yield of inulin or its P.D., namely: water pH and boiling time. Various decanting, centrifugation variants, the use of extraction co-solvents, such as methanol, acetone, ethanol, have been associated with the isolation of polymers with different P.D. Temperatures and spray techniques also influence a series of crystallographic features and rheological behaviors of the obtained product (TONELI & al, 2007 [20]; TONELI & al, 2010 [21]). However, chicory as a source of inulin has a number of disadvantages: low agronomic yield, and degradation of inulin from roots after harvesting, by a series of exohidrolase. However, some

studies have shown that the polydispersion degree of inulin from plants is clearly superior to that of inulins derived from enzymatic synthesis (WADA & al, 2005 [19]). In spite of these achievements, until now there is no effective industrial technology to produce branched inulin, with a high P.D. Genes for fructosyltransferases have been transferred from bacteria to plants such as tobacco, potato, corn, sugar beet or perennial ryegrass (*Lolium perenne*).

Generally, it was intended to obtain plants resistant to abiotic stress factors, such as drought, or plants capable of producing fructans at a higher level of agronomic and economic efficiency. The fructan synthesis has been directed to certain plants organs such as: leaves (tobacco, potato, sugar beet and *Lolium perenne* grass), tubers (potato) or seeds (corn). Intracellular accumulation of fructans was achieved in vacuoles in most plants, cytoplasm (at corn, potato and tobacco), plastids (potato and tobacco) or apoplasm (potato). Although vacuoles were considered to be the best site for intracellular levan accumulation (due to intracellular sequestration and lack of interaction with the intracellular environment), the results obtained were not encouraging. Synthesis of levan in cytoplasm was toxic to tobacco, corn and potato (CAIMI & al, 1996 [22]; CHALMERS & al, 2003 [15]; ROBER & al, 1996 [23]). Generally, inulin levels obtained in these plants were lower than those from plants naturally producing inulin (2.5-10% on dry weight basis). The best results were reported in plants grown *in vitro*, having sucrose as a source of carbon (VAN der MEER & al, 1994 [24]).

Transfer of genes for fructosyl transferases, from some plant species to other plant species, pursued either objectives related to the fructan synthesis mechanism or the biotechnological interest of plants. Gene transfer was primarily done from inuline-producing species to non-productive species, such as petunia, tobacco, potato or sugar beet. For example, gene transfer for 1-fructosyl-transferase from *Helianthus tuberosus* to sugar beet, leads to an accumulation of fructo-oligosaccharides of 40% in the plant's roots, based on its fresh mass, but the process has been accompanied by a decrease of the total soluble carbohydrate content of the root (SÉVENIER & al, 1998 [25]).

By concomitant gene transfer for 1-fructosyl transferase and fructan: fructan 6G fructosyl transferase to sugar beet, the total carbohydrate content and the accumulation of neoinulin, with a degree of polymerization of 3-5 at a rate of 90 mg/g (mass freshness), were preserved (WEYENS & al, 2004 [26]). With the exception of sugar beet, most transgenic plants accumulate significantly lower amounts of fructans (10 mg/g) compared to plants that naturally produce and accumulate fructans (60-150 mg/g) (CAIRNS, 2003 [27]). This is due, on the one hand, to the endogenous degradation of short-chain fructo-oligoglucides by invertase, but also to reduced sucrose affinity of plants fructosyl transferases in relation to their invertases. It is appreciated that the most promising transgenic plants used to produce inulin are: sugar beet and rice (MENSINK & al, 2015 [28]).

Inulin physical-chemical properties and applications.

The extraordinary applicative area of inulin molecules is due to both molecular flexibility (because the molecule chain has essentially a polyethylene oxide like structure) and the predominance of the furanose cycles (considered more flexible) than the more rigid pyranose rings (Figure 1) (BARCLAY & al, 2016 [29]).

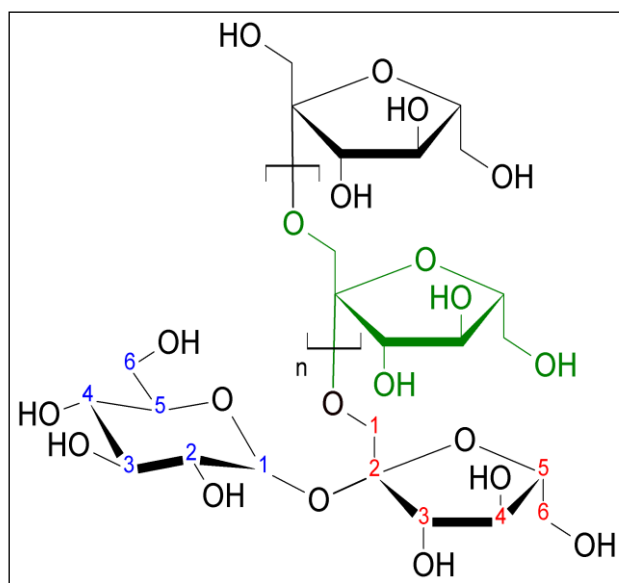


Figure 1. Structure of the inulin molecule

The morphology of inulin crystals (oblique or acicular) is dependent on the temperature of the solution from which it is recovered and the cooling rate. The two forms of crystals have different properties in terms of rheological behavior: acicular forms increase viscosity and the oblique ones improve lubricity (MENSINK & al, 2015 [28]). The three-dimensional structure of the inulin molecule adopt a spiral conformation, but the molecule is flexible.

Inulin conformations in solutions are similar to dextran, but depending on the degree of polymerization a number of other conformations are possible: zigzag, helical etc., (VEREYKEN & al, 2003 [30]). The solubility of inulins in water decreases as their molecular weight increases, but this relationship is mediated by their polydispersity index. Inulins are poorly soluble or insoluble in most organic solvents (ethanol, isopropanol), a property used to separate them. Temperature increases the solubility of all forms of inulin (BOUCHARD & al, 2008 [31]; GLIBOWSKI & al, 2011 [32]). The viscosity of inulin solutions generally increases with the increase of molecular weight and decreases by adding salts or temperature increase (NASKAR & al, 2010a [33]; NASKAR & al, 2010b [34]). The ability to form gels is higher for high molecular weight inulins. The power of these gels is provided by the amount of insoluble microcrystals remaining undissolved, but capable of interacting with both the solvent and other crystals, forming a network. Gels made up of smaller amounts of inulin tend to be weaker due to a lower concentration of crystalline material (RONKART & al, 2010[35]). The heat-formed gels are stronger and finer

than those formed by shear, but at the latter the structure is strongly dependent on the value of the mechanical stress applied to their formation. Also, the gelling process is dependent on the heating time, pH, the presence of solvents, the degree of polymerization etc. Some studies have shown a synergistic effect of inulin with other gelling agents, such as gelatin, alginate, maltodextrins and various types of starch, while other studies reported a competition for water with gelling agents (GONZALEZ-TOMÁS & al, 2008 [36]; KRONBERGA & al, 2011 [37]; MEYER & al, 2011 [38]). Generally, it is recognized that the remaining glucose blocks the chemical reactivity of the inulin, but it should be borne in mind that natural inulin, by the polydispersity character, may also contain ramifications ended with more reactive (reductive) carbohydrates or diglucides (STEVENS & al, 2001 [39]).

The presence of these reactive groups is higher in inulin with a low degree of polymerization (oligofructans), as well as those obtained by hydrolysis of larger molecules or affected by hydrolysis induced by the processing conditions (temperature, pH etc.) (MATUSEK & al, 2009 [40]). The reactive terminal groups can react with the amino group of the proteins, thus interacting with the Maillard reaction cycle. Blocking the participation in the Maillard reaction can be accomplished by adding sulfite or by adjusting the pH of the food systems in which inulin is used (BLECKER & al, 2003[12]; MARTINS & al, 2000 [41]). Inulin is stable to hydrolysis under normal temperature and pH conditions. Increasing temperature and deviations from neutral pH cause intensification of hydrolytic processes. Hydrolysis rates are related to the stability of the glycosidic bond. Generally, the glucosyl-fructosyl linkage is 4-5 times more resistant to acid hydrolysis than the fructosyl-fructosyl linkage, and the fructose terminal units are more easily cleaved than those within the chain (KURAKAKE & al, 2007 [17]).

Inulins are non-reducing carbohydrates but exceptionally, depending on the presence of some branches, which have terminal glucose units, they may be involved in a series of specific reactions, such as the reduction of the final acyclic ketone, to secondary alcohols. The high density of hydroxyl groups is of significant importance in the participation of inulin molecules to supramolecular organization forms, through hydrogen bonding, as is the case of gel formation (COUSSEMENT, 1999[42]).

Therapeutic properties. Fructans are believed to have bifidogenic effect in human nutrition, due to the fact that β -1,2 fructosyl linkages can not be hydrolysed by digestive enzymes in the upper gastrointestinal tract. Therefore, their calorific value is considered to be low. Their metabolism is accomplished by lactobacilli and bifidobacteria in the colon (by production of β -fructofuranosidases). These microorganisms are part of the beneficial flora of the colon and ensure the preservation of a favorable structure of the microbiota, by excluding populations with a potential pathogenic effect (*E. coli*, *Clostridium sp.*, *Salmonella sp.*, and others).

Recent researches suggest that intestinal disbiosis, a generic name under which intestinal floral imbalances

are found, may be the basis for diseases such as: irritable bowel syndrome, inflammatory bowel diseases such as Crohn's disease and ulcerative colitis, diarrheal syndromes associated to antibiotherapy, intestinal diverticulosis, even hepatic diseases but also for extraintestinal affections (allergies, obesity, diabetes etc.) (ROBERFROID, 1999 [43]). In addition, intestinal microbial metabolism byproducts such as short chain fatty acids (butyric acid, propionic acid) or lactic acid have significant effects in reducing serum cholesterol, increasing calcium and magnesium absorption, or preventing colon cancer (by inhibition of histone deacetylases through butyrates) (ZHANG & al, 2017 [44]; PHILLIPS & al, 2009 [2]). The antitumour effect of inulin could be systemic, according to animal studies. Thus, it was found that inulin suppressed the development of methyl-nitrosourea induced mammary carcinogenesis in mice (MARTINOD & al, 2009 [45]; TAPER & al, 1999 [46]). Inulin also has an immunomodulatory effect on the intestine by stimulating the production of immunoglobulin A and interleukin 10, indirectly diminishing the oxidative explosive activity of blood neutrophils and monocytes. It may also increase the ability of peripheral blood mononuclear cells to produce gamma interferon (ROLLER & al, 2004 [47]). Inulin induces the formation of glucagon-like peptides involved in stimulating insulin secretion and suppressing of hunger sensation (DELZENNE & al, 2007 [48]; SÉVENIER & al, 1998 [25]). This contributes indirectly to mechanisms by which cardiovascular risks are reduced, although there is evidence that inulin from food works in this direction even through other effects, namely: reducing the concentration of proatherogenic molecules such as p-cresylsulfate, increasing HDL cholesterol levels and reducing total cholesterol or serum triglycerides (HINRICHS & al, 2001 [49]; WADA & al, 2005 [19]; MEIJERS & al, 2010 [50]; RUSSO & al, 2010 [51]).

Levans can replace dextrans in medical applications such as blood plasma volume expansion or may have anti-tumor or immunomodulatory activity in mice (CALAZANS et al, 2000 [52]; SURIANO & al, 2018 [9]). Despite these evidence, a serious study of the therapeutic effects of inulin should take into account the polydispersity character of inulin molecules. From the perspective of the inulin degree of polymerization, the beneficial effect of oligofructoses is quite controversial.

Some authors have reported in various studies that short-chain carbohydrate ingestion, such as lactose, fructose, sorbitol and fructo-oligoglucides (up to 10 molecules of fructose), may be associated to some patients with symptoms of irritable bowel syndrome (O'BRIEN & al, 2003 [53]; TUCK & al, 2014 [54]). All these oligoglucides are osmotic active substances, insignificant attacked by hydrolases and thus they have the tendency to stay in the intestine for prolonged time, causing the increase of the amount of water in it. Their malabsorption leads to their exposure to intestinal microbiota species, that rapidly degrade them by formation of fatty acids and gases (hydrogen, carbon dioxide and methane) (DE GIORGIO & al, 2017 [55]; VAN der MEER & al, 1994 [24]).

The mechanisms by which oligo-fructoglucides produce the effects associated with metabolic syndromes are not fully understood. Acceleration of intestinal fructolysis can cause local inflammation of the tissues, characterized by weakening of occlusal junctions (these can be described as areas of apical adherence of intestinal wall cells whose role is to prevent passage of substances through intercellular space) and permeability increase of intestinal walls (ZHANG & al, 2017 [44]).

As a result, certain (unwanted) products of secondary metabolism of bacteria in the intestine (endotoxins) can enter the bloodstream causing metabolic endotoxemia. It is estimated that 50% of the Western population suffers from this process during the first five hours after taking the meal. The presence of these endotoxins in the blood stream contributes to the elevation of specific markers associated with systemic inflammation: interleukins 6 and 1-alpha, gamma interferon, triglycerides and insulin. Numerous studies have shown an increase in the value of these markers in animals fed excessively to fructose. The liver, through its numerous connections to the gastrointestinal system, is continuously exposed to endotoxins provided by it. Fatty liver disease has been correlated, in a number of studies, with bacterial overgrowth of the small intestine. In addition, it has been shown that more than 40 grams of fructose/day can cause intestinal discomfort in healthy patients. Until now fructose intolerance has been attributed to the fact that it is poorly digested in small intestine, but it has not been taken into account that excess fructose may favor changes in the normal structure of the intestinal microbiota, by favoring invasive bacterial species (PAN & al, 2018 [56]; ROBERFROID, 1999 [43]; RONKART & al, 2010 [35]; TONELI & al, 2008 [21]; KIM et al, 2017 [57]).

Industrial applications. Like other carbohydrates polymers, inulin has been used successfully to obtain pharmaceutical capsules capable of delivering the active substance to the target tissue and of preserving the interest pharmaceutical properties. This is due to the low chemical reactivity of inulin and its ability to form hydrogels. The maintenance of hydrogel stability, under various *pH* conditions, can be modulated by crosslinking with methacrylic anhydride, succinic acid or vinyl groups. Thus, due to the stability of inulin, in the stomach and small intestine, it has been used successfully to formulate drugs that target the colon. Inulin has also been used as a stabilizing agent for peptide or protein drugs, obtained from aqueous phase following thermal treatments (drying). This is due to the fact that inulin, through the many hydroxyl groups in the molecule, can supplement the hydrogen bonds that ensure the maintenance of the protein molecule conformation. These interactions are known to disappear as a result of water removal. In the case of alkaline phosphatase, the protective effect of inulin was dependent on the length of the polyglucid chain (TONELI et al, 2007 [20]).

In medical analysis, inulin is used to test renal function, namely glomerular filtration (due to the fact that it is not metabolised or resorbed to the renal tubules after intravenous administration, but it is excreted very rapidly

and can be dosed in the urine). The taste and neutral color of inulin, corroborated with special physical-chemical properties, recommended inulin for use in the food industry. This was done either in order to obtain functional products with optimal energy value (due to the therapeutic effects mentioned above), or in the direction of improving the texture and structure of these products. The energy value of inulin is estimated at about 1.5 kcal/g (ROBERFROID, 2002 [43]). Inulin has a sweetening power estimated at about 10% of that of sugar, although kestose type 1-oligo-inulin (a fructose triglycid) can replace fructose (SAEED & al, 2015 [58]). In addition, oligofructoses can have a quantitative synergy with a series of sweeteners such as acesulfame K or aspartame (WEIDMANN & al, 1997 [59]). The potential to form very versatile gels as well as the ability to work synergistically with other gelling agents makes inulin an effective substitute for lipids in certain foods. It is estimated that 1g of lipids can be replaced with 0.25g of inulin (COUSSEMENT, 1999 [42]).

Significant research in this direction has been done in the dairy industry. Thus, the possibility of obtaining *Sintiotic Petit-Suisse* cheese using two probiotic bacteria (*Lactobacillus acidophilus* and *Bifidobacterium animalis* subsp. *lactis*) and three prebiotics with inulin (medium chain inulin, short chain inulin and fructo-oligoglucides), added in various combinations, in a proportion of 10%, was studied. After 28 days of refrigeration, the sensory analysis revealed that the variant with the addition of oligofructose and medium chain inulin in equal proportions was the best (CARDARELLI & al, 2008 [60]). Similarly, simbiotic cottage cheese was made using *Lb. delbrueckii* as a probiotic and 8% medium-chain inulin as prebiotic. This addition did not alter the taste or texture of the cottage cheese after 15 days of storage at 5°C, compared to the control [ARAÚJO & al, 2010 [61]). Finally, other researchers have evaluated the effect of fat replacement with long-chain inulin, on texture and microstructure of fresh cheese made from goat's milk. The fat was substituted with inulin at a rate of 2-7%. Samples with inulin content had lower compression, rigidity, viscosity and adhesive strengths, except for 2% inulin. Electron microscopy investigations have shown that inulin breaks the casein-fat network, hence rheological properties depend on the inulin arrangement within the protein-fat network (SALVATORE & al, 2014 [62]). Inulin has been also used as a prebiotic to obtain assortments of yogurt (STIJEPIĆ & al, 2013 [63]).

Inulin can be used to improve the stability of foams, emulsions and creams. Applications to reduce water activity, with implications for controlling microbial growth in food, have also been studied. These properties are used in various branches of the food industry, such as dairy, bakery, meat, feed, creams and sauces. High degree of polymerization inulins are a very economical source of HFS (High Fructose Syrups), because they have a low glucose content.

Due to the large number of foods based on wheat flour and to their extensive consumption, these products are ideal vectors for enriching the diets with vegetal fibers, including inulin. Adding the inulin or inulin powder to the dough has

been done to replace the vegetal fat. The addition of the inulin gel has increased the moisture capacity of the doughs, as well as the "complex module" of doughs, compared to fat doughs. The bread volume, for doughs with inulin gels, was similar to that of doughs with fat, but significantly less if inulin is added as powder. Also, the inulin powder causes the hardness increase of the bread crust, compared to the bread obtained from the dough to which the inulin was added as a gel (PHILLIPS & al, 2009 [2]). The effects of inulin addition, on the rheology of wheat flour doughs, depend on the evaluation method (biaxial stretching as for the alveographic method, or the change in viscosity over time, by applying a constant mechanical stress, as in the farinographic method) (POPA & al, 2010 [64]).

Thus, the addition of inulin powder (Fibruline Instant) does not significantly change alveographic parameters: Extensibility (L), Resistance (P) and Mechanical Work (W) in doses up to 3%. Beyond these amounts, alveographic parameters are significantly worsened, probably due to changes in dough water distribution and inulin blockade of water required for the development of an extensive gluten network (TAMBA-BEREHOIU & al, 2017 [65]).

Inulin significantly increases the doughs farinographic stability (Figure 2) and decreases the softening degree (water retention capacity under mechanical stress conditions) (POPA & al, 2008 [66]).

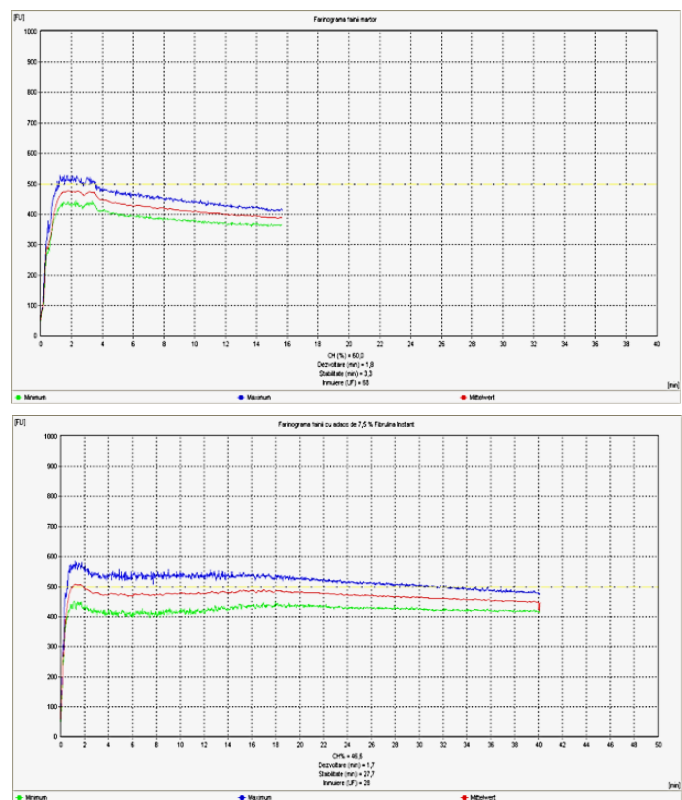


Figure 2. Effect of inulin powder addition on farinographic parameters (up-control, down-sample +7.5% Instant Fibruline, own researches).

The Falling Number parameter of the flour increases proportionally with the addition of inulin, due to the

increase of formed gels viscosity, as well as its stability (POPA & al, 2008 [66]).

The addition of inulin to pasta dough does not significantly alter its structural and textural properties. Inulin influences the swelling index and the firmness of the pasta, but does not alter its adherence and elasticity at boiling. In addition, inulin significantly degrades starch *in-vitro* digestion, thus lowering the glycemic index by up to 15% (BRENNAN & al, 2004 [67]; IVKOV & al, 2018 [68]).

Levans are more soluble than inulins and can form viscous water solutions, which is why they can be used as emulsifiers or encapsulating agents for a very wide range of products: biodegradable plastics, cosmetics, adhesives, textiles or detergents. Chemical derivatives of inulin have industrial uses, e.g. carboxymethyl inulin is used as a crust preventing agent, in the treatment of waste water (KIRBOGA & al, 2012 [69]; MARTINOD & al, 2009 [45]).

Conclusions

Inulins represent an extremely valuable vegetal resource with significant economic potential in terms of capacity to create added value. This is due to multiple directions of use, namely: improving diets by increasing fibers amount, optimization of caloric food density, providing a therapeutic support for certain diseases in the digestive system (although there are some controversies about intestinal fructolysis of oligofructoses), use in medical tests, formulations of drugs, shaping of the food products texture and structure, improving their stability, water treatment, obtaining biocomposite materials etc. The most common methods of obtaining inulins are those that allow it to be extracted from chicory in a manner similar to sugar obtaining. With the exception of sugar beet, most transgenic plants accumulate significantly lower amounts of fructans (10 mg/g) compared to plants that naturally produce and accumulate fructans (60-150 mg/g).

Conflict of interest disclosure

There are no known conflicts of interest in the publication of this article. The manuscript was read and approved by all authors.

References

1. J.M. CANTOR, (Eds.), Progress in food engineering research and development, Nova Publishers, Inc., New York, **Chapter 4:175- 196** (2008).
2. G.O. PHILLIPS, P.A. WILLIAMS, (Eds.), Handbook of hydrocolloids, Woodhead Publishing Ltd., Cambridge, Elsevier Inc., (2nd ed): 829-845, (2009).
3. A. BANGUELA, L. HERNÁNDEZ, Fructans: from natural sources to transgenic plants, *Biotecnología Aplicada*, 23(3): 202-210 (2006).
4. A.L. BONNEMA, L.W. KOLBERG, W. THOMAS, J.L. SLAVIN, Gastrointestinal tolerance of chicory inulin products, *Journal of the American Dietetic Association*, 110(6):865-868 (2010).
5. H. HIDAKA, M. HIRAYAMA, N.A. SUMI, A fructo-oligosaccharide-producing enzyme from *Aspergillus niger* ATCC 20611, *Agricultural and Biological Chemistry*, 52:1181-1187 (1988).
6. J.N. BEMILLER, Carbohydrate chemistry for food scientists, Woodhead Publishing AACC International, Cambridge, Elsevier Inc. (Third Edition): 253-259 (2019).
7. A.J. MOSHFEGH, J.E. FRIDAY, J.P. GOLDMAN, J.K.C. AHUJA, Presence of inulin and oligofructose in the diets of Americans, *The Journal of nutrition*, 129(7):1407S-1411S (1999).
8. S. DĂNĂILĂ-GUIDEA, R.V. DOBRINOIU1, L. VIȘAN, Research on the preparation a protocol of the direct organogenesis through “*in vitro*” culture techniques to *Helianthus tuberosus* (Jerusalem artichoke), *Scientific Papers. Series B, Horticulture*. LX:175-181 (2016).
9. F. SURIANO, A.M. NEYRINCK, J. VERSPREET, M. OLIVARES, S. LECLERCQ, T. VAN DE WIELE, N.M. DELZENNE, Particle size determines the anti-inflammatory effect of wheat bran in a model of fructose over-consumption: Implication of the gut microbiota, *Journal of Functional Foods*, 41:155-162 (2018).
10. P. BANCAL, N.C. CARPITA, J.P. GAUDILLÈRE, Differences in fructan accumulation in induced and field-grown wheat plants: an elongation-trimming pathway for their synthesis, *New Phytologist*, 120: 313-321 (1992).
11. S.N. RONKART, M. PAQUOT, C. FOUGNIES, C. DEROANNE, J.C. VAN HERCK, C. BLECKER, Determination of total water content in inulin using the volumetric Karl Fischer titration, *Talanta*, 70(5):1006-1010 (2006).
12. C. BLECKER, J.P. CHEVALIER, C. FOUGNIES, J.C. VAN HERCK, C. DEROANNE, M. PAQUOT, Characterisation of different inulin samples by DSC: influence of polymerisation degree on melting temperature, *Journal of Thermal Analysis and Calorimetry*, 71(1):215-224 (2003).
13. R.F. STEPTO, Dispersity in polymer science (IUPAC Recommendations 2009), *Pure and Applied Chemistry*, 81(2): 351-353 (2009).
14. I.M. VAN DER MEER, A.J. KOOPS, J.C. HAKKERT, A.J. VAN TUNEN, Cloning of the fructan biosynthesis pathway of Jerusalem artichoke, *Plant Journal*, 15: 489-500 (1998).
15. J. CHALMERS, X. JOHNSON, A. LIDGETT, G. SPANGENBERG, Isolation and characterisation of a sucrose:sucrose 1-fructosyl transferase gene from perennial ryegrass (*Lolium perenne*), *Journal of Plant Physiology*, 160:1385-1391 (2003).
16. A. HEYRAUD, M. RINAUDO, F.R. TARAVEL, Isolation and characterization of oligosaccharides containing d-fructose from juices of the Jerusalem artichoke – kinetic constants for acid hydrolysis, *Carbohydrate Research*, 128: 311-320 (1984).
17. M. KURAKAKE, K. OGAWA, M. SUGIE, A. TAKEMURA, K. SUGIURA, T. KOMAKI, Two

- types of β -fructofuranosidases from *Aspergillus oryzae* KB, *Journal of Agricultural and Food Chemistry*, 56(2):591-596 (2007).
18. A.G. HEYER, B. SCHROEER, S. RADOSTA, D. WOLFF, S. CZAPLA, J. SPRINGER, Structure of the enzymatically synthesized fructan inulin, *Carbohydrate research.*, 313(3-4): 165-174 (1998).
 19. T. WADA, J. SUGATANI, E. TERADA, M. OHGUCHI, M. MIWA, Physicochemical characterization and biological effects of inulin enzymatically synthesized from sucrose, *Journal of Agricultural and Food Chemistry*, 53(4): 1246-1253 (2007).
 20. J.T.C.L. TONELI, F.E.X. MÜRR, P. MARTINELLI, I.M. DAL FABBRO, K.J. PARK, Optimization of a physical concentration process for inulin, *Journal of Food Engineering*, 80(3): 832-838 (2007).
 21. J. TONELI, K. PARK, A. NEGREIROS, F. MURR, Spray-drying process optimization of chicory root inulin, *Drying Technology*, 28(3): 369-379 (2010).
 22. P.G. CAIMI, L.M. MCCOLE, T.M. KLEIN, P.S. KERR, Fructan accumulation and sucrose metabolism in transgenic maize endosperm expressing a *Bacillus amyloliquefaciens* sacB gene, *Plant Physiology*, 110: 355-363 (1996).
 23. M. ROBER, K. GEIDER, B. MULLER-ROEBER, L. WILLMITZER, Synthesis of fructans in tubers of transgenic starch-deficient potato plants does not result in an increased allocation of carbohydrates, *Planta*, 199: 528-36 (1996).
 24. I.M. VAN DER MEER, M.J.M. EBSKAMP, R.G.F. VISSER, P.J. WEISBEEK, S.C.M. SMEEKENS, Fructan as a new carbohydrate sink in transgenic potato plants, *Plant Cell*, 6:561-570 (1994).
 25. R. SÉVENIER, R.D. HALL, I.M. VAN DER MEER, H.J.C. HAKKERT, A.J. VAN TUNEN, A.J. KOOPS, High level fructan accumulation in a transgenic sugar beet, *Nature Biotechnology*, 16: 843-846 (1998).
 26. G. WEYENS, T. RITSEMA, K. VAN DUN, D. MEYER, M. LOMMEL, J. LATHOUWERS, Production of tailor-made fructans in sugar beet by expression of onion fructosyltransferase genes, *Plant Biotechnology Journal*, 2: 321-327 (2004).
 27. A.J. CAIRNS, Fructan biosynthesis in transgenic plants, *Journal of Experimental Botany*, 54: 549-567 (2003).
 28. M.A. MENSINK, H.W. FRIJLINK, K.VAN DER VOORT MAARSCHALK, W.L. HINRICHS, Inulin, a flexible oligosaccharide I: Review of its physicochemical characteristics, *Carbohydrate polymers*, 130: 405-419 (2015).
 29. T. BARCLAY, M. GINIC-MARKOVIC, P. COOPER, N. PETROVSKY, Inulin-a versatile polysaccharide with multiple pharmaceutical and food chemical uses, *Journal of Excipients and Food Chemicals*, 1(3): 27-50 (2016).
 30. I.J. VEREYKEN, J.A.VAN KUIK, T.H. EVERS, P.J. RIJKEN, B. DE KRUIJFF, Structural requirements of the fructan-lipid interaction, *Biophysical Journal*, 84(5): 3147-3154 (2003).
 31. A. BOUCHARD, N. JOVANOVIĆ, G.W. HOFLAND, W. JISKOOT, E. MENDES, D.J. CROMMELIN, G.J. WITKAMP, Supercritical fluid drying of carbohydrates: selection of suitable excipients and process conditions, *European Journal of Pharmaceutics and Biopharmaceutics*, 68(3): 781-794 (2008).
 32. P. GLIBOWSKI, S. PIKUS, Amorphous and crystal inulin behavior in a water environment, *Carbohydrate Polymers*, 83(2): 635-639 (2011).
 33. B. NASKAR, A. DAN, S. GHOSH, S.P. MOULIK, Characteristic physicochemical features of the biopolymer inulin in solvent added and depleted states, *Carbohydrate Polymers*, 81(3): 700-706 (2010a).
 34. B. NASKAR, A. DAN, S. GHOSH, S.P. MOULIK, Viscosity and solubility behavior of the polysaccharide inulin in water, water+ dimethyl sulfoxide, and water+ isopropanol media, *Journal of Chemical & Engineering Data*, 55(7): 2424-2427 (2010b).
 35. S.N. RONKART, M. PAQUOT, C. DEROANNE, C. FOUGNIES, S. BESBES, C.S. BLECKER, Development of gelling properties of inulin by microfluidization, *Food hydrocolloids*, 24(4): 318-324 (2010).
 36. L. GONZALEZ-TOMÁS, J. COLL-MARQUÉS, E. COSTELL, Viscoelasticity of inulin–starch-based dairy systems. Influence of inulin average chain length, *Food Hydrocolloids*, 22(7):1372-1380 (2008).
 37. M. KRONBERGA, D. KARKLINA, D. KLAVA, R. GALOBURDA, Evaluation of inulin gelling properties in new types of jellies, *Scientific Bulletin "Biotechnologies" – Series F.*, XV:70-76 (2011).
 38. D. MEYER, S. BAYARRI, A. TÁRREGA, E. COSTELL, Inulin as texture modifier in dairy products, *Food Hydrocolloids*, 25(8): 1881-1890 (2011).
 39. C.V. STEVENS, A. MERIGGI, K. BOOTEN, Chemical modification of inulin, a valuable renewable resource, and its industrial applications, *Biomacromolecules*, 2(1): 1-16 (2001).
 40. A. MATUSEK, P. MERÉSZ, T.K.D. LE, F. ÖRSI, Effect of temperature and pH on the degradation of fructo-oligosaccharides, *European Food Research and Technology*, 228(3): 355-365 (2009).
 41. S.I. MARTINS, W.M. JONGEN, M.A. VAN BOEKEL, A review of Maillard reaction in food and implications to kinetic modelling, *Trends In Food Science & Technology*, 11(9-10): 364-373 (2000).
 42. P.A. COUSSEMENT, Inulin and oligofructose: safe intakes and legal status, *Journal of Nutrition*, 129: 1412S-1417S (1999).
 43. M. ROBERFROID, Caloric value of inulin and oligofructose, *Br. Journal of Nutrition*, 87:139-143 (2002).
 44. D.M. ZHANG, R.Q. JIAO, L.D.KONG, High dietary fructose: Direct or indirect dangerous factors disturbing tissue and organ functions, *Nutrients*, 9(4): 335 (2017).
 45. A. MARTINOD, A. NEVILLE, M. EUVRAD, K. SORBIE, Electrodeposition of a calcareous layer: Effects of green inhibitors, *Chemical Engineering Science*, 64(10): 2413-2421 (2009).

46. H.S. TAPER, M. ROBERFROID, Influence of inulin and oligofructose on breast cancer and tumor growth, *Journal of Nutrition*, 129:1488S-1491S (1999).
47. M. ROLLER, G. RECHKEMMER, B. WATZL, Prebiotic inulin enriched with oligofructose in combination with the probiotics *Lactobacillus rhamnosus* and *Bifidobacterium lactis* modulates intestinal immune functions in rats, *Journal of Nutrition*, 134: 153-156 (2004).
48. N.M. DELZENNE, P.D. CANI, A.M. NEYRINCK, Modulation of Glucagon-Like Peptide 1 and Energy Metabolism by Inulin and Oligofructose: Experimental Data, *Journal of Nutrition*, 137: 2547S-2551S (2007).
49. W.L.J. HINRICHS, M.G. PRINSEN, H.W. FRIJLINK, Inulin glasses for the stabilization of therapeutic proteins, *International Journal of Pharmaceutics*, 215: 163-174 (2001).
50. B.K.I. MEIJERS, V. DE PRETER, K. VERBEKE, Y. VANRENTERGHEM, P. EVENEPOEL, P-Cresyl Sulfate Serum Concentrations in Haemodialysis Patients Are Reduced by the Prebiotic Oligofructose-Enriched Inulin, *Nephrology Dialysis Transplantation*, 25: 219-224 (2010).
51. F. RUSSO, G. RIEZZO, M. CHILOIRO, G. DE MICHELE, G. CHIMIENTI, E. MARCONI, B. D'ATTOMA, M. LINSALATA, C. CLEMENTE, Metabolic effects of a diet with inulin-enriched pasta in healthy young volunteers, *Current Pharmaceutical Design*, 16: 825-831 (2010).
52. G.M.T. CALAZANS, R.C. LIMA, F.P. DE FRANCA, C.E. LOPES, Molecular weight and antitumour activity of *Zymomonas mobilis* levans, *International Journal of Biological Macromolecules*, 27, 245-247 (2000).
53. C.M. O'BRIEN, A. MUELLER, A.G.M. SCANNELL, E.K. ARENDT, Evaluation of the effects of fat replacers on the quality of wheat bread, *Journal of Food Engineering*, 56(2-3):265-267 (2003).
54. C.J. TUCK, J.G. MUIR, J.S. BARRETT, P.R. GIBSON, Fermentable oligosaccharides, disaccharides, monosaccharides and polyols: role in irritable bowel syndrome, *Expert Review of Gastroenterology & Hepatology*, 8: 819-834, (2014).
55. R. DE GIORGIO, U. VOLTA, P.R. GIBSON, Sensitivity to wheat, gluten and FODMAPs in IBS: facts or fiction?, *Gut*, 65(1): 169-178 (2016).
56. Y. PAN, L.D. KONG, High fructose diet-induced metabolic syndrome: pathophysiological mechanism and treatment by traditional Chinese medicine, *Pharmacological Research*, 130: 438-450 (2018).
57. M. KIM, I.I. ASTAPOVA, S.N. FLIER, S.A. HANNOU, L.DORIDOT, A. SARGSYAN, M.A. HERMAN, Intestinal, but not hepatic, ChREBP is required for fructose tolerance, *JCI insight*, 2(24), e96703 (2017).
58. M. SAEED, I. YASMIN, I. PASHA, M.A. RANDHAWA, M. I. KHAN, M.A. SHABBIR, W.A. KHAN, Potential application of inulin in food industry: A review, *Pakistan Journal of Food Sciences*, 25(3): 110-116 (2015).
59. M. WEIDMANN, M. JAGER, Synergistic sweeteners, *Food Ingredients and Analysis International*, 6: 51-56 (1997).
60. H.R. CARDARELLI, F.C. BURITI, I.A. CASTRO, S.M. SAAD, Inulin and oligofructose improve sensory quality and increase the probiotic viable count in potentially synbiotic petit-suisse cheese, *LWT-Food Science and Technology*, 41(6): 1037-1046 (2008).
61. E.A. ARAÚJO, A.F. DE CARVALHO, E.S. LEANDRO, M.M. FURTADO, C.A. DE MORAES, Development of a symbiotic cottage cheese added with *Lactobacillus delbrueckii* UFV H2b20 and inulin, *Journal of Functional Foods*, 2(1): 85-89 (2010).
62. E. SALVATORE, M. PES, V. MAZZARELLO, A. PIRISI, Replacement of fat with long-chain inulin in a fresh cheese made from caprine milk, *International Dairy Journal*, 34(1): 1-5 (2014).
63. M. STIJEPIĆ, J. GLUŠAC, D. DURDEVIĆ-MILOŠEVIĆ, D. PEŠIĆ-MIKULEC, Physicochemical characteristics of soy probiotic yoghurt with inulin addition during the refrigerated storage, *Rom. Biotech. Letters*, 18(2): 8077-8085 (2013).
64. N.C. POPA, R. TAMBA-BEREHOIU, S. POPESCU, S. TAMBA, Investigations on predictive modelling of the farinographic parameters, *Scientific Bulletin "Biotechnologies" – Series F.*, 14: 50-57 (2010).
65. R. TAMBA-BEREHOIU, M.O. TURTOI, L.V. VISAN, C.N. POPA, Physico-chemical, rheological and technological characterization of some mixtures of wheat, oat, barley and millet flours, *Annals of the University Dunărea de Jos of Galați Fascicle VI-Food Technology*, 41: 102-114 (2017).
66. N.C. POPA, R. TAMBA-BEREHOIU, S. POPESCU, Modifications of some dough rheological descriptors through the addition of vegetal fibres with functional properties, *Annals of the Suceava University – Food Engineering*, VII: 25-33 (2008).
67. C.S. BRENNAN, V. KURI, C.M. TUDORICA, Inulin-enriched pasta: effects on textural properties and starch degradation, *Food Chemistry*, 86(2):189-193, (2004).
68. M. IVKOV, M. KOŠUTIĆ, J. FILIPOVIĆ, V. FILIPOVIĆ, Spelt pasta with addition of inulin as a functional food: sensory evaluation and consumer attitudes, *Rom. Biotech. Letters*, 23(3):13615-13624 (2018).
69. S. KIRBOGA, M. ÖNER, The inhibitory effects of carboxymethyl inulin on the seeded growth of calcium carbonate, *Colloids and Surfaces B: Biointerfaces*, 91: 18-25 (2012).