

Reformulación de galletas de masa corta: cambios en reología, textura y propiedades sensoriales

LAURA LAGUNA CRUAÑES

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REFORMULACIÓN DE GALLETAS DE MASA CORTA: CAMBIOS EN REOLOGÍA, TEXTURA Y PROPIEDADES SENSORIALES

Tesis Doctoral
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HACEN CONSTAR QUE:

el trabajo de investigación titulado **“Reformulación de galletas de masa corta: cambios en reología, textura y propiedades sensoriales”** que presenta Dña. Laura Laguna Cruaños por la Universidad Politécnica de Valencia, ha sido realizado en el instituto de Agroquímica y Tecnología de Alimentos (IATA-CSIC) bajo nuestra dirección y que reúne las condiciones para optar al grado de Doctor.

Valencia, abril de 2013.

Fdo.: Dra. Ana Salvador Alcaraz.

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*Contra viento y marea,
a los que fueron pirata*

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RESUMEN

REFORMULACIÓN DE GALLETAS DE MASA CORTA: CAMBIOS EN REOLOGÍA, TEXTURA Y PROPIEDADES SENSORIALES

El presente trabajo de tesis se ha centrado en la evaluación de las propiedades físicas y sensoriales de galletas tras su reformulación con nuevos ingredientes para crear productos más saludables utilizando técnicas reológicas, texturales y sensoriales.

La formulación de galleta consta de tres ingredientes fundamentales: harina, grasa y azúcar. Dada la demanda actual de los consumidores de alimentos saludables, el reemplazo de grasa y azúcar así como la incorporación de fibra en las galletas resulta de gran interés. Sin embargo, esta reformulación afecta significativamente a las propiedades de las galletas. En esta tesis mediante la aplicación de técnicas físicas y sensoriales se estudia la funcionalidad de los ingredientes básicos y de nuevos ingredientes con la finalidad de seleccionar el ingrediente óptimo que permita reformular obteniendo una galleta final de la máxima calidad y aceptación sensorial.

Las propiedades de viscoelasticidad lineal de la masa pudieron predecir aspectos de calidad tras el horneado como las dimensiones y la textura. Los ingredientes fuente de fibra utilizados son el almidón resistente, la fibra de manzana y la fibra de trigo. El almidón resistente confirió dureza a la masa mientras que las galletas resultaron más blandas, la fibra de trigo aumentó la resistencia a la deformación en la masa y la galleta, mientras que la incorporación de fibra de manzana no modificó significativamente las propiedades de la masa y galleta. El análisis sensorial descriptivo concluyó que la fibra que menos afectó a las propiedades físicas de la galleta fue la fibra de manzana, a pesar de que el color y aroma en el caso de la utilización de almidón resistente y fibra de trigo cambiaba menos respecto a la galleta control.

El estudio de la trayectoria oral de las galletas se realizó utilizando una técnica sensorial especializada denominada “predominio temporal de las sensaciones”. Se estudiaron galletas altas y bajas en grasa y con y sin adición de fibra de trigo. Se obtuvieron los atributos clave en el procesado oral. Se concluyó que el grado de dominancia de algunos de los atributos obtenidos podrían influir negativamente en la aceptabilidad por parte de los consumidores como ocurre en el caso de la sensación de sequedad bucal y dureza.

La reformulación de la galleta influyó en las propiedades de textura y el sonido emitido durante la fractura. El sonido emitido al romper las galletas y las curvas de fuerza-desplazamiento se relacionaron con los atributos y puntuación obtenidos mediante el análisis sensorial cuali y cuantitativo. Se observó que la utilización de inulina como reemplazante de la sacarosa proporcionó mejores resultados que el eritritol. La utilización de inulina como reemplazante de grasa también proporcionó características de textura y sonido similares a la galleta control, sin embargo, la utilización de hidroxipropilmetilcelulosa como reemplazante de grasa proporcionó galletas más duras y sonoras que la galleta control.

Un estudio más profundo de la funcionalidad del azúcar en galletas permitió dilucidar que el maltitol es un excelente reemplazante de la sacarosa en galletas. Para ello se estudiaron las diferentes interacciones de los componentes de las galletas con los diferentes azúcares empleados (sacarosa, eritritol y maltitol) en un sistema modelo, en la masa y en la galleta. Mediante técnicas de calorimetría diferencial se concluyó que los polioles (eritritol y maltitol) actúan plastificando el gluten modificando así su temperatura de transición vítrea. Las propiedades de la masa y la galleta al sustituir con eritritol se asemejan más a la masa y a la galleta que no contienen sacarosa, mientras que el maltitol presentó un comportamiento reológico y una textura similar a la sacarosa.

RESUM

REFORMULACIÓ DE GALLETES DE MASSA CURTA: CANVIS EN REOLOGÍA, TEXTURA I PROPIETATS SENSORIALS

El present treball de tesi s'ha centrat en l'avaluació de les propietats físiques i sensorials de galletes després de la seua reformulació amb nous ingredients per a crear productes més saludables utilitzant tècniques reològiques, texturals i sensorials.

La formulació de galleta consta de tres ingredients fonamentals: farina, greix i sucre. Donada la demanda actual dels consumidors d'aliments saludables, el reemplaçament de greix i sucre així com la incorporació de fibra en les galletes resulta de gran interès. No obstant això, esta reformulació afecta significativament les propietats de les galletes. En esta tesi per mitjà de l'aplicació de tècniques físiques i sensorials s'estudia la funcionalitat dels ingredients bàsics i de nous ingredients amb la finalitat de seleccionar l'ingredient òptim que permetia reformular obtenint una galleta final de la màxima qualitat i acceptació sensorial.

Les propietats de viscoelasticidad lineal de la massa van poder predir aspectes de qualitat després de l'enfornat com les dimensions i la textura. Els ingredients font de fibra utilitzats són el midó resistent, la fibra de poma i la fibra de blat. El midó resistent va conferir duresa a la massa mentres que les galletes van resultar més blanques, la fibra de blat va augmentar la resistència a la deformació en la massa i la galleta, mentres que la incorporació de fibra de poma no va modificar significativament les propietats de la massa i galleta. L'anàlisi sensorial descriptiu va concloure que la fibra que menys va afectar les propietats físiques de la galleta va ser la fibra de poma, a pesar que el color i aroma en el cas de la utilització de midó resistent i fibra de blat canviava menys respecte a la galleta control.

L'estudi de la trajectòria oral de les galletes es va realitzar utilitzant una tècnica sensorial especialitzada denominada 'predomini temporal de les sensacions'. Es van estudiar galletes altes i baixes en greix i amb i sense addició de fibra de blat. Es van obtenir els atributs clau en el processat oral. Es va concloure que el grau de dominància d'alguns dels atributs obtinguts podrien influir negativament en l'acceptabilitat per part dels consumidors com ocorre en el cas de la sensació de sequedat bucal i duresa.

La reformulació de la galleta va influir en les propietats de textura i el so emés durant la fractura. El so emés al trencar les galletes i les corbes de força-desplaçament es van relacionar amb els atributs i puntuació obtinguts per mitjà de l'anàlisi sensorial qualitat i quantitatiu. Es va observar que la utilització d'inulina com reemplaçant de la sacarosa va proporcionar millors resultats que l'eritritol. La utilització d'inulina com reemplaçant de greix també va proporcionar característiques de textura i so semblants a la galleta control, no obstant això, la utilització de hidroxipropilmetilcelulosa com reemplaçant de greix va proporcionar galletes més dures i sonores que la galleta control.

Un estudi més profund de la funcionalitat del sucre en galletes va permetre dilucidar que el maltitol és un excel·lent reemplaçant de la sacarosa en galletes. Per a això es van estudiar les diferents interaccions dels components de les galletes amb els diferents sucres emprats (sacarosa, eritritol i maltitol) en un sistema model, en la massa i en la galleta. Per mitjà de tècniques de calorimetria diferencial es va concloure que els poliols (eritritol i maltitol) actuen plastificant el gluten modificant així la seua temperatura de transició vítria. Les propietats de la massa i la galleta al substituir amb eritritol s'assemblen més a la massa i a la galleta que no contenen sacarosa, mentres que el maltitol va presentar un comportament reològic i una textura semblant a la sacarosa.

SUMMARY

SHORT-DOUGH BISCUIT REFORMULATION: CHANGES IN RHEOLOGY, TEXTURE AND SENSORY PROPERTIES

This thesis work has focused on the evaluation of physical and sensory properties of biscuits after reformulation with new ingredients to create healthier products using rheological, textural and sensory techniques.

The biscuit formulation consists of three basic ingredients: flour, fat and sugar. Currently, consumers demand healthy food. For that, fat and sugar replacement and the inclusion of fiber in the biscuits is of great interest. However, this reformulation significantly affects the properties of the biscuits. The functionality of the common and new ingredients was studied by physical and sensory techniques. After that, the optimal ingredient was selected for obtaining healthier biscuits.

The linear viscoelastic properties of the dough could predict quality aspects after baking as the dimensions and texture. The ingredients used as source of fiber were: resistant starch, apple fiber and wheat fiber. Resistant starch conferred to the dough hardness while the biscuits were softer, wheat fiber increased resistance to deformation in the dough and in the biscuit, while the apple fiber incorporation not significantly change the properties of dough or biscuit. The descriptive sensory analysis concluded that fiber least affecting the physical properties of the biscuit was apple fiber, although the color and flavor in the case of the use of resistant starch and wheat fiber were more similar to the control biscuit.

The study of oral path of biscuits was performed using a specialized sensory technique called "Temporal dominance of sensations". Biscuits were studied with high and low fat content and with and without addition of wheat fiber. Key attributes were obtained in oral processing. It was concluded that the degree of

dominance of some of the attributes obtained may adversely affect the acceptability by consumers as in the case of the sensation of dry mouth and hardness.

The reformulation of the biscuit influenced the textural properties and the sound emitted during fracture. The sound emitted by breaking biscuits and force-displacement curves related to the attributes and score obtained by qualitative and quantitative sensory testing. It was observed that the use of inulin as a replacement for sucrose gave better results than erythritol. The use of inulin as fat replacer also provided texture characteristics similar to the biscuit sound control, however, the use of hydroxypropylmethylcellulose as fat replacer provided biscuits and sound harder than the control biscuit.

A deeper study of the sugar biscuit functionality allowed elucidate that maltitol is an excellent replacement for sucrose in biscuits. The interactions of different components with different sugars biscuits (sucrose, erythritol and maltitol) in a model system, and the biscuit dough were studied. Using techniques of differential calorimetry was concluded that the polyols (erythritol and maltitol) act plasticizing gluten thus modifying its glass transition temperature. The properties of the biscuits with erythritol were more similar to those without any kind of sugar, however, maltitol biscuits showed similar rheological and texture similar to sucrose.

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INTRODUCCIÓN

INTRODUCCIÓN

1. Definición y clasificación de galletas

Según el Real Decreto 1124 (1982) se entiende por “galletas” los productos alimenticios elaborados por una mezcla de harina, grasas comestibles y agua, adicionada o no de azúcares y de otros productos alimenticios o alimentarios (aditivos, aromas, condimentos, especias, etc.) sometidos a un proceso de amasado y al posterior tratamiento térmico, dando lugar a un producto de presentación muy variada, caracterizado por su bajo contenido en agua.

Las galletas se diferencian de otros productos derivados de cereales en base a su contenido en agua. En general, se reconoce que las galletas poseen un contenido en agua inferior al 5%, a diferencia de otros productos horneados como el pan que posee un 35-40% de humedad o los bizcochos con un 15-30% de humedad (Wade, 1988).

La legislación española (Real Decreto 1124 (1982)) clasifica las galletas dentro de los siguientes grupos:

- Marías, tostadas y troqueladas.
- Cracker y de aperitivo.
- Barquillos con o sin relleno.
- Bizcochos secos y blandos.
- Sandwiches.
- Pastas blandas y duras.
- Bañadas con aceite vegetal.
- Recubiertas de chocolate.
- Surtidos.
- Elaboraciones complementarias.

Según Manley (1991) las galletas se pueden clasificar en base a la textura o dureza del producto final, al cambio de forma en el horno, a la extensibilidad de la masa, o a las diferentes formas de tratar la masa.

Otra clasificación (Wade, 1988) distingue entre dos tipos fundamentales de galletas: “de masa dura” (*hard dough*) y “de masa corta” (*short dough*), siendo una de las diferencias fundamentales entre estos dos tipos de galletas la existencia o no de largas cadenas de gluten que confieren a la masa extensibilidad (Manley, 1991). Cuando el gluten está desarrollado, la masa presenta un comportamiento viscoelástico dando lugar a masas duras, sin embargo, cuando la cantidad de grasa y azúcar es alta, el gluten no se puede desarrollar completamente y la masa se queda corta. Además, las galletas de masa corta aumentan su tamaño (*spread* o esparcimiento) durante los primeros estadios del proceso de horneado, mientras que las galletas de masa dura tienden a encoger longitudinalmente (Manley, 1991).

En España, las galletas de masa corta son más conocidas como “pastas de té” y en el Reino Unido como *short dough biscuit*. Este tipo de galleta se caracteriza por ser rica en grasa y azúcar con la presencia de pequeñas cantidades de agua, por lo que la masa no es elástica y rompe fácilmente bajo tensión (Manley, 1991).

2. Proceso de elaboración de galletas

La masa es el estado intermedio entre la harina y el producto terminado (Sai Manohar y Haridas Rao, 1999a). La calidad de la masa queda determinada por la cantidad y calidad de los ingredientes empleados. Cada masa tiene unas cualidades particulares de consistencia, elasticidad, resiliencia y moldeabilidad.

Al igual que existen numerosas formulaciones de galletas, también existen diversos procesos para formar la masa de galleta. En las galletas de masa corta el objetivo fundamental durante el amasado es que el gluten se desarrolle lo mínimo aunque debe lograrse la dispersión adecuada de los ingredientes (Baltsavias y col., 1999a). Hay fundamentalmente dos procesos de amasado:

- Método simple (*single-method*), donde se mezclan todos los ingredientes en una sola etapa (Pareyt y Delcour, 2008a).
- Método de punto pomada (*creaming-method*), donde primero se mezcla la mantequilla con el azúcar y los ingredientes minoritarios hasta alcanzar lo que se conoce en pastelería como “punto pomada” (*cream-up*) y, posteriormente, se añade el resto de ingredientes (Pareyt y Delcour, 2008a). En este caso, la grasa se combina con el azúcar ayudando a atrapar el aire. De hecho, la grasa envuelve individualmente los granos de azúcar impidiendo que se agreguen entre sí y formen terrones. Si no fuera así, cuando el azúcar se fundiese volvería a recristalizar formando partículas de mayor tamaño (Hutchinson, 1978). Esta etapa es determinante en la formación de la estructura del producto terminado y en la densidad de la masa.

Durante el amasado, la energía impartida a la masa ha de ser menor que la típicamente utilizada para el pan u otros productos de panadería, con el fin de evitar el desarrollo del gluten ya que la masa de galleta necesita tener buena extensibilidad, baja elasticidad y baja resistencia a la deformación (Cauvain y Young, 2006).

El tiempo de amasado también afecta a la masa, haciéndola más deformable, pero también puede afectar al gluten ayudándolo a desarrollarse (Baltsavias y col., 1999b).

Según Pareyt y col. (2008a), la distribución de los ingredientes dentro de la masa de galleta dependerá de la formulación empleada. De tal forma que si la concentración de grasa es alta, respondería al modelo propuesto por Baltsavias y Jurgens (1997a), donde la masa es un sistema bifásico compuesto por una fase grasa y una fase no grasa formada por una solución saturada de azúcar que envuelve las partículas de harina/gluten. Por otro lado, si el contenido en grasa es bajo, representaría el modelo presentado por Chevallier y col. (2000a) donde la masa de galleta es una suspensión de proteínas, complejos almidón-proteína y gránulos aislados de almidón en una fase líquida continua basada en una emulsión de lípidos en una solución concentrada de azúcar.

Durante el periodo de espera entre el amasado y el laminado de galletas ocurren numerosos cambios en la masa (Wade, 1988). Las galletas de masa corta cambian su consistencia en esta etapa. Aparentemente, parece que la masa se seca, pero los cambios que ocurren se deben a la lenta absorción del agua libre por los componentes hidrofílicos, como son la proteína y el almidón de la harina (Pareyt y col., 2008a). Con un tiempo de espera de alrededor de 30 minutos, la masa se estabiliza y se reducen las diferencias entre un lote y otro (Manley, 2000).

Posteriormente, la masa se lamina. Durante este proceso se recomienda girar la masa 90° cada vez que se lamina para evitar la obtención de galletas de forma ovalada tras el horneado, ya que el gluten se alinea en la dirección del laminado (Fustier y col., 2008), por lo que la longitud de la galleta disminuirá sólo en la dirección de la laminación mientras que la anchura aumentará (Thacker, 1993).

En el proceso de horneado se producen numerosos cambios que modifican radicalmente la estructura de la galleta como son la desnaturalización proteica, la fusión de la grasa, las reacciones de Maillard, la evaporación del agua y la

expansión de los gases generados (Chevallier y col., 2002). Esto se traduce en tres variaciones importantes. En primer lugar la disminución de la densidad del producto unida al desarrollo de una textura abierta y porosa. Posteriormente la reducción del nivel de humedad hasta 1-4% y finalmente un cambio en la coloración de la superficie (Manley, 2000).

Durante el horneado existe un solapamiento de procesos (Manley, 2000). La grasa es lo primero que funde y da a la masa un carácter plástico (Pareyt y col., 2008a); de hecho, las masas con mayor cantidad de grasa fundida durante la cocción se esparcirán más (Hoseney, 1994), retrasando por otra parte la acción de los agentes leudantes que liberarán gases y se expandirán. La expansión viene seguida de un colapso (Chevallier y col., 2000b), que marcará el diámetro final de la galleta. El almidón y las proteínas también sufren un proceso de calentamiento hinchándose y, en algunos casos, sufriendo una desnaturalización. También el agua presente en la masa se evapora contribuyendo a la expansión. La pérdida de humedad en la superficie de la galleta está relacionada con la temperatura en superficie. El azúcar contribuye a disminuir la viscosidad de la masa (Manley, 2000) y forma una estructura de masa no coagulada al subir la temperatura (al contrario que ocurre en otras masas como la de pan), por lo que durante la cocción la masa se convierte en una estructura de matriz azucarada.

El final del horneado se define por dos hechos: el color y el contenido en humedad, que están relacionados entre sí y se determinan muchas veces por un examen visual y determinación de la humedad, respectivamente (Wade, 1988).

Posteriormente al horneado, las galletas necesitan enfriarse para terminar de perder humedad y de estructurarse la matriz (Manley, 2000). De hecho, Burt y Fearn (1983) concluyeron mediante un análisis estereológico que la distribución de los componentes mayoritarios (grasa, proteína y almidón) era completamente homogénea en la galleta después de este enfriamiento.

3. Ingredientes de las galletas

3.1. Ingredientes mayoritarios

3.1.1. Harina

La harina es el ingrediente mayoritario de las galletas. La harina se produce tras la molienda del grano de trigo (botánicamente llamado cariósipide). En la Figura 1 se muestra un esquema simple del grano de trigo, formado básicamente por tres partes. Las capas exteriores, de color rojizo se llaman salvado, el centro blanco o amarillento endospermo y el diminuto embrión llamado germen (Manley, 2000).

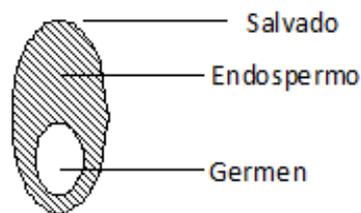


Figura 1. Partes de un grano de trigo.

La harina se compone principalmente de una mezcla de fragmentos de endospermo junto con gránulos de almidón y algunos fragmentos de proteína (Wade, 1988). En particular, las harinas de trigo débil son una mezcla de constituyentes como almidón (70-75%), proteínas (8-11%), lípidos, varios polisacáridos no almidonáceos como las pentosanas y una pequeña cantidad de agua (14%) (Fustier y col., 2008).

La mayoría de los gránulos de almidón presentes en la galleta se encuentran sin gelatinizar debido a la poca cantidad de agua presente en la masa y a la presencia de azúcar (Chevallier y col., 1999), de manera que el almidón estaría rodeado de los otros ingredientes y actuaría de “relleno” (Wade, 1988). Sin

embargo, este hecho no es homogéneo en toda la galleta. Chevallier y col. (2000a) afirmaron que el almidón está más hidrolizado en el centro que en la periferia y superficie de la galleta donde el gránulo se mantiene intacto y guarda su birrefringencia.

La proteína más importante de la harina es el gluten. Como ya se ha comentado, el contenido en gluten de las harinas utilizadas en la industria galletera es bajo (7-9%) (HadiNezhan y Butler, 2009). Una proporción adecuada de agua y harina, como ocurre en la fabricación de pan, hace que el gluten forme una masa viscoelástica (Pareyt y col., 2008a). Sin embargo, en el caso de las galletas de masa corta, donde hay poca cantidad de agua y sustancias que interfieren como la grasa o el azúcar, el gluten no es capaz de hidratarse (Gaines, 1990). Aun así, la presencia de gluten es uno de los factores que más afecta al diámetro de las galletas. De hecho, en la galleta el contenido en gluten se relaciona con el diámetro final de la misma, de tal forma que el diámetro de las galletas disminuye conforme aumenta el contenido de gluten (Pareyt y col., 2008b; Kaldy y col., 1993).

3.1.2. Grasa

La grasa es un ingrediente esencial en la fabricación de galletas y tras la harina es el segundo componente mayoritario en la formulación de la galleta (Sai Manohar y Haridas Rao, 1999b). El uso de grasa en la masa de galleta hace que la cantidad de agua necesaria para hacer la masa sea pequeña (Wade, 1988; Sai Manohar y Haridas Rao, 1999b), siendo la grasa el ingrediente responsable de la unión de todos los ingredientes (Pareyt y col., 2008a). Durante el amasado, la grasa actúa como lubricante y rodea la superficie de la harina inhibiendo la creación de una red cohesiva y extensible de gluten (Wade, 1988); además, la grasa presente en la masa de galleta rodea también los gránulos de almidón, rompe la continuidad de la estructura proteína-almidón (Ghotra y col., 2002) y afecta a la textura de la masa, de forma que la masa es menos elástica y no se encoge tras su laminación (Baltsavias, 1997b; Maache-Rezzonug y col., 1998). Por tanto, la grasa influye en el diámetro y en las

propiedades finales de textura de las galletas (Pareyt y col., 2009a), confiere a la galleta humedad y aumenta la fragilidad de la galleta (Maage-Rezzoug y col., 1998).

El preparado de grasa utilizado en la fabricación de galletas suele contener un 78% de materia grasa compuesta por grasa vegetal, aceites vegetales y aceites vegetales hidrogenados, aunque también contiene emulgentes (lecitina, mono y diglicéridos de ácidos grasos) que actúan rompiendo la grasa en partículas muy pequeñas (Oreopoulou, 2006). En la industria se le acuna con el nombre de “shortening” debido a que su adición inhibe la formación de una masa elástica, acortándola, de ahí que en inglés se traduzca como “short”, confiriéndole ciertas propiedades texturales al producto final (Pareyt y col., 2008a).

3.1.3. Azúcar

El azúcar mayoritariamente empleado en la elaboración de galletas es la sacarosa en forma cristalina, que es un disacárido no reductor (α -D-glucopiranosil-(1 \rightarrow 2)- β -D-fructofuranosa).

Desde el punto de vista sensorial, el azúcar en las galletas afecta al gusto, color, dimensiones, dureza y superficie de la galleta (Gallagher y col., 2003). Además, la cantidad y el tipo de azúcar influyen durante todo el proceso, desde el amasado hasta el envasado. En el proceso de mezclado de ingredientes, el azúcar compite con la harina por el agua inhibiendo la formación de gluten (Gallagher y col., 2003) y afectando, por tanto, a la consistencia de la masa (Olewnik y Kulp, 1984; Slade y Levine, 1994), que es fundamental en el momento del laminado y corte. Durante el horneado, el azúcar también influye en la gelatinización del almidón (Spies y Hosenev, 1982), en las reacciones de pardeamiento (Kulp y col., 1991), en la movilidad del gluten (Pareyt y col., 2009a), en la expansión de la galleta y en el carácter crujiente (Kulp y col., 1991). En el horneado no hay suficiente agua para disolver el azúcar añadido (Manley, 2000; Pareyt y col., 2008a), ya que el calor no se distribuye homogéneamente en toda la masa de galleta, de forma que los gránulos de

azúcar en el centro de la galleta se pueden observar en forma amorfa y en forma cristalina, mientras que los gránulos de azúcar en la superficie únicamente están en forma cristalina debido a que en la superficie el agua se evapora rápidamente durante el proceso de horneado (Chevallier y col. 2000a).

3.2. Ingredientes minoritarios de las galletas

3.2.1. Agua

El agua es un ingrediente clave durante el proceso de fabricación de las galletas a pesar de ser un ingrediente minoritario en el proceso de fabricación y ser casi totalmente eliminado durante el horneado (Pareyt y col., 2008a).

En la galleta el agua actúa como plastificante y disolvente, además influye en la viscosidad de la masa y en la textura una vez horneada. En la primera parte del amasado el agua actúa disolviendo algunos de los ingredientes llegando a dispersarse en la grasa, por eso la mezcla de la masa final tiene un color crema claro y una consistencia blanda, de ahí el nombre de “punto pomada” (Wade, 1988).

La cantidad de agua influye en la consistencia final de la galleta, de forma que, las galletas con una baja humedad son más frágiles, y a medida que se aumenta la cantidad de agua, el punto de fractura de la galleta disminuye, revelando una mayor elasticidad y deformabilidad (Baltsavias et al, 1999a). Por otra parte, el aumento en la cantidad de agua se ha asociado a masas más cohesivas y adhesivas dando lugar a galletas más duras (Sai Manohar y Haridas Rao, 1999a).

3.2.2. Sal

El contenido en sodio de la sal utilizada mejora las propiedades sensoriales al disminuir el sabor amargo y aumentar el dulzor (Keast y col., 2003). Su concentración más eficaz en las galletas es de 1-1,5% del peso de la harina, ya que a niveles superiores a 2,5% se hace desagradable al gusto (Manley, 2000).

3.2.3. *Leche en polvo*

La leche contribuye en la textura, gusto, color de la superficie y aporta un valor nutricional extra. La presencia de aminoácidos provenientes de la leche favorece las reacciones de pardeamiento durante el horneado, contribuyendo a la obtención del color y el aroma deseados (Wade, 1988).

Actualmente la mayoría de la leche utilizada es en polvo dada su facilidad de manejo y bajo contenido en humedad que prolonga su vida útil (Manley, 2000).

3.2.4. *Agentes leudantes*

Bicarbonato sódico (CO_3HNa)

En presencia de humedad, el bicarbonato reacciona con el agua produciendo anhídrido carbónico al formarse sal sódica y agua. Al calentarse, el bicarbonato libera algo de dióxido de carbono y permanece como carbonato sódico, actuando como agente esponjante, además controla el pH que puede afectar en el esparcimiento de la masa y en el color de la galleta (Manley, 2000).

Bicarbonato amónico ($CO_3(NH_4)$)

El bicarbonato amónico se descompone completamente por el calor desprendiendo anhídrido carbónico, amoníaco gaseoso y agua. Por tanto, se disuelve rápidamente produciendo un medio alcalino que hace que la masa sea muy blanda. Al igual que el bicarbonato sódico, también actúa como agente esponjante (Manley, 2000).

3.3. Ingredientes utilizados en la reformulación de galletas

El consumidor de hoy en día tiene una gran preocupación por su salud y bienestar (Berasategi, 2010), por eso, busca alimentos bajos en calorías y saludables (Baltsavias y col., 1997a).

Existen evidencias científicas que relacionan la ingesta de numerosos componentes esenciales y no esenciales de la dieta y la prevención de

diferentes enfermedades (Milner, 2002). La mayoría de los productos de panadería se pueden utilizar como vehículo de ingredientes nutricionalmente saludables (Sudha y col., 2007).

3.3.1. Fibra

El consumo de fibra en los países occidentales es bajo (Baixauli 2008a, 2008b). El interés en alimentos con alto contenido de fibra en las últimas décadas se ha incrementado, convirtiéndose en alimentos con un amplio mercado.

La fibra puede definirse como una mezcla compleja de diferentes sustancias de origen vegetal que son resistentes a la hidrólisis por las enzimas digestivas del ser humano (Salas-Salvadó y Megias-Rangil, 2004).

La fibra dietética puede clasificarse en soluble e insoluble proveyendo ambas funciones fisiológicas específicas y beneficios nutricionales. La fibra insoluble promueve el movimiento de la materia a través del sistema digestivo y la fibra soluble ayuda a disminuir el colesterol en sangre, así como regular los niveles de glucosa en sangre (Tosh y Yada, 2010). Ambos tipos de fibra han sido utilizadas en la fabricación de galletas.

El salvado de cereal como fuente de fibra para reemplazar harina en galletas ha sido uno de los más utilizados. Numerosos autores han utilizado granos de cebada en galletas como fuente de fibra (Prentice y col., 1978; Örtürk y col., 2002). Gujral y col. (2003) reemplazaron parte de la harina de trigo con salvado de trigo aumentando así la percepción sensorial y disminuyendo la resistencia a la fractura. Leelavathi y Rao (1993) también reemplazaron la harina de galleta por fibra cruda y tostada, consiguiendo substituir hasta el 30% sin disminuir la calidad sensorial de la galleta. Recientemente, Ellouze-Ghorbel y col. (2010) utilizaron diferentes fuentes de salvado de trigo (*Aestivium and Durum*) para enriquecer galletas. Sudha y col. (2007) utilizaron diferentes salvados de cereales obteniendo buena aceptación las galletas con un de 30% de salvado de avena en la formulación o 20% de salvado de cebada.

Además de salvado de cereales, en el intento de incorporar fibra a las galletas también ha sido muy popular la utilización de fibra de frutas, como por ejemplo, la fibra de manzana (Chen y col., 1988), la fibra de plátano (Fasolin y col., 2007) o la fibra de mango (Ajila y col., 2008). Incluso se han llegado a mezclar en una misma formulación diversos tipos de fibra: manzana, limón, salvado de trigo y fibra de trigo (Bilgiçli y col., 2007).

Sin embargo, se conoce bien que los consumidores perciben a menudo la fibra como un sabor fuerte y desagradable, con una textura gruesa, color oscuro y con una sensación de sequedad en la boca (Yue y Waring, 1998). Un tipo de fibra que no produce estos efectos indeseables asociados al empleo de las fibras tradicionales es el almidón resistente. El almidón resistente tiene un tamaño de partícula pequeño, lo que evita la textura y densidad propia asociada a los productos con alto contenido en fibra. Es de color blanco lo que permite su incorporación sin alterar el color y es de sabor suave. Además, posee beneficios fisiológicos similares a los de la fibra soluble. El almidón resistente se define como el almidón y la suma de los productos de degradación del almidón que no se absorben en el intestino delgado de individuos sanos (Asp, 1992). Existen cuatro tipos de almidón resistente: tipo I, es el almidón físicamente inaccesible encontrado de forma natural en los alimentos; tipo II, es el almidón nativo granular; tipo III, es el almidón retrogradado o cristalino y tipo IV, es el almidón químicamente modificado (Champ, 2004).

Aparicio-Sanguilán y col. (2007) utilizaron en galletas un producto rico en almidón resistente proveniente de banana sin obtener diferencias significativas de preferencia entre la galleta control y las galletas con almidón resistente.

3.3.2. Sustitutos de grasa

La Comunidad Económica Europea (CEE), en su política de nutrición, sugiere que únicamente el 20-30% de la energía ingerida debe de ser proveniente de la grasa (O'Connor, 1992), ya que existen evidencias de que una mayor ingesta está relacionada con enfermedades coronarias (LaRosa y col., 1990), además

de obesidad, cáncer y colesterol alto en sangre (Akoh, 1998). Por eso, la OMS en 2004 sugirió a la industria alimentaria reducir el contenido graso en los alimentos con el fin de disminuir la obesidad y los problemas derivados en el primer mundo.

Los sustitutos de grasa son sustancias de origen proteico o hidrocarbonado que pueden mimetizar las propiedades funcionales y sensoriales de la grasa pero con un menor contenido calórico (Zoulias y col., 2002a).

Los carbohidratos utilizados como sustitutos de grasa, como los almidones procesados, imitan la grasa al absorber el agua dando así lubricidad, cuerpo y una sensación placentera en boca en las galletas y otros productos horneados (Bath y col., 1992; Nonaka, 1997); además, todos ellos aportan entre 0 y 4 kilocalorías por gramo, es decir menos energía que las grasas (9 Kcal. por gramo).

Hasta el momento, se han investigado numerosas sustancias como sustitutos de grasa en galletas como los β -glucanos (Inglett y col., 1994), mezclas de povidexrosa, monoglicéridos y ésteres de ácidos grasos (Campdell y col., 1994, Sudha y col., 2007), maltodextrinas (Zoulias y col., 2002a, Sudha y col., 2007) o inulina (Zoulias y col., 2002ab; Zbikowska y col., 2008). Además, existen otras sustancias como mezclas de dextrinas y almidón, o derivados de celulosas que pueden ser utilizados potencialmente como sustitutos de grasa aunque no se han aplicado hasta el momento en galletas.

3.3.3. *Sustitutos de azúcar*

El alto consumo de azúcar está ligado a desórdenes de la salud como obesidad, problemas dentales o diabetes tipo II (Pareyt y col., 2009b). La reducción del azúcar en galletas es una buena manera de obtener un producto con menos calorías y más saludable (Drewnowski, 1998).

Diversos autores han estudiado el reemplazo de azúcar en galletas utilizando polioles (Olinger y Velasco, 1996; Zoulias, 2000), azúcares reductores como la fructosa (Sai Manohar y Haridas Rao, 1997), inulina (Gallagher y col., 2003), xilosa y glucosa, (Kweon y col., 2009) o arabinosilano (Pareyt y col., 2011). No

obstante, el entendimiento de la depreciación de la calidad al utilizar sustitutos de azúcar en galletas sigue siendo un reto para la industria.

4. Técnicas para evaluar la calidad de galletas

4.1. Reología

La palabra reología, etimológicamente, significa estudio del flujo (del griego reos: fluir y los: tratado, ciencia) y fue definida por Bingham en 1930 como “la rama de la física cuyo objetivo es el conocimiento fundamental y práctico de la deformación o flujo de la materia debido a la acción de fuerzas mecánicas externas” (Hernández y col., 2006).

Desde un punto de vista reológico se puede definir un comportamiento elástico (característico de los sólidos) y un comportamiento viscoso (característico de los fluidos) (Hernández y col., 2006). Por su parte el comportamiento viscoelástico es aquel que se caracteriza por poseer propiedades elásticas y viscosas simultáneamente. El comportamiento viscoelástico se puede medir instrumentalmente mediante ensayos reológicos oscilatorios y mediante ensayos de fluencia/relajación, entre otros.

La reología alimentaria es la extensión de esta disciplina a los productos alimentarios. De esta forma White (1970) define la reología alimentaria como “el estudio de la deformación y flujo de los materiales frescos, productos intermedios y productos finales de la industria alimentaria”.

En masas, el estudio de la reología es importante porque es un producto en constante cambio (Faubion y Faridi, 1986), es decir, aunque se dejase la masa reposar transcurrido un tiempo se podrían observar cambios; de igual forma que cuando se le aplica un proceso, como el laminado, se producen cambios en la viscoelasticidad de la misma. Por tanto, durante el procesado de las masas hay que tener en cuenta los valores óptimos de concentraciones de ingredientes, de tiempo de mezcla ingredientes, de espesor de laminado, etc., que es necesario controlar para que la calidad final del producto no se vea perjudicada (Faubion y Faridi, 1986).

Las propiedades viscoelásticas de la masa de galletas dentro de la zona lineal han sido estudiadas por numerosos autores. Baltsavias y col. (1997b) evaluaron las propiedades viscoelásticas lineales de la masa de galletas con diversas composiciones (variando la cantidad de los ingredientes de la masa:

harina, almidón, grasa, azúcar, agua y sal) concluyendo que la grasa y su estado (líquido o sólido) era un factor fundamental que afectaba a la rigidez y esparcimiento de la masa. Posteriormente, Papantoniou y col. (2003, 2004) estudiaron los efectos de los lípidos de la harina y su influencia en la viscosidad, y encontraron que la harina desgrasada poseía una mayor viscoelasticidad.

La combinación de ensayos oscilatorios y ensayos de fluencia-relajación en masa de galletas fue utilizada por Pedersen y col. (2004) para correlacionar la utilización de harinas de diferentes cultivos con los cambios observados en las dimensiones de galletas y en la viscoelasticidad de las masas. Posteriormente, estos autores también estudiaron los cambios en las propiedades viscoelásticas tras la adición de metasulfito de sodio o proteasas (Pedersen y col. 2005).

4.2. Textura

La textura de los alimentos se define como “la manifestación sensorial y funcional de la estructura, propiedades mecánicas y de superficie de alimentos percibidas por los sentidos de la visión, oído, tacto y cinestésicos” (Szczesniak, 2002). De esta definición, se concluye que la textura es una propiedad sensorial por lo que sólo puede ser juzgada, percibida y descrita por el ser humano. Sin embargo, instrumentalmente se pueden medir determinados parámetros físicos que proporcionan información sobre la textura de los alimentos.

Para los consumidores, la textura junto con el sabor y el color es una de las propiedades fundamentales que van a influir en la elección de unas galletas u otras (Mandala y col., 2006). De manera instrumental la textura se mide con un texturómetro, que es un instrumento desarrollado para medir el comportamiento mecánico de los alimentos. Se pueden realizar diferentes tipos de ensayos adaptando células de medida de diferente geometría (Hernández y col., 2006). Específicamente, para la medida de la textura en galletas se han utilizado ensayos de punción (Gaines, 1991, Sai Manohar y Haridas Rao, 1997,

Mandala y col., 2006), compresión (Sai Manohar, 1999b) y ensayos de flexión y rotura (Gaines, 1991; Baltsavias y col., 1999c, Saleem y col., 2005).

Además de la importancia de estudiar las propiedades mecánicas de las galletas, el estudio del sonido producido al romper o ser triturada es crucial para obtener un mayor entendimiento de la textura de las mismas. El estudio del sonido de alimentos ha sido estudiado de forma instrumental por varios científicos en relación con sus propiedades texturales (Drake, 1963, 1965ab; Drake y Halldin, 1974; Vickers y Bourne, 1976; Vickers y Wasserman, 1979).

Iles y Elson (1972) mostraron que los consumidores clasificaban los productos en el mismo orden según el sonido emitido y su preferencia. Desde entonces, la industria alimentaria ha considerado de gran interés el estudio de la emisión de sonido durante la producción y el almacenamiento (Roudat y col., 2002).

Entender y definir la terminología para describir las sensaciones asociadas a la emisión de sonido difiere según la lengua utilizada (Varela y col., 2008). De tal forma que, mientras que para la lengua japonesa existen numerosas expresiones (Yoshikawa y col., 1970) para la lengua castellana o el inglés son mucho más reducidas siendo *crocante* (para el inglés: *crunchy*) y *quebradizo* o *crujiente* (para el inglés: *crispy*) las generalmente más utilizadas (Varela y col., 2008). La diferencia entre estos dos grupos (*crocante* y *crujiente*) fue estudiada por Vickers (1984) que los separó en función de la frecuencia del sonido emitido. Frecuencias altas (*higher pitched sounds*) que producían sonidos agudos se relacionaron con alimentos crujientes (*crispy*) como, por ejemplo, una papa y frecuencias bajas (*lower pitched sounds*) se relacionaban con alimentos *crocantes* (*crunchy*) como, por ejemplo, una almendra.

La caracterización instrumental del sonido se realiza comúnmente mediante la utilización de un texturómetro y un micrófono acoplado al mismo, sometiendo a los alimentos a diferentes deformaciones: compresión, flexión o penetración (Castro-Prada y col., 2007). Esta combinación permite registrar las propiedades mecánicas y acústicas simultáneamente (Drake, 1963; Vickers, 1976). La combinación del análisis de fractura y las emisiones acústicas permite una mayor comprensión del carácter crujiente de los alimentos, de forma que se

estudia como es la fractura y que eventos de sonido la acompañan (Castro-Prada y col., 2007).

4.3. Propiedades térmicas

La mayoría de alimentos procesados han sufrido un tratamiento térmico, como ocurre durante el horneado de las galletas, produciéndose cambios en los ingredientes y su función, así como interacciones entre ellos.

Para la medida de estos cambios se utilizan técnicas de calorimetría diferencial de barrido (DSC), donde la muestra y una referencia se calientan de forma independiente midiéndose la diferencia en el flujo de calor para mantener una temperatura igual en ambas muestras (Sandoval y col., 2005).

La calorimetría diferencial de barrido ha sido ampliamente utilizada como técnica para la caracterización de los cambios térmicos asociados al almidón, los cuales poseen un gran impacto en la textura de los alimentos que lo contienen (Biliaderis y col., 1983).

La gelatinización del almidón ocurre cuando en exceso de agua se produce un cambio de un estado semi-cristalino a un estado amorfo (Sandoval y col., 2005). La determinación de esta entalpía (contenido de calor en un sistema por unidad de masa) se realiza integrando el área endotérmica del termograma obtenido (Sahin y Gülüm Sumnu, 2005).

Diversos autores han estudiado la gelatinización del almidón en galletas (Baltsavias y col., 1999a; Chevallier, 2002) concluyendo que ni la temperatura alcanzada en la cocción ni la cantidad de agua presente en la formulación es suficiente para una completa gelatinización del almidón contenido en la harina de las galletas, encontrándose mayoritariamente almidón sin gelatinizar en la superficie de las galletas (menor agua disponible) y almidón parcialmente gelatinizado en el centro de éstas.

Por otra parte, la transición vítrea en los alimentos está siendo estudiada por su relación con las características fisicoquímicas del alimento. En materiales complejos la medida de la transición vítrea requiere de un calorímetro diferencial de barrido con opción de termomodulación, que permite la

separación de las diferentes respuestas térmicas obtenidas (Kasapis, 2004). La transición vítrea es un cambio reversible en la región amorfa de un polímero desde o hacia una condición gomosa o desde o hacia una condición relativamente dura o quebradiza. La transición vítrea del gluten y su dependencia con el contenido en humedad fue observada por DSC por primera vez en 1984 (Levine y Slade, 1990). Posteriormente, en galletas tipo “crackers” se relacionó la transición vítrea (supuestamente del gluten) con los cambios en las propiedades mecánicas y con la pérdida del carácter crujiente (Nikolaidis y Labuza, 1996).

4.4. Propiedades sensoriales

Stone y Sidel (2004) definen el análisis sensorial de los alimentos como “el método científico usado para evocar, medir, analizar e interpretar las reacciones a determinadas características de los alimentos tal y como son percibidos por los sentidos de la vista, olfato, tacto, gusto y oído”.

Existen distintos tipos de pruebas sensoriales en función de la información que necesitemos obtener. En la reformulación de alimentos resulta imprescindible, por una parte, conocer los cambios sensoriales producidos por la adición de nuevos ingredientes realizando pruebas descriptivas (Meilgaard y col., 1991), así como conocer la aceptación de los nuevos alimentos reformulados por parte de los consumidores mediante pruebas de aceptación (van Kleef y col., 2006).

Entre todas las pruebas descriptivas, el análisis cuantitativo descriptivo (QDA, en sus siglas en inglés) fue desarrollado por Stone y col. (1974) y es una de las pruebas más utilizadas para caracterizar un producto, aportando una terminología propia que lo define. En general, el objetivo primordial de dicho análisis es encontrar un mínimo número de descriptores que contengan un máximo de información sobre las características sensoriales del producto. Este análisis se basa en la detección y la descripción de los aspectos sensoriales cuantitativos por grupos de catadores que han sido entrenados previamente y han elaborado una terminología estandarizada para describir el producto. Estos

jueces o catadores expertos deben dar valores cuantitativos proporcionales a la intensidad que perciban de cada uno de los atributos evaluados durante el análisis descriptivo (Stone y Sidel, 2004).

El análisis descriptivo ha sido extensamente empleado en la evaluación de la dureza, textura o aroma de galletas (Brown y col., 1998; Brown and Braxton, 2000, Burseg y col., 2009) así como en la reformulación de galletas con bajo contenido en sal y alto contenido en fibra (Vázquez y col., 2009).

Además del QDA, dentro de los ensayos descriptivos, existen diversos métodos que además incluyen la temporalidad en la masticación del alimento como el ensayo de Tiempo-Intensidad (Pineau y col., 2009), el ensayo Tiempo-Intensidad Dual (del inglés "*Dual-time Intensitive*") (Duizer y col., 1997) o el Perfil-Progresivo (Jack y col., 1994).

Un nuevo método sensorial llamado Predominio Temporal de las Sensaciones (del inglés *Temporal Dominance of Sensations*, TDS) presenta a los jueces una lista completa de atributos de los que tienen que elegir la sensación dominante en cada momento del tiempo de masticación así como su intensidad (Pineau y col., 2009).

Hasta el momento la técnica TDS se ha utilizado para estudiar la percepción de algunos vinos (Meillon y col., 2010), bebidas calientes (Le Révérend, 2008) y productos lácteos líquidos (Pineau y col., 2009). También se han utilizado en productos sólidos como los copos de trigo (Lenfant y col., 2009) o en *nuggets* de pollo (Albert y col., 2012). Hasta el momento la técnica TDS no se ha aplicado al estudio de la percepción de los diferentes atributos dominantes en galletas ni otros productos de panadería.

Las pruebas de aceptación se utilizan para medir la evaluación del nivel de agrado o desagrado de una muestra por los consumidores. Se utiliza una escala hedónica, siendo una de las más utilizadas la desarrollada por Jones, Peryam y Thurston (1955). Su principal ventaja es su facilidad de entendimiento con mínimas instrucciones y su versatilidad para ser usada en

numerosos productos (Stone y Sidel, 2004), entre los que se incluyen las galletas (Larrea y col., 2005, Aparicio-Sanguilán y col., 2007).

La posibilidad de reformular galletas tecnológicamente viables y conducir las hacia productos de mayor calidad nutricional, hace necesaria la utilización de las técnicas expuestas para el análisis de la masa y de la galleta final. Además, un estudio sensorial de las galletas nos permitirá no únicamente una valoración de sus propiedades instrumentales, sino de la percepción real de los cambios y aceptabilidad por el ser humano.

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OBJETIVOS

OBJETIVOS DE LA TESIS DOCTORAL

El objetivo general de la presente tesis fue evaluar las propiedades físicas y sensoriales de galletas reformuladas con nuevos ingredientes ricos en fibra y sustitutos de sacarosa y grasa para conocer su estructura y obtener galletas con mayor valor nutricional, menor contenido calórico y buena aceptación sensorial.

Para la consecución de este objetivo general se establecieron los siguientes objetivos parciales:

1. Evaluar los cambios reológicos (propiedades viscoelásticas lineales), texturales y sensoriales cuando parte de la harina se sustituye por concentraciones crecientes de almidón resistente.
2. Evaluar los cambios reológicos (propiedades viscoelásticas lineales), texturales y sensoriales cuando parte de la harina se sustituye por concentraciones crecientes de fibra de trigo de dos longitudes y de fibra de manzana. Influencia de la dosis, morfología y solubilidad de dichas fibras.
3. Aplicar nuevas técnicas sensoriales en el estudio del procesado oral de galletas ricas en fibra y bajas en grasa y su relación con la aceptación por parte del consumidor.
4. Evaluar los cambios en las propiedades sensoriales de las galletas cuando se sustituye parte de la grasa por un ingrediente alto en dextrinas.
5. Estudiar el efecto de la sustitución de grasa por inulina e hidroxipropilmetilcelulosa en las propiedades mecánicas y acústicas de las galletas durante la fractura y su correlación con el análisis descriptivo cuantitativo sensorial y con la aceptabilidad.

6. Estudiar el efecto de la sustitución de azúcar por inulina y eritritol en las propiedades mecánicas y acústicas de las galletas durante la fractura y su correlación con el análisis descriptivo cuantitativo sensorial y con la aceptabilidad.
7. Estudio de la funcionalidad de la sacarosa en las propiedades térmicas, reológicas y texturales en un sistema modelo, en la masa de galleta (producto intermedio) y en la galleta (producto final). Determinación de la funcionalidad del eritritol y el maltitol como sustitutos de la sacarosa.

ESTRUCTURA DE LA TESIS

ESTRUCTURA DE LA TESIS

El trabajo de investigación realizado ha dado origen a diversas publicaciones científicas que responden a los objetivos planteados y cuyo contenido se presenta en los distintos capítulos de la presente tesis doctoral. Las referencias de las publicaciones y el capítulo en que aparecen son:

Capítulo 1.

Laura Laguna, Ana Salvador, Teresa Sanz and Susana M. Fiszman. (2011). Performance of a resistant starch rich ingredient in the baking and eating quality of short-dough biscuits. *LWT - Food Science and Technology* 44, 737-746.

Laura Laguna, María J. Hernández, Ana Salvador and Teresa Sanz. (2012). Study on Resistant Starch Functionality in Short Dough Biscuits by Oscillatory and Creep and Recovery Tests. *Food Bioprocess and Technology* DOI 10.1007/s11947-012-0785-x.

Laura Laguna, Teresa Sanz, Sarab Sahi and Susana M. Fiszman. (2012). Role of fibre morphology in some quality features of fibre-enriched biscuits. *International Journal of Food Properties* (aceptado).

Laura Laguna, Paula Varela, Ana Salvador and Susana Fiszman. (2012). A new sensory tool to analyse the oral trajectory of biscuits with different fat and fibre contents. *Food Research International* (aceptado).

Capítulo 2.

Laura Laguna, Paula Varela, Ana Salvador, Teresa Sanz, Susana M. Fiszman. (2012). Balancing texture and other sensory features in reduced fat short-dough biscuits *Journal of Texture Studies* 43, 235-245.

Laura Laguna, Paula Varela, Cristina Primo, Ana Salvador, and Teresa Sanz. HPMC and INULIN as fat replacers in biscuits: sensory and instrumental evaluation. LWT - Food Science and Technology (enviado).

Capítulo 3.

Laura Laguna, Cristina Primo-Martín, Ana Salvador and Teresa Sanz. (2012). Inulin and erythritol as sucrose replacers in short dough cookies. Sensory, fracture and acoustic properties. Food Bioprocess and Technology (enviado).

Laura Laguna, Katleen J.R. Vallons, Albert Jurgens, Teresa Sanz (2012). Understanding the effect of sugar and sugar replacement in short dough biscuits. Food Bioprocess and Technology, DOI: 10.1007/s11947-012-0968-5.

Los capítulos de la tesis se han estructurado en base a la funcionalidad del ingrediente utilizado en la reformulación de las galletas.

El capítulo 1 aborda los objetivos relacionados con el reemplazo de parte de la harina por fibra. Como fuentes de fibra se utilizaron almidón resistente, fibra de trigo y fibra de manzana. Se evalúan las propiedades reológicas de la masa para determinar la funcionalidad de las fibras sobre la estructura de la masa y de las galletas, así como las propiedades de textura y los cambios sensoriales que se producen en las galletas. Además, se aborda el estudio de la percepción sensorial de la textura durante la masticación y su relación con la aceptabilidad sensorial utilizando nuevas herramientas sensoriales no aplicadas anteriormente a este producto.

El capítulo 2 se centra en la resolución de los objetivos asociados a la sustitución de parte de la grasa empleada en la formulación de las galletas por nuevos ingredientes a base de carbohidratos. Se utilizaron: un ingrediente alto en dextrinas, hidroxipropilmetilcelulosa e inulina. En este capítulo se aborda el estudio del reemplazo parcial en las características de fractura analizando los

eventos de fuerza simultáneamente con los de sonido, tanto de forma instrumental como sensorial.

Por último, el capítulo 3 se centra en los objetivos relacionados con la reducción de la sacarosa. Se estudiaron los cambios en las propiedades físicas y sensoriales al utilizar inulina, maltitol y eritritol como sustitutos de sacarosa. Se evalúan las propiedades de fractura y sonido y se determinan los atributos sensoriales que mejor describen y caracterizan dichas galletas. Por otro lado, para un mayor entendimiento de los cambios que se producen en la estructura del producto al utilizar diferentes azúcares, se realiza un estudio más profundo con el diseño de un sistema modelo que permite predecir cuál es el sustituto de sacarosa más adecuado que aporta propiedades similares la sacarosa sin modificar la calidad de la galleta.

CAPÍTULO 1

PERFORMANCE OF A RESISTANT STARCH RICH INGREDIENT IN THE
BAKING AND EATING QUALITY OF SHORT-DOUGH BISCUITS

Laura Laguna, Ana Salvador, Teresa Sanz and Susana M. Fiszman

LWT- Food Science and Technology 44(2011) 737-746

Abstract

The effect of replacing part of the wheat flour with a resistant starch rich ingredient (RSRI) – a source of functional fibre with potential health benefits – was studied in short dough biscuits. A control with no replacement and 3 formulations in which 20, 40 and 60g of flour per 100g were replaced by an RSRI (samples 20RSRI, 40RSRI and 60RSRI) were prepared. From a technological point of view, the RSRI level influenced the consistency of the raw dough and the ease of sheeting and cutting. Regarding the eating quality of the final product, addition of the RSRI increased the breaking strength and crumbliness and reduced the resistance to penetration. In the RSRI biscuits, both the surface and the crumb were paler. The sensory acceptance of the 20RSRI biscuits did not differ significantly from that of the control. 40RSRI reduced the acceptability of the colour, appearance and texture without altering the taste, sweetness and overall acceptance. Neither of these two levels significantly reduced the consumption intention. However, 60% flour replacement produced biscuits with lower sensory acceptability and a significant reduction in consumption intention. In general, the results could be interpreted in terms of the protein-diluting effect of the added ingredient and changes in the water-retention capacity of flour mixtures containing RSRI. The present results proved that resistant starch rich ingredients (RSRI) have good potential for developing fibre-rich biscuits without changing their general features.

Key words: resistant starch, biscuits, flour replacement, baking performance, eating quality, acceptability

1. Introduction

A new trend is that consumers are demanding foods which display two main properties: the first is the traditional nutritional aspects of the food, the second is that additional health benefits are expected from its regular ingestion (Aparicio-Sanguilán, Sáyago-Avendi, Vargas-Torres, Tovar, Ascensio-Otero & Bello-Pérez, 2007).

The effect of dietary fibre on promoting health and preventing disease has been an issue of interest for many years and has become a subject of renewed research (Shahidi, 2000). The intake of fibre and fibre-containing foods remains low in many populations worldwide (Loening-Baucke, Miele, & Staiano, 2004). The interest in foods with high fibre contents has increased in recent decades and the importance of this food constituent has led to the development of a large market for fibre source ingredients in products such as bread, snacks, muffins or a number of types of biscuits that are conveniently consumed at breakfast (Nilsson, Ostman, Holst & Bjorck, 2008). As new ingredients emerge, there is a need to understand their functionality and their effects on formulation. This way of increasing fibre levels can be useful for ensuring that the population receives adequate amounts of fibre (Baixauli, Salvador & Fiszman, 2008a). Most bakery products can be used as vehicles for different nutritionally rich ingredients, permitting diversification (Sudha, Vetrmani & Leelavathi, 2007).

It is well known that consumers often perceive fibre as having a strong flavour, being unpalatable and possessing a coarse texture and a poor, dry mouth feel (Yue & Waring, 1998); these and other negative attributes such as dark colour and a masking of flavour are often associated with high-fibre baked products. Sources of soluble dietary fibre, such as oat bran, have been the focus of numerous new products. However, the level of oat bran in commercial baked products is usually low, possibly because bran adversely affects product texture compared to the original formulations (Hudson, Chiu & Knuckles, 1992).

Like fibre, resistant starch (RS) is being examined for both its potential health benefits and its techno-functional properties in foods. RS is the sum of starch

and products of starch degradation not absorbed in the small intestine of healthy individuals (Asp, 1992). Four types of RS have been identified (Champ, 2004): RS type I is a physically inaccessible starch found in starchy foods which are not fractionated and refined – mostly pulses and some cereals. RS type II refers to native resistant starch granules. RS type III comprises retrograded starches (Eerlingen, Jacobs, & Delcour, 1994; Eerlingen, Van den Broeck, Delcour, Slade, & Levine, 1994). Finally, RS type IV is made up of chemically modified starches with a far higher number of modifications than the usual chemically-modified starches authorised in Europe; type IV is authorised in Japan. RS has been shown to provide benefits such as increased digestive tract activity and the production of desirable metabolites like short-chain fatty acids in the colon (Yue & Waring, 1998).

Moreover, RS has become commercially available as RS-rich ingredients that can be used to produce foods of improved quality. Compared to conventional fibres, it has many advantageous features. It is a natural white source of dietary fibre, has a bland flavour and gives a better appearance, texture and mouth feel than other typical fibres (Baixauli, Salvador, Hough & Fiszman, 2008; Eerlingen, van Haesendonck, de Paepe, & Delcour, 1994).

Biscuits are the most popular bakery items which are consumed by nearly all levels of society. This is mainly due to their being ready to eat, of good nutritional quality and available in different varieties at an affordable cost.

The addition of fibre to biscuits has been studied for different fibre sources such as coconut residue (Khan, Hagenmaier, Rooney & Mattil, 1976), brewers' spent grain (Prentice, Kissell, Lindsay & Yamzaki, 1978), wheat bran (Leelavathi & Rao, 1993) and rice bran (Babcock, 1987; Saunders, 1989; Skurray, Wooldridge & Nguyen, 1986). Sudha et al. (2007) studied the effects of the addition of different cereal brans (wheat, rice, oat and barley); the biscuits with wheat bran (20%), oat bran (30%) and barley bran (20%) were highly acceptable. However, few papers deal with RS enrichment of biscuits. Aparicio-Sanguilán et al. (2007) used an experimental RS-rich product (RSRP) obtained

from lintnerized banana in biscuits; an affective test showed no difference in preference between the RSRP biscuits and the control sample without the RSRP. However, they did not perform any tests to ascertain the changes induced by this ingredient in the physical properties of the dough, during the baking procedure, or in the final product.

All the studies mentioned have shown the possibility of using biscuits as effective carriers of different sources of fibre in order to improve the daily fibre intake of human beings. Short dough biscuits constitute a simple system which consists of three major ingredients (flour, sugar and fat) and a small amount of water; they have high acceptability and are very popular in many countries. Furthermore, an increasing number of commercial resistant starch-rich ingredients is available.

The aim of this study was to evaluate the physical, textural and sensory changes that take place in short dough biscuits when increasing proportions of the flour are replaced by an RS-rich ingredient (RSRI). Additionally, the resistant starch was analysed in order to verify the actual RS content of the biscuits with the different formulations.

2. Materials and methods

2.1. Materials

Four formulations were prepared using the same quantity of all the ingredients except the wheat flour and RS. The proportions of these two ingredients were 100:0 (control), 80:20, 60:40 and 40:60; the latter three samples were named 20RSRI, 40RSRI, and 60RSRI respectively. The biscuit ingredients (percentages given on a dough basis) were: a) soft wheat flour suitable for biscuits (Belenguer,S.A., Valencia, Spain) (composition data provided by the supplier: 15% moisture, 11% protein, 0.6% ash; alveograph parameters $P/L=0.27$, where P =maximum pressure required and L =extensibility; and $W=134$, where W =baking strength of the dough) and b) RSRI, a source of

resistant starch (Hi-maize 260, National Starch Food Innovation, Manchester, UK, composition data provided by the supplier 10% moisture, 58% dietary fibre according to AOAC 991.43 method), together making up 50g/100g, c) shortening 30g/100g (St. Auvent, Vandemoortele France), d) sugar 15g/100g (Azucarera Ebro, Madrid, Spain), e) milk powder 0.3 g/100g (Central Lechera Asturiana, Peñasanta, Spain), f) salt 0.1 g/100g, g) sodium bicarbonate 0.1 g/100g (A. Martínez, Cheste, Spain), h) ammonium hydrogen carbonate 0.06 g/100g (Panreac Quimica, Barcelona, Spain) and i) tap water 4.44 g/100g; the water level thus remained constant in all the formulations.

2.2. Moisture content

The % moisture content of flour and RSRI was determined according to the Approved Method 44-15.02 (AACC International, 2009)

2.3. Alkaline water-retention capacity

The % alkaline water retention capacity (%AWRC) of the flour, the RSRI and the flour/RSRI mixtures (proportions 100:0, 80:20, 60:40 and 40:60) was calculated according to the Approved Method 56-10.02 (AACC International, 2009).

2.4. Biscuit preparation

The shortening was creamed in a mixer (Kenwood Major Classic, UK) for 4 minutes at minimum speed to obtain a homogenous cream. After this, the sugar was added and mixed in for 2 minutes at speed 4. The milk powder, previously dissolved in all the water, was added and mixed in for 2 minutes at the minimum speed. Finally, the flour (or flour/RS), sodium bicarbonate and ammonium hydrogen carbonate were mixed in together at minimum speed for 2 minutes. The dough was then sheeted using a rolling pin over a 300 x 240 x 16 mm (length x width x height) rectangular frame to ensure a sheeted dough of even height. The sheeted dough was allowed to rest for 30 minutes at 4°C before cutting it into rectangular pieces measuring 80 x 30 x 16 mm (length x width x height). Twelve pieces were placed on a perforated tray. The biscuits were

baked in a conventional oven for 6 min at 175 °C, then the trays were turned 180°, bringing the side that had been at the back to the front of the oven to ensure homogenous cooking, and baked for a further 6 min at the same temperature. The oven and the oven trays were always the same, the trays were placed at the same level in the oven and the number of biscuits baked was always the same. After cooling, the biscuits were packed and stored in heat-sealed metalized polypropylene bags. The biscuit samples were evaluated on the following day in all cases.

2.5. Dough characteristics

2.5.1. Density measurements

The dough density was calculated in three replicates as the weight of a piece of dough divided by the nominal dough piece nominal volume ($3.84 \times 10^{-4} \text{ m}^3$), expressed in g.cm^{-3} .

The sheeted dough (16-mm thick) from the different formulations was analysed. A TA-XT.plus Texture Analyzer equipped with the Texture Exponent software (version 2.0.7.0. Stable Microsystems, Godalming, UK) was used. A test speed of 1 mm.s^{-1} and a trigger force of 5g were used in all the tests. Each test was conducted on six replicates of each formulation.

2.5.2. Wire cutting measurements

Rectangles of biscuit dough measuring 80 x 30 (length x width), the same size as the biscuits, were sheared transversally through the middle with a wire cutter. The mean cutting force on the plateau region (N) was measured.

2.5.3. Sphere penetration measurements

Dough discs with a diameter of 45 mm were penetrated to a depth of 10 mm with a 0.5 mm-diameter spherical stainless steel probe (P/0.5). The maximum force (N) attained during penetration was measured.

2.5.4. Flat disc extensional compression measurements. Dough discs, 45 mm in diameter, were compressed up to 50% of their initial height using a 75 mm-

diameter aluminium plate (P/75). The maximum force (N) and the final diameter (mm) of the dough discs after compression were measured. The diameter gain was calculated (final diameter (mm) – 45mm).

2.6. Differential scanning calorimetry (DSC)

DSC measurements were performed with a Q2000 modulated DSC (TA-Instruments Inc., USA). Measurements were performed in the doughs and in the corresponding biscuits of the control and the 60RSRI samples. Freeze-dried samples were weighed and distilled water added at a 1:3 (w/v) sample to water ratio in large volume DSC pans (TA-Instruments Inc., USA). The samples were heated from 5 to 130°C at 10°C/min. The enthalpy was expressed in J/g of dried sample and in J/g of dried wheat starch.

2.7. Biscuit evaluation

2.7.1. Moisture content and aw

The moisture content of the biscuits was determined in three replicates of each formulation according to the Approved Method 44-01 (AACC International, 2009).

Water activity (aw) was determined in three replicates of each formulation, using a Decagon AquaLab meter (Pullman, WA, USA) calibrated with a saturated potassium acetate solution (aw=0.22).

2.7.2. RS and total dietary fibre (TDF) content

The RS content of the commercial ingredient (RSRI) and of the biscuits with different RSRI concentrations was determined according to the Approved Method 32-40 (AACC International, 2009). The method was carried out with a Megazyme Kit. Three replicates of biscuits from each formulation and of the resistant starch ingredient Hi-Maize 260 were incubated with alpha amylase and amyloglucosidase for sixteen hours in a shaking water bath, after which denatured alcohol was added and the RS from the samples was recovered with consecutive centrifugations and washed with denatured alcohol. The RS, which

was recovered in pellet form, was then dissolved (with KOH and subsequently with an acetate buffer) and incubated with amyloglucosidase. The D-glucose obtained was measured using a glucose oxidase/oxidase reagent in a spectrophotometer.

The total dietary fibre content of the doughs and the biscuits was determined in three replicates by the Approved Method 32-07 (AACC International, 2009), using a FOSS Fibretec E 1023 Filtration module and Shaking Water Bath 1024 system.

2.7.3. Physical characteristics of the biscuits

The biscuit density was calculated in six replicates as the weight of a biscuit divided by its volume, expressed in g.cm^{-3} .

The biscuit length and width were measured by placing 10 biscuits edge-to-edge (both vertically and horizontally). The biscuit thickness was measured by stacking 10 biscuits. Measurements were expressed in mm as the mean value/10 of three different trials. Changes in the dimensions were expressed as gains (+) in comparison with the initial dimensions of the biscuits before baking (80-mm long x 30-mm wide x 16-mm thick). Each biscuit was also weighed individually before and after baking.

2.7.4. Colour

Measurement of the upper surface and internal (crumb) colour of the biscuits was carried out with a Konica Minolta CM-35000d spectrophotometer. To measure the crumb colour, the biscuits were cut perpendicularly with a finely-serrated knife and the cut surface was measured. Four replicates of each formulation were measured. The results were expressed in accordance with the CIELAB system with reference to illuminant D65 and a visual angle of 10°. The parameters determined were L^* ($L^* = 0$ [black], $L^* = 100$ [white]), a^* ($-a^*$ = greenness, $+a^*$ = redness), b^* ($-b^*$ = blueness, $+b^*$ = yellowness).

Chroma (C_{ab}^*) is the attribute that allows the degree of difference in comparison to a grey colour of the same lightness to be determined for each hue, so it is

considered the quantitative attribute of colourfulness; Hue (h_{ab}) is the attribute according to which colours have been traditionally defined as reddish, greenish, etc. These two parameters were defined by the following equations:

$$C^*_{ab} = [(a^*)^2 + (b^*)^2]^{1/2}$$

$$h_{ab} = \arctan [b^*/a^*]$$

The total colour difference (DE^*) between the control biscuit and the different RSRI biscuits was calculated as follows:

$$DE^* = [(L^*_c - L^*_s)^2 + (a^*_c - a^*_s)^2 + (b^*_c - b^*_s)^2]^{1/2}$$

where subscript c = control and subscript s = samples containing the RSRI.

The values used to determine whether the total colour difference was visually obvious were the following (Francis & Clydesdale, 1975):

$$\Delta E^* > 3: \text{ colour differences are obvious to the human eye.}$$

2.7.5. Biscuit texture analysis

The texture of the biscuits was measured using the Texture Analyzer described above. A test speed of 1mm/s was used for all tests. Ten replicates of each formulation were conducted.

Breaking strength. Biscuits were broken using the three point bending rig probe (A/3PB). The experimental conditions were: supports 50 mm apart, a 20 mm probe travel distance and a trigger force of 20g. The force at break (N) and the gradient of the initial steep slope of the curve (N.mm) were measured.

Crumbliness. The biscuits were cut into 2-cm sized cubes with a finely-serrated knife. The cubes were compressed to 50% of their initial height using a 75-mm diameter aluminium plate (P/75) with a trigger force of 10g. The maximum force (N) during compression was taken as the Crumbliness Index.

Bite test. Penetration tests were conducted with the upper Volodkevich Bite Jaw (VB), penetrating the sample (whole biscuit) to 10 mm; a trigger force of 20 g was set. Two 'bites' were made in each biscuit (one third in from each end), so

a total of 20 values were registered for each formulation. The maximum force at penetration (N) was measured.

2.7.6. Consumer sensory analysis

A total of 103 untrained panellists (consumers) aged from 15 to 70 years who consumed this type of biscuit frequently took part in the study. 77 were female and 26 male. Each consumer received four biscuits, one for each RSRI content (control, 20RSRI, 40RSRI and 60RSRI), presented individually in a single session following a balanced complete block experimental design. The biscuits were coded with random three-digit numbers.

Consumer acceptance testing was carried out using a nine point hedonic scale (9 = like extremely; and 1 = dislike extremely). The consumers had to score their liking for the 'appearance', 'texture', 'colour', 'sweetness', 'taste ', and 'overall acceptance' of each biscuit sample.

2.8. Statistical analysis

Analysis of variance (one way-ANOVA) was applied to study the differences between formulations; least significant differences were calculated by the Tukey test and the significance at $p < 0.05$ was determined. These analyses were performed using SPSS for Windows Version 12 (SPSS Inc., USA).

3. Results and discussion

The source of resistant starch (RS) used was a thermally-modified Type II based on the *ae-VII* corn hybrid; the modification renders the native granule more stable when the starch is held at a high temperature in the presence of limited water, according to Haralampu (2000). Observation of this ingredient suspended in an excess of water and heated to 100°C for 5 minutes, viewed under the microscope using polarised light, showed that the starch granules had not lost their birefringence (data not shown), which is an indication of their resistance to gelatinisation.

3.1. *Dough characteristics*

The rheological properties of biscuit doughs are important, as they influence the machinability of the dough as well as the quality of the biscuits. Chevallier, Colonna, Buléon and Della Valle (2000) identified the structure of short dough as a suspension of proteins, starch-protein associations and isolated starch granules in a continuous sugar solution phase in which lipids are emulsified in a concentrated sugar solution. The fat has a lubricating function in the dough (so very little water is required to achieve the desired consistency) and restricts gluten formation, implying that physical properties of biscuit dough depend on the distribution of water and fat in the system. The %AWRC values for the flour, the 80/20, 60/40 and 40/60 flour/RSRI mixtures, and RSRI were 74.59, 81.02, 93.16, 106.9 and 129.8 g/g on a dry basis, respectively. Since these values rose significantly as the proportion of RSRI increased, the presence of the RSRI could be expected to reduce the water availability in the system, producing drier, harder doughs.

The biscuit dough density could also affect the dough's rheological properties, as it is considered an indicator of the amount of air incorporated into the dough during mixing (HadiNezhad & Butler, 2009). In the present study, the density of the control dough was significantly lower ($1.24 \pm 0.02 \text{ g/cm}^3$) than that of the three formulations with partial replacement of the flour with different levels of the RSRI; no significant differences were encountered between these three ($1.35 \pm 0.02 \text{ g/cm}^3$). It may therefore be considered that the air incorporation efficiency was greater when no RSRI was present.

Table 1. Dough characteristics obtained with penetrometer, wire cutter and 50% compression.

Dough Sample	Plateau force (N) (wire cutter)	Max force (N) (sphere penetration)	Compression up to 50%	
			Max force (N)	Diameter increase (mm)
Control	0.26 ^a (0.03)	1.54 ^a (0.21)	29.5 ^a (2.5)	+11 ^a
20RSRI	0.27 ^a (0.03)	1.40 ^a (0.04)	24.6 ^a (0.29)	+11 ^a
40RSRI	0.52 ^b (0.07)	1.98 ^b (0.12)	42.6 ^b (4.5)	+12 ^a
60RSRI	0.59 ^b (0.06)	2.81 ^c (0.32)	45.4 ^b (2.9)	+7 ^b

Values in parentheses are standard deviations. Means in the same column without a common letter differ ($p < 0.05$) according to the Tukey's test.

The wire cutting test involves pushing a wire through the specimens from an initial indentation to a steady-state cutting stage. The wire probe has the advantage of presenting a constant contact area with the sample, minimizing the friction effects (in comparison with blade cut, for instance) (Dunn, Burton, Xu, & Atkins, 2007). The force – displacement relationship obtained for the wire cutting test depends on a combination of fracture, plastic/viscous deformation and surface friction effects. The difference in force in the cutting path depends on the cohesive forces between the dough constituents. Typical force – displacement curves were obtained for the four formulations with two differentiated phases: in the first, the wire indents into the dough; in the second, the material starts to separate and a steady-state cutting phase proceeds to form a plateau (Goh, Charalambides & Williams, 2005). The force values of these plateaux (Table 1) represent dough-cutting ease and are also related to dough consistency. The 40RSRI and 60RSRI doughs showed significantly

higher cutting force plateau values than the control and the 20RSRI doughs. The curves for the highest RSRI level also showed higher fluctuation in the plateau values (Figure 1) which could be attributed to secondary sample microcracking. The plateau zone corresponds to the traction and separation of the material in the wire's path; the cut surface of the samples with the RSRI had a grainier visual appearance and was drier to the touch. These changes in consistency as the level of RSRI increases could be attributed to the lower moisture level and higher water retention in the doughs containing the RSRI.

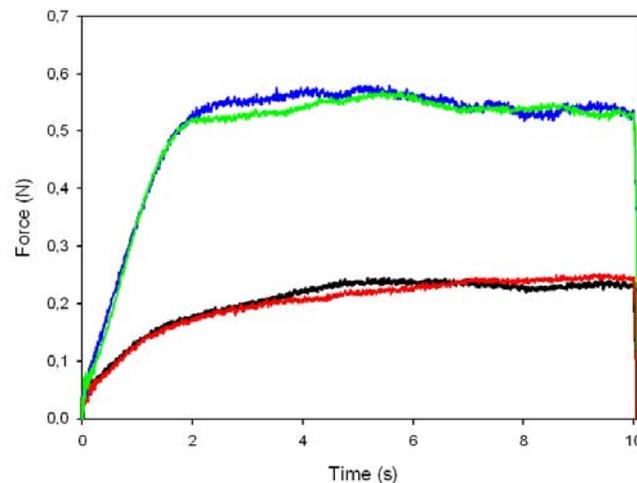


Figure 1. Effect of partial replacement of wheat flour with and RSRI on the wire shear resistance of biscuit dough. Black: control, red:20RSRI, blue: 40RSRI, green: 60RSRI.

The results for sphere penetration and flat disc extensional compression showed a similar evolution for RSRI addition (Table 1), which increased the consistency of the biscuit dough. In general, it seemed that a low level of RSRI (20 RSRI sample) hardly affected the textural characteristics of the dough but that higher RSRI levels had significant effects on dough rheology.

Flat disc extensional compression is a test that indicates dough yielding and viscous behaviour.

Yielding occurred under very small strain, where the systems predominantly behaved like solids; under a large strain they behaved more like shear thinning liquids; Baltasvias, Jurgens, and van Vliet (1999a) have stated that yielding behaviour is strongly influenced by intact flour particles that act as defects in the material.

The differences in extensional compression values can be attributed to differences in the amount of material compressed (HadiNezhad & Butler, 2009), since the samples containing the RSRI showed higher density; in addition, the differences could be explained by taking into account the higher AWRC of the RSRI samples: as more water was retained, its mobility and availability for gluten development were restricted; also, the RSRI samples contained less wheat protein, which contributes dough viscosity when hydrated. In a biscuit system model approach based on gluten–starch blends, Pareyt Wilderjans, Goesaert, Brijs and Delcour (2008) also found that dough hardness decreased when more gluten was added.

From a technological point of view, it is not advisable for the dough to be too stiff; however, none of the formulations presented any problems during mixing or rolling.

Gaines (1990) suggested that some functional associations of soft wheat proteins occur during biscuit dough mixing, affecting dough consistency to some (slight) extent. The diluting effect of the RSRI on the wheat proteins of the flour should give a less cohesive or structured dough; however, the higher AWRC of the flour/RSRI mixtures compared to flour seems to have had a major effect in the present case, producing harder doughs.

3.2. RS and Total Dietary Fibre content

The analyses indicated that the RSRI contained 39.1% RS as determined enzymatically. As expected, the RS content of the biscuits increased with the addition of RSRI (Table 2). These results confirm that the RSRI made a significant contribution to the final RS content of the formulated biscuits and that

baking did not alter the indigestibility of this ingredient. The TDF contents in biscuits (Table 2) were compared with the corresponding values in

doughs (4.40, 11.38, 13.88, and 15.40 g/g on a dry basis for control, 20RSRI, 40RSRI and 60RSRI respectively) showing a good recovery of TDF after baking. The biscuits have high fibre content and are a promising way to formulate fibre-enriched food.

Table 2. Biscuit characteristics for control and resistant starch-rich ingredient (RSRI) formulations.

Biscuit sample	Moisture (g/100g)	a_w	Water loss(g/100g)	RS content (g/100g)	TDF (g/100g)
Control	4.37 ^a (0.01)	0.50 ^a (0.0)	5.0 ^a (0.2)	0.78 ^a (0.08)	6.03 ^a (0.59)
20RSRI	5.51 ^b (0.49)	0.59 ^c (0.0)	4.2 ^b (1.2)	6.04 ^b (0.05)	13.67 ^b (0.75)
40RSRI	5.09 ^{ab} (0.05)	0.56 ^b (0.01)	4.0 ^b (0.4)	12.32 ^c (0.48)	17.41 ^c (1.02)
60RSRI	5.21 ^{ab} (0.13)	0.55 ^b (0.01)	4.2 ^b (0.2)	14.10 ^d (0.37)	22.83 ^d (2.6)

Values in parentheses are standard deviations. Means in the same column without a common letter differ ($p < 0.05$) according to the Tukey test.

3.3. DSC measurements

In order to evaluate the degree of starch gelatinization after cooking and to understand the contribution of starch to the structural characteristics of the biscuits, DSC measurements were performed. Representative DSC thermograms corresponding to the doughs and the biscuits of the control and the 60RSRI samples are shown in Fig. 2.

All the samples showed an endothermic peak at 70°C, which corresponds to the melting of starch crystals. The presence of this peak in the biscuits implied that part of starch granules from the wheat flour remained ungelatinized after the baking process.

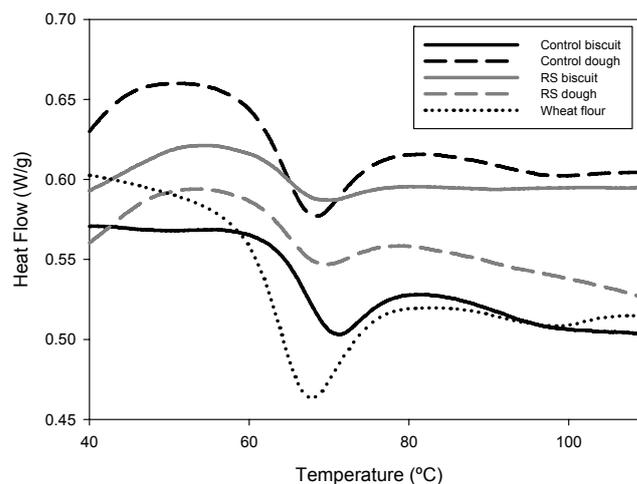


Figure 2. DSC thermograms of control dough and biscuit, and 60RSRI dough and biscuit

The temperature and the enthalpy of melting of starch crystals, expressed per g of dried sample and per g of dried starch, are shown in Table 3. In the dough samples the peak appeared at a lower temperature (68.34 °C and 69 °C, control and 60RSRI, respectively) than in the biscuits (71.31 °C and 70.07 °C, control and 60RSRI, respectively). This increase in the gelatinization temperature is explained due to the heat-moisture treatment that occurs during biscuit baking. Heat-moisture treatment of starches has been shown to produce an increase in the gelatinization temperature (Hoover, Vasanthan, Senanayake Martin, 1994; Jacobs & Delcour, 1998).

Table 3. Temperature and enthalpy of starch gelatinization (ΔH) o doughs and biscuits corresponden to the control and a resistant starch rich ingredient formulation.

Type of sample	T (°C)	ΔH (J/g dried sample)	ΔH (J/g dried starch)
Control dough	68.34	3.50	8.10
Control biscuit	71.31	2.25	5.21
60RSRI dough	69.59	1.43	8.50
60RSRI biscuit	70.07	1.12	6.65

The control dough showed higher enthalpy per weigh of dried sample than the 60RSRI, which is associated to the lower starch content present in the 60RSRI (wheat flour was replaced by RS). As expected, the enthalpies expressed per weigh of dried wheat starch were very similar in both doughs (8.1 and 8.5 for the control and 60RSRI, respectively), as the difference in both doughs were related to the different amount of raw wheat starch present.

The comparison of the enthalpies per weight of the dried samples, or per weight of dried wheat starch, reflected a decrease in the transition energy in the biscuits in comparison to the corresponding doughs (same wheat starch content), which reflects and increase in the extent of starch gelatinization the biscuits in comparison to the corresponding doughs. The decrease in the enthalpy was higher in the control than in the 60RSRI samples, which reflects the lower starch content available for gelatinization in the 60RSRI.

The thermograms also reflected the presence of a peak at 100°C attributed to dissociation of amylase-lipid complexes in all the samples.

3.4. Biscuit evaluation

3.4.1. Moisture content and water activity

Both the moisture content and the water activity were higher in the biscuits with the RSRI than in the control (Table 2). This could be attributed to the higher water retention capacity of the system, which increased in line with the proportion of the RSRI (see section 3.1). Aparicio-Sanguilán et al. (2007) also obtained an increase in moisture and water activity for biscuits enriched with a RS-containing product extracted from bananas; the increase was related to each material's relative abundance of amorphous starch zones, which had a considerable influence on water absorption.

3.4.2. *Physical characteristics*

Biscuits comprise a matrix and the biscuits' properties are mainly determined by the volume of air spaces and fat globules, as well as the level of inhomogeneities (Błaszczak Fornal & Ramy, 2004). The baking process transforms the dough into a cellular solid with a characteristic final texture (Chevallier et al., 2000). Because of the CO₂ gases produced by the raising agents and water evaporation, biscuits undergo a substantial expansion early in baking. The degree of spread is controlled by the spread rate and the set time which, in turn, depend on the level of water in the dough that is free to act as a solvent and the strength of the dough (Ram & Singh, 2004). Levine and Slade (1990) stated that “good” biscuits facilitate expansion without significant functional network formation followed by structural collapse into a rubbery, thermoplastic polymer system. Pareyt et al. (2008) and Kweon, Slade, Levine, Martin and Souza (2009), among others, have recorded changes in biscuit geometry during baking by taking time-lapse photographs at intervals through a glass window in the oven door; these experiments have demonstrated the two-step process mentioned above.

The RSRI used in the present work consists of granules of high-amylose hybrid maize starch; these high-amylose starches are resistant to the activity of enzymes. Brown (1996) stated that granules of enzyme-resistant high-amylose maize starch are not completely gelatinised even at the boiling point of water. This fact agrees with the DSC experiments where lower enthalpy associated to starch gelatinization was found in the RSRI containing dough than in the control dough, as RSRI did not gelatinize at that temperature. Consequently, the effect of adding the RSRI to formulations will mainly depend on two factors: its greater water-holding capacity and the lower quantity of wheat proteins as flour replacement increases; differences as a result of the ungelatinised starch granule content of the dough would not be expected.

In the present formulations, the control biscuit presented the highest length and width values (Table 4). In the RSRI biscuits, the higher the amount of RSRI the

lower the spread, both in length and width, and no change in the thickness of the biscuits was observed (values very near to 16.4 for all formulations) (data not shown). The biscuit's dimensions are determined by the spread rate, which is controlled by the dough consistency and biscuit set time (Miller & Hosenev, 1997). Pareyt et al. (2008) showed that the spread rate decreased linearly with the gluten level. The RSRI substitution had a diluting effect on the wheat proteins. The moisture content and the AWRC data (see section 3.1.) for the different flour/RSRI mixtures showed that the water, which acts as a plasticiser (Slade, Levine, Ievolella & Wang, 1993), is in a lowering level and is increasingly retained as the RSRI level in the system rises; a less pronounced dissolution of the sucrose conducted to a less pronounced volume creation during baking. This has a knock-on effect in delaying the onset of spread, another important factor as Pareyt et al. (2008) showed. Sudha et al. (2007) found that depending on the fibre type (oat, barley, wheat or rice bran) added to a biscuit formulation, the resistance to extension varied; they attributed this effect to interaction between the polysaccharides and the protein in the wheat flour.

The presence of the RSRI reduced the water loss during cooking (Table 2). This result is in accordance with the higher moisture content and water activity obtained for the biscuits of all the formulations containing RSRI. These results were in agreement with the AWRC values for RSRI systems (see section 3.1), indicating that water management of the system was altered by the incorporation of the RSRI to replace part of the wheat flour. In addition, it has been taken into consideration that RSRI-containing dough had a lower moisture level.

Table 4. Biscuit physical and texture characteristic for control and resistant starch-rich ingredient (RSRI) formulations.

Biscuit sample	Length gain (mm)	Width gain (mm)	3- Point Break Test		Crumbliness (N)	Volodkevich Max Force (N)
			Force at break (N)	Gradient (N mm)		
Control	+13.7	+5.5	17.2 ^a (1.4)	9.6 ^a (0.9)	3.4 ^a (1.1)	12.21 ^a (0.26)
20RSRI	+13.0	+9.1	7.5 ^{ab} (0.7)	4.9 ^c (0.2)	2.6 ^b (0.6)	7.53 ^c (0.50)
40RSRI	+10.0	+7.3	7.0 ^{bc} (0.8)	4.5 ^c (0.3)	1.9 ^b (0.4)	8.65 ^b (0.24)
60RSRI	+6.10	+5.2	5.9 ^c (0.6)	6.5 ^b (1.1)	2.2 ^b (0.4)	8.52 ^b (0.54)

Values in parentheses are standard deviations. Means (N=10) in the same column with the same letter do not differ significantly ($p < 0.05$) according to the Tukey test.

3.4.3. Colour

The golden-brown colour of biscuits is mainly caused by reducing sugars-aminoacid interaction or a Maillard-type reaction, which forms brown polymers or melanoidins. The colour parameter values (L^* , C^* , H^* , a^* , b^*) of the biscuits' upper surface and crumb are shown in Table 5. The surface and crumb of the control biscuit were significantly darker ($L^* = 65.99$) and slightly yellower than in biscuits made with RSRI. The addition of the RSRI communicated lightness both to the surface and to the crumb. Baiuxauli, Salvador and Fiszman (2008) had already reported that muffins made with increasing amounts of an RSRI were whiter than the control formulation without the RSRI; they attributed this to a diluting effect of the RSRI on the pigmented elements (egg yolk) of the formulation. This could be the case here with the pigmented elements of the biscuit shortening. In addition, the samples with the RSRI contained a lower level of wheat proteins and had a higher moisture content, contributing to a reduction in the Maillard reaction. Gallagher, Kenny, and Arendt (2005) reported that as the levels of protein substitution increased from 5 to 15% in a short dough biscuit formulation, L^* values decreased. Ozturk, Koksel and Ng (2009)

also found that crust colour values decreased at 30% addition in several RSRI-supplemented breads

In terms of the total colour difference (DE^*) between the control biscuit and the biscuits containing the RSRI, the only sample with a crumb colour DE^* value higher than 3 was sample 60RSRI. On the upper surface, all the samples that contained the RSRI had a DE^* of over 4 (data not shown), meaning that all of them were paler than the control. 60RSRI biscuits would be perceived as whiter, having higher values of L^* , lower values of b^* and a less saturated colour (lower C^* values). This means that there was a reduction in the typical golden or very light brown colour caused by Maillard reactions in the external crust; although Maillard reactions take place throughout the biscuit dough, browning takes place more intensely on the external surface, promoted by high temperatures and a low moisture content. According to Hadiyanto, Asselman, van Straten, Boom, Esveld and van Boxtel (2007), baking of most products is finished before caramelisation (and carbonisation) reactions at temperatures above 150 °C start: even though oven temperatures above 150°C are being used, the temperature of the surface seldom exceeds this value.

No significant differences in hue (H^*) and redness (a^*) were found between the formulations.

However, the colour values of the biscuit surface were darker than the crumb in all cases, as expected. It is interesting that the addition of the resistant starch did not alter the colour of the biscuits to any drastic extent. Adding other fibres such as wheat, oat or rice bran darkens the biscuits considerably, causing colour value differences of over 3 even at 10% concentrations (Sudha et al., 2007)

Table 5. Biscuit colour parameter values for control and resistant starch-rich ingredient (RSRI) formulations

Biscuit sample	Crumb				Upper surface					
	L*	C _{ab} *	H _{ab} *	a*	b*	L*	C _{ab} *	H _{ab} *	a*	b*
Control	66.9 ^a (1.6)	23.7 ^a (0.7)	84.5 ^a (0.3)	2.3 ^a (0.2)	24.7 ^a (0.28)	66.0 ^a (3.2)	31.0 ^a (1.7)	81.3 ^a (1.3)	4.7 ^a (0.9)	30.6 ^a (1.6)
20RSRI	68.9 ^{ab} (1.8)	22.4 ^b (0.8)	83.8 ^a (0.8)	2.6 ^a (0.4)	24.3 ^a (0.72)	70.4 ^b (1.6)	30.6 ^a (1.8)	80.7 ^a (1.6)	5.0 ^a (0.5)	30.2 ^a (1.6)
40RSRI	69.3 ^b (3.8)	22.4 ^b (0.5)	83.8 ^a (0.3)	2.5 ^a (0.1)	22.7 ^b (0.83)	70.8 ^b (1.3)	26.8 ^b (1.7)	82.7 ^a (0.7)	3.5 ^b (0.5)	27.5 ^{bc} (1.6)
60RSRI	71.1 ^c (2.4)	22.8 ^b (0.8)	83.6 ^a (0.3)	2.4 ^a (0.1)	22.2 ^b (0.5)	71.8 ^b (0.9)	27.7 ^b (1.4)	82.8 ^a (0.5)	3.4 ^b (0.5)	26.6 ^c (1.6)

Values in parentheses are standard deviations. Means in the same column without a common letter differ ($p < 0.05$) according to the Tukey test. Means (N=4) in the same column with the same letter do not differ significantly ($p < 0.05$) according to the Tukey test. L*: Lightness; C_{ab}*: chroma; H_{ab}*: a*: redness; b*: yellowness.

3.4.4. *Texture analysis*

The texture of biscuits can be interpreted in terms of the state of their principal ingredients. The process of baking a short dough induces a large decrease in product bulk density, leading to a cellular solid with a thin coloured surface and a porous inner structure. Kulp, Olewnik and Lorenz (1991) determined that starch granules remained in their native condition during biscuit baking and did not form a continuous structure. According to Slade and Levine (1994), biscuit sugar is in a concentrated solution that delays or prevents starch gelatinisation/pasting during biscuit baking. Baltsavias, Jurgens and van Vliet (1999b) showed that irrespective of composition, starch gelatinisation is slight due to the limited water content coupled with the low baking temperature, giving a crisp texture. Also, proteins do not aggregate and hydrate enough to form a gluten network (Chevallier, Della Valle, Colonna, Broyart, & Trystram 2002).

In this case, the quantity and type of both fat and sugar was the same in the four formulations, the changes in the properties of the doughs and the biscuits should be interpreted in terms of the water distribution and lower protein content of the formulations with the RSRI. According to Eliasson and Larsson (1993, p. 263) water plays a complex role, since it determines the conformational state of biopolymers, affects the nature of interactions between the various constituents of the formulation and contributes to dough structuring. If the proportion of water is too low, the dough becomes brittle, not consistent (Sai Manohar & Haridas Rao 1999). The presence of increasing proportions of the RSRI has the effect of decreasing total dough moisture level because wheat flour had a higher moisture level than RSRI; also RSRI diluted the flour proteins which needs to be taken into account when interpreting the results. Soft wheat flour proteins are not functionally inert in biscuit dough, especially during baking, when more associations of proteins probably occur due to increasingly hydration and proximity, affecting biscuit texture (Gaines 1990).

3.4.4.1. 3-Point Break Test. All the biscuits fractured in tension and the fracture took place relatively close to their central zone, where maximum stress

occurred. This implies that the condition regarding the distance between supports was satisfied. The control biscuit slope (Fig. 3) is typical of the force-deformation curves described for materials exhibiting brittle fracture, characterized by an essentially elastic response and a small fracture strain. However, a somewhat shoulder-shaped part appeared in the curve just before fracture; this could imply some plastic deformation in the vicinity of the crack. This behaviour is highly dependent on the proportion of the biscuit ingredients. Nevertheless, what is important is the comparison between the control and the other formulations.

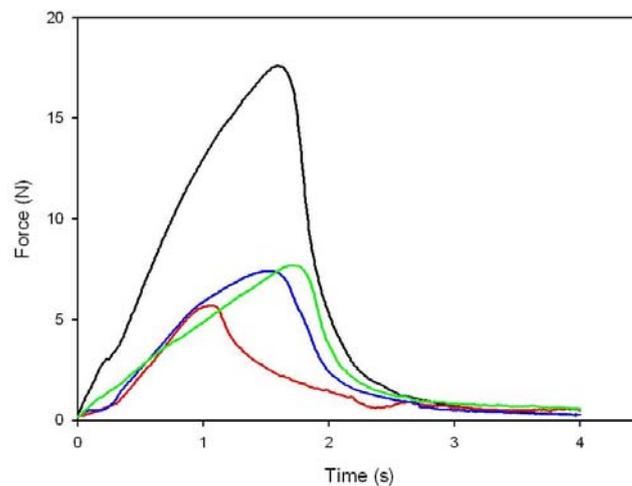


Figure 3. Effect of partial replacement of wheat flour with and RSRI on the beaking strength of biscuits. Black: control, red:20RSRI, blue: 40RSRI, green: 60RSRI.

The highest value of force at break corresponded to the control biscuit (Table 4), indicating a more interconnected, structured matrix, probably due to the higher level of wheat proteins in the control samples. The biscuits containing the RSRI showed curves characterized by a lower initial force gradient and lower peak force values, indicating lower resistance to snapping. In particular, for 60RSRI biscuit, bending strength is even lower and the peak value was

obtained earlier, indicating a weaker structure and shorter elastic response. Additionally, it can be seen that the force values did not drop as sharply as in the control sample after reaching the fracture stress values. This could imply that the crack propagated more slowly and the probe had to travel further to cause fracture. No significant differences in density were found between the biscuits of the four formulations (all values were around 1.20 g.cm^{-3}) indicating that any difference in air retained by the dough before baking (reflected in different dough density) was compensated by more moisture retention after baking (less weight loss) due to the presence of the RSRI. The greater the quantities of the RSRI replacing part of the flour, the lower the gluten content and the more destructured the system.

The dough properties (see section 3.1) showed that the consistency of the 20RSRI dough was very similar to that of the control, while the compression and wire cutting test values for samples 40RSRI and 60RSRI reflected harder doughs. Obviously, during baking, the lack of a sufficient quantity of proteins to give the dough structure and cohesion became an important factor in the final texture of the biscuit, although the greater moisture content of the RSRI formulations had a tenderising effect. On replacing 30 % of the flour in a short dough biscuit formulation with native wheat starch, Baltsavias, Jurgens and van Vliet (1999b) found slightly lower fracture stress; also, the stress-strain curve showed a small shoulder just before fracture which they attributed to a stronger local permanent deformation as a result of a somewhat different fat distribution. Biscuits with increasing levels of different fibre sources become harder, as seen in their increasing breaking strength values, especially, as reported by Sudha et al. (2007), when 30 and 40% of rice and barley bran are added. The gradient values of the curves indicated that the control sample was stiffer than the samples with the RSRI.

The compression curves of the cube-shaped biscuit samples showed that the biscuits with the RSRI were weaker than the control samples (Fig. 4). This test

was used to determine the greater or lesser crumbliness of the samples by assessing their cohesiveness.

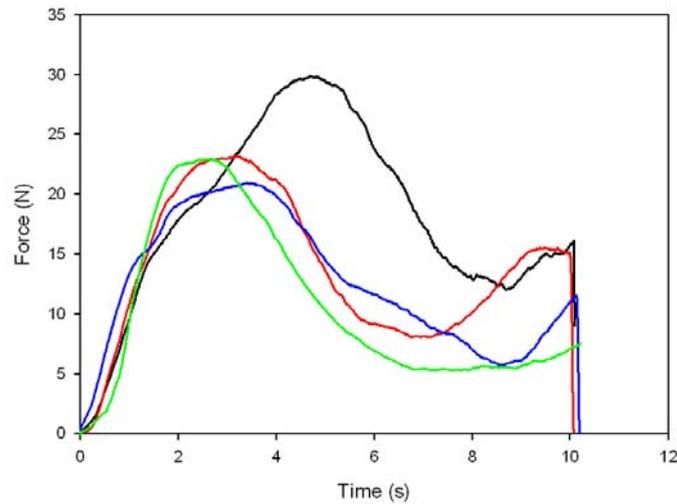


Figure 4. Effect of partial replacement of wheat flour with an RSRI on the crumbliness index of biscuits. Black: control, red:20RSRI, blue: 40RSRI, green: 60RSRI.

A less cohesive structure that collapsed under low compression force values would indicate greater crumbliness (Table 4). In other words, the presence of the RSRI enhanced the crumbliness of the biscuit. This could be because the formulations containing the RSRI had less flour protein present so formed less entangled matrices. It should be noted that no significant differences were found between the values for any of the samples containing the RSRI, which could be interpreted as meaning that they have a similar degree of lack of cohesion within the biscuit matrices. It should be emphasized that for this type of product, this characteristic is not negative from the point of view of the texture perceived by the consumer. A certain degree of 'tenderness' is appreciated in biscuits with a high fat content. Yue and Waring (1998) reported that a sensory panel described 40% total-dietary-fibre resistant-starch biscuits as having a tender, shortbread-like texture; also, the hardness value obtained with a 3-point break test was lower than for the control biscuit without resistant starch. Brown,

Langley and Braxton (1998) found that there was a strongly significant inverse correlation between hardness and crumbliness in a series of biscuits exhibiting texture differences, as assessed by ordinary consumers (n=19).

Volodkevich Bite Jaws imitate the force of biting the biscuit. The control biscuit showed a typical penetration curve with a maximum corresponding to the point when the probe penetrated the sample; the values then fell, indicating relaxation of the stress, and finally rose again as the increasing dimensions of the probe penetrated the sample (Fig 5).

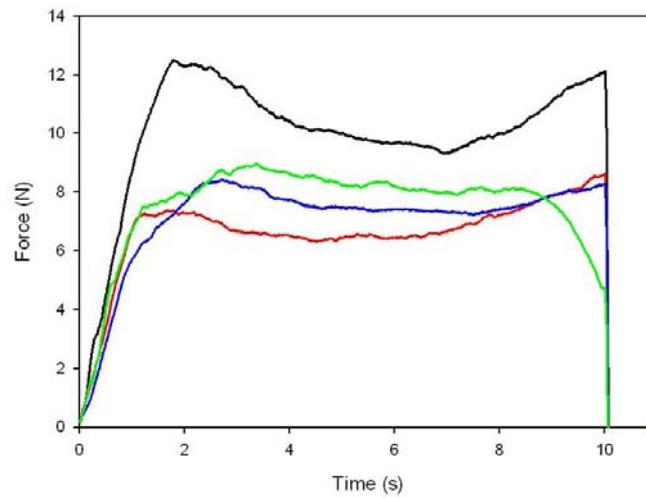


Figure 5. Effect of partial replacement of wheat flour with an RSRI on penetration behaviour (Volodkevitch Bite). Black: control, red: 20RSRI, blue: 40RSRI, green: 60RSRI.

The penetration force values of the samples containing the RSRI were lower than those of the control and there were practically no differences between them (Table 3); also, the

curves for the RSRI biscuits lacked a 'defined' profile pattern in the sense that they were more variable in shape and a number of them had lost the initial penetration peak. As with the crumbliness results, no significant differences were found between the results for the RSRI samples, indicating greater crumb

tenderness than in the control sample. None of these methods distinguished between different levels of the RSRI. Gaines, Kassuba and Finney (1996) used a puncture test on various biscuits and reported that the fracture force rose with increasing flour protein content. In the present case, the RSRI dilutes the protein content of the flour.

3.4.5. Sensory analysis

Four biscuits (one of each formulation) were monadically given to consumers who were free to taste as much as necessary to rate the acceptability of each of the six attributes to be analysed. None of the consumers mentioned any fatigue or sense of fullness that could affect their scores. The mean sensory acceptance scores for the 'appearance', 'colour', 'texture', 'taste' and 'sweetness', and 'overall acceptance' of biscuits with different RSRI levels are presented in Figure 6.

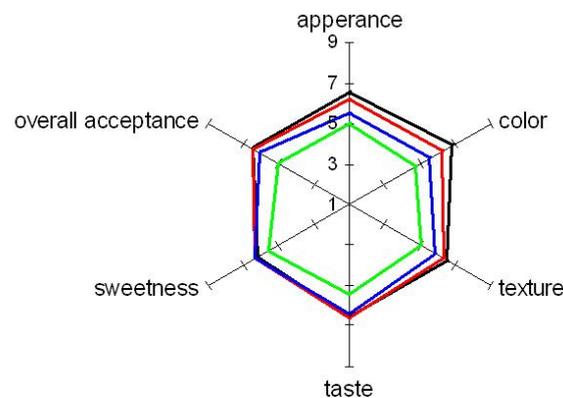


Figure 6. Sensory hedonic tests of biscuits prepared with different levels of an RSRI. 1=dislike extremely. Black: control, red: 20RSRI, blue: 40RSRI, green: 60RSRI.

The acceptance scores for the control and 20RSRI biscuits were not significantly different for any of the sensory attributes. 40RSRI obtained lower scores only for appearance, colour and texture, whereas the taste, sweetness and overall acceptance values showed no significant differences compared to the control and 20RSRI samples. The 60RSRI biscuit was the least acceptable sample. None of the consumers observed the dry mouth feel, lack of

smoothness in the biscuit surface or dark crumb colour reported by Sudha et al. (2007) for biscuits prepared with different bran sources.

Consumption intention values, expressed as the percentage of consumers that answered “yes” to the question Would you consume this product?, were 76, 74, 66, and 43% for the control, 20, 40, and 60RSRI biscuits respectively, a consumption intention of over 50% for all the formulations except the 60RSRI biscuits. Baixauli, Salvador, Hough et al. (2008) stated that consumers generally identified fibre-enriched products with dark products, so when using an alternative fibre such as the RSRI, which is white (the final products were not darker), it is advisable to give consumers more information on what these fibres are and thereby provide them with better criteria on which to base their choice.

This study has shown the good potential of a resistant starch rich ingredient (RSRI) for developing fibre-rich biscuits with the aim of developing products that increase dietary fibre intake by substituting this ingredient for part of the flour in the formulation. Replacing 20% of the wheat flour with the RSRI did not affect the dough rheology, while higher proportions gave stiffer doughs. In the baked biscuits, replacing part of the flour with the RSRI gave crumblier, tenderer, less hard textures and shapes that spread less. Consumer acceptance of the biscuits was good and significant differences were practically non-existent except in the 60RSRI formulation, which was significantly less acceptable.

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STUDY ON RESISTANT STARCH FUNCTIONALITY IN SHORT DOUGH
BISCUITS BY OSCILLATORY AND CREEP AND RECOVERY TEST

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Abstract

The effect of wheat flour replacement by a resistant starch rich ingredient (RSRI) on the structure of short dough biscuits was studied by oscillatory and creep and recovery tests to determine linear viscoelastic properties. The RSRI was substituted for the flour at three different levels: 20, 40 and 60% (w/w). The use of RSRI increased the elastic and the viscous moduli but did not influence $\tan\delta$. The compliance values during the creep test were adjusted successfully to the Burger model. The creep and recovery test revealed an increase in elasticity and resistance to flow and a decrease in deformability with RSRI, but the differences were only significant at the 40% and 60% levels. The RSRI did not affect relative recovery, thus no effect on the type of structure is expected. Deformability was positively correlated with biscuit spread during baking.

Key words: short dough, biscuit, rheology, oscillatory, creep-recovery

1. Introduction

Biscuits can be good fibre carriers because they are widely consumed. Previous work showed the possibility to incorporate resistant starch, as a source of fibre in biscuits, with good consumer acceptability. The resistant starch rich ingredient (RSRI) was added to short dough biscuits, replacing wheat flour, and its effect on dough and biscuit texture was studied by empirical instrumental tests and by sensory analysis (Laguna et al., 2011). The RSRI increased the consistency of the raw dough and the ease of sheeting and cutting. Replacing 20% of the wheat flour with RSRI did not affect the dough texture significantly, while higher proportions gave stiffer dough's. In the baked biscuits, replacing part of the flour with the RSRI gave crumblier, tenderer, less hard textures and shapes that spread less. Resistant starches are broadly researched due to their important benefits in human nutrition (Hodsagi et al. 2011).

To increase the knowledge about the inner structure of the dough and the relation to the microstructure, fundamental rheological measurements are required (Menjivar 1990)

Oscillatory and creep-recovery are fundamental tests that measure the linear viscoelastic properties of a substance. The stress applied to the sample in the linear viscoelastic region is low enough not to produce an irreversible change in the structure, so information about the unaltered system structure is obtained.

In an oscillatory test the sample is submitted to an oscillatory shear movement of a certain frequency and stress (or strain) amplitude inside the linear region and the strain (or stress) produced is measured (Barnes 2000).

The linear viscoelastic properties of biscuit doughs have been studied by a number of authors. Baltasavias et al. (1997) evaluated the linear viscoelastic properties of short-doughs with various compositions. They concluded that fat was a crucial structural component; decreasing the fat content or replacing solid fat with liquid oil caused a marked decrease in the stiffness of the dough and greater dispersal of the fat in the dough. Also, the authors (Baltasavias et al.,

1997) studied the effect of flour replacement by native starch and observed a quantitative decrease in the viscoelastic modulus and the absence of gluten was expected to affect water distribution and, thereby, the rheological properties of the non fat phase. The contribution of other flour components like damaged starch, which increase short dough consistency, or pentosans, which decrease G' , was difficult to establish, as it was not possible to separate the effects due to the gluten, pentosans and damaged starch. As for the role of sucrose, it increased the liquid-like properties of the dough, because sucrose modifies the properties of the non-fat phase via its influence on the amount of solvent.

The effect of endogenous flour lipids on the structure of semi-sweet doughs and short doughs has been studied by oscillatory tests (Papantoniou et al., 2003; Papantoniou et al., 2004). The employment of defatted flour produced higher viscoelasticity and the microstructure of the defatted short dough biscuits revealed that their gluten protein was more hydrated and developed. It was suggested that polar lipids form bonds with the protein molecules and help to control the access of water to the proteins (Papantoniou et al., 2004).

Creep and recovery are transient tests in which an instantaneous stress in the lineal viscoelastic region is applied to the sample for a certain time (creep) and after removal of the stress the deformation is measured (recovery). The system deformation per unit stress is called compliance and is measured over time.

The mathematical treatment of the theory of creep and recovery uses model substances such as springs and dashpots (Schramm 1994). One of the most widely used model is the four-components Burguer model, comprising the association in series of the Maxwell model (association of spring and dashpot in series) and the Kelvin-Voigt model (parallel association) (Dolz et al., 2008).

Pedersen et al., (2004) used oscillatory and creep-recovery tests to study the linear viscoelastic properties of semi-sweet biscuit doughs made with flours from eight different cultivars and related them to changes in biscuit dimension. The decrease in biscuit length was correlated to the phase angle δ the farinograph and the creep recovery parameters. Pedersen et al. (2005) also

evaluated the effect of adding sodium meta-bisulphite and a commercial protease. An increase in dough extensibility and a decrease in elasticity were observed. Biscuit contraction and spread were mostly correlated to % recovery of the dough and to protein and gluten content.

However, in spite of the volume of studies on rheology and texture of dough, we are not aware of any detailed studies that have described the oscillatory or creep and recovery behaviours of short dough biscuits containing resistant starches, with established mathematical models.

The aim of the present study was to understand the functionality of RSRI in the short dough biscuit structure by studying the linear viscoelastic properties (oscillatory and creep and recovery tests) of the dough. The rheological results were described according to mathematical models and related to previous results from empirical tests carried out in the biscuit dough and in baked biscuits.

2. Material and methods

2.1. Short dough biscuit ingredients

Four formulations were prepared using the same quantity of all the ingredients except the wheat flour and RSRI (a resistant starch rich ingredient) as shown in Table 1.

2.2. Short dough biscuit preparation

The shortening was creamed in a mixer (Kenwood Major Classic, UK) for 4 minutes at minimum speed (60 rpm) to obtain a homogenous cream. After this, the sugar was added and mixed in for 2 min at speed 4 (255 rpm). The milk powder, previously dissolved in all the water, was added and mixed in for 2 min at the minimum speed. The flour (or flour/RSRI), sodium bicarbonate and ammonium hydrogen carbonate were then mixed in together at minimum speed

for 2 min. Finally, the dough was sheeted to a thickness of 4 mm and kept in a refrigerator for 24 hours.

Table 1. Ingredients used in the biscuit performance (percentages given on a flour basis).

<i>Ingredients (%)</i>	<i>control</i>	<i>20RS</i>	<i>40RS</i>	<i>60RS</i>
Flour ^a	100	80	60	40
Fat (shortening)	60	60	60	60
Sugar	30	30	30	30
Milk	0.576	0.576	0.576	0.576
Salt	0.2	0.2	0.2	0.2
Sodium bicarbonate	0.2	0.2	0.2	0.2
Amonium bicarbonate	0.115	0.115	0.115	0.115
Water	9	9	9	9
RSRI ^b	0	20	40	60

^asoft wheat flour suitable for biscuits, composition data provided by the supplier: 15% moisture, 11% protein, 0.6% ash; alveograph parameters $P/L=0.27$, where P=maximum pressure required and L=extensibility; and $W=134$, where W=baking strength of the dough.

^bHi-maize 260, composition data provided by the supplier: 10% moisture, 58% dietary fibre according to AOAC 991.43 method.

2.3. Rheological experiments

Oscillatory and creep and recovery tests were performed at 25°C in a controlled stress rheometer (AR-G2, TA Instruments, Crawley, England) with the temperature controlled by a Peltier system. The rheometer was equipped with a 20mm roughened parallel plate with a gap of 1.5 mm. Dough cylinders of 13

mm internal diameter were placed on the rheometer. The samples were allowed to rest in the measurement position for a 10 min equilibration time.

2.3.1. Oscillatory tests

A stress sweep test was carried out to determine the linear viscoelastic region, after which frequency sweeps were conducted from 10-0.01Hz at stress amplitude of 20 Pa within the linear viscoelastic region. The storage modulus (G'), loss modulus (G'') and loss tangent ($\tan \delta = G''/G'$) values were recorded.

2.3.2. Creep and recovery tests

An instantaneous stress, σ_0 , was applied within the linear viscoelastic region and maintained for 1200s. The stress was then removed and the sample was allowed to relax for 1200s. The system deformation per unit stress, called compliance, was measured by the following formula: $J(t) = \gamma(t) / \sigma_0$.

Each measurement was carried out 3 times in doughs prepared on different days. To protect against dehydration, vaseline oil (Panreac, Spain) was applied to the exposed surfaces of all the samples.

For the entire rheological values non-linear curve fitting is applied to the data using the Levenberg-Marquardt algorithm (KaleidaGraph 4.0, Synergy Software). This is a non-linear iterative method, so an initial value for each parameter is supplied. They are improved in each interaction until a correct order of magnitude previously established.

2.4. Statistical analysis

Analysis of variance (one way-ANOVA) was applied to study the differences between formulations; the least significant differences were calculated by the Tukey test and the significance at $p < 0.05$ was determined. Principal component analysis (PCA) was used to correlate the sensory and instrumental parameters. These analyses were performed using SPSS for Windows Version 12 (SPSS Inc., USA).

3. Results and discussion

3.1. Viscoelastic properties

3.1.1. Oscillatory test: frequency sweep test

The influence of flour replacement by RSRI on the frequency sweeps carried out in the linear region are shown in Figure 1a. In all the doughs, the frequency dependence was typical of weak gels, with higher values for G' than G'' and weak dependence of both moduli on frequency within the available frequency range studied.

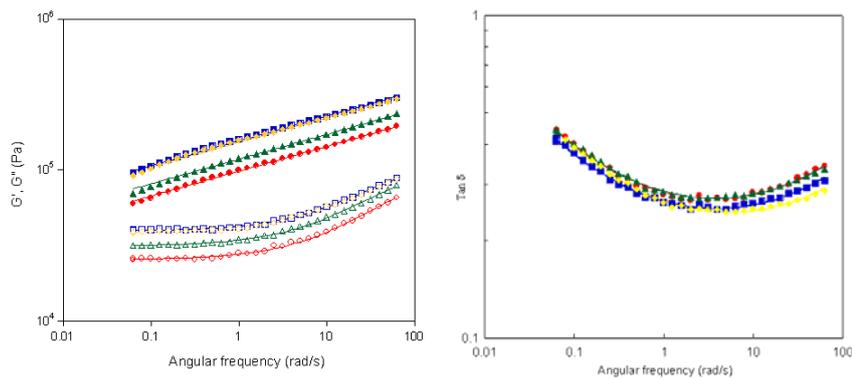


Figure 1. A) G' (closed symbols) and G'' (open symbols) as a function of frequency and B) $\tan \delta$ as a function of frequency for the different formulations. Red: control; green: 20RSRI, yellow: 40RSRI, blue: 60RSRI

The different formulations showed a similar frequency dependence of both G' and G'' . In all the formulations the frequency dependence of G' correspond to straight lines in the log-log plot (Figure 1a), therefore $G'=f(\omega)$ can be fitted to power law equations (Dolz et al., 2006):

$$G' = K' \omega^{n'} \quad (1)$$

The power law index, n' , is related to the slope of the straight lines in Fig 1a. The values obtained for all the formulations were very similar, as it was expected due to the parallelism of the lines in Figure 1a. We calculated then a mean value, $n = 0.164 \pm 0.002$, and fitted the functions again fixing n (with

R>0.996). The values obtained for K' are shown in Table 2. They correspond to the values of storage modulus for $\omega = 1$ rad/s.

The G'' values also showed a similar dependence with frequency in all the samples, which were adjusted to the following empirical equation (Dolz et al., 2006):

$$G'' = K'' - Z \exp(10 - T \omega^{n''}) \quad (2)$$

In order to simplify and make easier the discussion, we reduced the number of parameters in eq. (2). As the values obtained for Z and n'' were similar, we refitted the functions fixing Z and n'', using the mean values, $Z = 2.81 \pm 0.23$ Pa and $n'' = 0.80 \pm 0.02$ ($R > 0.998$). Values obtained for K'' and T are shown in Table 2. The parameter T gives information about dependence with frequency. Their values are not statistically different, as it was expected when observing Figure 1a. The parameter K'' corresponds to the G'' values for very low frequencies.

Replacing flour with RSRI increased the values of G' and G'', which is reflected in the increase in the parameters K' and K''. The control dough showed the lowest values of both parameters, followed in increasing order by 20RSRI, 40RSRI and 60RSRI. However, the differences with the control sample were only significant for the higher levels of flour replacement (40RSRI and 60RSRI).

Tan δ values versus frequency are shown in Figure 1b. The tan δ values reflect the existence of more solid behaviour (tan δ closer to 0) at higher frequencies (shorter time) and a more liquid behaviour (tan δ closer to 1) at lower frequencies (longer time). The frequency dependence of tan δ can be fitted to the following equation:

$$\log(\tan\delta) = A(\log \omega)^2 + B(\log \omega) + C \quad (3)$$

As the values obtained for A were similar, the average value of A were calculated being 0.154 ± 0.003 ($R > 0.996$). The values of B and C are shown in Table 2.

Table 2. Values of the parameters obtained from the equations 1, 2 and 3 adjusting the frequency dependence of G' , G'' and $\tan \delta$.

Type of dough	Oscillatory test				ω_c	
	$G' = K' \omega^n$ (eq.1)* K' (Pa s ^{0.164})	$G'' = K'' \cdot 2.81 \exp(10 - T \omega^0)$ (eq.2)** K'' (Pa)	T (s ^{0.8})	$\log(\tan \delta) = A(\log \omega)^2 + B(\log \omega) + C$ (eq.3) B		C
Control	99538 ^a (2237.3)	87662.5 ^b (562.1)	0.04 ^a (0.004)	0.176 ^a (0.002)	1.256 ^a (0.002)	3.728
20RSRI	116840 ^a (1414.2)	92542 ^c (859.1)	0.05 ^a (0.001)	0.183 ^a (0.002)	1.325 ^a (0.002)	3.198
40RSRI	162035 ^b (9524.7)	102060 ^a (1301.1)	0.07 ^a (0.024)	0.177 ^a (0.003)	1.256 ^a (0.003)	3.755
60RSRI	150100 ^b (2177.9)	101035 ^a (998.0)	0.04 ^a (0.013)	0.228 ^b (0.002)	1.322 ^a (0.002)	5.516

In the same column, values with the same letter are not statistically different according to the Tukey test ($p < 0.05$).

* $n' = 0.164$

** $n'' = 0.8$

The value of frequency at which $\tan\delta$ reaches a minimum (ω_c) was calculated according to equation (4) as suggested by Sopade et al. (2004). The obtained values of ω_c are shown in Table 2.

$$\frac{d(\log \tan \delta)}{d(\log \omega)} = 0 \quad (4)$$

In general, no differences in B and C values were found among the samples (Table 2), indicating that although the substitution of RSRI affected the G' and G'' values, no effect on the relationship of the two moduli was observed. As Fig. 1 shows, the increment in RSRI produced an increment in G' and G'' but the relation between G' and G'' ($\tan\delta=G''/G'$) was kept constant. The fact that the viscoelastic properties (values of $\tan\delta$ were not altered indicates that no significant structural change occurs). Therefore it could be stated that the type of organization of the structure was not significantly affected by RSRI incorporation.

3.1.2. Creep and recovery tests

The effect of replacing flour on average of three replicates with RSRI on the compliance values ($J= \gamma/\sigma$) as a function of time in the creep and recovery tests is shown in Figure 2.

All the systems showed both viscous and elastic characteristics (some instantaneous deformation and a certain percentage of recovery). As in the oscillatory tests, the creep and recovery tests showed that the viscoelastic characteristics were affected by flour replacement. A decrease in the compliance values was observed as the replacement of flour by RSRI increased, indicating a greater resistance to deformation. Lower compliance values are indicative of stronger structural matrices, which show higher resistance to deformation.

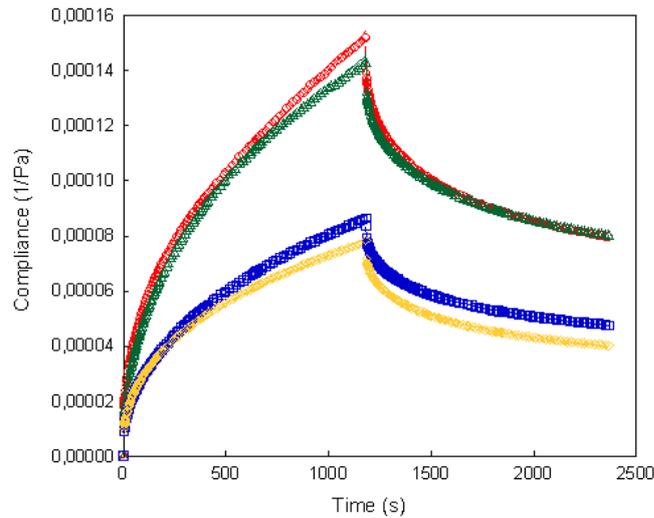


Figure 2. Compliance versus time in a creep and recovery test for the different formulations. Red: control; green: 20RSRI, yellow: 40RSRI, blue: 60RSRI

3.1.2.2. Creep test

The creep data ($0 \leq t \leq 1200\text{s}$) were satisfactorily adjusted to the Burger model (eq 5), always yielding values of $r^2 \geq 0.994$. The Burger model is made up of four components that comprise the association in series of the Maxwell model and the Kelvin-Voigt model (Steffe, 1996; Barnes, 2000).

$$J(t) = \frac{1}{G_0} + \frac{1}{G_1} \left[1 - \exp\left(\frac{-t G_1}{\eta_1}\right) \right] + \frac{t}{\eta_0} \quad (5)$$

In the Burger model (eq. 5), as García-Loredo et al. (2011) described, $J(t)$ represents the overall compliance at any time t , G_0 is the instantaneous elastic modulus of the Maxwell element and G_1 is the elastic modulus of the Kelvin-Voigt element. The dashpot of the Maxwell element represents the residual viscosity (η_0) and the dashpot associated with the Kelvin-Voigt element is the internal viscosity, η_1 (Dolz et al., 2008). These values allow the internal structure of different systems to be compared, yielding a mechanical model with

behaviour in response to deformation that is similar to that of such systems (Dolz et al., 2008).

The values obtained for G_0 , η_0 , G_1 and η_1 are reported in Table 3. The replacement of wheat flour by RSRI reinforced the structure, raising the values of all the parameters in the Burger model.

Table 3. Creep and recovery parameters

Type of dough	Creep parameters				Recovery parameters				
	$G_0 \times 10^{-4}$ (Pa)	$\eta_0 \times 10^{-7}$ (Pa s)	$G_1 \times 10^{-4}$ (Pa)	$\eta_1 \times 10^{-6}$ (Pa s)	$J_{INF} \times 10^5$ (Pa ⁻¹)	$J_{KV} \times 10^5$ (Pa ⁻¹)	$J_{MAX} \times 10^5$ (Pa ⁻¹)	$J_{SM} \times 10^5$ (Pa ⁻¹)	%R
Control	6.15 ^{ab} (0.49)	1.33 ^a (1.33)	2.07 ^a (0.16)	2.22 ^a (0.16)	4.43 ^b (0.69)	10.24 ^b (0.86)	15.2 ^d (0.14)	5.3 ^c (0.03)	70.5 ^a (4.9)
20RSRI	6.05 ^a (0.64)	1.61 ^b (0.06)	1.81 ^a (0.04)	2.88 ^a (0.13)	3.88 ^{ab} (0.33)	10.01 ^b (0.03)	14.3 ^c (0.28)	4.1 ^b (0.02)	73.0 ^a (1.4)
40RSRI	9.35 ^{bc} (0.64)	2.53 ^c (0.06)	3.31 ^b (0.11)	5.05 ^b (0.13)	2.59 ^{ab} (0.57)	5.77 ^a (0.53)	8.6 ^b (0.06)	2.9 ^a (0.02)	70.5 ^a (6.4)
60RSRI	9.15 ^c (1.06)	2.94 ^d (0.01)	3.58 ^b (0.23)	4.60 ^b (0.29)	1.95 ^a (0.13)	5.57 ^a (0.16)	7.8 ^a (0.06)	2.5 ^a (0.02)	75.0 ^a (1.4)

In the same column, values with the same letter are not statistically different according to the Tukey test (p<0.05).

As occurred in the previous section two different groups were found: the control and 20RSRI and the 40 RSRI and 60 RSRI. The effect of RSRI was significant for concentrations higher than 20%, as observed in the oscillatory results however due to a saturation effect it was not a structural change in the 40RSRI and 60RSRI short dough.

In particular, the significantly higher G_0 and G_1 values in 40RSRI and 60RSRI indicate a harder and more rigid structure. The increase in G_0 indicates an increase in the elastic properties and therefore higher recoverability, as G_0 corresponds to the initial instantaneous deformation. The significantly higher η_0 and η_1 indicate a higher resistance to flow. In short, the addition of RSRI

produces greater opposition to deformation, which becomes significant at the 40% flour replacement level. Results extracted from the Burger model parameters agree with the higher G' and G'' obtained with increased RSRI concentration.

Recovery test

After removing the stress, the reduction in material deformation was measured (the recovery part of the test).

Three well differentiated zones comprise the recovery response (Dolz et al., 2008): the first recovery (J_{SM}) is nearly instantaneous and corresponds to the deformation suffered by the spring of the Maxwell element, the second recovery (J_{KV}), due to the Kelvin-Voigt element, is slower and tends to an asymptote for $t \rightarrow \infty$. Lastly, the residual deformation (J_{INF}), which is due to sliding of the Maxwell dashpot, determines permanent deformation.

The compliance values during the recovery test were satisfactorily adjusted to the following empirical equation (eq. 6), employed in previous works (Dolz et al., 2008; Bayarri et al., 2009):

$$J(t) = J_{INF} + J_{KV} \exp(-B t)^c \quad (6)$$

Where B and c are parameters related to the recovery rate of the system. They were very similar in all the dough formulations, with mean values of $B = 0.045 \pm 0.005 \text{ s}^{-1}$ and $c = 0.43 \pm 0.01$.

The J_{SM} (initial shear compliance) (eq. 7) values were calculated as follows:

$$J_{SM} = J_{MAX} - (J_{INF} + J_{KV}) \quad (7)$$

where J_{MAX} is the compliance value for the longest time (1200s) in the creep transient analysis, which corresponds to the maximum deformation.

Lastly, the final percentage recovery of the entire system was calculated by equation 8 (Dolz et al., 2008):

$$\%R = \frac{J_{MAX} - J_{MIN}}{J_{MAX}} \times 100 \quad (8)$$

The J_{MAX} , J_{INF} , J_{KV} , J_{SM} and %R obtained are shown in Table 3. J_{MAX} decreased significantly as the RSRI concentration increased, revealing the lower deformability provided by substituting RSRI. The instantaneous recovery (J_{SM}) and the secondary recovery (J_{KV}) also decreased significantly with RSRI concentration. As in the creep test, two patterns of behaviour were observed: one formed by the control and 20RSRI and the other by 40RSRI and 60RSRI. As it was mentioned before the effect of RSRI was significant for concentrations higher than 20%, as observed in the oscillatory and creep results. Differences between 40RSRI and 60RSRI were not found due to a saturation effect.

A finding that should be highlighted is that the %R was not significantly affected by the presence of RSRI, indicating that the RSRI did not affect the recovery response. Higher initial deformation produced higher residual deformation and vice versa, irrespective of the sample formulation. The system was deformed to a greater or lesser extent and the degree of recovery was proportional to the initial deformation. This result indicates that replacement of wheat flour by RSRI reinforced the dough structure, which became more elastic and viscous (higher values for the Burger model and G' and G'') and possessed a high resistance to deformation, but had a very small effect on the type of structure, which is why the %R and $\tan \delta$ values did not change.

The effect of RSRI on the dough structure could be summarized as a reinforcement of the structure without any significant alteration in the type of structure. It seems that RSRI only exerts a concentration effect on the dough structure and has no effect on the type of network. Pedersen et al. (2004) also found no differences in % recovery when comparing semi-sweet biscuit doughs from different cultivars, although the maximum strain and recovery were strongly affected by the cultivar. In some cultivars the effect was associated with the protein content, a higher protein content being associated with an increase in extensibility and recovery. Cultivars with the same protein content (range

from 10.2-11.3%) and water absorption also revealed differences in extensibility, indicating that differences in the structure of the gluten (gluten content 17.7-23.4%) or other components also contribute to this property (Pedersen et al., 2004). In other cultivars, however, extensibility was not affected or was negatively correlated with protein content. In creep tests of dough made from durum wheat with different protein levels (from 8.9 up to 14.7%) a high correlation between protein content and maximum strain has been observed (Edwards et al., 1999).

Correlation between the linear viscoelastic properties of the dough and empirical measurement of dough and biscuit texture and spread properties was obtained.

In order to understand how the oscillatory and creep and recovery properties of the dough relate to the dough texture and biscuit spread and texture properties obtained in a previous study (Laguna et al., 2011) principal component analysis (PCA) was carried out.

The first two components that explained 93.32% of the variance are shown in Figure 3. The first component (PC1) explained 74.99% of the variance and the second component (PC2) explained 18.83%. PC1 showed a positive correlation with all the biscuit texture parameters (crumbliness, Volodkevich maximum force, force at break and gradient), with the parameters related to biscuit spread (length gain and width gain) and with all the dough recovery parameters (J_{SM} , J_{MAX} , J_{INF} and J_{KV}). The creep parameters (G_0 , G_1 , η_0 and η_1), the oscillatory parameter A and the dough texture parameters (maximum force for dough penetration, maximum force for dough compression and the plateau force after wire-cutting the dough) correlated negatively with PC1. The control and 20RSRI formulas appeared separately in the positive part of PC1, associated with higher values for the biscuit texture parameters, biscuit spread and recovery dough parameters, while 40RSRI and 60RSRI appeared in the negative part, associated with higher values for the dough texture parameters, creep parameters and the oscillatory parameter K' .

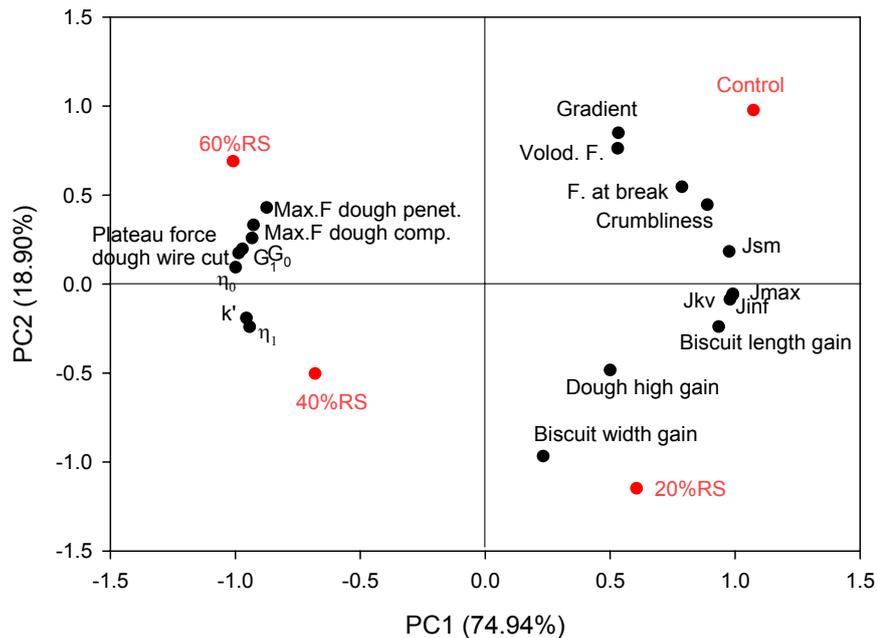


Figure 3. PCA 2D loading plot of the two principal components explaining 93.82% of the variance.

The effect of RSRI substitution on the dough properties can be explained by the combined effect of two factors: 1) the lower gluten content due to wheat flour replacement and 2) the effect of RSRI on the moisture content, dough density and water availability.

Gluten provides extensibility properties to the dough (Edwards et al., 1999). The control formula, with the highest amount of gluten (11.4% of gluten, flour basis), showed the highest deformation in response to the applied stress (maximum value of J_{MAX}). This higher deformation was associated with a less rigid system (lower K' and creep parameter values). As the gluten in the formula was reduced by RSRI substitution (20%RSRI contains 9.12% gluten in flour basis, 40% RSRI contains 6.84 and 60%RSRI contains 4.56 gluten in flour basis), deformability decreased and the parameter K' and the creep parameters increased, so the system's rigidity increased. This explains why the dough

recovery parameters appear separately from the parameter K' , the creep parameters and the dough texture parameters in the PCA diagram. The control and 20RSRI showed the highest deformability (higher J_{MAX} values) and the lowest K' and creep and texture parameter values (lower rigidity). In contrast, 40RSRI and 60RSRI showed the lowest values for recovery parameters (least deformation) and the highest values for K' and the creep and texture parameters, indicating their greater rigidity.

The increase in rigidity and decrease in deformability associated with wheat flour replacement by the RSRI can also be associated with the lower moisture content of RSRI in comparison to flour (Laguna et al., 2011), which produces drier doughs and an increase in dough density associated with the addition of RSRI. In addition, RSRI increases the water retention capacity (the %AWRC values for the flour, the 80/20, 60/40 and 40/60 flour/RSRI mixtures were 74.59, 81.02, 106.9 and 129.8 g/g on a dry basis, respectively), reducing the water available for gluten development. Although water is a minor component of the biscuit dough, the level of water in the formula affects gluten development, spread, final moisture and eating quality (Lai and Lin, 2006).

In biscuits, substituting RSRI decreased the texture parameters. The control and 20RSRI appeared to be related to the highest biscuit texture parameter values, while 40RSRI and 60RSRI were associated with lower values.

The effect of the RSRI on biscuit texture may be associated with the decrease in gluten functionality (through the diluting effect of RSRI and lower water availability), combined with the increase in water retention properties. The decrease in gluten functionality made the biscuit matrix less structured and gave it a softer, less tough texture, while the RSRI created more amorphous zones in it (Laguna et al., 2011). The higher moisture and water activity in the biscuits with the RSRI may also explain the softer biscuit texture in the presence of RSRI. Higher moisture is associated with a softer biscuit texture.

Another important biscuit property which was significantly affected by wheat flour replacement by the RSRI was the spread rate. The control and 20RSRI

showed higher spread (gain in width and length), while 40RSRI and 60RSRI showed the lowest. The higher spread rate was positively associated with the recovery parameters, which reflect deformability and are negatively associated with the oscillatory parameter A and the creep parameters, and these in turn reflect the elastic properties. The effect of RSRI on the spread rate cannot be explained in terms of gluten content and gluten functionality reduction, as previous studies have stated that the spread rate decreases linearly with the gluten level (Donelson, 1988). Therefore, the effect on the spread rate has to be associated with RSRI functionality in the system rather than with decreased gluten functionality. Gaines and Finney (1989) associated a faster spread rate and later setting with the employment of soft wheat flour instead of hard wheat flour, as soft wheat dough was less viscous during baking. The presence of RSRI decreases deformability and increases system elasticity and resistance to flow, which may explain the decrease in spread.

4. Conclusions

This study showed the effect of RSRI in short dough biscuit. The values obtained for linear viscoelastic properties of the short dough biscuit revealed that replacement of wheat flour by RSRI increases system rigidity and reduces deformability. However, $\tan \delta$ and % recovery were not altered, implying that the RSRI was not changing the type of structure but exerting a concentration effect on the dough structure. This effect of the RSRI can be associated with lower gluten functionality and lower water availability on dough in presence of RSRI. The spread of the biscuits could be predicted from the dough properties, as higher dough deformation correlated positively to biscuit length and width gain.

The comparison among small (oscillatory and creep-recovery) and large deformation (texture) measurements revealed that both can be correlated and explained dough changes during baking.

For all the tests, two samples groups (control-20RSRI and 40RSRI-60RSRI) were found and the results can be related with the RSRI quantity that affects the gluten content and water disponibility.

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ROLE OF FIBRE MORPHOLOGY IN SOME QUALITY FEATURES OF FIBRE-
ENRICHED BISCUITS

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Abstract

The effect of replacing 5% and 10% of the flour in a biscuit formulation with two wheat fibres (of different lengths) and apple fibre (differences in morphology and proportion of soluble fraction) was studied. All the fibres decreased the flour pasting properties. The longer wheat fibre produced the greatest increase in the G' and G'' viscoelastic moduli of the dough compared to the control (no fibres added). The biscuit texture properties were measured using the 3-point break test and cone penetrometry: wheat fibre biscuits were more resistant to breaking while apple fibre produced a crumbly biscuit. The biscuits' matrices were observed by C-Cell (image analysis): the apple fibre biscuits were more airy and less compact than the wheat fibre biscuits. Sensory analysis by a trained panel showed minor changes in the apple fibre biscuits compared to the control and greater hardness in the wheat fibre biscuits.

Key words: biscuit, fibres, physical and sensory changes

1. Introduction

Nowadays, consumers are increasingly interested in healthy food products. In the 1980s, dietary fibre was identified as an important component of a healthy diet and the food industry began to look for palatable ways to increase the fibre content of its products (Aparicio-Sanguilán, 2007). In the past decade, research has documented connections between dietary fibre intake and decreased risks of chronic diseases (Truswell, 1993). For this reason, fibre has received much attention in both scientific and informative literature.

However, the intake of fibre and fibre-containing foods remains low in many populations worldwide. Interest in foods with high fibre contents has increased in recent decades and the importance of this food constituent has led to the development of a large market for fibre-rich ingredients to be used in baked products such as snacks, muffins or a number of types of biscuits that are convenient to consume at breakfast (Nilsson et al., 2008). Because of their diversity, bakery products are used as a source of different nutritionally-rich ingredients (Sudha, 2007). Biscuits are a very popular bakery item which is consumed by nearly all levels of society. Some of the reasons for such wide popularity are their ready-to-eat nature, affordable cost, good nutritional quality, availability in different tastes and longer shelf life (Ajila, 2008).

Dietary fibres can be classified into insoluble or soluble and each fraction provides specific physiological functions and nutritional benefits: insoluble fibre promotes the movement of material through the digestive system and soluble fibre helps to lower blood cholesterol and regulate blood glucose levels (Tosh et al., 2010). Both types have been used in biscuits to increase the daily intake of fibre, mainly using cereal brans, which are rich in insoluble fibre, and gums such as pectin, which are soluble fibres.

Cereal bran as a source of fibre to replace flour in biscuits has been studied by many authors. Brewers' spent grain has been studied in sugar biscuits (Prentice et al., 1978) and in wire cut biscuits (Örztürk et al., 2002). Gujral et al., (2003) replaced part of the wheat flour with wheat bran and coarse wheat flour in

biscuits; the latter replacement increased the sensory scores and lowered the fracture strength. Leelavathi and Rao (1993) studied the replacement of flour by raw and toasted wheat bran, reporting that the former, up to 30%, could be used as a substitute for flour in the preparation of high-fibre biscuits without affecting the overall biscuit quality. Recently, Ellouze-Ghorbel et al. (2010) used different sources of wheat bran (*Aestivium* and *Durum*) to enrich biscuits.

Sudha et al. (2007) reported on the influence of different cereal brans on the sensory quality of biscuits; they obtained acceptable biscuits by incorporating 30% of oat bran or 20% of barley bran into the formulation.

Some studies have been carried out using fruit fibres: apple fibre (Chen, 1988), banana (Fasolin et al., 2007) and mango dietary fibre (Ajila et al., 2008). Bilgiçli et al. (2007) replaced wheat flour with apple fibre, lemon fibre, wheat fibre and wheat bran; however, they did not compare the effect of the different solubilities of the fibres used.

More recently, a new generation of fibres has been studied, including inulin, oat beta-glucan enriched fraction (BGEF) and potato fibre and resistant starch (Brenan et al., 2004). Aparicio-Sanguilan et al. (2007) obtained biscuits with resistant starch from lintnerized banana and Tuohy (2001) added guar gum, obtaining biscuits with a prebiotic effect.

All the papers mentioned above studied the physicochemical changes in the dough or the biscuit (as well as physiological changes) as being dependent on the source of fibre employed; in general, they did not mention the morphology of the fibre as a factor to take into consideration.

Prentice et al. (1978) and Örtürk et al. (2002) stated that fibre length could be expected to have an important effect on the properties of the dough and the biscuits, but on comparing these two studies, the final conclusion about the influence of different fibre sizes in biscuits was not clear.

The aim of the present work is to study the influence of dose, fibre length and proportion of soluble fraction on short dough biscuits using an apple fibre and two wheat fibres of different lengths to replace part of the flour.

2. Materials and methods

2.1. Ingredients

The three fibres used (Vitacel, J. Rettenmaier & Söhne, Germany, kindly supplied by IRS Iberica, Barcelona, Spain) were wheat fibre (WF) of two different sizes (200 and 101) and apple fibre (AF). The main fibre characteristics provided by the supplier are shown in Table 1.

Table 1. Fibre characteristics as provided by the supplier.

Fibre	Bulk density (g/l)	WBC gH ₂ O/g d.s.	Average particle size (% max)	IDF/SDF (%)
WF-200	75-100	8.3	>250 µm: 5% >75 µm: 5-30% >32 µm: 50-80%	94.5/2.5
WF-101	260-335	4.8	>250 µm: 1% >75 µm: 30% >32 µm: 50%	94.5/2.5
AF-401	450-465	5	>400µm: max 0.5% >150µm: max 40% >32µm: max 80%	75/25

WBC: Water Binding Capacity

IDF: Insoluble Dietary Fibre

SDF: Soluble Dietary Fibre

The amounts of flour (plain soft wheat flour suitable for biscuits: Golden Dawn, Allied Mills, UK) (composition data provided by the supplier: 14.4% moisture, 9.2% protein, 0.6% ash) used in the different recipes were 100, 95 or 90 g, corresponding to replacement of 0, 5 or 10 g of the flour with the different fibres. The codes employed to identify the samples were control (no flour replacement, no fibre) and 5% or 10% followed by the fibre code, meaning that 5% or 10% of the flour had been replaced by that fibre (Table 1); for example, 10%WF-200 means that 10% (10g) of flour was replaced with WF-200 fibre. A total of 7 samples was prepared: control, 5%WF-101, 5%WF-200, 5%AF, 10%WF-101, 10%WF-200 and 10%AF.

The remaining ingredients were (flour weight basis): a) shortening 32.15%: non-hydrogenated vegetable fat for frying (Bako, UK), b) powdered sugar 29.45% (British sugar Plc, UK), c) milk powder 1.75% (Dairy Crest, UK), d) salt 1.05%, f)

sodium bicarbonate 0.35%, e) ammonium hydrogen 0.2% and f) tap water 11%, adjusted for each formula: (control: 11.00%, 5%WF101: 11.60%, 5%WF200:11.73%, 5%AF:12.84%, 10%WF101:12.17%, 10%WF200:12.46%, and 10%AF:13.36%).

2.2. Biscuit preparation

The flour and fibre were pre-blended for 15 minutes in a double cone mixer (KEK-Gardener, UK). The fat, sugar, milk powder, leaving agents, salt and water were mixed in a mixer (Fit Hobart mixer, USA) for 30 seconds at low speed (no.1), the bowl was scraped down and they were mixed again for 3 minutes at a higher speed (no.3). The flour or the flour/fibre mix was then added and mixed in for 20 seconds at speed 1 then, after scraping down the bowl once more, for a further 40 seconds at speed 1.

After a 10-minutes resting period in a plastic bag, the dough was sheeted and moulded to 6.5 cm diameter x 0,5 cm thick (with docking and logo) in a single step using a RTech Minilab sheeting line (Rtech Ltd., Warrington UK); 15 biscuits were placed on each 48x21.5 cm perforated tray and baked in a tunnel oven (Spooner UK) with two sections at different temperatures, 220 and 200 °C, for a total of 5.3 minutes.

2.3. Flour pasting properties

The pasting properties of the flour and the flour/fibre mixes were measured using a Rapid Visco Analyser (RVA-4, Newport Scientific, Australia). Flour or a flour/fibre mix (3.5 g) was added to 25 mL of water. 3.28 and 3.22 g of flour alone (representing the removal of 5% and 10% of the flour respectively) in 25 mL of water were also tested.

Rapid initial stirring was carried out by applying a 960 rpm stirring step for the first 10s of the test, followed by decreasing and stabilizing the stirring to 160 rpm for the rest of the test. A reference maize starch (Colflo 67, National Starch) was employed to calibrate the RVA before testing each batch of each formulation.

The temperature profile consisted of an initial holding time of 1 min at 25 °C, raising the temperature to 95 °C at a rate of 14 °C/min, a holding stage at 95 °C for 3 min and lowering the temperature to 25 °C at a rate of 14 °C/min, followed by a final holding period of 2 min at 25 °C. The paste viscosity was expressed in centipoises (cp; 12 cp \cong RVU - rapid viscosity units). The following parameters were determined: peak viscosity, through viscosity, breakdown, final viscosity, setback, peak temperature and pasting temperature. At least two determinations were carried out in each sample.

2.4. Dough rheology properties

Strain sweep tests were performed using a strain-controlled ARES rheometer (TA Instruments, UK) fitted with parallel plates (25 mm diameter, 2 mm gap). The measurements were performed at 25 °C. The elastic modulus (G'), viscous modulus (G'') and phase angle (δ) were obtained.

The rheological analysis was always carried out just after moulding the dough. Each formulation was prepared twice, on different days, and 4 samples of each preparation were measured. The results were expressed as the average of the 8 determinations performed per formulation.

2.5. Biscuit properties

2.5.1. Texture and sound emissions

The texture of the biscuits was measured using a Texture Analyzer TA.TX.plus (Stable Micro Systems, Godalming, UK). Data management was performed using Texture Exponent software (version 2.0.7.0. Stable Microsystems, Godalming, UK). Twenty replicates of each formulation were conducted.

Breaking strength. The biscuits were broken using the three point bending rig probe (A/3PB). The experimental conditions were: supports 50 mm apart, a 20 mm probe travel distance and a trigger force of 20g. The max force (N), the area (N.sec), and the displacement at rupture (mm) were measured.

An acoustic envelope detector (AED) coupled to the texturometer was used for sound recording; the experimental conditions were adapted from Varela et al.(2006). The gain of the AED was set at 1. A Bruel and Kjaer free-field microphone (8-mm diameter), calibrated using a Type 4231 Acoustic calibrator (94 and 114 dB SPL-1,000 Hz), was placed in a frontal position in order to gain a better acoustic signal, at a distance of 4 cm and an angle of 45° to the sample. A built-in low pass (anti-aliasing) filter set the upper calibrated and measured frequency at 16 kHz. Ambient acoustic and mechanical noise were filtered by a 1 kHz high-pass filter. The AED operates by integrating all the frequencies within the band pass range, generating a voltage proportional to the sound pressure level (SPL). The data acquisition rate was 500 points/s for both force and acoustic signals. All tests were performed in a laboratory with no special soundproofing facilities at an ambient temperature of $22 \pm 2^{\circ}\text{C}$.

The texture and sound parameter measured for each formulation was the number of sound peaks and SPLmax (dB). Each sound graph was simultaneously displayed with the correspondent force/displacement graph and after choosing the real peaks (rather than those due to external noise), the statistics were calculated.

Cone penetrometry. The test was performed under the following experimental conditions: a test speed of 5mm/s, a trigger force of 5g and a cone travelling distance of 12mm. The parameters obtained from the cone penetrometry probe were area under the curve (N/sec), total number of peaks, maximum force (N), maximum time (sec) at break, number of peaks at one sec and force at one sec (N).

2.5.2. Image analysis

Two biscuit from each formulation were cut on a horizontal plane and the surface of the biscuit was removed. The exposed surface was photographed with a C-Cell imaging system (Calibre Control International, Campden & Chorleywood Food Research Association, UK).

2.5.3. Scanning Electron Microscope

The fibres and biscuit samples were examined without any further preparation using a Carl Zeiss EVO 60 Scanning Electron Microscope (Cambridge UK); the pressure was controlled at 60Pa.

The biscuits were broken in two to obtain a fracture surface, then the sample was trimmed behind the fracture surface to produce a strip of biscuit a few millimetres wide for examination. The biscuit sample, with the fractured surface uppermost, was stuck to an aluminium SEM stub with silver 'DAG' cement.

2.5.4. Dimensions

Biscuit thickness was measured by stacking 10 biscuits vertically against the biscuit thickness ruler, sliding the gauge to rest on top of the pile and recording the average thickness.

Biscuit 'length' was measured by arranging the biscuits along the length ruler with the stamped word parallel to its long edge and recording the average length; the biscuits were then rearranged with the written word perpendicular to the long edge on the ruler and the average 'width' was measured. These measurements were expressed in mm as the average value/10 of two replicates.

2.5.5. Moisture and fibre determination

The % moisture content of the flour, flour/fibre mixtures and biscuits was determined according to Approved Method 44-15.02 (AACC International, 2009).

The total dietary fibre content of the doughs and biscuits was determined in three replicates by Approved Method 32-07.01 (AACC International, 2009), using a FOSS Fibretec E 1023 Filtration module and Shaking Water Bath 1024 system.

2.6. Sensory Analysis

Selection of terms and panel training. A panel of eight assessors (between 25 and 38 years old) skilled in quantitative descriptive analysis (QDA) was trained to select the descriptors using the checklist method.

Terms were selected and discussed in an open session with the panel leader. The assessors were given a brief outline of the procedures and a list of attributes and representative samples, and were asked to choose and write down the most appropriate attributes to describe all the sensory properties of the biscuits, or to suggest new ones. The panel leader collected and wrote all the attributes on a board. The panel then discussed the appropriateness of the selected attributes, their definitions and the procedures for assessing them. At the end of the session a consensus on the list of attributes (colour; thickness; flour, butter and toast odour; visual structure; manual hardness; crumbliness; fragility; hardness; crunchiness; doughy, flour, butter and apple taste) and procedures had been chosen; this procedure was proposed by Stone and Sidel (2004) in order to obtain a complete sensory description of a product.

The panellists attended twelve 1-hour training sessions. Training involved two stages: in the first stage, different samples were tested by the panellists to gain a better understanding of all the descriptors and different tastings were carried out until the panel was homogeneous in its assessments, as explained here below under Statistical Analysis. In the second stage, the panellists used 10-cm unstructured scales to score the intensity of the selected attributes. The assessors were instructed to score the external appearance first, followed by odour, then the manual properties of the biscuit, then in-mouth texture and, finally, taste. The panel's performance was evaluated by principal component analysis, using the Pearson correlation matrix, until there were no outliers in the group between the different training sessions.

Formal assessment. A balanced complete block experimental design was carried out in duplicate (two sessions) to evaluate the samples. The intensities of the sensory attributes were scored on a 10 cm unstructured line scale. Seven

samples were evaluated per session. In each session, the samples were randomly selected from each cooking batch and served in random order, each on a separate plastic tray identified with random three-digit codes. The panellists were instructed to rinse their mouths with water between sample evaluations.

Testing was carried out in a sensory laboratory equipped with individual booths (ISO 8589, 1988). Data acquisition was performed using Compusense five release 5.0 software (Compusense Inc., Guelph, Ont., 158 Canada).

2.7. Statistical Analysis

Analysis of variance (one way-ANOVA) was applied to study the differences between formulations; least significant differences were calculated by the Tukey test and the significance at $p < 0.05$ was determined. These analyses were performed using SPSS for Windows Version 12 (SPSS Inc., USA).

For each descriptor, two-way ANOVA was applied to check panel performance considering assessors, samples and their interaction as factors.

Analysis of variance (one-way ANOVA) was applied to the trained panel in order to study the effect of formulation; least significant differences were calculated by Tukey's test ($p < 0.05$).

3. Results and discussion

3.1. Fibre morphology and hydration properties

The morphology of the different fibres was studied by SEM. The two wheat fibres both had an elongated shape but were different in length, WF-200 being longer than WF-101 (Figures 1a and 1b, respectively); the apple fibre had a completely different morphology, as its particles were shorter and rounded in shape (Figure 1c). These observations were in accordance with the size data provided by the supplier (Table 1).

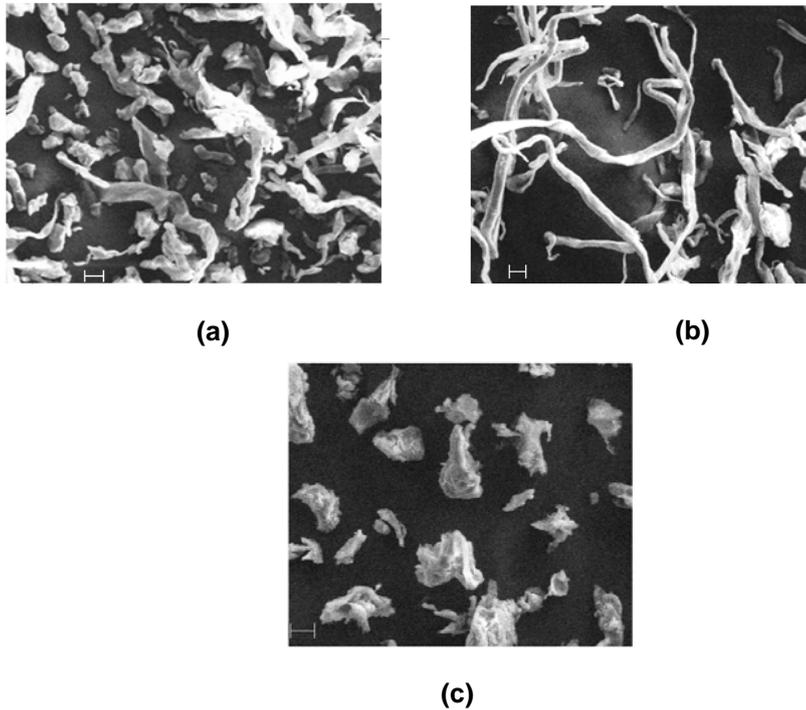


Figure 1. Microphotographs of the different fibres (a) Wheat fibre 101; (b) Wheat fibre 200; (c) Apple fibre. (Scale=20 μ m)

Mongeau et al. (1978) studied the water holding capacity of insoluble dietary fibres in relation to particle size and affirmed that at a lower particle size, fewer pores and holes remained in the wheat fibre to hold water; in accordance with this, the longer wheat fibre (WF-200) should be able to bind more water than the shorter wheat fibre (Table 1). Chaplin et al. (2003) investigated the interaction between dietary fibre and water binding and affirmed that fibre can bind water directly in a number of ways. For instance, polysaccharide single chains are able to interact with other chains to form junction zones which enclose large amounts of water, strongly bound and fairly static. They found that these junction zones were denser and less flexible than single chain zones.

This situation effectively increases the fibre diameter of the junction zones, reducing the pore size, increasing the capillarity and reducing the tendency for the water to exit the cavities. That could also explain why WF-200 has a higher water binding capacity (WBC) than WF-101, as the long chains could form more junctions, entrapping the water better.

The morphology of the apple fibre (AF-401) indicated a completely different scenario. Whilst wheat fibre is almost totally insoluble, apple fibre contains 25% soluble fibre. Although it does not contain long or long-shaped fibres, its soluble fraction (pectin) is highly hydrophilic (Tosh et al., 2010), exhibiting a WBC value approaching that of the longer wheat fibre (WF 200). Thibault et al. (2001) reported that the hydration properties of fibres depend on their chemical and physical structures (surface area and particle size) and on their processing history.

In turn, all these factors that could affect the water status in the dough would affect its technological characteristics (such as handling properties) and the final properties of the biscuits.

While water is a minor component in a biscuit dough formula, it plays an important role during dough and biscuit preparation (Pareyt et al., 2008). The water level used in the recipe affects gluten development in the dough, biscuit spread during baking, moisture retention and the eating quality of the finished product (Lai et al., 2006).

In the present study the fibres absorbed high amounts of water, preventing proper hydration of the ingredients and making the dough powdery or causing it to fall apart. In consequence, the proper water level for each formulation was determined by a mixograph in preliminary tests (data not shown).

3.1.2. Flour pasting properties

The parameters generated by the RVA describe starch swelling, gelatinization, and gelling ability. During one RVA cycle (heating- temperature maintenance - cooling), the starch granules in excess water swell and absorb moisture, leading

to disruption of the hydrogen bonds and leaching of amylose from the starch granules. Finally, during the cooling cycle stage, the starch molecules reassociate and form a gel (Brennan et al., 2004). The changes in rheological properties observed during starch gelatinization depend on the presence of swollen starch granules in a dispersed amylase-amylopectin matrix and on the amylose-amylopectin interaction (Eliasson et al., 1986), which would also be affected by the addition of fibre.

The maximum peak viscosity was obtained with flour alone (100% = 3.5 g). In the samples containing 5% and 10% less flour (3.28 and 3.11g of flour respectively in the same amount of water), the peak viscosity values decreased as expected. When these percentages of flour were replaced with any of the fibres, the values recovered to some extent, varying according to the type of replacement (Table 2). The closest values to the 100% flour sample corresponded to 5%WF-200, where they were higher than for the 5%-less flour-only sample, so this fibre makes the mix more viscous, probably by absorbing part of the available water. This sample was followed by 5%AF and 5%WF-101. Whilst the composition of WF-200 and WF-101 was the same, their differing physical shapes led to different behaviour. The samples with 10% replacement followed the same trend (WF-200>AF>WF-101), but in this case they all presented higher peak viscosity values than the flour-only sample with 10% less flour.

All the pasting parameter values indicated that the effects on these properties of replacing part of the flour with fibres could be

Table 2. Pasting properties of the flour and flour/fibre mixtures.

Ingredients	Flour/fibre (total=3.5g)	Peak viscosity (cp)	Trough (cp)	Breakdown (cp)	Final viscosity (cp)	Setback (cp)	Peak T (°C)	Pasting T (°C)
Flour	3.5/0.00	133.75 ^e (0.6)	64.46 ^{de} (2.0)	69.29 ^e (1.4)	136.71 ^{cd} (1.9)	72.25 ^{cd} (0.01)	5.60 ^a (0.01)	85.18 ^a (0.67)
Flour (5% less)	3.28/0.00	120.79 ^d (0.9)	60.17 ^{bc} (1.2)	60.63 ^d (0.3)	133.67 ^e (2.8)	73.50 ^d (1.65)	5.60 ^a (0.09)	86.40 ^b (0.01)
Flour (10% less)	3.11/0.00	97.96 ^a (0.3)	52.00 ^a (0.1)	45.96 ^a (0.3)	113.88 ^a (0.2)	61.88 ^a (0.18)	5.50 ^a (0.05)	87.60 ^c (0.57)
5%WF-200/Flour	3.28/0.17	124.29 ^e (0.9)	65.58 ^e (0.1)	58.71 ^d (0.9)	136.17 ^{cd} (0.1)	70.58 ^{bcd} (0.01)	5.57 ^a (0.05)	86.45 ^{bc} (0.07)
5%WF-101/Flour	3.28/0.17	117.63 ^{cd} (1.4)	59.00 ^b (0.6)	58.63 ^d (0.8)	126.21 ^b (1.5)	67.21 ^b (0.88)	5.60 ^a (0.01)	86.35 ^{ab} (0.01)
5%AF/Flour	3.28/0.17	120.00 ^{cd} (0.7)	62.00 ^{bcd} (0.6)	58.00 ^d (0.1)	130.25 ^{bc} (1.3)	68.25 ^{bc} (0.71)	5.43 ^a (0.05)	86.28 ^{ab} (0.04)
10%WF-200/Flour	3.11/0.34	117.00 ^c (0.6)	63.67 ^{cd} (0.1)	53.33 ^c (0.7)	130.21 ^{bc} (2.5)	66.54 ^b (2.42)	5.47 ^a (0.01)	86.38 ^b (0.04)
10%WF-101/Flour	3.11/0.34	101.04 ^a (1.0)	53.46 ^a (1.1)	47.58 ^{ab} (0.1)	112.54 ^a (1.2)	59.08 ^a (0.12)	5.47 ^a (0.01)	87.20 ^{bc} (0.14)
10%AF/Flour	3.11/0.34	109.21 ^b (0.5)	59.08 ^b (0.2)	50.13 ^b (0.8)	126.88 ^b (0.3)	67.79 ^b (0.06)	5.47 ^a (0.09)	86.35 ^{ab} (0.01)

Values in parentheses are standard deviations.

Means in the same column without a common letter differ ($p < 0.05$) according to the Tukey test.

Peak viscosity: the maximum viscosity developed during the heating portion of the test

Trough: minimum viscosity after the peak normally occurring around the start of sample cooling

Breakdown: peak viscosity minus trough viscosity

Final viscosity: the viscosity at the end of the peak test

Setback: final viscosity minus trough viscosity

Peak temperature: temperature at which peak viscosity occurred

Pasting temperature: temperature at which viscosity first increased by at least 25 cp over 20 sec.

explained by the different WBCs of the fibres, which make less water available for the flour.

Brennan et al., 2004 studied flour replacement with resistant starch (RS) and inulin; as the level of these two fibres rose, the peak viscosity values fell, an effect that these authors attributed to a reduction in the amount of gelatinizable starch. In the present case, when the flour was replaced with fibre the pasting values were slightly higher than those for the reduced flour alone samples (5% or 10% less), so although the starch content was lower, the fibre helped to recover (and in some cases exceed) the viscous properties of the starch (flour) that had been replaced.

3.2. Dough properties

Dough properties depend on the contributions of the different ingredients as starch, proteins and the water present, which in turn influence the handling properties. If the dough is too firm or too soft, it is not easy to handle; the dough must be sufficiently cohesive to hold together during the different processing steps and viscoelastic enough to separate cleanly when cut by the mould (Gujral et al., 2003).

Rheological properties. The G' and G'' values were independent of the applied strain up to a critical value (δ_c) which defines the onset of non-linear response. In all the doughs, G' was always higher than G'' (Table 3). The replacement of flour by all the fibres resulted in an increase in both G' and G'' values, the highest increase being found with 10% replacement by WF-200; 5%WF-200, 10%WF-101 and AF doughs showed similar elasticity (G') results. Therefore, the main difference was that the longest fibre (WF-200) gave the dough the most elasticity.

Table 3. Influence of different fibres and flour replacement levels on linear viscoelastic properties during strain sweeps at 25°C. G' (storage modulus), G'' (loss modulus), |G*| (complex modulus), tan delta and critical strain γ_c .

Sample	G'	G''	G*	Tan delta	γ_c
Control	223483 ^a (18567)	128615 ^a (12114)	257853 ^a (22117)	0,58 ^d (0,01)	0,054 ^{bc} (0,01)
5%WF101	277455 ^b (22114)	154886 ^{ab} (12623)	317773 ^{ab} (25253)	0,56 ^{cd} (0,01)	0,058 ^c (0,00)
5%WF200	359857 ^c (24016)	195714 ^c (14302)	409544 ^{cd} (27855)	0,54 ^{bc} (0,01)	0,051 ^{abc} (0,01)
5%AF	226500 ^a (21079)	129250 ^a (12632)	260871 ^a (24436)	0,57 ^{cd} (0,01)	0,059 ^c (0,00)
10%WF101	365914 ^c (16145)	196430 ^c (7204)	415346 ^{cd} (16385)	0,54 ^b (0,02)	0,039 ^{ab} (0,01)
10%WF200	661200 ^d (35968)	321200 ^d (13627)	751351 ^d (83974)	0,49 ^a (0,02)	0,037 ^a (0,01)
10%AF	317000 ^{bc} (32501)	169714 ^{bc} (19533)	359661 ^{bc} (37443)	0,53 ^a (0,02)	0,045 ^{abc} (0,01)

Values in parentheses are standard deviations.

Means in the same column without a common letter differ ($p < 0.05$) according to the Tukey test.

A decrease in tan delta (values closer to 0) was also found for the 10% replacement with WF-200, implying greater predominance of the elastic as opposed to the viscous component.

The effect of the fibres on the onset of non-linear response (γ_c) was only significant for the 10% fibre addition level, which reduced the γ_c values, meaning that the sample became less resistant to the applied strain.

3.3. Biscuit properties

Many chemical and physicochemical reactions occur during baking, like protein denaturation, some loss of the starch's granular structure, fat melting, Maillard reactions and browning, dough expansion, water evaporation and the production and thermal expansion of gases (Chevallier, 2002). After baking, the dough will be transformed into a solid structure where each ingredient has different roles: the flour influences water binding and limits the dough's expansion, fat gives aeration and lubrication and interferes with gluten

development and sugar influences dough viscosity and gluten development (Pareyt et al., 2008).

In the present work the flour was replaced by fibres with different properties and compositions; in consequence, the dough properties had changed and the properties of the biscuit would conceivably also be affected.

3.3.1. *Texture analysis*

3-Point break test. All the biscuits fractured under tension and the fracture took place relatively close to their central zone (in the lower surface), where the maximum stress occurred. This implied that the condition regarding the distance between supports was satisfied. The curves obtained from the 3-point bending test (not shown) were similar to others previously reported for materials with brittle fracture patterns (Saleem, 2005): they were characterized by an initial elastic response followed by a small fracture strain.

No significant differences were found in the values for maximum force (hardness) and displacement on breaking into two pieces, although they were higher for the biscuits containing WF-200, followed by WF-101 (Table 4). The samples with AF presented similar maximum force values to those of the control sample. Consequently, a clear relationship was found between fibre size and hardness: the greater the fibre size, the higher force required to break the biscuit. This was observed for both levels of fibres replacing part of the flour.

The number of sound peaks, an indication of crispness, showed that the wheat fibre reduced this attribute (at both replacement levels), which means that there was less microcracking and probably denotes a more compact biscuit matrix, whereas the apple fibre retained or increased biscuit crispness. This result is of great technological importance when deciding which fibre to use to enrich a biscuit, as it shows that a smaller particle size will give a crisper, crumblier texture.

Displacement at rupture presented no significant differences between fibres (Table 4), however, it can be observed that the control biscuit broke before the

biscuits with flour replacements, meaning that the control biscuits presented a lower capacity for elastic deformation.

Saleem et al. (2005) reported a correlation between the moisture content and the curves obtained by 3-point bending, pointing out that for less moisture the load (N) was higher; in the present case, no correlation between moisture content and force was found with fibre enrichment; this discrepancy is probably because those authors studied changes in moisture higher than 1%, whereas in the present case the maximum difference between the moisture contents of the samples was 0.49%.

Previous authors (Sudha et al., 2007; Gujral et al., 2003) have observed increased breaking strength when flour is replaced by fibre. Brennan et al. (2004) used different soluble and insoluble fibres for flour replacement in biscuits, observing a slight increase in the breaking strength probe values for those containing potato peel (insoluble) and no increase for inulin and beta-glucans (soluble). However, none of these studies discussed the particle size of the fibres added.

Cone penetrometry test. The area under the curve could be an indication of the sample's resistance to cone penetration as well as of the toughness of the sample; 10%WF-200 was the toughest biscuit (Table 4).

All the force deformation curves showed numerous peaks that could be understood as the breaking events that occurred when the probe passes through the layers within the product structure. A more compact structure will be reflected by fewer peaks and an airy or layered structure by a high number of peaks.

Table 4. Instrumental texture characteristics, composition data and dimensions of control and fibre-added biscuits.

Sample	3-Point break test				Cone penetrometry				Composition data				Dimensions	
	Max Force (N)	Rupture (mm)	No. of sound peaks	Area (N/s)	No. of peaks at 1 s	Force at 1 s (N)	Moisture (%)	Fibre (%)	Height (cm)	Length (cm)	Width (cm)			
Control	13.70 ^{ab} (4.36)	0.38 ^a (0.2)	3.17 ^{abc} (1.5)	20.4 ^{bc} (6.7)	15.40 ^{cd} (2.41)	13.53 ^a (2.95)	3.75 ^c (0.03)	5.90 ^a (0.55)	0.77 ^a (0.01)	6.75 ^a (0.02)	6.80 ^a (0.02)			
5%WF-101	17.42 ^{bc} (2.33)	0.42 ^a (0.1)	2.77 ^{ab} (1.8)	25.8 ^{cd} (5.6)	12.79 ^b (2.15)	18.94 ^{bc} (3.99)	3.54 ^{ab} (0.02)	8.96 ^b (0.03)	0.78 ^a (0.05)	6.76 ^a (0.03)	6.75 ^a (0.01)			
5%WF-200	23.24 ^d (5.32)	0.53 ^a (0.1)	2.91 ^{ab} (1.4)	21.5 ^{bcd} (7.7)	13.77 ^{bc} (2.33)	18.15 ^{bc} (1.96)	3.65 ^b (0.03)	8.86 ^b (0.14)	0.79 ^a (0.04)	6.66 ^a (0.01)	6.74 ^a (0.07)			
5%AF	14.16 ^{ab} (2.99)	0.44 ^a (0.1)	5.10 ^c (2.0)	13.0 ^a (3.9)	16.89 ^d (2.08)	14.23 ^a (3.72)	3.67 ^{bc} (0.02)	9.70 ^{bc} (0.71)	0.75 ^a (0.03)	6.74 ^a (0.01)	6.80 ^a (0.01)			
10%WF-101	21.03 ^{cd} (4.37)	0.48 ^a (0.1)	3.63 ^{abc} (1.4)	27.7 ^{ab} (8.8)	12.35 ^b (3.30)	16.48 ^{ab} (4.86)	4.03 ^a (0.02)	11.08 ^{bc} (0.86)	0.78 ^a (0.07)	6.74 ^a (0.02)	6.79 ^a (0.01)			
10%WF-200	24.18 ^d (3.84)	0.52 ^a (0.2)	1.94 ^a (0.8)	29.8 ^a (8.5)	9.71 ^a (2.35)	21.60 ^c (2.69)	3.90 ^a (0.03)	11.08 ^{bc} (0.86)	0.79 ^a (0.01)	6.70 ^a (0.06)	6.78 ^a (0.02)			
10%AF	12.39a (2.54)	0.41 ^a (0.2)	4.25 ^{bc} (1.8)	15.5 ^{ab} (8.8)	14.86 ^{cd} (1.68)	16.48 ^{ab} (5.09)	3.70 ^{bc} (0.08)	11.67 ^c (0.32)	0.75 ^a (0.02)	6.75 ^a (0.03)	6.75 ^a (0.04)			

Values in parentheses are standard deviations.

Means in the same column without a common letter differ ($p < 0.05$) according to the Tukey test

In order to study the biscuit matrix properly, as well as the mean resistance of each biscuit, the number of peaks and the force at one second were also measured (Table 4). Fewer peaks and a higher maximum force were observed for the 10%WF-200 biscuit, while the AF fibre provided the biscuit with a high number of peaks, related to an airy structure and lower maximum force as the cone penetrated, which could be related to its having a brittle structure and being the easiest to fracture.

The differences between WF-200 and WF-101 could be attributed to their different fibre size, as the long WF-200 could form chain entanglements, creating an internal network that provides strength and more resistance at the breaking point. Despite this, the WF (WF-101 and WF-200) samples were very similar to each other.

3.3.2. *Biscuit images*

Baltsavias et al. (1999) considered the biscuit a cellular solid, a model structure of connected beams or plates. In the present study, each peak found by the cone penetrometry was related to a microfracture in the biscuit matrix, so it could be said that the control biscuit and AF biscuit contained more of Baltsavias' plates, created by air inside the biscuit matrix. In order to confirm this theory, the structure shown by images was studied.

The photographs of 10%WF-200 proved it to have a highly compact structure, while the control had more air pockets and holes in the biscuit matrix (images not shown). Blaszcak et al. (2004) explained that biscuit matrix properties are mainly determined by air spaces and fat globules. These observations are in accordance with the texture results obtained in the present study: more microcracking during cone penetrometry occurred in the control samples, favoured by air pockets, whereas the wheat fibres created a more compact structure. Images of the AF biscuits could not be obtained due to the brittleness of the samples, which made them impossible to cut perpendicularly without breaking into several pieces.

3.3.4. Scanning Electro Microscopy

No evident differences due to replacing flour with fibres were observed in the SEM photographs of the biscuits' matrices (images not shown). The photographs showed two kinds of fat globules in the biscuit matrix, as described by other authors (Blaszczak et al., 2004); the matrix observed was mainly protein with embedded fat globules and starch granules.

3.3.5. Dimensions

No significant differences were found in the biscuits' height, width or length due to the addition of fibres. Some reduction in the biscuits' dimensions due to fibre addition has been observed in previous works; Brennan et al. (2004) suggested that the fibres may act as biscuit dough mixture stabilizers at up to 10% of replacement, enabling the reformulated biscuit dough to retain its diameter during baking. Higher replacement of flour (60%) with resistant starch has been related to a lower gluten content (Laguna et al., 2010).

3.3.6. Moisture and fibre content

The analysis showed no significant differences in the final moisture content of the biscuits (Table 4). Each recipe had a different water content, so the water loss rate during the baking process was also different. The dough with the highest water loss was AF (at both 5% and 10%). This could be attributed to the apple fibre's being unable to retain as much water as the cellulosic material of the wheat fibre.

The fibre content (Table 4) showed the expected differences due to the percentages of fibre added.

3.4. Sensory analysis

The appearance of the wheat fibres was that of white powders with a neutral flavour and odour; the apple fibre had a slightly fruity odour and a beige-brown colour.

Figure 2 shows the scores obtained, displaying only the attributes which presented significant differences compared to the control. The trained panel scored the 5%WF samples as being less crunchy and flaky (a more compact biscuit structure), with a lighter colour, harder (manual and in-mouth) and with a more doughy mouthfeel (Figure 2a). For the biscuits with 10% of the flour replaced by the two wheat fibres (WF-200 and WF-101), these effects were stronger (Figure 2b). Comparing the wheat fibre samples at the same percentage of replacement, the longer of the two (WF-200) produced biscuits that differed more from the control than the shorter one (WF-100). These results were in agreement with the instrumental texture results.

The sensory textural profile of the AF biscuits presented no significant differences compared to the control, as was expected in view of the instrumental measurements. However, the colour and taste attributes did differ (Figure 2c): the AF samples were darker, with a toast odour which masked the floury and butter taste and with a certain apple taste.

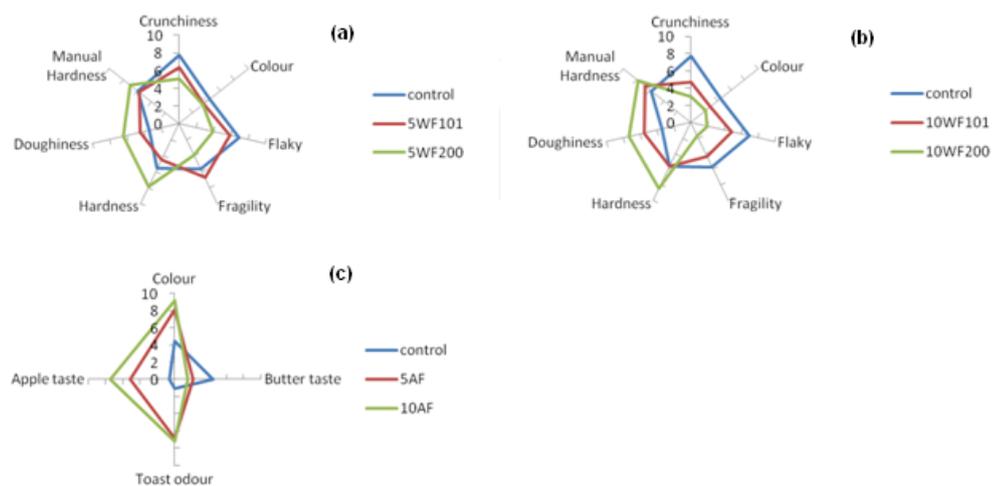


Figure 2. Mean descriptive sensory scores for biscuits (a) control, 5%WF-200, and 5%WF-101; (b) control, 10%WF-200, and 10%WF-101; (c) control, 5%AF and 10%AF.

Conclusions

This study has shown the influence of the morphology of the different fibres on the dough and biscuit matrix properties, making proper selection of fibre size and shape essential when formulating a fibre-enriched biscuit.

The apple fibre needed more water to reach the correct water level for dough handling, although all the fibre-enriched biscuits had a similar final water content. The AF biscuits had similar texture properties to the control but the fibre gave them a fruity taste. The biscuits with wheat fibre were neutral in flavour but harder in texture; this was attributed to the high water binding capacity of these elongated fibrils, which created a compact biscuit matrix structure. The medium length wheat fibre (WF-101) biscuits were not as hard as those with the longer wheat fibre (WF-200).

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A NEW SENSORY TOOL TO ANALYSE THE ORAL TRAJECTORY OF
BISCUITS WITH DIFFERENT FAT AND FIBRE CONTENTS

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FOOD RESEARCH INTERNATIONAL

Abstract

Reformulating traditional products such as biscuits can be a useful tool for providing the population with healthier snacks. However, it involves changes in the eating characteristics of the final product. This study focuses on the oral perception of these biscuits, using the Temporal Dominance of Sensation (TDS) technique with two different amounts of fat (60g and 30g fat/100g flour) and fibre (4g and 8g fibre/100g flour). The TDS data obtained with a trained panel showed that hardness was the first dominant attribute in all the formulations during the mastication process. The dominance of the other parameters appeared to depend more on the fat and fibre contents, as crispness appeared with high-fat biscuits and crunchiness with low-fat, high-fibre ones, while both attributes were perceived in intermediate formulations. In the high-fibre formulations, grittiness and dry mouthfeel appeared during chewing and dry mouthfeel was dominant. At the end of the mastication all the biscuits were perceived as pasty. A fat mouthfeel was also perceived with both high-fat and low-fat biscuits, with or without the addition of a low level of fibre. Biscuits with high fibre and low fat contents were perceived as less crisp. Penalty analysis based on JAR scales, showed that excessive hardness and dry mouthfeel were the most penalizing attributes causing significant drop in biscuit acceptability.

Keywords: oral processing; temporal dominance of sensations; biscuit; consumer perception

1. Introduction

Biscuits are consumed at almost every level of society. The reasons for the widespread popularity of these baked products include their being ready-to-eat, affordable, of good nutritional quality, available in different tastes and having a long shelf life (Ajila, Leelavathi, & Rao, 2008). However, biscuits contain high levels of fat and nowadays consumers demand reduced-calorie foods with health benefits (Handa, Goomer, & Siddhu, 2012).

Reducing fat and adding fibre to biscuits without affecting the sensory characteristics is a significant challenge. Fat imparts shortening, richness and tenderness, improving flavour and mouthfeel (Pareyt & Delcour, 2008). Fibre generally gives the product some flavour but makes it unpalatable, with a coarse texture and a poor, dry mouth feel (Yue & Waring, 1998). In consequence, lowering the fat content and increasing the fibre are expected to greatly influence mechanical behaviour in the mouth, which has implications for product acceptability (Hutchings & Lillford, 1988).

Brown & Braxton (2000) related a preference for "Rich Tea"-type biscuits with their relative ease of oral breakdown, using the Time Intensity Procedure. Time intensity (TI) is the method most often used to study the evolution of sensations with time. However, only one attribute is evaluated at a time and the number of attributes is limited (Pineau, *et al.*, 2009). Other dynamic sensory methodologies that have been used to describe changes in sensory attributes during the eating process include Dual Time Intensity, which measures two sensory attributes simultaneously (Duizer, Bloom, & Findlay, 1997), or Progressive Profiling, where the panellist scores the same attributes several times during eating (Jack, Piggot, & Paterson, 1994). However, these techniques cannot give information about the dominant attributes in real time while the product is being eaten.

A new sensory method called Temporal Dominance Sensation (TDS) presents the panellists with a complete list of attributes, from which they choose the dominant one at each point in time, at the same time also scoring its intensity

up to the moment the sensation ends (Pineau *et al.*, 2009). Labbe *et al.* (2009) and Pineau *et al.* (2009) compared the TI and TDS techniques in gels with different levels of odorants and in dairy products, respectively, concluding that although both techniques resulted in closely-matched behaviour patterns, TDS improved the sequencing of sensations over time. Albert, Salvador, Schlich, and Fiszman (2012) compared the results of TDS (with untrained panellists) and descriptive sensory profiling (with a trained panel). They showed that TDS gave similar results, making it possible to monitor the behaviour of the food piece when it is broken down and physically transformed by the organs of the mouth.

The objectives of this study were: (1) to conduct an in-depth study of the mechanical phenomena occurring in mouth during the oral processing of biscuits using TDS, (2) to study the impact of fibre addition and fat reduction on the perception of texture over time during in-mouth handling, (3) to achieve a better understanding of the factors affecting consumer acceptance.

2. Materials and Methods

2.1. Biscuit Ingredients and Preparation

Six samples (3x2 design) were tested. The percentages of the biscuit ingredients are shown in Table 1. The code letters for the samples were C (control, complete formulation), LF (50% less fat), LW and HW (respectively 4% and 8% added fibre replacing flour), LWLF (low fibre and low fat) and HWLF (high fibre and low fat).

Ingredients. The ingredients employed to prepare the biscuits were: soft wheat flour suitable for biscuits (Belenguer, S.A., Valencia, Spain) (composition data provided by the supplier: 15% moisture, 11% protein, 0.6% ash; alveograph parameters $P/L=0.27$, where P is the maximum pressure required and L is the extensibility; W, the baking strength of the dough, was 134), shortening (St. Auvent, Vandemoortele France), fibre (wheat fibre with a fibre length of 200 μ m, (Rettenmaier Ibérica, Barcelona, Spain), sugar (Azucarera Ebro, Madrid,

Spain), milk powder (Central Lechera Asturiana, Granda, Spain), salt, sodium bicarbonate (A. Martínez, Cheste, Spain), ammonium hydrogen carbonate (Panreac Quimica, Barcelona, Spain) and tap water.

Table 1. Biscuit formulations.

Ingredients (g/100g flour)	C	LF	LW	LWLF	HW	HWLF
Flour	100	100	96	96	92	92
Fat	60	30	60	30	60	30
Wheat fibre	0	0	4	4	8	8
Sugar	30	30	30	30	30	30
Milk powder	1.8	1.8	1.8	1.8	1.8	1.8
Salt	1	1	1	1	1	1
Sodium bicarbonate	0.4	0.4	0.4	0.4	0.4	0.4
Ammonium bicarbonate	0.2	0.2	0.2	0.2	0.2	0.2
Water	9.3	9.3	9.3	13	9.3	15

Preparation. The sugar, milk powder (previously dissolved in all the water), leavening agents and fat were mixed in a mixer (Kenwood Major Classic, UK) for 30 seconds at low speed (60 rpm). After scraping down the bowl, the dough was mixed for a further 3 minutes at a higher speed (255 rpm). The flour or flour/fibre mixture was added and mixed in for 1 minute at a speed of 60 rpm. The dough was then allowed to rest for 10 minutes at room temperature and finally rolled to a thickness of 10mm in a dough laminating machine (Parber, Zamudio, Spain).

The dough was cut into circular pieces (30 mm in diameter and 10 mm in height). Fifty-four pieces were placed on a perforated tray. The biscuits were baked in a conventional oven for 25 min at 175 °C. The oven and the oven trays were always the same, the trays were placed at the same level in the oven and the number of biscuits baked was always the same. After cooling to ambient temperature, the biscuits were packed and stored in heat-sealed metalized polypropylene bags.

2.2. Sensory Analysis

2.2.1 Temporal Dominance of Sensations (TDS)

Selection of terms and panel instruction and training. Thirteen assessors with previous experience in quantitative descriptive analysis of short dough biscuits participated in this study.

Four 1-hour preliminary sessions were conducted in order to explain the TDS technique and the notion of temporality of sensations and give the assessors the chance to test the data collection software and familiarize themselves with it. In the first session the subjects described two very different biscuits (C and HWLF) and generated a list of terms, mainly focusing on texture change descriptors over the mastication period. In the second session, the most frequently cited attributes were selected and their definitions and the protocol for measuring them were developed (Table 2).

Table 2. Attributes definitions generated by the trained panel.

Attribute	Description
Hardness	Force required breaking the biscuit with the incisors. From not to very.
Crispness	High pitched sound produced when the product brittles under the teeth during mastication, as a potato chip, with multiple fractures at low work of force From not to very.
Crunchiness	Low pitched sound produced at biscuit fracture during mastication, as in an almond. From not to very.
Pastiness	Mouthfeel of ball or paste formation. From not to very.
Fat mouthfeel	Film fat/oil feeling in mouth. From not to very.
Grittiness	Describe the presence of small dry particles which tend to scrape off the tongue. From not to very.
Dry mouthfeel	Related with the feeling for dryness in the mouth. From not to very.

During the third and fourth sessions the panellists were able to understand the dominance and sequence concepts and participated in a simulated TDS session with several samples of biscuits in order to solve questions and get used to the computer program and methodology.

In the computer screen (Fig. 1) the complete list of the selected attributes is presented. After pressing the Start button, the panellists are asked to choose the dominant sensations (scoring their intensity although intensity is not the key information recorded in a TDS task) over the time of consumption. Once the sample is completely swallowed they should press the Stop button. To make the task affordable, it is recommended that the list contains less than 10 attributes; in addition, the panel training should be oriented to the identification of the different sensory qualities (i.e. the sensory attributes) to improve dominant attribute selection (Pineau *et al.*, 2012).

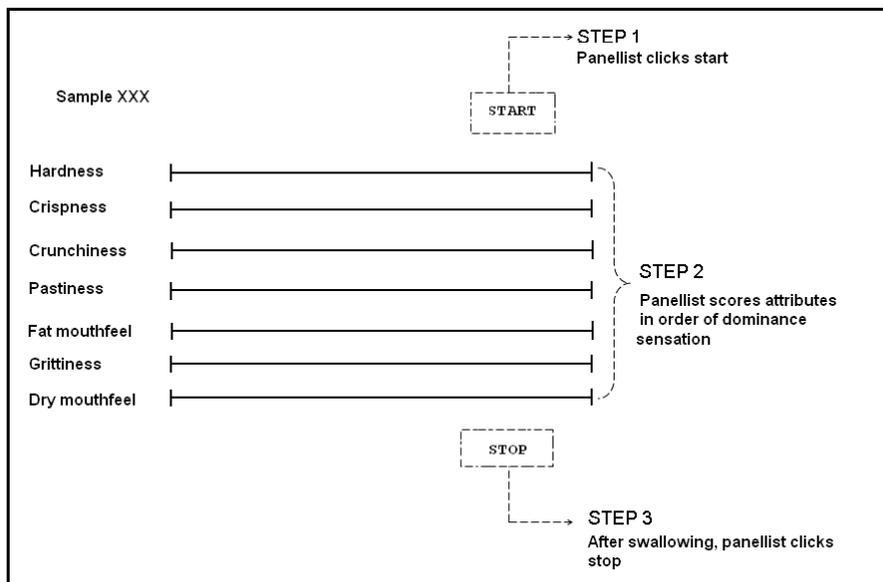


Figure. 1. Example of a computer screen for performing Temporal Dominance of Sensations in biscuits

Formal assessment. The TDS evaluation took place over three sessions held on three different days in order to conduct three replications. The samples were

presented in a sequential monadic series on plastic trays and the panellists were instructed to bite off half a biscuit. On the computer screen, the panellists were presented with a list of the seven attributes, each associated with an 100mm unstructured scale anchored from weak to strong.

Temporal Dominance of Sensations data analysis. The data was collected with Fizz Software version 2.45 (Biosystems, Couternon, France). As explained by Lenfant et al. (2009), the attribute chosen as dominant and the times when the dominance started and stopped were collected for each panellist run. As the duration of mastication up to swallowing differed from one subject to another and the sensory perception time scales differ as a result, the data were normalized by adjusting them according to each subject's individual duration of mastication (Albert et al., 2012). An example of the raw data can be seen in Fig. 2, which represents one sample and its dominant attributes as chosen by one panellist over the consumption period. In this example, the panellist reported 'hardness' for seconds 3 to 11, 'crispness' for seconds 11 to 27 and so on. The dominance rates across the panel for each sensation at different time points were then plotted for each sample, and these data were represented by smoothed curves.

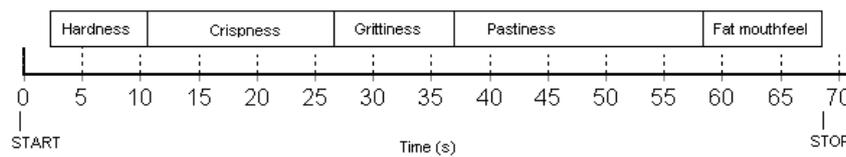


Figure 2. Example of raw TDS data for one biscuit

In the TDS curves, the period when a sensation was dominant for a product at panel level (dominance rate) was computed at each point of time (Lenfant et al. 2009), then the intensity is not taken into account, TDS curves of all the attributes are shown in the same graph.

When the TDS curves were plotted, two additional lines were drawn for the chance and significance levels. The chance level refers to the dominance rate

that an attribute could obtain by chance. Its value is inversely proportional to the number of attributes ($P_0 = 1/p$, where p is the number of attributes). The significance level is the minimum value this proportion should equal if it is to be considered significantly ($p < 0.05$) higher than P_0 . It is calculated as follows:

$$P_s = P_0 + 1.645 \sqrt{\frac{P_0(1-P_0)}{n}}$$

where P_s is the lowest significant proportion value ($\alpha = 0.05$) at any point in time for a TDS curve and n is the number of subjects x number of replications.

Rosner (1995) recommended that $np(1-p) > 5$ (where n = number of trials and p = probability of success). In the present study, 13 panellists performed 3 replications of each product and 7 attributes were used.

2.2.2. Consumer test

All the consumer sessions were held mid-morning, before lunchtime (11.00 - 13.00). A total of 100 consumers aged from 18 to 65 years who frequently consume this type of biscuit agreed to take part in the study. The consumers evaluated the six samples in a single session. The biscuits were coded with random three-digit numbers following a balanced complete block experimental design. Overall liking, texture liking and flavour liking were scored on nine-point hedonic scales (from 1=dislike extremely to 9=like extremely).

The adequacy of four of the attributes was measured with “just-about right” (JAR) scales. These scales usually have five points to assess whether there is too little, too much or a “just-about-right” level of an attribute (Lawless & Heymann, 1998). The end-points are anchored with labels that represent levels of the attribute that deviate from a respondent’s theoretical ideal point in opposite directions, while the central point is the ideal (Rothman, 2007). In this study, the adequacy of the ‘hardness’, ‘dry mouthfeel’, ‘fat mouthfeel’ and ‘pastiness’ levels of each sample were scored on bipolar JAR scales (from 1=much too little to 5=much too much, with 3=just about right).

Crispness intensity was measured on a 9-point intensity scale rather than a JAR scale, as crispness is an example of a well-liked attribute that “you can never have too much of”, so in this case JAR scales do not help to understand the adequacy or attribute level, as it may never be possible to reach the theoretical “ideal level” (Rothman, 2007).

2.3. Instrumental texture analysis

The texture of the biscuits was measured instrumentally using a TA.TX.plus Texture Analyzer (S Table Micro Systems, Godalming, UK). Penetration tests (12 mm diameter cylindrical probe P/0.5), were conducted with whole biscuits, setting a distance of 10mm, a test speed of 1mm/s and a trigger force of 0.19N.

Twelve biscuits corresponding to two batches of each formulation prepared on different days were measured.

The maximum force (N) as a measure of hardness, the number of force peaks (with a threshold of 0.01N) as an index of crispness/grittiness and the gradient of the initial steep slope of the curve (N/sec) as a measure of biscuit deformability were measured.

In order to take a picture of the samples after breaking, two biscuits from each formulation were placed in Petri dishes before the penetration test. The dishes with the broken biscuits were then placed in a flatbed scanner (HP 4300c, Hewlett Packard, USA) and the images, with a resolution of 199 pixels per inch, were saved.

2.4. Data analysis

Analysis of variance (two-way ANOVA with fat, fibre and their interaction as factors) was applied to the consumer liking scores, instrumental texture analysis results and TDS mean maximum intensity scores to study the differences between the formulations. The least significant differences were calculated by Tukey's test ($p < 0.05$). These analyses were performed using XLSTAT 2009.4.03 statistical software (Microsoft, Mountain View, CA).

The JAR results were analysed by penalty analysis (PA), using XLSTAT software, to identify potential directions for product improvement on the basis of consumer acceptance by highlighting the most penalizing attributes in liking terms. The respondent percentages (x-axis) were plotted against the penalties (y-axis), and an attribute was considered significant when the respondent percentage was higher than 20% (Xiong & Meullenet, 2006) and the penalty score (drop in overall liking) was higher than 1. Penalty analysis was used in order to gain an understanding of the attributes that most affected liking ratings (Plaehn & Horne, 2008). This technique is used to relate JAR scales to liking data, particularly in order to understand which side of the JAR scale is linked to lower hedonic ratings. The usefulness of the method is that it provides guidance for product reformulation or a better understanding of attribute adequacy in relation to liking in terms of direction, with the assumption that the maximum hedonic score will occur at the “just about right” point (Rothman, 2007).

Principal Component Analyses (PCA) of the instrumental analysis scores and mean maximum intensity scored in the TDS tests were plotted using XLSTAT software.

The results of attribute dominances over time were considered along with instrumental texture parameters and other sensory measurements that helped to understand the biscuits' eating quality.

3. Results and Discussion

3.1. Texture changes during oral processing of the biscuits

When the TDS curves rise from between the chance and significance levels to above the latter, they are considered consistent at panel level. In the present study, in the TDS curves (Fig. 3) the x-axis corresponds to the standardized time (%) and the y-axis to the dominance rate (%).

A general overview of the TDS curves showed that hardness is the first sensation perceived for all the biscuits at the beginning of the mastication period. Subsequently, depending on the level of fat, the biscuits were perceived as being crisp (those with the full level of fat: samples C, LW and HW) or crunchy (those with the low fat level: samples LF, LWLF and HWLF). At the end of the mastication period, dry mouthfeel appeared as the dominant sensation for the samples with added fibre. This sensation was more accentuated for the low-fat biscuits. Fat mouthfeel was detected at a late stage with samples C, LW and LF.

3.1.1. Hardness

This was the first dominant sensation perceived for all the biscuits at the beginning of the mastication period (Figure 3).

The sensory perception of hardness was based on the force required to break the biscuits with the incisors, in accordance with Lillford (2011), who states that the first fracture in a biscuit occurs between the teeth.

In agreement with the sensory perception of hardness, the instrumental measurement of hardness (maximum force during penetration) showed significant differences ($p\text{-value}<0.005$) with different fat and fibre contents (Table 3). The samples with a low fat content (LF, LWLF, and HWLF) were significantly ($p<0,05$) harder than the full-fat biscuits and hardness was increased by the addition of fibre (Figure 4) as the interaction in table 3 shows. The first explanation for these two facts is that when the fat content is reduced the flour particles become more hydrated, making their components more accessible to water, so the gluten also becomes more hydrated and a tougher dough is obtained, resulting in harder biscuits (Ghotra, Dyal, & Narine, 2002). The second is that fibre consists of single polysaccharide chains that can interact with each other to form junction zones which can enclose large amounts of strongly bound and fairly static water, making the junction zones denser and less flexible than the single chain zones (Chaplin, 2003).

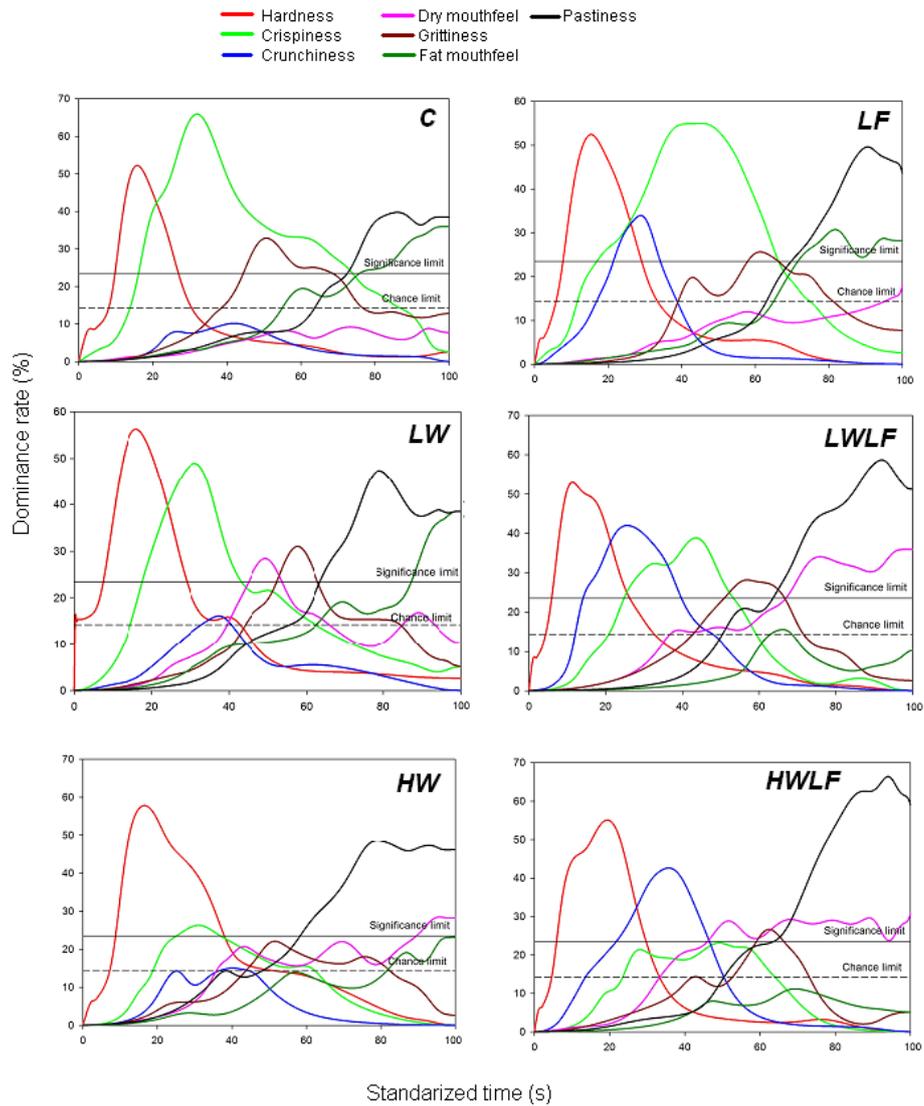


Figure 3. Normalized TDS curves by sample

This phenomenon effectively increases the fibre diameter in the junction zones, reduces the pore size, increases the capillarity and reduces the tendency for the water to exit the cavities, reinforcing the biscuit matrix. As a result, biscuits with

a high fibre content will be harder and more compact (Laguna, Sanz, Sahi, & Fiszman, 2012).

Table 3. Texture parameters obtained with cylinder penetration for control and reformulated biscuits.

Biscuit formulation	Max. Force (N)	Number of peaks	Curve initial slope (N/sec)		
C	56.18 ^a (9.94)	116.96 ^c (19.64)	26.94 ^a (11.30)		
LF	135.34 ^c (21.92)	100.00 ^{bc} (23.29)	63.59 ^b (15.75)		
LW	76.41 ^b (10.78)	86.49 ^{ab} (15.57)	26.32 ^a (7.74)		
LWLF	172.59 ^d (24.41)	83.96 ^{ab} (22.70)	72.01 ^b (12.42)		
HW	86.62 ^b (11.65)	79.00 ^a (22.89)	30.67 ^a (5.10)		
HWLF	228.76 ^e (44.45)	70.14 ^a (18.36)	83.00 ^c (16.07)		
Anova of Instrumental parameters	Fat	F	633.10	8.56	435.9
		p	< 0.0001	0.004	< 0.0001
	Fibre	F	79.54	33.53	10.65
		p	< 0.0001	< 0.0001	< 0.0001

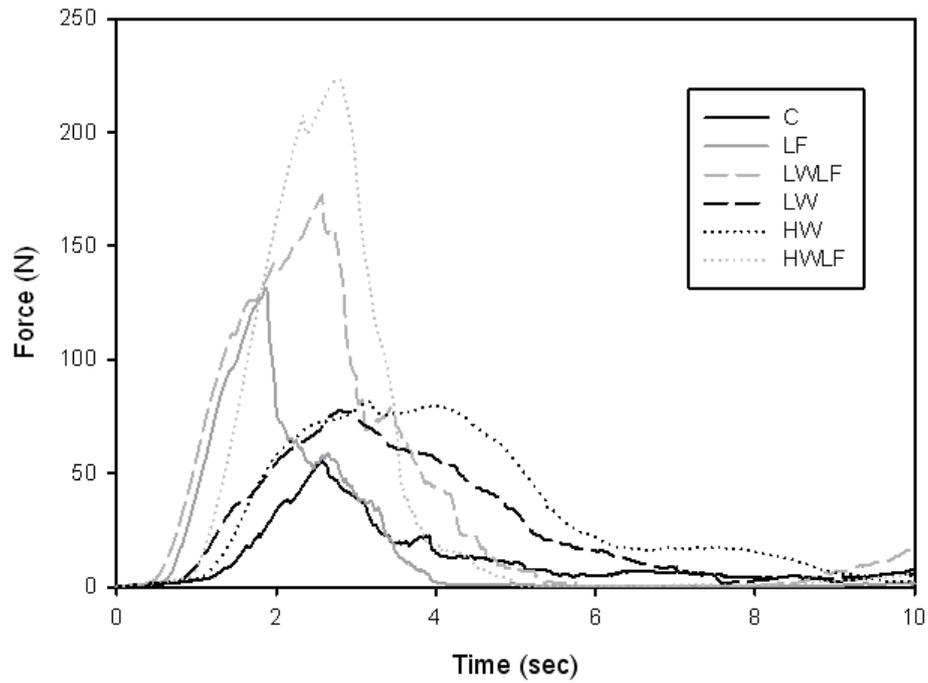


Figure 4. Force vs. time instrumental penetration curves. Effect of fat content and different percentages of flour replacement by fibre on cylinder penetrometer resistance

The mean TDS maximum intensity scores and their correlation to the textural parameters was studied through PCA (Figure 5). The first two components explained 98.5% of the total information. The mean maximum intensity of hardness in the TDS tests correlated well with the initial slope and maximum force of the texture parameters and was related with samples with added fibre and a low fat content (LWLF and HWLF).

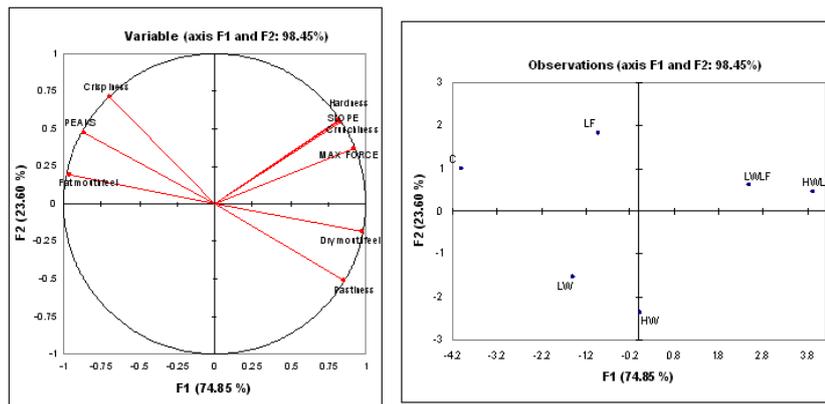


Figure 5. Principal Component Analysis of TDS maximum intensity and instrumental parameters. Only TDS attributes with significant differences (hardness, crispiness, crunchiness, dry mouthfeel, fat mouthfeel and pastiness) were used in the analysis. (a) variables plot, (b) product plot.

3.1.2. Crispness and crunchiness

In the TDS curves, the crispness and crunchiness attributes appeared at different moments in the oral process, depending on the fat and fibre level of the sample. Both attributes have been unequivocally related with fracture properties in previous works (Vincent, 1998). Crispness was dominant for full-fat biscuits and crunchiness for low-fat, high-fibre ones, while intermediate formulations presented both attributes at different times.

Crispness. This was the second dominant sensation for full-fat biscuits (C, LW and HW). Within the full-fat sample group (C, LW and HW), the sensation of crispness differed over time (Figure 3). For biscuit C, crispness was dominant between 16% and 74% of the chewing time. The sensation appeared to last time as the fibre level increased. In the biscuits with the higher fibre content (HW), the dominant sensation only appeared for between 25% and 37% of the chewing period.

Observing the instrumental texture profiles, and also when visually observing the breakage pattern (Figure 6), the full-fat samples were more fragile than the low fat biscuits, probably because the fat interrupted the gluten network,

creating smaller pieces. This type of breakage pattern is usually perceived sensorially as crispness and is typically accompanied by high-pitched sounds (Vickers, 1984). As can be observed from the ANOVA values (Table 3), the breaking slopes of these three samples (C, LW and HW) did not differ significantly but the number of peaks decreased with fibre addition. In a full-fat biscuit, therefore, fibre can be said to have more effect on the number of peaks than on resistance at break (the initial slope of the curve).

Crunchiness. In samples with a low fat content and the addition of two levels of fibre (LWLF and HWLF), crunchiness was the second dominant attribute (Figure 3). Both biscuits had a more compact structure than the full-fat samples and broke into larger pieces in the penetration tests (Figure 5). This kind of breakage pattern is commonly associated with the sensory perception of crunchiness and is usually accompanied by a low-pitched sound (Vickers, 1984).

As the instrumental texture analysis showed (table 3) that the initial slope was higher (see also Figure 4) for the low fat biscuits (LF, LWLF, HWLF) than for the full-fat samples (C, LW, HW). This slope pattern is associated with hardness or rigidity. Also, the curve of the low fat biscuits fell rapidly after the first rupture, signalling a clear cut, whereas the curve of the full-fat biscuits declined gradually. Consequently, fat was the main factor that significantly influenced the break (Table 3).

The biscuits LWLF and LF were the only samples in which both crunchy and crisp attributes were perceived as dominant. The two attributes were superimposed during part of the mastication period, indicating that some panellists found crispness dominant and others crunchiness, or that the same panellist may have selected both attributes, one after the other, during mastication. Samples LWLF and LF were precisely the ones with an intermediate texture profile: they formed small (crisp) and large (crunchy) crumbs, but could also be understood as breaking more cleanly but later forming crisper crumbs.

The samples without added fibre were related to crispness (C and LF) whilst the samples with added fibre (both levels, low and high) were related more to crunchiness and to the instrumental parameter of maximum force (Figure 4).

In Table 4, the mean maximum intensity perceived showed that crispness diminished with fibre addition and was more influenced by fibre, as the ANOVA F statistic showed (F (fibre) on crispness= 55.865; F(fat) on crispness=0.272), whereas crunchiness was more influenced by the fat content (F (fat) on crunchiness= 193.15; F (fibre) on crunchiness=3.54).

3.1.3. *Dry mouthfeel, pastiness and fat mouthfeel*

These attributes were predominant during the next phase of mastication.

Dry mouthfeel and grittiness. As it can be observed in figure 3, these two attributes were the predominant sensations during chewing. Saliva enters the mouth and helps the jaw to form the bolus. The samples with a high fibre content have a higher saliva demand because the fibre can absorb more water than flour, so a dry mouthfeel was created. The samples without added fibre in the formulation (C and LF) elicited a predominantly gritty sensation, more so and for longer in sample C, but no dry mouthfeel was perceived. The rest of the samples showed intermediate predominance (in some cases not significant) of both attributes. In the samples with the lower level of fat (LWLF and HWLF) the dry mouthfeel predominated for much longer, reaching the end of the chew in both cases.

Dry mouthfeel intensity was scored higher by the panellists when the fat content was reduced and fibre was added. For this reason, dry mouthfeel was situated in the opposite quadrant (figure 5) to the C and LF samples in the PCA and described samples LWLF and HWLF better than the other attributes.

Grittiness did not present significant differences in TDS intensity.

Table 4. Mean maximum intensity obtained by TDS panellists

	Hardness	Crispiness	Crunchiness	Dry mouthfeel	Grittiness	Fat mouthfeel	Pastiness
C	2.65 ^a (2.27)	7.19 ^b (1.81)	0.77 ^a (1.44)	1.67 ^a (2.41)	3.24 ^a (3.27)	4.41 ^c (3.06)	2.89 ^a (3.23)
LF	6.35 ^b (2.95)	6.69 ^b (2.69)	4.35 ^b (3.89)	2.89 ^a (2.97)	3.40 ^a (3.67)	3.36 ^{bc} (3.12)	3.62 ^a (3.07)
LW	2.29 ^a (1.67)	3.63 ^a (2.82)	0.68 ^a (1.69)	3.19 ^{ab} (3.44)	3.37 ^a (3.52)	3.10 ^{bc} (3.02)	4.45 ^{ab} (3.31)
HW	2.71 ^a (1.90)	2.19 ^a (2.29)	0.83 ^a (1.59)	4.95 ^{bc} (3.46)	3.57 ^a (3.39)	2.17 ^{ab} (2.69)	6.41 ^{bc} (2.96)
LWLF	7.28 ^{bc} (2.56)	3.75 ^a (3.01)	6.61 ^c (3.01)	5.44 ^{cd} (3.15)	3.37 ^a (3.35)	1.32 ^a (2.11)	5.94 ^{bc} (2.80)
HWLF	8.33 ^c (1.31)	3.11 ^a (3.04)	6.06 ^{bc} (3.48)	7.05 ^d (2.33)	3.91 ^a (3.79)	1.23 ^a (2.12)	6.56 ^c (2.68)

Values in parentheses are standard deviations.

Means in the same column with the same letter do not differ significantly ($p>0.05$) according to Tukey's test.

Pastiness. The intensity of pastiness followed the same trend as dry mouthfeel: low intensity in biscuits C<LF<LW, followed by HW, LWLF and HWLF (Table 4), and appeared as the dominant attribute at the end of the mastication period.

The control biscuit (C) was the sample in which pastiness appeared latest (after 70% of the chewing period) and had the lowest dominance rate (fewer panellists selected it). In the other samples, pastiness appeared earlier and more panellists selected it as dominant, indicating that other proportions of fibre and fat produced a denser matrix and a bolus that was more difficult to insalivate.

Fat mouthfeel. Both C and LF (the samples with no added fibre) presented a dominance of this attribute at the end of mastication. It was noticeable that the perception of a fat mouthfeel did not depend on the fat level, so it could be hypothesised that a “cleaner” perception of fat appeared in the absence of added fibre, which captures the saliva to form a bolus suitable for swallowing. In fact, the only sample with additional fibre that presented any dominance of fat

mouthfeel was LW, which contained the full level of fat and the lower level of added fibre.

High fat mouthfeel intensity scores (figure 5) were either influenced by fat or by fibre addition (F (fibre) on fat mouthfeel = 13.77; F F(fat) on fat mouthfeel =12.45), which is why this attribute appears close to C, LF and LW and at a distance from LWLF and HWLF in the PCA.

With regard to the time of mastication needed for each formulation, theoretically a high value of hardness, as in samples LF, LWLF and HWLF, should increase the number and length of chewing cycles during a mastication sequence (Foster, Woda, & Peymon, 2006; Hiimae & Palmer, 1999), while biscuits with a high fat content, which adds extra lubrication, should require fewer chewing cycles (Foster, *et al.*, 2011). However, no significant differences in mastication time were found between the biscuits under study. The explanation could be that the presence of fat in the biscuits that were not hard (C, LW, HW) interrupts the biscuit matrix, creating a higher number of particles with a greater chance of being retained in non-chewed compartments (Flynn, *et al.*, 2011). Biscuits with both high fibre and low fat (HWLF) formed fewer particles than C when broken (Figure 6).

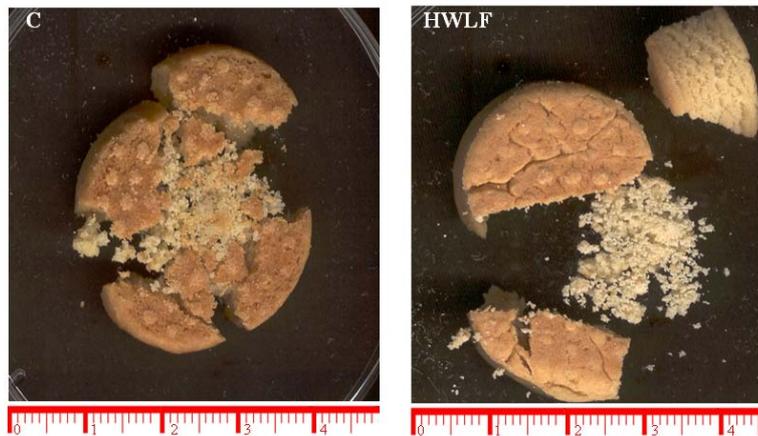


Figure 6. Examples of different break patterns in short dough biscuits. C(full-fat with no added fibre), HWLF (low-fat high-fibre biscuits)

Also, a fat biscuit melts in the mouth (Pareyt & Delcour, 2008), covering it and elongating the “not-clean” mouthfeel, and the panellists had been instructed not press the “stop” button until they felt that their mouth was completely clean. In short, the expected result for the low fat formulations (LF, LWLF and HWLF) was a higher number of chewing cycles, which proved to be the case, but in the high-fat formulations the fat helped to create more biscuit particles and mouth covering effects that compensated for the chewing time.

3.2. Consumer tests

3.2.1. Liking and intensity tests

Consumers scored their overall, texture and flavour liking and their perception of crispness intensity. The reason for choosing the attribute of crispness instead of crunchiness was based on previous studies (Varela, Salvador, Gámbaro, & Fiszman, 2008) that demonstrated how Spanish consumers know and understand the concept of crisp (in Spanish “crujiente”), but rarely use the term crunchy (in Spanish “crocante”) and could misunderstand it.

Figure 7 shows the mean consumer scores. As expected, there was a general trend of higher overall acceptance scores for full fat than for low fat biscuits. Additionally, for the same fat level, the addition of fibre decreased the consumers’ liking scores. The ANOVA results showed that the effects of fibre and fat were significant for all the attributes (Table 5). Fat mainly had a greater impact on texture liking whilst fibre has a greater impact on flavour liking and crispness intensity. The fibre-fat interaction was only significant for flavour liking. This result suggested that some masking phenomenon might occur between fat and fibre on the biscuits’ flavour perception. Fiber is a water (saliva)-demanding ingredient, so flavour perception during mastication in higher-fibre formulations may be different from the lower-fibre ones

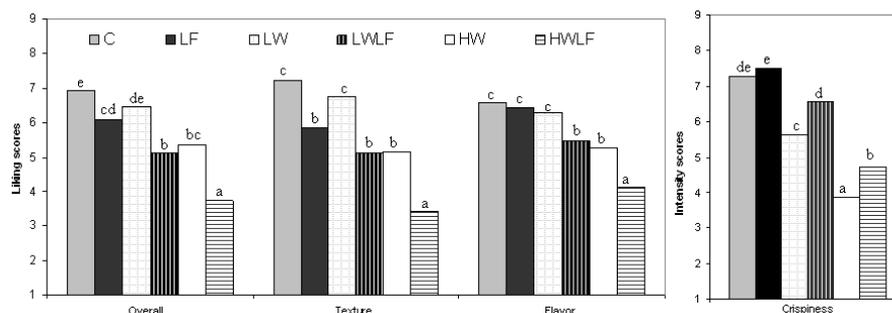


Figure 7. Consumer tests of biscuits prepared with different levels of fat and fibre. (a) Overall, texture and flavour liking (b) crispness intensity

Table 5. ANOVA of attribute ratings (100 consumers)

Consumer's attribute	Fat		Fiber		Fibre*fat	
	F-ratio	p-value	F-ratio	p-value	F-ratio	p-value
Overall liking	65.85	< 0.0001	54.50	< 0.0001	2.06	0.128
Texture liking	106.94	< 0.0001	78.79	< 0.0001	0.53	0.587
Flavour liking	22.23	< 0.0001	49.14	< 0.0001	3.72	0.025
Crispy intensity	15.76	< 0.0001	110.69	< 0.0001	1.88	0.162

Significant p-values (5% level) are highlighted in bold.

3.2.2. Attribute adequacy and its relation to liking – Penalty analysis

Penalty analysis (attributes 'hardness', 'dry mouthfeel', 'fat mouthfeel' and 'pastiness') was used to gain an understanding of the attributes that most affected liking ratings (Plaehn & Horne, 2008).

Figure 8 displays the significant penalties (drops in overall liking) by proportion of consumers. The number of consumers used as the cut-off point was 20% and the mean drop considered was 1 liking point.

A biscuit with many attributes in the upper right-hand corner of the penalty plot is considered worse than one with a few attributes in the lower left-hand corner.

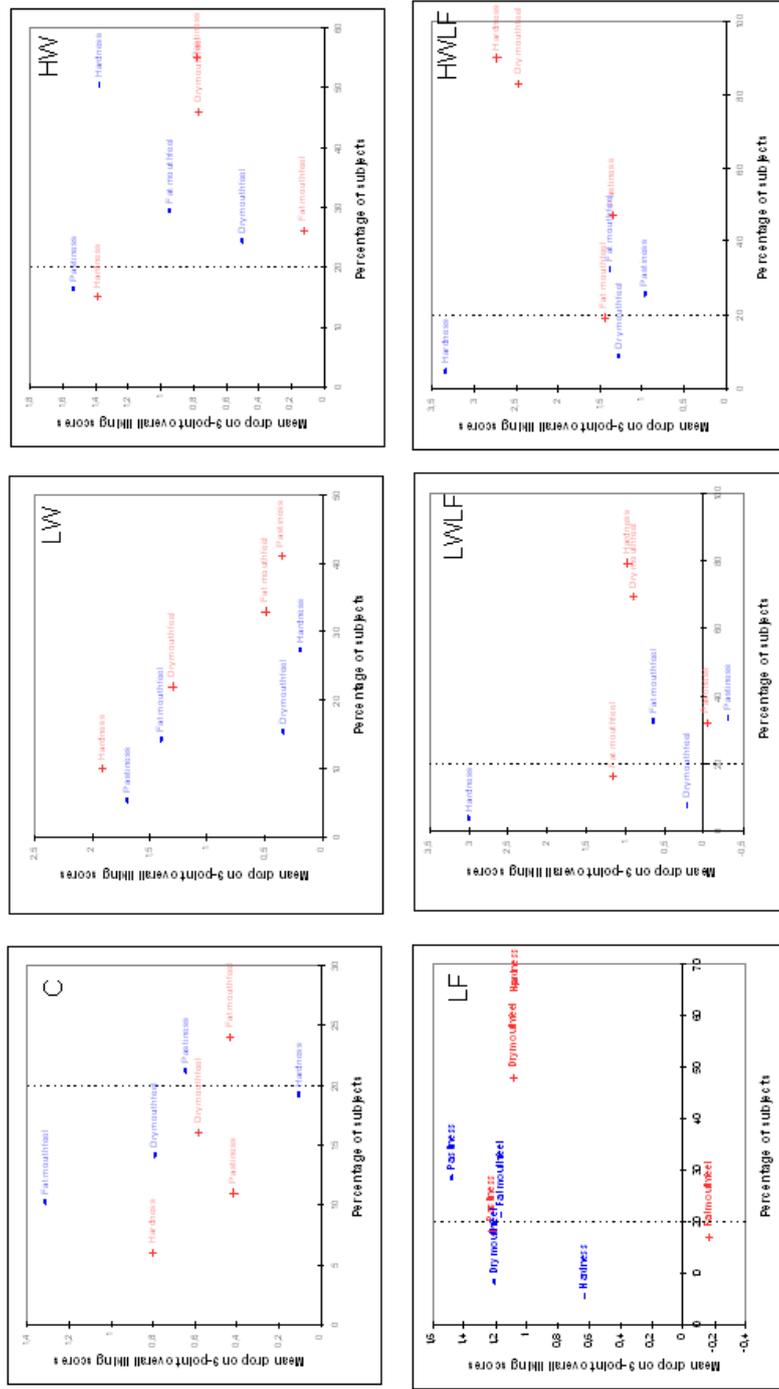


Figure 8. Penalty analysis. Representation of significant penalties (drops in liking) by proportion of panelists. The cut-off point was 20% of consumers stating that an attribute was "not enough" (-) or "too much" (+). It is only above this point (>20% of consumers) that is important to take the deviation into account

The “desired response” is attributes located in the lower left area of the penalty plot, meaning that only a few consumers say the attribute level is not right and the impact on overall liking is small. The C and LW biscuits were in this favourable situation, with low percentages of consumers finding attributes that deviated from their ideal and therefore with low penalties. The opposite position, the upper right-hand corner, contains the attributes that a new reformulation would aim to change: those which are responsible for the largest drops in liking and which high percentages of consumers rate as “not right”.

In general, the most penalising attributes that deviated from the ideal “right point” were “too much” hardness and “too much” dry mouthfeel, found for all the low-fat samples and especially for the high fibre and low fat sample (HWLF). Over 80% of consumers thought that the attributes of HWLF were “not right” and it was penalized with a drop in liking of more than 2.5 points. Another attribute related to the low acceptability of this sample was pastiness, with almost 50% of consumers finding it “too much” and a penalty of 1.5 points. Fat mouthfeel was a penalising attribute for 30% of consumers, with an associated drop in overall liking of around 1.5.

HWLF was the least-appreciated biscuit for texture, flavour and overall liking, and the consumers considered it had quite low crispness. The consumers' adequacy ratings were in good agreement with the findings of the TDS analysis, where dry mouthfeel was dominant at the mid-mastication stage and pastiness appeared as highly dominant at the end of the oral process before swallowing (65-100%). Also, crispness and fat mouthfeel did not appear as significant attributes of this sample at any point of the oral process, meaning that this product lacks those typical sensations of this class of biscuits.

Interestingly, the HW biscuits were penalized with a drop of 1.4 points in their liking; according to penalty analysis hardness was the only attribute that would be responsible for this liking drop. However, they were harder than C via instrumental analysis, and no difference in intensity of hardness was registered by the TDS (trained panel); so there is probably another attribute (not

measured) that may have driven the consumers' perception. In addition, assessment of attribute intensity by consumers is subjected to some limitations since they were not "calibrated" as trained panellists did. When looking at the TDS profile, it is clear that HW was the least complex sample, with no dominance of crispness or crunchiness and with pastiness driving most of the in-mouth perception from the early mastication stage. Its instrumental texture was also characterized by a low number of force peaks, signalling a lack of fracture events related to crispness. The consumers probably perceived it as not being hard enough because hardness is often linked to crispness in their minds, and a sample that collapsed in the mouth immediately with no perception of crispness, leading to a dryish paste, made them wish for a harder and hopefully crisper biscuit.

This is the first study of dynamic method for biscuits and reveals that with the use of TDS is easy to highlights differences in mouth processing among samples. However, more study is needed in this area in order to correlate better which time-attributes may be critical for the consumers' acceptance. In addition, further studies should cover other food matrices.

As limitations, in the present study it has been noticed that panellists complained about the difficulty to keep in mind all the attributes simultaneously during an evaluation, as also reported by other authors (Pineau et al. 2009). The panellists found easy to click the dominant sensation of each attribute but less easy to think in its intensity.

Additionally, each panellist had different paths of mastication (force, number of bites, mouth size or time needed to swallow), so is important to standardize all the experimental conditions as good as possible.

4. Conclusions

The present study proposes new tools for a better understanding of biscuit eating quality. As food ingestion is a dynamic action, this paper has contributed

a dynamic approach by obtaining TDS data and trying to link them to consumers' liking scores.

The fat and fibre levels modulated the dominance of the sensations experienced during the biscuit oral assessment; knowledge of the occurrence and intensity of these characteristics would be a valuable tool to know biscuit eating quality.

The adequacy of some sensory attributes assessed by consumers through JAR scales and penalty analysis, together with the dynamic sensory profiles measured by a trained panel, provided insights which could give clear pointers for biscuit reformulation. Studies correlating perceived sensations and consumer acceptance would be of high interest.

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CAPÍTULO 2

BALANCING TEXTURE AND OTHER SENSORY FEATURES IN REDUCED
FAT SHORT DOUGH BISCUITS

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Abstract

Shortening in short dough biscuits was replaced by a new ingredient, N-Dulge, (a mixture of tapioca dextrin and tapioca starch) at two different levels (10%, 20%). The texture was balanced by replacing part of the flour with resistant starch (RS) (20%, 40%). Nine different formulations were tested by a trained panel and by consumers. The results were also correlated with textural characteristics. The trained panel results showed that the fat replacer increased the hardness and crumbliness and that these effects were balanced out by the addition of resistant starch. These results correlated well with the instrumental texture parameters. The consumers did not find significant differences in acceptability between the control biscuits (no replacement) and the biscuits with 10% fat replaced by N-Dulge and 20% of flour replaced by resistant starch.

Keywords: biscuit, descriptive analysis, fat replacement, resistant starch.

1. Introduction

Reducing fat in the everyday diet has become a public health issue and a concern for most consumers (Zoulias *et al.* 2002a). In the USA and Europe, fat consumption constitutes about 40% of the total daily calorie intake, whereas health specialists recommend that it should not exceed 30% (Zoulias *et al.* 2002b). The reason for this recommendation is that high fat intake is associated with various health disorders such as obesity, cancer, high blood cholesterol and coronary heart disease (Akoh 1998). The WHO (2004) has suggested that the food industry should reduce the fat content of processed foods to decrease the high obesity rate in the first world.

Fat is an essential ingredient in short dough biscuits and is the largest component after flour (Manohar *et al.* 1999). The major functions of fats in this baked system are imparting shortening, richness, and tenderness and improving flavour and mouthfeel (Pareyt *et al.* 2008). To reduce the fat content and still obtain acceptable biscuits, fat replacers can be used. Fat replacers are substances of carbohydrate or protein origin which can be used in some foods to imitate the functional and sensory properties of fat with considerably fewer calories. Carbohydrate-based fat replacers like processed starches imitate fat by binding water and providing lubricity, body and a pleasant mouth sensation in biscuits and similar baked products (Bath *et al.* 1992; Nonaka 1997). Replacement of 50% of fat by soluble β -glucan and amyloextrins derived from oat flour produced cookies that were not significantly different from full-fat ones. At higher substitution levels, moistness and overall quality decreased (Inglett *et al.* 1994). The tenderness of biscuits decreased with increased fat substitution by replacers based on pectins (a blend of gums) or oats. Fat substitute use produced a significant increase in moisture content. Variations in volume and crumb firmness were associated with the type of fat substitute and level of fat replaced (Conforti *et al.* 1996).

A blend of polydextrose, monoglycerides, diglycerides and acid esters is another fat mimetic that has been tested in oatmeal biscuits (Campbell *et al.*

1994). Up to 100% of the fat was replaced and a sensory panel rated five attributes (first bite, first chew, chew down, chewed down mass and residual). The panel found that the 100% blend affected the first chew and residual characteristics of the biscuit but that lower substitution percentages did not have a significant effect on any of the five sensory categories.

Zoulias *et al.* (2002a) studied the textural properties of biscuits using five types of fat mimetics (Litesse, an improved maltodextrose; C*deLight, maltodextrin with a low dextrose equivalent; Dairytrim, a rich oat β -glucan product; Raftiline, an inulin product; and SimpleseDry, a blend of microparticulated whey proteins). C*delight, Raftiline and Simplese gave low-fat cookies a more tender texture, measured through instrumental analysis, but no sensory evaluation was carried out in this study.

Sudha *et al.* (2007) reduced fat from 20% (control) to 10%, 8% and 6% in soft dough biscuits, replacing it with polydextrose or maltodextrin. The biscuits' hardness increased with the replacement and the sensory parameters of texture, taste and flavour were affected. These results show that besides being responsible for texture, fat also imparts flavour, taste, mouthfeel and lubricity to the biscuits.

Reducing the fat content of biscuit recipes affects the overall product quality less than a comparable reduction of sugar, allowing for equal or greater calorie savings, as studied by Drewnowski *et al.* (1998) in a number of types of biscuit.

A new fat replacer, N-Dulge, was launched in 2009. N-Dulge is the commercial name of a mixture of tapioca dextrin and starch intended for fat reduction in sweet baked products like cookies. As its components have undergone physical processing but no chemical processing, it meets clean-label requirements. The label declaration is "dextrin starch".

The addition of the new ingredient addition in a food product, requires a sensory evaluation by a trained descriptive panel (Meilgaard *et al.* 1991). The evaluation needs to include the appearance of any new sensory features (Baixauli *et al.*

2008) and to correlate these effects with consumer perceptions. Consumer research in the early stages of new product development makes it possible to go further and deeper into understanding consumer needs, often well beyond what could be understood without them (Van Kleef *et al.* 2005).

A number of consumer studies have been carried out on added-ingredient biscuits. For instance, good results have been obtained with extruded orange pulp (Larrea *et al.*, 2005), resistant starch-rich lintnerized banana starch (Aparicio-Sanquilán *et al.* 2007) and King palm (Vieira *et al.* 2008), but fewer studies have used trained descriptive panels (Brown *et al.* 1998; Brown and Braxton 2000; Martínez *et al.* 2002; Vázquez *et al.* 2009; and Burseg *et al.* 2009). The same applies to fat replacement studies. Zoulias *et al.* (2002b) studied the replacement of fat using a trained sensory panel and found that the hardness and brittleness estimated by the panel were in good agreement with measurements derived from a texturometer. Moreover, a previous study replacing flour with apricot kernel flour obtained similar properties to those of the control biscuits with up to 40% fat replacement (Seker *et al.* 2010). However, once the performance of a new ingredient has been described, normally no effort is made to solve or compensate for the appearance of an unbalanced sensory profile.

The two main objectives of the present study were 1) to assess the effect of different levels of shortening replacement by a dextrin-rich ingredient on the sensory profile of short dough biscuits and 2) to counterbalance any sensory changes produced by the fat replacement and obtain similar sensory properties to those of the full fat biscuits. They were addressed in two stages.

2. Materials and methods

2.1. Ingredients

The biscuit formulations employed in the two stages of this study (fat replacement and counterbalancing sensory features) are shown in Table 1. The

percentages of shortening replacement (10% and 20%) were selected after a preliminary study which showed that 25% or more of shortening replacement made the dough unmanageable.

The ingredients were: a) soft wheat flour suitable for biscuits (Belenguer, S.A., Valencia, Spain) (composition data provided by the supplier: 15% moisture, 11% protein, 0.6% ash; alveograph parameters $P/L=0.27$, where P =maximum pressure required and L =extensibility; and $W=134$, where W =baking strength of the dough), b) ND (N-Dulge FR, National Starch Food Innovation, Manchester, UK, a mixture of tapioca dextrin and tapioca starch), c) RSRI, a source of resistant starch (Hi-maize 260, National Starch Food Innovation, Manchester, UK, composition data provided by the supplier 10% moisture, 58% dietary fibre), d) shortening (78% fat, St. Auvent, Vandemoortele France), e) sugar (Azucarera Ebro, Madrid, Spain), f) milk powder (Central Lechera Asturiana, Peñasanta, Spain), g) salt, h) sodium bicarbonate (A. Martínez, Cheste, Spain), i) ammonium hydrogen carbonate (Panreac Quimica, Barcelona, Spain) and j) tap water.

Fat replacement with a dextrin-rich ingredient: two formulations were prepared using the same quantity of all the ingredients except for 10% and 20% of the shortening, which was replaced with ND (samples 10ND and 20ND, Table 1).

Counterbalancing atypical sensory features: High levels of fat in biscuits produce a very soft texture (Pareyt *et al.* 2008) and low fat levels make the texture harder. In order to decrease this atypical effect, a resistant starch rich ingredient was added, as a previous study (Laguna *et al.* 2010) showed that biscuits with flour replacement by resistant starch exhibit a softer, crumblier texture. As shown in Table 1, four formulations were prepared with 10% and 20% of the shortening replaced by ND and 20% and 40% of the flour by a resistant starch rich ingredient (RSRI) (samples 10ND-20RSRI, 10ND-40RSRI, 20ND-20RSRI and 20ND-40RSRI). The control with no substitutions and two additional formulations with only the two levels of flour replacement by RSRI

Table 1. Biscuit formulations prepared with different levels of shortening replacement with n-dulce (nd) and with both fat replacement with nd and flour replacement with a resistant starch rich ingredient (RSRI).

Ingredient (g)	Control	10ND	20ND	10ND20RSRI	10ND40RSRI	20ND20RSRI	20ND40RSRI	20RSRI	40RSRI
Flour	100	100	100	80	60	80	60	80	60
Shortening	60	54	48	54	54	48	48	60	60
Sugar	30	30	30	30	30	30	30	30	30
Milk	0.58	0.58	0.58	0.58	0.58	0.58	0.58	0.58	0.58
Salt	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Sodium bicarbonate	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Ammonium bicarbonate	0.115	0.115	0.115	0.115	0.115	0.115	0.115	0.115	0.115
Water	9	9	9	9	9	9	9	9	9
ND	0	6	12	6	6	12	12	0	0
RSRI	0	0	0	20	40	20	40	20	40

(20% and 40%: samples 20RSRI and 40RSRI respectively) were also prepared for comparison purposes.

2.2. Biscuit preparation

The sugar, milk powder (previously dissolved in all of the water), leavening agents and shortening were mixed in a mixer (Kenwood Major Classic, UK) for 30 seconds at low speed (#1). The dough was scraped from the sides of the bowl and mixed for 3 minutes at a higher speed (#3). The flour or the flour/ND, flour/RSRI or flour/ND/RSRI mixture was then added and mixed for 1 minute at speed #1.

The dough was sheeted with an 8 mm high rolling pin and allowed to rest for 30 minutes at 4°C before cutting it into rectangular pieces measuring 50 x 30 x 8mm (length x width x height). Twenty-five pieces were placed on a perforated tray. The biscuits were baked in a conventional oven for 4 min at 175 °C then the trays were turned 180° and baked for a further 4.5 min at the same temperature. Turning the trays brought the side that had been at the back to the front of the oven to ensure homogenous cooking. The oven and the oven trays were always the same, the trays were placed at the same level in the oven and the number of biscuits baked was always the same. After reaching ambient temperature, the biscuits were packed and stored in heat-sealed metalized polypropylene bags. The biscuit samples were evaluated on the following day in all cases.

2.3. Sensory Analysis

Testing was carried out in a sensory laboratory equipped with individual booths (ISO 8589, 1988). Data acquisition was performed using Compusense five release 5.0 software (Compusense Inc., Guelph, Ont., 158 Canada).

2.3.1 Descriptive analysis

Selection of terms - A panel of eight assessors, aged between 25 and 36 years and skilled in quantitative descriptive analysis, was trained to select the descriptors using the checklist method (Lawless *et al.*, 1998).

Terms were selected and discussed in an open session with the panel leader. The assessors were first given a brief outline of the procedures and a list of attributes and representative samples. They were then asked to choose and write down the most appropriate attributes to describe all the sensory properties of the biscuits or to suggest new ones. The panel leader collected and wrote all the attributes on a board and the panel discussed the appropriateness of the selected attributes, their definitions and preliminary discussions on how to assess the products. At the end of this session a consensus on the list of attributes was reached (Table 2). This procedure was proposed by Stone and Sidel (2004) in order to obtain a complete description of a product's sensory properties.

Panel training.-The panellists attended twelve 1-hour training sessions. Training involved two stages. During the first stage, different samples were tasted by the panellists to attain a better understanding and final agreement of how to measure all the descriptors and different tastings were conducted until the panel was homogeneous in its assessments. At these sessions, external references were provided as an aid to identifying some attributes.

For example, for floury odor a Petri dish with flour was available and for buttery odor a mixture made of shortening and milled biscuit was provided. In this way, the panellists understood the extremes of the scales.

During the second stage, the panellists used 10 cm unstructured scales to score the selected attributes of the biscuits. Assessors were instructed to score the odors first, followed by color, shape regularity, hardness (hand), and brittleness (hand). They then had to take a bite from the edge of the biscuit in order to score the flavour, crunchiness and pasty mouthfeel. Panel performance and consensus were evaluated by principal component analysis, using the Pearson correlation matrix (Cliff and King, 1999).

Table 2. Attributes, scale extremes and definitions used in the descriptive sensory analysis of the biscuits by a trained panel.

Attribute	Scale extremes	Definitions
Color	Light/Dark	Evaluation of the overall color of the biscuit
Shape regularity	Weak/Strong	Evaluation of uniformity or irregular edges and cracks in the surface of the biscuit
Floury odor Buttery odor	Weak/Strong	Evaluation of odors by smelling the biscuit once
Hardness	Weak/Strong	Evaluation of biscuit hardness when held at both ends and broken in two
Crumbliness	Weak/Strong	Evaluation of the biscuit crumbs formed during the hardness test
Floury taste Buttery taste	Weak/Strong	Evaluation of flavours perceived while chewing the biscuits
Crunchiness	Weak/Strong	Evaluation of noise when the biscuits are bitten into by the incisors.
Pastiness	Weak/Strong	Evaluation of the degree of perception of pastiness in the mouth in relation with salivation and difficulty in swallowing

Formal assessment. A balanced complete block experimental design was carried out in duplicate (two sessions) to evaluate the samples. The intensities of the sensory attributes were scored on a 10 cm unstructured line scale. Nine samples were evaluated per session. In each session, the samples were randomly selected from each cooking batch and served in random order, each on a separate plastic tray identified with a random three-digit code. The panellists were instructed to rinse their mouths with sparkling water between

sample evaluations, as sparkling water was found to remove shortening from the mouth between samples more efficiently than still water.

2.3.2. *Consumer test*

A total of 100 frequent short dough biscuits' consumers (untrained) aged from 18 to 65 years took part in the study. To avoid fatigue, the consumers evaluated 7 samples over three sessions (three biscuits at each session). The biscuits were coded with random three-digit numbers following a balanced complete block experimental design. Consumer acceptance testing was carried out using a nine point hedonic scale (9 = like extremely; and 1 = dislike extremely). The consumers scored their liking for the 'appearance', 'hardness', 'color', 'sweetness', 'taste', and 'overall acceptance' of each sample.

2.4. *Biscuit texture analysis*

The texture of the biscuits was measured using a TA.TX.plus Texture Analyzer (Stable Micro Systems, Godalming, UK). A test speed of 1mm/s was used for all the tests. Ten biscuits from each formulation were measured.

Breaking strength. The biscuits were fractured using the three point bending rig probe (A/3PB). The experimental conditions were as follows: distance between supports: 40 mm, probe travel distance: 20 mm, trigger force: 20g. The force at break (N) was taken as the breaking strength.

Bite test. Penetration tests were conducted with the upper Volodkevich Bite Jaw (VB), penetrating the sample (whole biscuit) to a distance of 5mm, again with a trigger force of 20g. Two 'bites' were made in each biscuit (one third from each end), so a total of 20 values was registered for each formulation. The parameters measured were the area under the curve (N.sec), taken as the resistance to penetration, and the number of peaks, taken as an index of crunchiness.

2.5. Color

Measurement of the upper surface color of the biscuits was carried out with a Konica Minolta CM-35000d spectrophotometer. Four replicates of each formulation were measured. The results were expressed in accordance with the CIELAB system with reference to illuminant D65 and a visual angle of 10°. The parameters determined were L* (L* = 0 [black], L* = 100 [white]), a* (-a* = greenness, +a* = redness), b* (-b* = blueness, +b* = yellowness).

The total color difference (ΔE^*) between the control biscuit and the different ND, ND/RSRI biscuit was calculated as follows:

$$\Delta E^* = [(L^*_c - L^*_s)^2 + (a^*_c - a^*_s)^2 + (b^*_c - b^*_s)^2]^{1/2}$$

where subscript c = control and subscript s = samples containing the ND or ND/RSRI.

The value used to determine whether the total color difference was visually obvious was (Francis & Clydesdale, 1975):

$$\Delta E^* > 3: \text{color differences are obvious to the human eye.}$$

2.6. Statistical analysis

Two-way ANOVA was applied to each descriptor to check panel performance, considering the assessors, the samples and their interaction as factors.

Analysis of variance (one way-ANOVA) was applied to the consumer test in order to study the effect of formulation; least significant differences were calculated by Tukey's test and the significance at $p < 0.05$ was determined.

Pearson correlation was calculated for the instrumental analysis, descriptive analysis and consumer test data.

All the statistical analyses were performed with XLSTAT 2009.4.03 statistical software (Microsoft).

3. Results and discussion

3.1. Sensory Analysis

3.1.1. Quantitative descriptive analysis

Step 1, N-Dulge substitution

Compared with the control sample, replacing 10% or 20% of the shortening with ND (samples 10ND and 20ND, respectively) affected the textural attributes most. A significant increase ($p < 0.05$) in hardness and crunchiness scores and a significant decrease ($p < 0.05$) in crumbliness were found (Fig. 1a). These results were in accordance with previous works which have reported that a reduction in fat levels leads to harder biscuits (Lai and Lin, 2006). Brown *et al.* (2000), studying rich tea biscuits, stated that a biscuit must be somewhat hard in order to be perceived as crunchy, but soft enough to obtain the expected crumbly texture. These authors also found an inverse relation between hardness and crumbliness, while crunchiness appeared as an independent characteristic related to hardness and crumbliness.

Pastiness was not significantly affected by replacing shortening with ND. The perception of pastiness arises when the crumbs generated by fragmentation of the biscuits begin to absorb saliva and form a paste which is difficult to handle in the mouth. The pastiness values of N-Dulge biscuits were low and did not present significant differences compared with the control samples ($p < 0.05$).

The texture of biscuits can be interpreted in terms of the state of their main ingredients. The process of baking short dough leads to a cellular solid matrix with a porous inner structure. Kulp *et al.* (1991) determined that due to the limited water content, the starch granules remained in practically their native condition during biscuit baking rather than forming a continuous structure, thus providing a crispy/crunchy texture. When fat is mixed with the flour before hydration it prevents the gluten from forming a network. A gluten network is undesirable in biscuit dough as it leads to toughness (Sudha *et al.* 2007). According to Baltsavias *et al.* (1999), fat can form a more continuous phase when the system is rich in fat, or can be dispersed when present at lower levels.

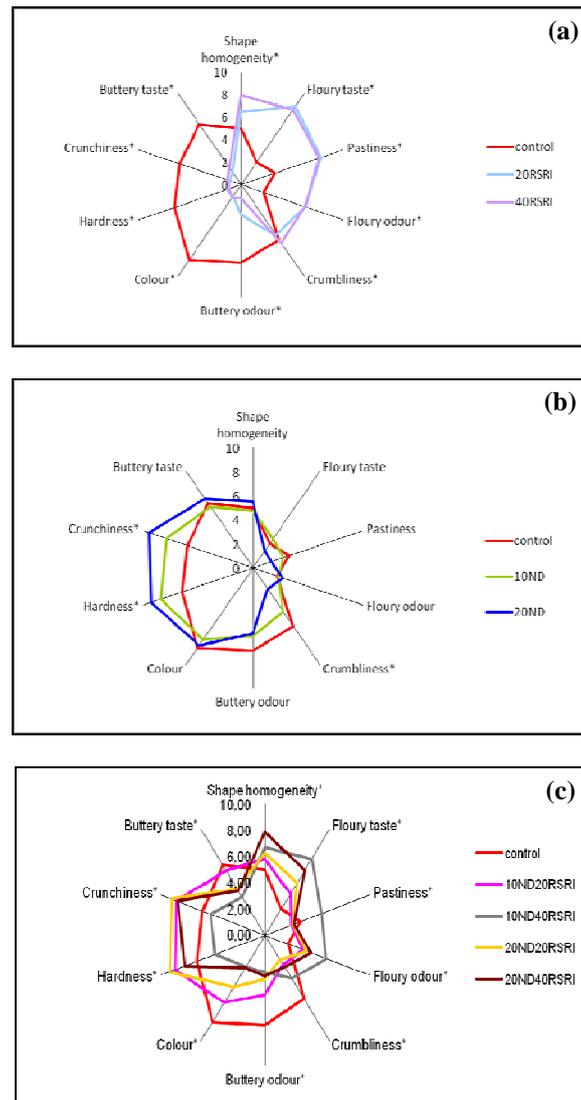


Fig 1. Average (n=2) descriptive sensory scores. Control formulation compared with (a) biscuits with 10 and 20% N-Dulge (ND) shortening replacement, (b) biscuits with 20 and 40% flour replacement with resistant starch-rich ingredient (RSRI) and (c) biscuits with 10 and 20% ND shortening replacement along with 20 and 40% RSRI flour replacement (*)Attributes with significant differences.

When a sufficient amount of fat is present, it can surround and isolate the proteins and the starch granules, thereby breaking the continuity of the protein and starch structure and giving more tender biscuits (Ghotra *et al.* 2002). In the present work, it has been shown that the fat was increasingly replaced by ND, that continuous fat phase begun to be dispersed, affecting then the final texture by the decrement of biscuit tenderness.

Fat replacement with ND did not significantly affect the odor or flavour attributes. This means that up to 20% shortening replacement by ND was not detectable in terms of odor and taste. Drewnowski *et al.* (1998) reported that fat was often difficult to detect in foods since no single attribute can be unambiguously associated with fat content. Thus, in taste terms, ND could act as good fat replacer in this matrix at up to 20% substitution.

Biscuit color was not affected by the ND substitution, as the color scores for 10ND and 20ND biscuits did not present significant differences compared with the control biscuits. The golden-light brown color of biscuits is mainly caused by an interaction between reducing sugars and amino acids (Maillard-type reaction) which forms brown polymers or melanoidins. N-Dulge is composed of starch and dextrins, which would not promote additional browning as they do not contribute to the Maillard reaction.

Step 2, balancing textural parameters

In order to counterbalance the textural changes observed with N-Dulge substitution (hardness, crunchiness and crumbliness), part of the flour was replaced by a resistant starch-rich ingredient (RSRI). Laguna *et al.* (2010) found that unlike fat replacement, replacement of wheat flour by an RSRI reduced biscuit hardness and increased crumbliness. These authors studied the instrumental texture of biscuits with different levels of flour replaced with resistant starch and obtained products with crumblier, more tender textures. In this regard, an RSRI could be considered a good additional ingredient to restore the tenderness associated with a full-fat biscuit recipe.

Resistant starch is also a source of fibre with interesting health benefits (Yue and Waring, 1998). Resistant starch is white, is a natural source of dietary fibre and has a bland flavour, giving a better appearance, texture and mouthfeel than other typical fibres (Baixauli *et al.*, 2008).

To present the effects of RSRI on the biscuits' sensory profile, biscuits were formulated with 20% and 40% of the flour replaced by RSRI. Figure 1b clearly shows biscuit's hardness and crunchiness decreased significantly and crumbliness increased. Other sensory features introduced by RSRI will be commented on below.

With these results in mind, biscuits were formulated with 10% and 20% of the shortening replaced by ND and 20% and 40% of the flour replaced by RSRI (Table 1). In this way, reduced-fat fibre-enriched short dough biscuits were obtained. The replacement percentages were selected on the basis of the above-mentioned study (Laguna *et al.* 2010).

The significant effects of RSRI addition on the sensory profile of the ND biscuits are shown in Figure 1c. To a large extent, the addition of the RSRI balanced out the effects on texture associated with substituting ND for the shortening, bringing the textural sensory characters closer to those of the control. The effect was more significant in the biscuits with a higher percentage of the RSRI (40% wheat flour replacement) and lower ND substitution (10% shortening replacement). The RSRI significantly lowered the hardness and crunchiness scores and decreased the crumbliness. These textural improvements were accompanied by other sensory changes: a significant increase in pasty mouthfeel compared to the control sample, together with an increase in floury flavour and odor and a decrease in buttery flavour and odor (Fig. 1c). A decrease in buttery flavor and odor was also found in the RSRI-only samples (Fig. 1b), despite having the same shortening content as the control sample. This could be explained by this starch's ability to bind some aroma compounds (Van Ruth *et al.* 2003; Seuvre *et al.* 2006). On the other hand, the more floury flavor found in the RSRI-substitution samples could mask other flavors such as

the buttery one. In the ND-only substitution biscuits, even at the lowest final shortening content (20ND), the perception of buttery flavor was not significantly different from that of the control biscuit, meaning that the flavor was still perceptible at the same levels. Thus, it could be concluded that the changes in flavor perception were due to the RSRI.

The ND/RSRI-substitution biscuits had significantly lighter color (Fig. 1c), lacking brown color when compared with the control. RSRI is whiter than wheat flour, which could have helped make the dough lighter from the start. In addition, the total amount of proteins available for Maillard browning is lower in RSRI than in the same amount of flour, so less browning during baking is to be expected (Laguna *et al.* 2011). Also, ND/RSRI-substitution biscuits had a significantly more regular shape; the addition of RSRI could reduce the elasticity and deformability of the dough; also, the gluten content is lower when less wheat flour is present, which would explain why the shape of these biscuits was significantly more regular than in the control sample.

3.1.2. Consumer test

The mean scores for liking of appearance, color, texture, taste, sweetness and overall liking are shown in Figure 2.

For the seven formulations evaluated, all the mean scores were between 3.5 and 6.8. The control biscuit obtained the maximum scores for appearance and liking of color. However, sample 10ND20RSRI scored highest for overall liking, texture and liking of sweetness (although not significantly different from the control scores), meaning that this reformulated biscuit with less fat and therefore fewer calories (fat provides 9 kcal/g and carbohydrates only 4 kcal/g) was the most acceptable.

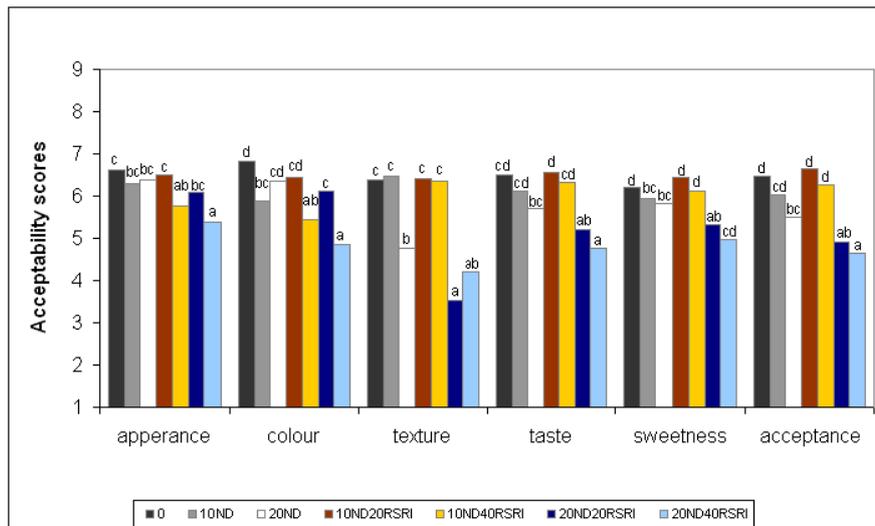


Figure 2. Mean consumer acceptance scores for control and reformulated Biscuits. Same letter indicates no significant differences between means

The samples with the lowest scores for texture liking were 20ND20RSRI and 20ND40RSRI. Texture is a critical attribute both in consumer preference and to match consumer expectations with respect to a particular biscuit type or brand (Brown et al. (1998). So, these samples also obtained the lowest scores for overall acceptance.

Seker et al. (2010) studied the sensory effect of apricot kernel flour (40.2% fat and 21.8% protein) as a fat replacer in cookies (control formulation contained only 40 g fat/100 g flour), obtaining liking scores that did not significantly differ from the control. Zoulias et al. (2002b) reported that the sensory limit for fat replacement in a cookie formulation with 40 g of fat per 100 g flour was 50% because of the decrease in overall quality. This statement is in agreement with Drewnowski et al. (1998), who reported that the acceptability ratings of sensory panels were relatively unaffected by a 25% reduction of fat in a number of biscuit formulation. In the present study, the liking scores of the 20% replacement of shortening by N-Dulge in the consumer test were lower, the worst-affected attribute being texture; these samples received

the highest hardness scores for the trained panel (see the Quantitative Descriptive Analysis section). Zoulias *et al.* (2002a) also noted that a high hardness value is an unpleasant characteristic for this category of product.

The biscuits with the highest replacement of fat (20%) by ND and highest replacement of flour (40%) by RSRI obtained the lowest scores. Laguna *et al.* (2011) reported that biscuits with 60% of flour replacement by RSRI were too soft and were rated lower than the control biscuit by the consumers. Brown and Braxton (2000) obtained a similar response for soft biscuits; their preference mapping showed a consistent dislike of the samples of rich tea-type biscuits that had been softened by leaving the biscuits in a moist atmosphere for 1 h prior to testing.

3.2. Biscuit texture analysis

The instrumental texture analysis results for the biscuits are shown in Table 3. The biscuit with 20% shortening replacement by ND showed the highest breaking strength and bite test penetration resistance values. Zoulias *et al.* (2002a,b) also found that the hardness of low-fat biscuits increased with the use of fat mimetics (polydextrose, maltodextrine with low dextrose, oat product rich in β -glucans, oligofructose and a blend of microparticulated whey proteins and emulsifiers). Similarly, Seker *et al.* (2010) studied fat replacement by apricot kernel flour in a wire-cut cookie and found that the force necessary to break the biscuit increased with fat replacement.

The increase in hardness due to fat replacement is associated with gluten development. Ghotra *et al.* (2002) noted that in the absence of shortening, the water interacts with the flour protein to create more cohesive and extensible gluten, while if the fat level is high, the fat surrounds the proteins, shortening their development. Pareyt *et al.* (2008) reported that in this type of biscuit little if any gluten is formed, giving a very soft texture. In the present study, the higher the fat level the softer the biscuits (lower bite test area and breaking strength values).

Table 3. Physical properties of the biscuits

Sample	Width (cm)	Length (cm)	Stack height (cm)	Breaking strength (N)	Bite test	
					Area (N.sec)	Number of peaks
Control	3.7 ^a (0.3)	5.4 ^a (0.1)	1.1 ^a (0.1)	7.3 ^b (0.6)	60 ^c (6)	22.0 ^c (4.0)
10ND	3.8 ^a (0.2)	5.9 ^d (0.1)	1.3 ^{ab} (0.3)	19.4 ^d (1.2)	111 ^e (18)	31.1 ^d (4.8)
20ND	3.9 ^b (0.1)	5.6 ^{ab} (0.1)	1.2 ^{ab} (0.1)	42.6 ^e (2.9)	255 ^g (18)	24.8 ^c (3.7)
20RSRI	3.6 ^a (0.5)	5.8 ^{cd} (0.1)	1.8 ^b (0.1)	4.1 ^a (0.4)	30 ^b (2)	12.8 ^b (3.7)
40RSRI	3.57 ^a (0.04)	5.6 ^{bc} (0.1)	1.3 ^{ab} (0.1)	2.7 ^a (0.2)	17 ^a (2)	5.4 ^a (3.0)
10ND- 20RSRI	3.77 ^a (0.03)	5.8 ^{cd} (0.1)	1.3 ^{ab} (0.1)	11.8 ^c (1.2)	71 ^c (7)	23.3 ^c (3.2)
10ND- 40RSRI	3.6 ^c (0.1)	5.6 ^{ab} (0.1)	1.3 ^{ab} (0.1)	9.9 ^c (0.8)	60 ^c (6)	12.3 ^b (2.7)
20ND- 20RSRI	3.7 ^a (0.1)	5.5 ^{ab} (0.1)	1.3 ^b (0.1)	23.4 ^e (1.7)	124 ^f (11)	25.5 ^c (4.2)
20ND- 40RSRI	3.5 ^a (0.1)	5.7 ^{bcd} (0.1)	1.3 ^{ab} (0.1)	20.3 ^d (1.9)	91 ^d (8)	23.2 ^b (3.8)

Values with the same letter differ significantly and/or do not differ significantly

the instrumental texture parameters measured (number of peaks, area of curve and maximum peaks).

Negative correlations were found between instrumentally measured biscuit width and shape regularity in the sensory test, indicating that a slightly barrel-shaped biscuit was detected as a lack of regularity. This same attribute correlated positively with consumer liking of the appearance, meaning that wider biscuits obtained higher consumer acceptance, which could be explained as a preference for a more homemade appearance rather than geometrically perfect biscuits.

Instrumental color ΔE^* correlated negatively with color consumer liking. Less acceptance was expressed when the biscuit differed from the control color, mainly due to L^* values.

Since peaks in biting test represent fracture events, the expected clear association between the number of peaks and the perception of crunchiness was found. It was also found that sensory hardness correlated significantly and positively with resistance to biting (obtained from the number of peaks and area under the curve) and breaking strength (the maximum peak with the 3-point bending rig), indicating that the texture scores of the trained panel correlated well with instrumental texture parameters. Color as measured by the trained panel correlated positively with the number of peaks, which could be explained by the fact that the crunchier biscuits were also the darkest (more dehydrated) ones.

Table 4. Pearson correlation coefficients between instrumental results and sensory descriptive (qda) and consumer scores.

Variable	INSTRUMENTAL						QDA						CONSUMER					
	Width	ΔF^{**}	Number of peak (Ebs test)	Area of curve (lbs test)	Maximum Peak (2-year)	Shape like grassy	Days like grassy	Hardness	Color	Crymelness	Overall acceptance	Appearance	Color	Taste	Texture	Softness		
INSTRUMENTAL																		
Width	1	-0.149	0.405	0.707	0.405	-0.834	0.514	0.804	0.494	0.145	0.802	0.449	0.314	0.379	0.341			
Length	0.074	1	0.084	-0.159	-0.117	0.002	-0.113	-0.245	-0.139	0.131	-0.055	0.077	0.071	0.274	0.340			
ΔF^{**}	-0.149	1	-0.174	-0.138	-0.041	0.513	0.052	-0.391	0.175	-0.295	-0.597	-0.652	-0.338	-0.431	-0.371			
Number of peak (Ebs test)	0.405	-0.174	1	0.414	0.439	-0.449	0.908	0.777	0.862	-0.350	0.474	0.379	-0.234	-0.245	-0.307			
Area of curve (lbs test)	0.707	-0.138	0.414	1	0.945	-0.359	0.715	0.412	0.732	-0.477	0.251	-0.013	-0.053	-0.257	-0.250			
Maximum Peak (2-year)	0.405	-0.041	0.439	0.945	1	-0.359	0.746	0.335	0.774	-0.410	0.154	-0.127	-0.250	-0.359	-0.421			
QDA																		
Days like grassy	-0.834	0.513	-0.449	-0.359	-0.359	1	-0.500	-0.544	-0.402	-0.351	-0.904	-0.659	-0.447	-0.479	-0.397			
Hardness	0.514	0.052	0.908	0.715	0.746	-0.500	1	0.773	0.991	-0.434	0.349	0.145	-0.358	-0.359	-0.404			
Color	0.804	-0.391	0.777	0.412	0.335	-0.544	0.773	1	0.715	0.075	0.743	0.401	0.245	0.135	0.053			
Crymelness	0.494	0.175	0.862	0.732	0.774	-0.402	0.991	0.715	1	-0.501	0.244	0.042	-0.335	-0.440	-0.445			
CONSUMER																		
Overall acceptance	0.145	-0.295	-0.350	-0.477	-0.410	-0.351	-0.434	0.075	-0.301	1	0.505	0.591	0.656	0.931	0.912			
Appearance	0.802	-0.597	0.474	0.331	0.154	-0.904	0.349	0.743	0.244	0.505	1	0.899	0.497	0.531	0.453			
Color	0.449	-0.652	0.379	-0.013	-0.127	-0.659	0.145	0.401	0.042	0.591	0.899	1	0.434	0.420	0.513			
Taste	0.344	-0.338	-0.234	-0.053	-0.230	-0.447	-0.358	0.245	-0.325	0.656	0.497	0.634	1	0.890	0.846			
Texture	0.379	-0.421	-0.245	-0.257	-0.399	-0.479	-0.359	0.135	-0.440	0.931	0.531	0.420	0.890	1	0.957			
Softness	0.341	-0.371	-0.307	-0.290	-0.421	-0.397	-0.404	0.053	-0.445	0.912	0.453	0.513	0.846	0.957	1			

Values in bold are significant.

QDA, quantitative descriptive analysis

Lastly but no less importantly, overall liking was highly correlated to the consumers' texture, taste and appearance liking scores. This suggests that these three parameters were important drivers of liking for the consumers evaluating the products. In particular, texture and taste liking showed a great drop in all biscuit formulations with 20%ND when compared to the control sample. The reformulated biscuit with 10% shortening replaced by N-Dulge along with 20% of flour replaced by resistant starch overcame this drop, showing acceptability values that were not significantly different to those of the control sample (Fig. 2).

4. Conclusion

This study shows how sensory analysis can be used successfully to ascertain how to balance the effects of new ingredients. Texture was the main issue when a percentage of fat was removed: the biscuits got harder and crumblier.

In the present study, the addition of resistant starch to replace part of the flour proved to be a good way of balancing out the detrimental effects of fat replacement on texture. In the case of 10% shortening replacement by N-Dulge (a mixture of tapioca dextrin and tapioca starch), this addition achieved samples with good acceptability.

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**HPMC AND INULIN AS FAT REPLACERS IN BISCUITS: SENSORY AND
INSTRUMENTAL EVALUATION**

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and Teresa Sanz

Abstract

Because a high fat content in foods is associated with health disorders, consumers demand low fat products, but without any loss of their texture quality. The instrumental texture and sensory properties of biscuits in which 15% and 30% of the fat had been replaced by two different carbohydrate-based fat replacers (inulin and hydroxypropyl methylcellulose) were studied. The instrumental texture measurements showed that inulin and hydroxypropyl methylcellulose (HPMC) biscuits were harder and the sound emissions were higher than for the control biscuits. The trained sensory panel rated the biscuit with 15% fat replacement by inulin as crisper than the control. The consumer study revealed that fat replacement up to 15% with inulin or HPMC provided acceptable biscuits, but higher replacement decreased the overall acceptability.

Keywords: Biscuits, fat replacers, inulin, HPMC, texture.

1. Introduction

In biscuits, fat is a very important ingredient, contributing to the texture, mouthfeel, flavour and overall perception of them (Zoulias, Oreopoulou, & Tzia, 2002a). However, an excess of energy intake and the consequent high amount of fat (especially saturated fat) is associated with health disorders such as obesity, cancer, high blood cholesterol and coronary heart disease (Akoh, 1998). In fact, the total fat content should not be higher than 30% of daily energy intake. Saturated fats should not be more than 10% and monounsaturated and polyunsaturated 20% of total energy intake (USDA 2000).

For this reason concerned consumers want to reduce the fat content of their food, without affecting flavour and texture (Nonaka, 1997). In fact, mechanical properties of biscuits are largely dependent on the fat component of the formulation (Baltsavias, Jurgens, & van Vilet, 1999) because fat interacts with the other ingredients to develop and mould texture, mouthfeel and overall sensation of lubricity of the product (Giese, 1996; Stauffer, 1998). Fat replacement in biscuits has been studied by many different authors (Röbke, Ktenioudaki, & Gallagher, 2011; Zbikowska & Rutkowska, 2008; Zoulias et al. 2002a; Zoulias, Oreopoulou, & Kounalaki, 2002b; Sudha, Srivastava, Vetrmani, & Leelavathi 2007; Laguna, Varela, Salvador, Sanz, & Fiszman, 2011). Nevertheless, the problem associated to the loss of texture quality remains unsolved. Nowadays, the consumer appreciation is the criteria for the food choice and a loss in desirable quality parameters is a problem (Roudaut, Dacremont, Pamies, Colas, & Meste, 2002). In biscuits, texture plays a decisive role in appreciation. Specifically for some types of biscuits crispness has a main role for biscuit acceptability.

Fat has an important role during the manufacturing process as well as in the final texture of biscuits. During dough mixing, fat competes with the water for the flour surface surrounding the starch and protein (Pareyt & Delcour, 2008), producing tender biscuits (Baltsavias, Jurgens, & van Vliet, 1999). Replacing fat

without affecting product quality characteristics is a challenging task (Röβle et al., 2011). “Fat replacers” are substances of carbohydrate or protein origin which can imitate the functional and sensory properties of fat while “fat substitute” is referred to the substances that provides identical physical and sensory properties to fats, as well as fat replacers, but does not provide calories (Lindsay 2000).

Inulin has been used as a fat replacer in the dairy industry (Güven, Yasar, Karaka, & Hayaloglu, 2005), in spreads and fillings, desserts and dressings (Franck, 2002; Roberfroid, 2005), in bread making (O’Brien, Mueller, Scannell, & Arendt, 2003) and in biscuits (Zoulias et al., 2002ab; Zbikowska et al., 2008). Zoulias et al. (2002a) used compression tests to compare the biscuit with or without fat replacer. An increase in the maximum stress of no fat replacer biscuits was obtained respect to the inulin biscuits; however, they did not compare with full fat biscuits. Additionally, an increase in maximum stress was associated with harder biscuits. Zbikowska et al. (2008) also used inulin as fat replacer. A sensory panel described texture biscuits attributes and found inulin biscuits as less crispy and harder in comparison with full fat biscuits.

Both manuscripts reveal that inulin could be use as fat replacer: however, none of these studies conducted a study of the instrumental and sensory properties to understand the influence of inulin on texture.

Hydroxypropyl methylcellulose (HPMC), a cellulose ether, has also been used as fat replacer in many food applications. It imparts creaminess, lubricity, air entrapment and moisture retention to baked goods, frozen desserts, dry mix sauces, pourable and spoonable sauces, dressings (Akoh, 1996), gluten free breads (Sabanis and Tzia, 2011; Mariotti et al. 2012;) and it has been used as films in processed fruits and vegetables (Krochta and De Mulder-Johnston, 1997; Park 1999; Villalobos 2006). To date, HPMC has not been used as a fat replacer in biscuits.

The aim of this work was to study the effect of partial fat replacement with inulin and HPMC in biscuits. Instrumental texture was studied through simultaneous fracture and sound emission measurements and the sensory properties (appearance, texture and sound) of the biscuits were studied using both a trained panel and a consumer panel.

2. Material and methods

2.1. Materials

Two different fat replacers were used: inulin (Frutafit[®] HD, provided by Sensus Roosendaal, The Netherlands; composition data as provided by the manufacturer: carbohydrates > 99,5%, inulin > 90%, fructose < 10%), and hydroxypropyl methylcellulose (Methocel F4M Food Grade, Dow Chemical Company, Midland, MI, USA; composition data as provided by the manufacturer: methoxyl and hydroxypropyl substitution of methyl cellulose 29% and 6.8% respectively, viscosity 4000 mPa·s in a 20g/L solution). The biscuit formulations are shown in Table 1.

2.2. Biscuit preparation

Biscuit preparation followed the same procedure as Laguna, Salvador, Sanz and Fiszman (2011) with the following modifications: in the biscuits with HPMC, the HPMC was dissolved in water at 2% and then added after the flour and mixed for 30 seconds at speed 60 rpm and the dough was sheeted to 10 mm in a dough laminating machine (Parber, Zamudio, Spain) and cut into rectangular pieces measuring 50 x 30 x 10mm (length x width x height). Twenty-five pieces were placed on a perforated tray. The biscuits were baked in a conventional oven for 4 min at 175 °C. The trays were then turned 180°, bringing the side that had been at the back to the front of the oven to ensure even cooking, and baked for a further 4.5 min at the same temperature. The oven and the oven trays were always the same, the trays were placed at the same level in the oven each time, and the number of biscuits baked was always the same. After

reaching ambient temperature, the biscuits were packed and stored in heat-sealed metalized polypropylene bags.

Table 1. Ingredients of biscuits with varying fat levels(15 and 30 indicate % of fat replacement).

Ingredients (g/100g flour)	Control	Inulin15	Inulin30	HPMC15	HPMC30
Flour	100	100	100	100	100
Fat	32.15	27.33	22.50	27.33	22.50
Fat replacer	0	4.82	9.65	4.82*	9.65*
Sugar	29.45	29.45	29.45	29.45	29.45
Milk	1.75	1.75	1.75	1.75	1.75
Salt	1.05	1.05	1.05	1.05	1.05
Sodium bicarbonate	0.35	0.35	0.35	0.35	0.35
Ammonium bicarbonate	0.2	0.2	0.2	0.2	0.2
Water	11	11	11	11	11

*g of a dispersión of HPMC in water at 20 g/L

2.3. Moisture content and water activity

The moisture content of the biscuits was determined in four replicates of each formulation. The samples were grinded in a mill and transferred (2-3 g) to a weighted moisture dishes and were placed in an oven at 105°C for 24 h. The difference in weight before and after the oven was calculated and the moisture content in a percentage was register. Water activity (a_w) was determined in four milled biscuits replicates of each formulation as the RH of the air in equilibrium with the outer part of the samples in a sealed measuring chamber using a chilled-mirror dew point technique at 22°C (Aqua Lab Series 3, Decagon Devices, Pullman Wash., U.S.A.).

2.4. Simultaneous fracture and sound emission of the biscuits

The acoustic emission of the biscuits was measured during fracturing at 20 mm/s using a Texture Analyzer (TA-XT Plus, Stable Micro Systems Ltd., Surrey, UK) inside an isolated acoustic chamber with a prepolarized free-field ½" microphone in combination with a Deltatron preamplifier microphone (Brüel Kjaer, Nærum, Denmark) as described by Primo-Martín, Sözer, Hamer, & van Vliet (2009). The biscuit was placed on a flat platform with a slit (width 4 mm, length 60 mm). Test pieces were fractured between a wedge (30° cutting angle, 32 mm wide) and the platform. The wedge shaped probe and a deformation speed of 20 mm/s (and a 65 kHz data sampling rate) were chosen to simulate biting with the front teeth (Vincent, Jeronimidis, Khan, & Luyten, 1991). The distance between the microphone and the sample was 7 cm. Extensive analysis of the sound was performed using MatLab (Matworks 7.0.4) as described by Castro-Prada, Primo-Martín, Meinders, Hamer, & Van Vliet (2009). The sound emitted was characterized in terms of the number of sound pulses and mean intensity (dB), both of which are related to the perception of crispness (Castro-Prada et al., 2009). The total energy (W/m^2), and maximum intensity (dB) were also measured. The mechanical parameters measured were the number of force drops (drop in force higher than 0.5N), slope of the first event (gradient of the curve up to the first major peak) (N/s), and maximum force (N).

2.5. Sensory Analysis

Sensory testing was carried out in a sensory laboratory equipped with individual booths (ISO 8589, 1988). Data acquisition was performed using Compusense five release 5.0 software (Compusense Inc., Guelph, Ont., Canada).

The samples analyzed by sensory analysis were: control, inulin15, HPMC15, and HPMC30. The inulin30 biscuits were not subjected to sensory testing as

they were not considered edible due to their hardness on the first bite with the incisors.

2.5.1. Consumer test

A total of 100 consumers (untrained, 50 males and 50 females) recruited in the centre of research (IATA) and the university of Valencia, aged from 18 to 65 years and frequent consumers of short dough biscuits, took part in the study. The consumers evaluated the 4 samples in one session. The biscuits were coded with random three-digit numbers and the sample presentation followed a balanced complete block experimental design. Consumer acceptance was rated on nine-point hedonic scales (1 = dislike extremely and 9 = like extremely). For each sample, the consumers scored the attributes acceptability in the following order 'overall acceptability', 'appearance', 'colour', 'odour', 'texture', 'hardness', 'crispness', 'dry mouthfeel', 'taste', 'sweetness' and 'buttery taste'.

2.5.2. Quantitative Descriptive Analysis

The QDA method (Stone et al., 1974; Stone and Sidel, 1998, 2003) provides a complete word description of a product and it was done as follow.

Selection of terms. A panel of eight assessors, aged between 25 and 40 years and with wide experience in quantitative descriptive analysis, was trained to assess the biscuit samples. The assessors selected the descriptors using the checklist method (Lawless & Heymann, 1998). Terms were selected and discussed in an open session with the panel leader. The panel leader collected all these attributes that the trained

panel have selected and wrote them on a board and the panel discussed the appropriateness of the selected attributes, their definitions and how to assess the products. This procedure was proposed by Stone and Sidel (2004) to obtain a complete description of a product's sensory properties. At the end of this session a consensus on the list of descriptors was reached (Table 2). The selected attributes focus on different aspects of crispness, including sound quality, sound intensity, and sound duration.

For some difficult attributes as the quality of the sound that refers to its pitch the panellists had a piece of nougat ("turrón") as a reference for a low-pitched sound (tough) and a potato crisp for high-pitched sound (brittle).

Panel training. The panellists attended twelve 1-hour training sessions. Training involved two stages: during the first stage, different samples were tasted by the panellists to attain a better understanding and final agreement on how to measure all the descriptors. Different sessions were conducted until the panel was homogeneous in its assessments.

Panel performance was evaluated by principal component analysis, using the Pearson correlation matrix, until the group no longer contained any outliers from one training session to another (King, Hall, & Cliff, 2001).

Formal assessment. A balanced complete block experimental design was carried out in duplicate (two sessions) to evaluate the samples. The intensities of the sensory attributes were scored on 10-cm unstructured line scales. All four samples were evaluated in each session. The samples were randomly selected from each cooking batch and served in random order, each on a separate plastic tray identified with a three-digit

code. The panellists were instructed to rinse their mouths with water between sample evaluations.

2.6. Statistical analysis

Analysis of variance (one way-ANOVA) was applied to study the differences between formulations. Least significant differences were calculated by the Tukey test and the significance at $p < 0.05$ was determined.

Two-way ANOVA was applied to each descriptor to check panel performance, considering the assessors, the samples and their interaction as factors. Analysis of variance (one way-ANOVA) was applied to the consumer test in order to study the effect of formulation. Least significant differences were calculated by Tukey's test and the significance at $p < 0.05$ was determined.

Pearson correlation was calculated to correlate the sensory data with the instrumental parameters.

All the statistical analyses were performed with XLSTAT 2009.4.03 statistical software (Microsoft).

Table 2. Attributes, scale extremes and definitions used in the descriptive sensory analysis of the biscuits by a trained panel.

Parameter	Attribute	Scale extremes	Definitions
Colour	Intensity of yellowness	Low/High	Evaluation of the overall colour of the biscuit
	Intensity of toasted colour		
Odour	Buttery	Weak/Strong	Evaluation of odours by smelling the biscuit once
	Toasted		
Surface appearance	Porosity	Weak/Strong	Evaluation of holes and flatness of biscuit surface
	Convexity		
Manual texture	Hardness	Weak/Strong	Evaluation on grasping both ends and breaking biscuit in two
	Sound intensity at break		
Matrix appearance	Matrix aeration	Weak/Strong	Evaluation of the biscuit matrix on observing the inside of half a biscuit
Flavour	Buttery	Weak/Strong	Evaluation of the flavour perceived while chewing the biscuits
	Toasted		
In-mouth texture	Hardness on first bite	Weak/Strong	Placing the biscuit between the incisors, evaluation of its hardness.
	Sound at break on first bite	Weak/Strong	Evaluation of the noise produced when biting into the biscuits with the
	Quality of sound	High-pitched	
	Hardness during mastication	Weak/Strong	
	Sound duration during mastication	Short/long	Evaluation of the duration of the sound when chewing with the molars, keeping the lips closed
	Moisture	Low/High	Evaluation of the degree of perception of pastiness in the mouth in relation with salivation and difficulty in swallowing

3. Results and discussion

3.1. Moisture content and water activity

It is well known that moisture content, water activity and water distribution have a strong effect on the perception of crispness and on the mechanical signatures of dry, brittle, cellular foods such as biscuits. Food crispness has a sigmoid relationship with water content and activity (Peleg, 1994; Heidenreich, Jaros, Rohm & Ziemsa, 2004).

To compare the crispness behaviour of the different biscuits, the first step was to determine whether the moisture content and water activity could affect the sensory appreciation of crispness. The moisture and water activity values of the biscuits are shown in Table 3. The moisture contents ranged from 2.81 to 4.53 g water/100g product and the a_w values from 0.12 to 0.24. The water content-activity range of the biscuits was found on the plateau where crispness does not depend on them.

Table 3. Moisture content and water activity of the biscuits (15 and 30 indicate % of fat replacement)

Biscuit sample	Moisture (g water/100g product)	a_w
Control	4.53 ^a (0.20)	0.240 ^a (0.003)
Inulin15	3.07 ^c (0.03)	0.160 ^c (0.002)
Inulin30	2.81 ^c (0.10)	0.120 ^d (0.004)
HPMC15	3.89 ^b (0.10)	0.210 ^b (0.002)
HPMC30	4.58 ^a (0.12)	0.250 ^a (0.002)

Values in parentheses are standard deviations. Means (N = 4) in the same column with the same letter do not differ significantly ($p < 0.05$) according to the Tukey test.

a_w : water activity

3.2. *Texture and acoustic properties*

Representative force-time curves and the simultaneously recorded sound obtained during the fracture process are shown in Figure 1.

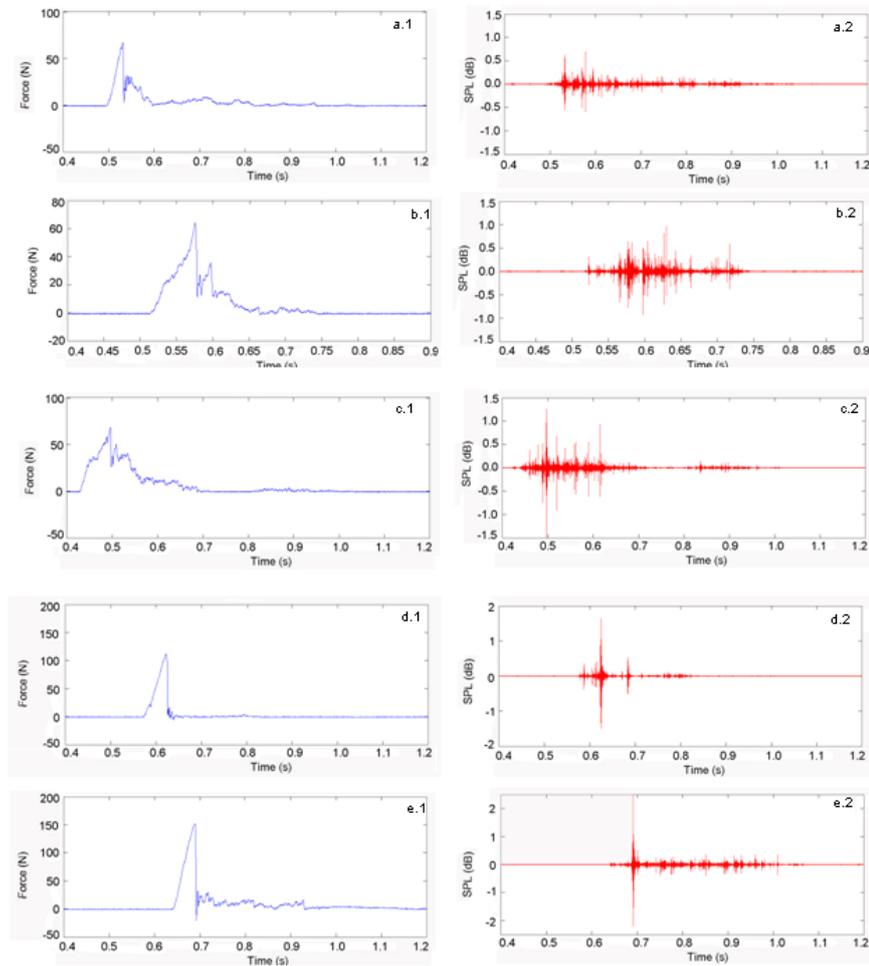


Figure 1. Wedge fracture probes for a) control b) HPMC15, c) HPMC30, d) Inulin15, e) Inulin30; in each case, 1 is the force graph and 2 the sound graph

Representative force-time curves and the simultaneously recorded sound obtained during the fracture process are shown in Figure 1. For all the samples, the biscuit fracture showed one big force event due to a strong top layer (crust)

that was harder to break. This area lost more water during baking because it reached a higher temperature than the interior (Pareyt and Delcour, 2008). Immediately below the crust there was a cellular structure (crumb or biscuit matrix) represented by much smaller force drops.

When the probe started to touch the top surface of the biscuit, the force started to increase and small sound pulses were emitted. Continuous fracture of the crust resulted in high intensity sound pulses. Finally, the sound pulses decreased when the biscuit split into two parts.

Although the size of the peaks was different, the replacement of fat did not noticeably affect the general profile of the force and sound curves, since the fat replacement biscuits were also characterized by the presence of multiple force and sound events.

To quantify the differences in the biscuit profiles objectively, specific parameters were obtained from the sound and force curves. They are presented in Table 4.

Fat replacement produced a significant increase ($p < 0.05$) in the maximum force during fracturing, which correlates with hardness. The magnitude of the force depended on the type of fat replacer and on its concentration; higher fat replacement produced higher maximum force. The increase in maximum force was higher when fat was replaced with inulin. For 15% fat replacement by HPMC no significant differences with the control were found. If the maximum force reached is considered to represent the overall hardness of the biscuit, the inulin-30 biscuits were the hardest.

Baltsavias et al. (1999), also found that reducing the fat content increased the fracture stress of biscuits. Similarly, Ghotra, Dyal, & Narine (2002) and Pareyt & Delcour (2008) stated that the fat surrounds and isolates the gluten and starch, breaking the protein-starch continuity; as a result, higher fat biscuits are shorter, less hard and more inclined to melt in the mouth.

A gradient up to a point where the curve suddenly falls can be seen in all the curves (Figure 1); this behaviour was measured as the slope of the first event

Table 4. Data obtained from texture and acoustic measurements

Biscuit sample	Sound data				Force data			
	Number of sound pulses	Total energy [W/m ²]	Maximum intensity [dB]	Mean intensity [dB]	Maximum Force [N]	Number of force drops	Slope of the first event [N/s]	
Control	391.0 ^a (137.6)	7.8E-07 ^a (3.1E-07)	92.1 ^a (2.5)	64.1 ^a (1.8)	70.9 ^a (10.4)	16.4 ^{ab} (2.6)	2052.3 ^a (998.3)	
Inulin15	455.6 ^{ab} (230.1)	2.99E-06 ^c (7.8E-07)	99.6 ^b (1.9)	70.1 ^c (1.1)	126.6 ^c (19.6)	18.3 ^b (2.4)	2727.8 ^a (1276.1)	
Inulin30	551.3 ^{ab} (250.4)	4.2E-06 ^{bc} (2.9E-06)	100.2 ^b (3.9)	70.7 ^{bc} (3.6)	183.6 ^d (32.5)	16.3 ^{ab} (2.7)	2255.6 ^a (1251.3)	
HPMC15	474.0 ^{ab} (152.3)	1.1E-06 ^a (4.6E-07)	93.2 ^a (1.7)	65.5 ^a (1.9)	80.3 ^{ab} (15.8)	15.3 ^a (1.6)	1706.5 ^a (600.5)	
HPMC30	560.1 ^b (155.4)	2.1E-06 ^b (5.4E-07)	98.1 ^b (3.4)	68.7 ^b (1.2)	92.4 ^b (20.2)	14.8 ^a (3.2)	1958.4 ^a (860.3)	

Values in parentheses are standard deviations. Means (N=10) in the same column with the same letter do not differ significantly ($p < 0.05$) according to the Tukey test.

and is related with fragility. No significant differences between the control and fat substitute biscuits were found in the slope of the first event.

The sound-related parameters revealed significantly ($p < 0.05$) higher total sound energy and mean intensity values, especially in the inulin15 biscuits, in comparison to the control. The maximum intensity values were also higher for inulin15, inulin30 and HPMC30 than for the control. No significant differences in the number of sound pulses were found among the different samples except for HPMC30, which showed a higher number than the control biscuit.

In short, if a crisp product is characterised by a brittle fracture at a low fracture force and distinguishable fracture events with the emission of sound (Luyten, Plijter, & Van Vliet, 2004; van Vliet, Visser, & Luyten, 2007), the fat replacement biscuits cannot be considered crisper: a higher maximum force was required to break the biscuit and their higher sound emission implies that while they are harder, they are not necessarily crisp.

3.3. Sensory analysis

3.3.1. Quantitative descriptive analysis

Although this paper mainly focuses on texture properties, fat replacement also affected appearance, odour and flavour. The scores for the samples evaluated by the trained panel are presented in Table 5.

Crispness is perceived through a combination of the noise produced and the force required to break down a product during eating (Duizer, Campanella, & Barnes, 1998). In this study, each panellist had to split crispness into a number of attributes, assessing the fracture and the sound emitted during the fracture separately. The panellists also differentiated between hardness by hand (snapping the biscuits) and by mouth (with the incisors), providing different hardness and sound intensity scores. The hardness at break was differently perceived by hand and mouth. In-mouth, the hardness at break and the sound intensity increased with fat replacement; whilst these differences were not

appreciable by hand. The in-mouth sensory results were reflected by the instrumental results.

The panellists found that the inulin15 biscuit had the highest-pitched fracture sound, followed by HPMC15 and the control, while the HPMC30 produced the lowest-pitched sound.

During mastication, each biscuit formulation exhibits its own “breakdown path”. The panellists probably characterized these differences as hardness and sound duration during mastication. The control biscuit was the softest. This is in agreement with Baltsavias et al. (1999), who stated that the incorporation of fat into a biscuit dramatically decreased the fracture stress. They also asserted that this decrease is due to air cell size reduction; however, in the present study the panellists scored the full-fat biscuit (control) as having a more aerated matrix. The sound duration during in-mouth mastication was independent of the quantity of fat and the inulin15 biscuit had the longest sound.

As the toasted colour increased, so did the toasted odour. The inulin15 biscuit had the most toasted colour. This could be attributed to a greater presence of reduced sugar (provided by the inulin), which would increase the reducing sugar/aminoacid interaction for the Maillard-type reaction (Chevallier, Colonna, Buleón, & Della Valle, 2000) which forms brown polymers or melanoidins. Additionally, the low moisture content of inulin15 would enhance the Maillard reaction.

The yellowness cited by the trained panel was only significantly different for the biscuit with the highest fat replacement, HPMC30, and may have been due to pigment dilution in the presence of the HPMC solution, although no differences were found between the control, HPMC15 and inulin15.

Table 5. Average (n = 2) descriptive sensory scores for fat replacement. Control formulation and reformulated biscuits.

Biscuit sample	Colour Intensity of yellow-ness	Intensity of toasted colour	Buttery odour	Toasted odour	Crust properties Porosity	Convexity	Crumb properties Matrix aeration	Crispness (manual) Manual texture: hardness	Manual sound: intensity	Flavour Buttery	Toasted Flavour	Hard-ness on first bite	Sound at first bite	Quality of sound (in-mouth)	Hardness during mastication	Sound duration during mastication	Moisture
Control	5.3 ^b (2.6)	2.6 ^a (1.9)	6.7 ^c (2.2)	2.63 ^a (1.6)	3.1 ^a (2.0)	4.4 ^a (2.13)	6.2 ^b (2.2)	4.7 ^a (1.9)	4.6 ^a (2.9)	6.5 ^c (2.6)	2.0 ^a (1.5)	2.9 ^a (2.0)	4.9 ^a (2.1)	4.8 ^a (2.2)	2.2 ^a (1.6)	4.2 ^{ab} (2.3)	5.8 ^{bc} (2.6)
Inulin 15	6.4 ^b (1.3)	7.8 ^b (1.6)	4.5 ^{ab} (3.0)	7.0 ^b (2.2)	3.0 ^a (1.9)	3.0 ^a (1.9)	4.0 ^a (2.2)	3.9 ^a (1.9)	4.9 ^a (3.1)	4.2 ^b (2.8)	6.8 ^b (2.7)	5.2 ^b (2.8)	7.6 ^b (2.1)	7.0 ^b (2.3)	5.6 ^b (2.3)	7.5 ^c (1.9)	2.2 ^a (1.9)
HPM/C15	5.9 ^b (1.2)	6.8 ^b (1.7)	3.0 ^{ab} (1.9)	5.7 ^b (2.4)	5.6 ^b (2.2)	4.5 ^a (2.5)	4.9 ^{ab} (2.1)	6.4 ^b (2.1)	4.5 ^a (2.9)	3.7 ^b (2.0)	5.4 ^b (2.7)	6.6 ^b (1.9)	6.6 ^b (2.0)	5.4 ^{ab} (2.9)	4.3 ^b (1.9)	5.1 ^b (2.4)	4.3 ^b (2.3)
HPM/C30	2.9 ^a (1.6)	2.35 ^a (2.0)	2.2 ^a (1.6)	1.67 ^a (1.3)	8.1 ^c (1.2)	6.7 ^b (1.9)	5.7 ^{ab} (2.5)	6.6 ^b (2.3)	3.8 ^a (2.3)	1.9 ^a (1.5)	1.4 ^a (1.2)	5.6 ^b (2.6)	5.1 ^a (2.9)	3.9 ^a (2.8)	4.0 ^b (2.4)	3.2 ^a (2.1)	6.9 ^c (2.4)

The perception of moisture was significantly different between the samples; the highest was HPMC30 with no significant differences with control biscuit. Lillford (2011) affirmed that the presence of fat in cakes gave a high sensory perception of greater moistness. Consequently, HPMC was able to imitate this moist mouthfeel.

Fat mimetics are less flavourful than the fat they replace because, as they usually absorb water, they are able to carry soluble flavours but not lipid-soluble flavour compounds (Akoh, 1988). These statements are in accordance with the panellists' scores, where the fat replacement was found to decrease the buttery odour and flavour, which is associated to the reduced fat content per se; this difference was greater in the HPMC biscuits.

3.3.2. Consumer test

T3.3.2. Consumer test

The consumers assessed their overall acceptance and their liking for the biscuits' appearance, colour, odour, texture, hardness, crispness, dry mouthfeel, flavour, sweetness and buttery taste. Figure 2 shows the mean scores of the four formulations for each attribute. Control, inulin15 and HPMC15 obtained similar scores for almost all the attributes, while HPMC30 scored lower mean values. Focusing on texture, the inulin15 scores showed a less acceptable texture than the control and the HPMC15 biscuit but achieved good overall acceptance (the highest score, in fact), meaning that although texture is considered a critical attribute (Brown, Langley, & Braxton, 1998), it did not affect the overall acceptance

Dry mouthfeel, sweetness and buttery taste obtained low liking scores in comparison with the other attributes. The dry mouthfeel liking score for inulin15 was low; this dry mouthfeel was also reported by Forker, Zhan, & Rohm (2011) and Conforti, Charles, & Duncan (1997) as a negative perceived attribute. In the present study, high replacement with HPMC was not rated differently from the control biscuit. Forker et al. (2011) stated that the impression of sweetness

increased with fat replacement, but no significant differences between the concentrations or type of fat replacers used (inulin or HPMC) were found in the present study. However, the buttery taste was different for high fat replacement (HPMC30).

On the basis of these results, consumers did not consider control, inulin15 and HPMC15 to differ significantly, whereas HPMC30 was not acceptable.

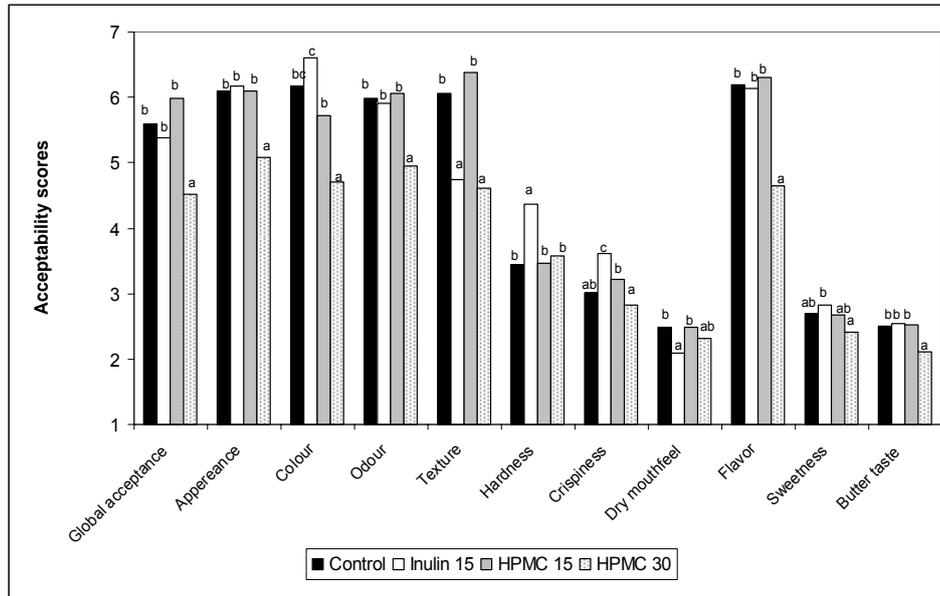


Figure 2. Mean consumer acceptance scores for the control biscuit and biscuits with fat replaced by Inulin and HPMC.

Previous works have also obtained acceptable scores for low fat biscuits. Seker, Ozboy-Ozbas, Ozturk, & Koksel (2010) studied the sensory effect of apricot kernel flour in biscuits to replace up to 40% of fat, obtaining acceptability scores that were not significantly different from the control. Zoulias et al. (2002b) reported a sensory limit for fat replacement in a biscuit formulation of 50% because of the decreasing overall quality. Drewnowski, Nordenstenb, & Dwyer (1998) reported that the acceptability ratings of sensory panels are relatively unaffected by a 25% reduction in fat. These results probably denote that biscuit formulations are very variable (different kinds of fat, quantities of

flour or fat replacers), so each formulation has its own characteristics and consumer expectations.

4. Conclusions

This work is a preliminary study about the suitability of inulin and HPMC as fat replacers in biscuits. The relationship between texture properties perceived by humans and measured instrumentally is evaluated. Higher degrees of fat replacement gave biscuits of unacceptable sensory-texture properties. The level and type of fat replacer influence the texture characteristics of biscuits and their sensory qualities. Inulin and HPMC increase the hardness and maximum sound intensity, however, inulin cannot achieve fat replacement percentages higher than 15%. HPMC can be used at higher fat replacement rates while keeping the dough workable, but concentrations higher than 15% cannot be employed if good sensory properties are to be retained.

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CAPÍTULO 3

**INULIN AND ERYTHRITOL AS SUCROSE REPLACERS IN SHORT DOUGH
COOKIES. SENSORY, FRACTURE AND ACOUSTIC PROPERTIES**

Laura Laguna, Cristina Primo-Martín, Ana Salvador and Teresa Sanz

Abstract

The effect of sucrose replacement by erythritol and inulin was studied in short dough cookies using instrumental and sensory analysis. Two levels of replacement were used (25 and 50% of total sucrose content). Descriptive sensory analysis showed that the sucrose replacement affects visual and texture cookies characteristics, being the differences perceived by mouth greater than by hand. In general, sucrose substitutes produced a less crispy cookie and lower consumer acceptability, with the exception of 25% sucrose replacement by inulin. Matrix aeration attributes such as open and crumbly obtained by trained panel were important properties, and correlated positively with consumer acceptance and negatively with maximum force at break (hardness). Inulin cookies sensory properties were more similar to the control than the erythritol cookies. Also, consumer overall acceptance decreased significantly with sucrose replacement by erythritol. The analysis of texture and sound revealed that inulin cookies were softer while erythritol cookies were harder in comparison with control cookies; despite this difference, inulin cookies had similar sound characteristics to erythritol cookies.

Keywords: cookie, sucrose replace, inulin, erythritol, texture, sound.

1. Introduction

Sucrose is the main sugar utilised in the cookie industry. High sugar levels are undesirable for health reasons such as dental problems, obesity and type II-diabetes (Pareyt and others 2009). Nowadays, consumers are concerned about health (Berasategi 2010) and they are looking for non-caloric and healthy products (Baltasavias and Jurgens 1997). For this reason the interest in research about reduction of sucrose content in cookies without reducing sensory quality has increased (Drewnowski 1998).

In general, sugar affects flavor, dimensions, color, hardness, surface characteristics (Gallagher and others 2003) and crispness (Kulp and others 1991). The absence of sucrose in cookies increased the gluten network providing elasticity to the dough and the cookies, which is an undesirable effect (Laguna and others, 2012).

The effect of sucrose replacement in cookies has been studied by many authors. Olinger and Velasco (1996) replaced sugar in baked products such as cakes and cookies with polyols. Lactitol cookie was the closest sample to the control cookie from a sensorial point of view. Sai Manohar and Haridas Rao (1997) employed reducing sugars as sucrose replacers in cookies and studied their rheological characteristics. They concluded that liquid glucose and invert syrup produced greater changes in adhesiveness and stickiness, while high fructose corn syrup had a greater effect on the color of cookies. Zoulias and others (2000) studied also the effect of sugar replacement by different polyols, fructose, and acesulfame-K in cookies that contain polydextrose as fat replacer. They found that cookies prepared with maltitol and lactitol were closed to control cookie. On the other hand, xylitol affected negatively the flavor, fructose provided a bitter aftertaste and mannitol was not suitable as sucrose replacer for cookies. In general, all the polyol-containing cookies were less sweet, than acesulfame-K-cookies, which improved the perceived flavor and general acceptance. Gallagher and others (2003) used an oligosaccharide to replace 20-30% of the sugar in cookies. They obtained softer eating cookies and

different surface color attributes. Kweon and others (2009) studied the functionality of xylose, glucose, fructose and sucrose in the cookie, concluding that whereas formulations with sucrose optimize the flour performance for the cookie, the formulation with xylose exaggerates the worst aspect of cookie flour functionality. Recently, Pareyt and others (2011) used arabinoxylan oligosaccharides as potential sucrose replacers in sugar-snap cookies to replace up to 30% sucrose. They obtained cookies with a diameter and height comparable to their control sample; however, the color of the cookies was darker in comparison with the control.

Inulin, made from the chicory plant, is a fructosyl-fructose-linked oligomeric carbohydrate with a degree of polymerization ≥ 10 (De Vries 2003). It is moderately water soluble and in water can form a short, spreadable gel network (Zahn and others 2010). Inulin possesses the advantage that may act as dietary fibre with prebiotic effects (Meyer et al. 2011). Inulin has been used as a sucrose replacer in low-sugar milk chocolate with prebiotic properties (Farzanmehr and Abbasi 2009). There are no studies about the employ of inulin as sucrose replacer in cookies.

Erythritol is a 4-carbon sugar alcohol, white, anhydrous, non-hygroscopic, crystalline substance with a mild sweetness and an appearance similar to sucrose (Perko and DeCock 2008). Erythritol is not caloric and due to its small molecular size is absorbed in the small intestine and not fermented as other sugars alcohols having high digestive tolerance (Perko et al. 2008). Erythritol has been used as a sucrose replacer in bakery products such as chiffon cake (Lin and others 2003) or Danish cookies (Lin and others 2010) obtaining good stability during baking, increasing cookie lightness, and giving acceptable hedonic scores up to 50% sucrose replacement.

Texture is one of the most important sensory properties determining the quality of cookies. Texture is a sensory and functional manifestation of the structural, mechanical and surface properties of foods, detected through the senses of sight, hearing, touch, and kinesthetics. Only human beings can perceive and

describe texture; instruments are just able to measure certain physical parameters that can be correlated to the sensory parameters (Szczesniak 2002).

Up to now, none of the previous studies in sucrose reduction in cookies has focused on the change of sensory and physical properties. As far as texture is one of the main cookie characteristic determining quality and consumer acceptability, a complete sensory and instrumental texture study in sucrose reduced cookies became of special interest.

Inulin and erythritol may be considered as sucrose substitutes with advantageous properties. Inulin possesses the advantage that may act as dietary fibre with prebiotic effects (Meyer et al. 2011) and erythritol has better digestive tolerance than other polyols.

The hypothesis of this work was to understand the functionality and to evaluate the suitability of inulin and erythritol as partial sucrose replacers in cookies through the study of the instrumental and sensory properties of texture. Sensory analysis, including hedonic and descriptive tests were carried out and their relationship with instrumental properties evaluated.

Material and Methods

Ingredients

The cookie formulations used are shown in Table 1. Five samples were tested: a typical short dough composition as the control or full fat cookie (control), and two sucrose replacers (erythritol and inulin) at two different sucrose replacement levels of 25% and 50% were used. The cookies were namely 25 SE (25% of sucrose replacement by erythritol), 50SE (50% of sucrose replacement by erythritol), 25SI (25% of sucrose replacement by inulin) and 50SI (50% of sucrose replacement by inulin).

The ingredients were: a) soft wheat flour suitable for cookies (Belenguer, S.A., Valencia, Spain) (composition data provided by the supplier: 15% moisture, 11% protein, 0.6% ash; alveograph parameters $P/L=0.27$, where P =maximum pressure required and L =extensibility; and $W=134$, where W =baking strength of the dough); b) shortening (78% fat, St. Auvent, Vandemoortele, France); c) sucrose (Azucarera Ebro, Madrid, Spain); d) inulin (Frutafit® HD; inulin > 90%, fructose < 10%); e) erythritol (Zerose™ erythritol 16957); f) milk powder (Central Lechera Asturiana, Granada, Spain); g) salt, h) sodium bicarbonate (A. Martínez, Cheste, Spain); i) ammonium hydrogen carbonate (Panreac Quimica, Barcelona, Spain) and j) tap water.

Table 1. Ingredients used in the biscuit performance (percentages given on a flour basis).

Ingredients	100S	75SE	50SE	75SI	50SI
Flour	100	100	100	100	100
Shortening	32.15	32.15	32.15	32.15	32.15
Sucrose	29.45	22,08	14,72	22,08	14,72
Sucrose replacer	0	7.37	14.73	7.37	14.73
Milk powder	1.75	1.75	1.75	1.75	1.75
Salt	1.05	1.05	1.05	1.05	1.05
Sodium bicarbonate	0.35	0.35	0.35	0.35	0.35
Ammonium hydrogen carbonate	0.2	0.2	0.2	0.2	0.2
Water	11	11	11	11	11

Cookie preparation

The sucrose or inulin/sucrose or erythritol/sucrose mixture, the milk powder (previously dissolved in all the water), the leavening agents and the shortening were mixed in a mixer (Kenwood Major Classic, Hampshire, UK) for 30 s at low speed (60 rpm). The dough was then scraped from the sides of the bowl and mixed for 3 minutes at a higher speed (255 rpm). The flour was then added, and

mixed for 1 minute at low speed (60 rpm). After a 10 min resting period in a plastic bag, the dough was sheeted in a laminating machine (Parber, Zamudio, Spain) to 10 mm and cut into rectangular pieces measuring 50 x 30 x 10mm (length x width x height). Twenty-five pieces were placed on a perforated tray. The cookies were baked in a conventional oven for 4 min at 175 °C. The trays were then turned 180°, bringing the side that had been at the back to the front of the oven to ensure even cooking, and baked for a further 4.5 min at the same temperature. The oven and the oven trays were always the same, the trays were placed at the same level in the oven each time, and the number of cookies baked was always the same. After reaching ambient temperature, the cookies were packed and stored in heat-sealed metalized polypropylene bags. The cookie samples were evaluated on the following day in all cases.

Sensory Analysis

Testing was carried out in a sensory laboratory equipped with individual booths (ISO 8589, 1988). Data acquisition was performed using Compusense five release 5.0 software (Compusense Inc., Guelph, Ontario, Canada).

Descriptive analysis

Selection of terms and panel training - A panel of nine assessors, aged between 25 and 45 years and skilled in quantitative descriptive analysis, was trained to select the descriptors using the checklist method (Lawless and Heyman 1998).

The assessors were first given a brief outline of the procedures and a list of attributes and representative samples; they were then asked to choose and write down the most appropriate attributes to describe all the sensory properties of the cookies or to suggest new ones. The panel leader collected and wrote all the attributes on a board and the panel discussed the appropriateness of the selected attributes, their definitions and preliminary discussions on how to assess the products. At the end of this session a consensus on the list of attributes was reached. This procedure was proposed by Stone and Sidel

(2004) in order to obtain a complete description of a product's sensory properties

The evaluated terms (Table 2) were yellowness and toast surface intensity color; toast and buttery odor; porosity and convexity of surface appearance; snapping by hand: hardness and sound intensity at break; once the cookie was split in two parts, the panelist observed: matrix aeration. After this, panelists tasted the cookie and selected: buttery, sweet and toast flavor. The in-mouth texture attributes chosen were: hardness on first bite, sound at breaking on first bite, quality of sound, hardness during mastication, sound duration during mastication and finally, moisture mouth feel.

The panelists attended twelve 1-hour training sessions. Training involved two stages: during the first stage, different samples were tasted by the panelists to attain a better understanding and final agreement of how to measure all the descriptors, and different tastings were conducted until the panel was of one mind in its assessments.

Table 2. Attributes, scale extremes and definitions used in the descriptive sensory analysis of the biscuits by a trained panel.

Parameter	Attribute	Scale extremes	Definitions
Color	Yellowness intensity	Low/High	Evaluation of the overall color of the biscuit
	Toast intensity		
Odor	Buttery	Weak/Strong	Evaluation of odors by smelling the biscuit once
	Toast		
Surface appearance	Porosity	Weak/Strong	Evaluation of holes and flat surface of the biscuit
	Convexity		
Manual texture	Hardness	Weak/Strong	Evaluation by breaking in two by both ends and broken
	Sound intensity at break		

Matrix appearance	Matrix aeration	Weak/Strong	Evaluation of the biscuit matrix observing the inside of half a biscuit
Flavor	Buttery	Weak/Strong	Evaluation of the flavor perceived while chewing the biscuits
	Sweet		
	Toast		
In-mouth texture	Hardness on first bite	Weak/Strong	Placing the biscuits between the incisors, evaluation of the hardness.
	Sound at break first bite	Weak/Strong	Evaluation of the noise produced when biting into the biscuits with the incisors
	Quality of sound	High-pitched/ Low-pitched	Evaluation of the noise produced when biting, being high pitched as a chip and low pitched as a candy nougat
	Hardness during	Weak/Strong	Evaluation of the necessary for required during the mastication
	Sound duration during mastication	Short/long	Evaluation of the duration of the sound when chewing with the molars and lips closed
	Moisture	Low/High	Evaluation of the degree of perception of pastiness in the mouth in relation with salivation and difficulty in swallowing

Formal assessment.-A balanced complete block experimental design was carried out in duplicate (two sessions) to evaluate the samples. The intensities of the sensory attributes were scored on a 10 cm unstructured line scale. Four samples were evaluated per session.

50SI cookie was not subjected to sensory testing as they were considered not sensory acceptable due to the high hardness in the first bite with incisors.

In each session, the samples were randomly selected from each baked batch and served in random order, each on a separate plastic tray identified with a random three-digit code. The panelists were instructed to rinse their mouths with water between sample evaluations.

Consumer test

A total of 100 consumers (untrained), aged from 18 to 65 years, who were frequent consumers of short dough cookies took part in the study. The consumers evaluated four samples in one session. The cookies were coded with random three-digit numbers; sample presentation followed a balanced complete block experimental design. Consumer acceptance testing was carried out using nine-point hedonic scales (1 = dislike extremely and 9 = like extremely). The consumers scored their liking for the 'overall acceptance', 'appearance', 'color', 'odor', 'texture', 'hardness', 'crispness', 'dry mouthfeel', 'taste', 'sweetness' and 'butter taste' of each sample.

Moisture content and water activity

The moisture content of the cookies was determined in four replicates of each formulation according to the Approved Method 44-40.01 (AACC International 2009) placing the samples in an oven at 105°C in order to achieve constant weight. Water activity (aw) was determined in four replicates of each formulation as the relative humidity of the air in equilibrium with the outer part of the samples in a sealed measuring chamber using a chilled-mirror dew point technique at 22°C (Aqua Lab Series 3, Decagon Devices, Pullman WA, U.S.A.).

Simultaneous fracture and sound emission of the cookies

Acoustic emission of the cookies was measured during fracturing using a Texture Analyzer with a load cell of 30Kg (TA-Xt Plus, Stable Micro Systems Ltd., Surrey, UK) inside an isolated acoustic chamber with a prepolarized free-field one-half-in" microphone in combination with a Deltatron preamplifier microphone (Brüel Kjær, Nærum, Denmark) as described by Primo-Martín and

others (2009). The cookie was placed on a flat surface with a slot aperture (width 4 mm, length 60 mm). Test pieces were crushed between a wedge (30° cutting angle, 32 mm wide) and the surface. The wedge shaped probe, and a deformation speed of 20 mm/s (and 65 kHz data sampling rate) were chosen to simulate biting with the front teeth (Vincent 1998). The distance of the microphone to the sample was 7 cm. Extensive analysis of the sound was performed using MatLab (Matworks 7.0.4) as described by Castro-Prada and others (2009). Emitted sound was characterized in terms of the number of sound events and mean intensity (dB) which are related to crispness perception (Castro-Prada and others 2009). From the force curves, represented in x-axis with time and y-axis as Sound Pressure Level (SPL), the number of force drops, the slope at rise first event (N/s), and the maximum force (N) were studied.

Statistical analysis

Analysis of variance (one way-ANOVA) was applied to texture, sound and consumer acceptability to study the effect of the formulation; least significant differences were calculated by the Tukey's test and the significance at $p < 0.05$ was determined.

Two way analysis of variance (ANOVA) was applied to each descriptor to check panel performance, considering the assessors, the samples and their interaction as factors.

Pearson correlation was calculated to correlate the sensory data with the instrumental data.

All the statistical analyses were performed using XLSTAT 2009.4.03 statistical software (Microsoft, Mountain View, CA).

Results and Discussion

Quantitative descriptive analysis

The scores for the four samples evaluated by the trained panel are presented in Figure 1. Among all the attributes studied, ten were statistically different: toast color intensity, surface porosity, manual hardness at break, matrix aeration, buttery and sweet flavor, hardness at first bite, sound intensity in mouth at break in the first bite, sound duration during mastication and moisture mouthfeel.

The highest toast intensity color was for the 50SE cookies, followed by the control cookies, although no significant differences between these two samples were found.

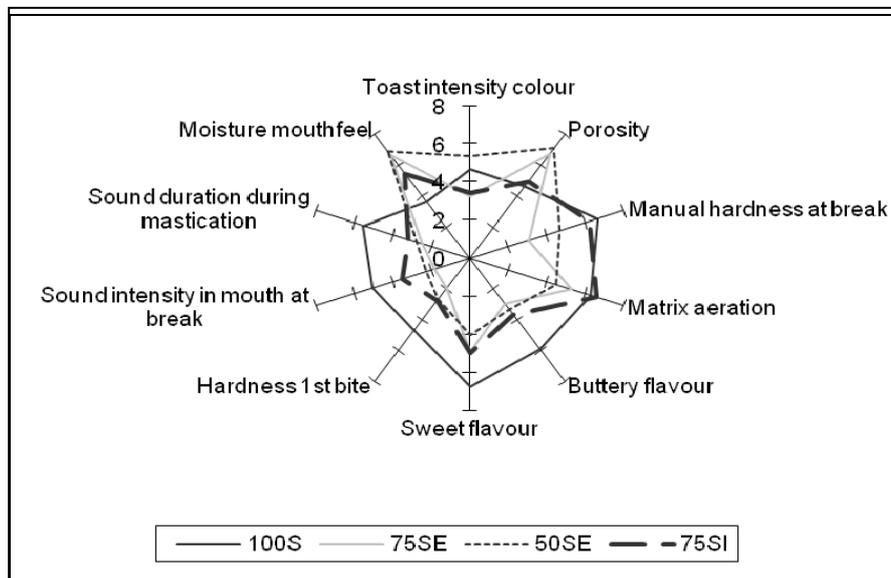


Figure 1. Average (n = 2) descriptive sensory scores for sucrose replacement. Control formulation compared with erythritol and inulin replacement.

In short dough recipes, the water is insufficient to dissolve all the added sugars, so crystal sugars are found, being the size of the crystal an important quality

aspect (Manley 2000). In this study crystal sugars were observed on the cookie surface. The panelists defined the cookie porosity as the holes and spots formed due to crystal sugars found in the cookie surface. The highest score for porosity was given to the erythritol cookies (50SE) obtaining an increase in porosity with the sucrose replacement. The “porosity” found by the panelists in the erythritol sample may be explained due to the fact that erythritol could not be dissolved in the water employed in this cookie formula and remained as intact granules on the cookie surface. So higher concentrations of erythritol implied higher number of holes or spots presented on the cookie surface. Also, the erythritol cookie was significantly different from inulin and control cookie, whose surfaces were evaluated as flat.

Manual hardness at break was also one of the differentiate attributes. The control and 25SI cookies were the hardest samples, followed by the 50SE and 25SE.

In the matrix aeration two groups were found, control-25SI and 50SE-25SE cookies. Matrix aeration refers to the entrap air in the cookie matrix. It seems that erythritol did not allow air to be entrapped in the dough, whilst the control and inulin did.

The sweet flavor and butter flavor were marked by the trained panel as significantly different. Both flavors were described as significantly higher for the control sample compared to the sucrose replaced samples. It is known that sweetness is determined by the sugar used and both sucrose replacers used were less sweet. The erythritol is 60-70% less sweet than sucrose (Perko and DeCock 2008) and the inulin used is mainly composed by long chained polymers of fructose having less sweetness (approximately 10% of the sweetness of sucrose) (Orafti 1996). No changes in the fat of the cookie formula were made so, it seems that sucrose increased the butter taste, as the control cookie was given the most buttery taste, followed by 25% of sucrose replacement (in both replacers: inulin and erythritol) and finally the least buttery taste was the 50% erythritol cookie.

The hardness on first bite was in relation with manual hardness, but higher differences can be observed between samples in-mouth, than by hand. Panelists scored the control cookie as the hardest while sucrose replacement cookies were perceived as softer, without significant differences among them. However, theoretically, and as it will be discussed further on in this work, the sucrose replacement enhanced gluten cross-linking increasing the instrumental cookie breaking strength (Pareyt and others 2009). It was noticed that panelists perceived the crust (top layer of the cookie) as harder in the presence of sucrose. The moisture mouthfeel was found to be higher in the inulin/sucrose and erythritol/sucrose cookies than the control; this fact probably contributed to the hardness perceived in the control sample.

The sound intensity at break in the first bite and the sound duration during mastication was strong for the control and inulin cookies, and weak for the erythritol cookies. There will be further discussion about this subject in the next section.

The results from the descriptive analysis tests provide a complete sensory description of the effect of sucrose replacement in cookies, and the basis for determining those sensory attributes that are important to acceptance.

Consumer test

Apart from being nutritious, the food product sensory properties should be acceptable by consumer to guaranty success (Scholtz and Bosman 2005). Sensory consumer tests are essential to evaluate food acceptability. The mean consumer scores for global acceptance and consumer liking of appearance, color, odor, texture, hardness, crispness, dry mouth feel, taste, sweetness and butter taste for the control sample and sucrose replacement cookies are presented in Figure 2.

In all the attributes scored, the acceptability of the 25SI cookie did not differ from the control cookie. The opposite behavior occurred for 50SE which was different in almost all the attributes except in hardness and dry mouthfeel.

Additionally, these attributes had lower acceptability scores than the other attributes for all samples. In general, the 25SE cookies were less appreciated than the control and the 25SI, but not as low as 50SE.

The consumer's scores for crispness, sweetness and butter taste were lower than the scores of the other sensory attributes evaluated. The differences found among formulations with different sugars and different quantities of them showed that consumers were only capable of perceiving the difference between the control and 50SE. In a previous work, Taylor and others (2008) used tagatose as sucrose replacer obtaining a lower score in sweetness acceptability for a level of replacement or higher.

In conclusion, consumers preferred the control and inulin cookies (25SI) than the erythritol (both 25SE and 50 SE) cookies.

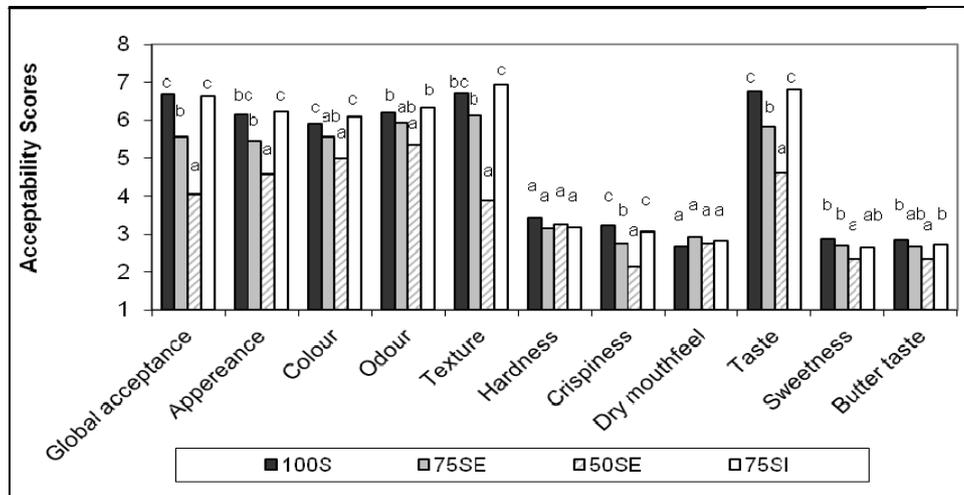


Figure 2. Mean consumer acceptance scores for control, and sucrose replacement biscuit by erythritol and inulin

Instrumental measurements: changes in moisture and fracture due to sucrose replacement

Moisture and water activity

The moisture content and water activity have a large effect on crispness perception and on the mechanical signature of brittle cellular foods. The moisture and water activity values of the cookies are shown in Table 3. In order to compare the crispness behavior of the different cookies, the first step is to determine if the moisture and water activity values of all the cookies are in the range which does not affect crispy sensory appreciation. Food crispness has a sigmoid relationship with water content and water activity (Peleg 1994). The moisture contents range from 3.92 to 5.62 and a_w from 0.205 to 0.345. These low values guarantee independence with the crispness characteristics.

Table 3. Biscuit characteristics for control and sucrose replacement biscuits.

Biscuit sample	Moisture (%d.b.)	a_w
100S	3.92 ^a (0.28)	0.22 ^b (0.004)
75SE	5.62 ^c (0.18)	0.34 ^c (0.003)
50SE	4.55 ^b (0.12)	0.28 ^d (0.006)
75SI	4.01 ^a (0.11)	0.22 ^b (0.005)
50SI	4.08 ^a (0.10)	0.20 ^a (0.002)

Values in parentheses are standard deviations. Means (N = 4) in the same column with the same letter do not differ significantly ($p < 0.05$) according to the Tukey test.

a_w : water activity

Fracture and sound properties

Representative force-time curves and the simultaneously recorded sound obtained during the fracture process of the different types of cookies are shown in Figure 3. The general pattern of cookie fracture was characterized by an increase in force followed by a sudden drop. In the control cookies the force time curve is characterized by a jagged profile composed of multiple peaks of varying amplitude, revealing the presence of a brittle cellular structure (Vincent 1998). The force drops were accompanied by sound events.

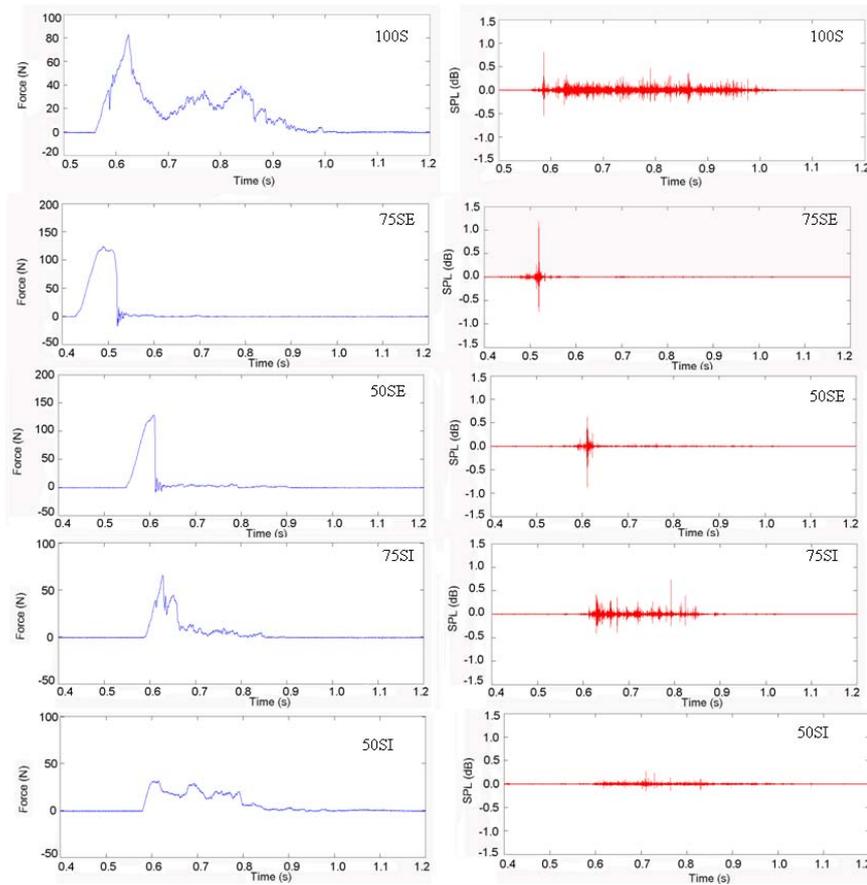


Figure 3. Force (left side) and sound pressure (right side) as a function of time during the biscuit deformation for the different formulations.

The replacement of sucrose affected the curve profiles obtained. In both the sucrose replaced cookies a decrease in the jaggedness of the force curves was observed, which indicates a decrease in the brittleness, especially in the cookies with erythritol as sucrose replacer (25SE and 50 SE).

The corresponding sound curves also showed a decrease in the number of sound events. In the erythritol curves practically no sound was registered during fracturing.

In the inulin cookies (25SI and 50 SI) a lower force at break and a more jagged profile was observed in comparison with the erythritol cookies.

To objectively quantify the differences in the profiles of all the cookies, specific parameters were obtained from the curves (Table 4).

In all of the force curves it can be appreciated a gradient up to a point where the curve falls; this behavior was measured as the slope of the first event and it is related to the sample deformation. The slope values revealed no significant differences among the control and erythritol samples. In the inulin cookies, the 50% level of replacement (50SI) showed significantly lower slope values, indicating less fragile and more ductile behavior.

The sucrose replaced by erythritol (25SE and 50SE) showed significantly higher values of maximum force achieved during fracturing. This implies that instrumentally the erythritol cookies were harder cookies. Inulin had the opposite effect to erythritol, and the 50% replacement (50SI) showed a significant decrease in maximum force; however, the 25% replacement cookies (25SI) were not significantly different to the control.

The sound data reveal that despite being the hardest cookies, the mean intensity of sound emitted by the erythritol cookies was very low.

For both the 25% and 50% replacement of sucrose by erythritol, the number of sound events and the mean sound intensity were significantly lower than the control. The erythritol force curves (Figure 3b and 3c) showed a profile composed of a unique peak with a round top, instead of a sharp point. This

indicates less number of microfractures (less jaggedness profile) associated to a more viscous material, and explains the lack of sound emitted during fracturing.

Sound data corresponding to the inulin cookies (25SI and 50SI) revealed a significantly higher number of sound events than in the erytritol cookies (25SE and 50SE). No significant differences were found in the number of sound events among the 25SI cookie and the control cookie, although the mean sound intensity was significantly lower in the 25SI samples than in the control cookie.

The sound-related parameters revealed significantly higher values of total sound energy and mean intensity between full sugar cookies (control) and the formulations with sugar replacement.

Table 4. Data obtained from texture and acoustic measurements.

Biscuit sample	Force data			Sound data	
	Force drops	Slope rise first event [N/s]	Max force [N]	Sound events	Mean intensity [dB]
100S	15,4 ^{abc} (2,9)	1999,6 ^a (460,2)	88,2 ^a (12,88)	667,4 ^a (128,1)	66,2 ^a (1,2)
75SE	16,9 ^{ab} (2,7)	1981,3 ^a (452,3)	120,8 ^b (6,2)	199,7 ^b (118,5)	61,8 ^b (1,7)
50SE	17,7 ^a (2,1)	1939,6 ^{ab} (878,2)	135,3 ^c (12,3)	225,4 ^b (165,1)	61,6 ^b (2,6)
75SI	14,7 ^{bc} (2,2)	1900,0 ^a (306,9)	79,1 ^a (15,8)	573,2 ^a (117,5)	63,0 ^b (1,3)
50SI	12,7 ^c (2,1)	1332,6 ^b (365,9)	42,2 ^d (16,8)	419,0 ^c (124,6)	55,5 ^c (2,7)

Values in parentheses are standard deviations. Means in the same column with the same letter do not differ significantly ($p < 0.05$) according to the Tukey test.

Despite the fact that no definition was given for a “crispy attribute” from the trained panel, in figure 1 it can be observed that the trained panel related the sound intensity with the sound event, fitting in the definition of crispy products (fracture and sound emitted at low force at break). The panel agreed that the crispiest cookie was the control cookie, followed by the 25SI cookie, the 25SE cookie and the 50SE cookie.

In summary, the instrumental data based on the fracture behavior and the accompanied sound emission revealed two different behaviors between erythritol and inulin. Partial replacement of sucrose by erythritol was associated with lower sound events and a higher force required for breaking the cookie. Inulin cookies, however, were softer in texture but produced sound properties closer to the control cookie.

A food product is considered crispy when it fractures with multiple fracture events at a relative low work of mastication, and these different successive fracture events are accompanied by sound emission (Luyten and others 2004). According to this definition, the instrumental results showed that inulin will perform better than erythritol as a partial sucrose replacer in order to maintain the crispness properties of the cookie.

Correlation between sensory and instrumental values

Correlation between sensory and instrumental values

The correlations between sensorial (descriptive and acceptability) and instrumental parameters were studied.

The sound events measured by instrumental analysis correlated well with the sound at break first bite in mouth scored by the trained panel ($r=0.959$). This could mean that the panelist scored as the cookie with more sound the samples that have more sound events, following the order: control>25SI>50SE>25SE.

The maximum force obtained instrumentally was negatively correlated with sensory matrix aeration ($r=-0.993$). In the present work, the creaming method was used to mix the ingredients, meaning that the sugar is mixed with the fat

and water to form a cream-like mixture, and as Pareyt and Delcour (2008) cited, it is at this stage when the air is incorporated and the sugar helps to cream the air into the fat. As the results showed, the sucrose (control) and the inulin cookies (25SI) had the highest levels of matrix aeration, compared to the erythritol cookies (25SE, 50SE) that had a dense matrix. The cookies scored with the highest matrix aeration by the trained panel (control and 25SI) were also the ones with the highest number of sound events obtained by instrumental analysis. As more air is entrapped in the cookie matrix (sensorial test) more sound events were found (instrumental test). The matrix aeration was also positively correlated with consumer's acceptance: global acceptance and appearance ($r=0.972$ and 0.987 , respectively).

The liking scores of crispness and sweetness by consumers were not statistically correlated (Pearson correlation) with the instrumental data obtained. However, these scores followed the same trend than the number of sound events and the mean intensity of sound (i.e., when the sound events and the mean intensity of sound increased, an increase in crispness was found by consumers).

Conclusions

From the collected data of this study it could be concluded that sucrose replacement affects the cookie's appearance as well as the cookie matrix. As a consequence, changes in sensory and instrumental data are observed. The trained panel found significant differences in toast color, surface porosity, manual hardness, matrix aeration, butter and sweet flavors, sound break at first bite, sound duration during mastication, and moisture mouth feel; the 25SI cookies were the closest sample to control sample. Consumer acceptance decreased with erythritol replacement, especially at 50% sucrose replacement, except for hardness and dry mouthfeel where no significant differences among the samples were found. The instrumental data collected shows that the 25SI

cookies were softer and more brittle (compared to control cookie) and the 25SE and 50SE cookies were harder and tougher.

This study has shown that although sucrose replacement using erythritol produces suitable dough from a processing point of view, erythritol is not suitable from a sensory point of view (too hard cookie). Inulin could be used to replace up to 25% of sucrose in short dough cookies without having a detrimental effect on consumer perception of the product. Further studies will be needed to achieve a complete sucrose replacement without quality loss.

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UNDERSTANDING THE EFFECT OF SUGAR AND SUGAR REPLACEMENT
IN SHORT DOUGH BISCUITS

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Abstract

Sucrose is the main sugar used in short dough biscuit formula and it plays an important role in the biscuit manufacturing as well as in the biscuits final quality. However, for health reasons, high levels of sucrose are undesirable, making sucrose replacement an important issue to study. The present study focused on sucrose reduction and its replacement by polyols (erythritol and maltitol) in short dough biscuits. The effects were investigated in a model system composed of gluten and different sugars (sucrose, maltitol and erythritol), in biscuit dough and in baked biscuits. Modulated thermal analysis showed that sucrose decreases the glass transition temperature, however for both polyols studied, no transition was found due to a plasticization effect. The gelatinization of starch in the biscuits was not affected by the sugar or quantity of sugar used. Temperature sweeps of short dough revealed that the presence of sugar delays the transitions. Furthermore, G^* increased with sucrose replacement, with the smallest changes for the maltitol-containing biscuits compared to the control. Finally, texture and dimension analysis were carried out. Sugar-free and erythritol-containing biscuits were compact, elastic and resistant to the breaking force compared to the control biscuits and the maltitol-containing biscuits.

Keywords: Thermal properties, rheology, biscuit, sugar, polyols

1. Introduction

Biscuits are popular bakery items, consumed by nearly all levels of society. They are ready to eat, of good nutritional quality and available in different varieties at an affordable cost. Short-dough biscuits comprise widely diverse products. Generally, they are rich in both fat and sugar while containing only a small amount of water. Sucrose is the main sugar utilized in the biscuit industry. Nowadays, however, such high sugar levels are considered undesirable (Gallagher 2003) for several health reasons such as dental problems, obesity, type II-diabetes, high blood cholesterol and coronary disease (Pareyt 2009a). Decreasing the amount of sugar added to biscuits is a good way to obtain a healthier and lower-calorie product (Drewnowski 1998).

From a sensorial point of view, sugar affects flavor, dimensions, color, hardness and surface of the biscuit finished product (Gallagher 2003). Additionally, the quantity and type of sugar used will have an effect during the whole biscuit preparation procedure (from dough mixing to packaging). In the mixing process, sucrose competes with flour for the available water, inhibiting the gluten development (Gallagher 2003). Moreover, it affects the dough consistency (Olewnik et al. 1984) which plays a role in the sheeting step. During baking, sugar has an effect on gelatinization of starch (Spies et al. 1982), browning reaction (Kulp et al. 1991), gluten mobility (Pareyt et al. 2009b), biscuit spread, crispness, and surface characteristics (Kulp et al. 1991).

Due to all the sugar functions in biscuits, sugar replacement is rather difficult to achieve. The effects of sugar reduction and sugar replacement in biscuit have been studied by many authors. Olinger and Velasco (1996) replaced sugar in baked products such as cake and biscuit with lactitol, maltitol, isomalt, sorbitol and polydextrose. They describe changes in the spread, crust color, and tacky surface. Sai Manohar et al. (1997) applied reducing sugars in biscuits and studied their dough rheological characteristics and biscuit quality, concluding that among the different reducing sugars, high fructose corn syrup imparted the

best color to biscuits. Zoulias et al. (2000) studied the effect of sugar replacement by different polyols (maltitol, lactitol, sorbitol, xylitol, mannitol), fructose and acesulfame-k, observing that the biscuits prepared with maltitol and lactitol resembled the control biscuit, while also sorbitol-containing biscuits had acceptable properties. However, xylitol and fructose negatively affected the flavor and mannitol restricted the spread and imparted unpleasant flavor and appearance, making them unsuitable for biscuits. In general, all the polyol-containing biscuits were less sweet than the control while the sweetness of biscuits containing acesulfame-k was increased improving their perceived flavor and general acceptance. Lately, Gallagher et al. (2003) used an oligosaccharide, Raftilose, to replace 20-30% of the sugar resulting in softer biscuits and different surface color attributes. Kweon et al. (2009) studied the effects of xylose, glucose, fructose and sucrose on flour functionality in biscuits, concluding that while sucrose optimizes the flour performance, xylose negatively affected biscuit quality. Recently, Pareyt et al (2011) used arabinoxylan oligosaccharides as a potential sucrose replacer in sugar-snap biscuit to replace up to 30% sucrose obtaining comparable diameters and heights; however, darker colors were found compared to the control.

Although the effect of sugar reduction and replacement on dough and biscuit properties has been widely studied, understanding of ingredient interactions and fundamental concepts is lacking. Deeper knowledge of the functionalities of sugar and sugar replacers in the different processing steps is essential in order to improve the quality of low-calorie biscuits. Therefore, the objective of this paper is to study the feasibility of using various sugars (sucrose, erythritol and maltitol) in short dough biscuits from three viewpoints: 1) in a model system composed of hydrated gluten with sugar 2) in biscuit dough and 3) in baked biscuits. The obtained results will provide essential information on the role of sugars in biscuit manufacturing and quality and on the suitability of erythritol and maltitol as sugar replacers. In addition, understanding of sugar ingredient interactions in short dough biscuits will be obtained.

2. Materials and methods

2.1. Model system

Ingredients of model system

Gluten water model systems were prepared according to the formulations presented in table 1 where: a) granulated sucrose provided by Suiker Unie (Breda, the Netherlands) b) Erythritol (Cargill, Amsterdam, the Netherlands), c) Sweet pearls (Maltitol, Roquette Freres, Lestrem, France), d) gluten, Vital gluten Protinax ©.

Table 1. Model system formulations.

Ingredients (g)	S0	S0.5	S1	E0.5	E1	M0.5	M1
Gluten	1	1	1	1	1	1	1
Water	1	1	1	1	1	1	1
Sugar	-	0.5	1	0.5	-	0.5	-
Maltitol	-	-	-	-	-	0.5	1
Erythritol	-	-	-	0.5	1	-	-

All the systems contains gluten:water in a ratio of 1:1 in g units. S0: system without sucrose; S0.5: system with 0.5g of sucrose; S1: system with 1g of sucrose; E0.5: system with 0.5g of erythritol; E1: system with 1g of erythritol; M0.5: system with 0.5g of maltitol; M1: system with 1g of maltitol.

The model system ingredients quantities were chosen to study the effect of different sugars in gluten. In table 1 the ratio between the ingredients are shown. As can be seen in the table 1, the samples were labeled in such way that the first letter refers to the sugar used being S= sucrose, E= erythritol and M=maltitol; the following number corresponds to the sugar-ratio selected. The first sample, S0, is a 1:1:0 mixture of gluten,water and no sugar, this gluten:water ratio was remained constant in all the formulations. In the other samples each sugar was used in two ratios: 0.5 and 1, respect to the gluten and water mixture. For example, S0.5 corresponds to a mixture (on a gram basis) of gluten, water and sucrose at 1:1:0.5 and so on.

Preparation of model system

The model systems were prepared according to a modification of the method described by Kalichevsky et al. (1992), in which gluten is hydrated and mixed using approximately 200ml of liquid nitrogen, 100 ml of water or the mixture of sugar and water, was sprinkled into liquid nitrogen and milled in a Retsch Grindomix GM 200 (Retsch Benelux, Belgium) for 10 seconds at 8 r.p.m. by an electric mill. More liquid nitrogen was added and the gluten powder was milled together with the water, again for 10 seconds. Subsequently, the mixture was transferred to a plastic container, rested overnight at 4°C and finally transferred into a metal tray.

Next, all the samples were dried in an oven at 30 °C for 24 hours. The samples were then stored for 3 days over P₂O₅ for optimal drying, and subsequently over saturated salt solutions of various relative humidities for at least two weeks in order to obtain a variety of water contents. The salts used were KNO₃ ($a_w=0.936$), KCl ($a_w=0.843$), KI ($a_w=0.69$), Ca(NO₃)₂ ($a_w=0.51$), K₂CO₃ ($a_w=0.432$), MgCl₂ ($a_w=0.328$), CH₃CO₂K ($a_w=0.22$) and LiCl ($a_w=0.113$). One part of each sample was transferred to DSC pans while the other part was transferred into a separate dish for water activity measurement. The water activity was measured using a Decagon Aqua Lab meter (Pullman, WA, USA) calibrated with a 8.57 molal lithium chloride solution ($a_w=0.500$).

The equilibrium moisture content of the samples was determined gravimetrically by drying in an oven at 105 °C for at least 24 hours.

Modulated Differential Scanning Calorimetry (MDSC)

Modulated calorimetric measurements were carried out using a MDSC Q2000 (TA Instruments, New Castle, USA).

The samples (10-15 mg) were scanned from -40 °C to 120 °C at a rate of 2°C/min and the cycle was repeated two times in order to provide a good resolution of the transition phenomena. The period and the amplitude of

modulation were 100s and 0.5°C respectively. The glass transition temperature (T_g) was determined as the mid-point temperature in the reversing heat flow signal, using the automatic T_g analysis tool available in the Universal Analysis software (TA instruments, New Castle, USA).

2.2. Dough and biscuit performance

Biscuit ingredients

Two different sucrose replacers were used: erythritol and maltitol.

The biscuit formulations are based on a short dough biscuit recipe from Manley (1991) and shown in table 2. The sweetener quantities were calculated based on their molarities in order to maintain the same molar concentration of sugar. The ingredient ratios used were such as to avoid a slightly cohesive dough powder and the final dough obtained was poorly elastic but extensible enough to allow an easy shaping of the material. All the reformulations were calculated such that the ratio between flour and other ingredients (except sucrose) was kept the same, see table 2.

The sample names in table 2 are composed of a number and a letter, the number corresponds to the level of sugar used, taking 17.14g of sucrose in the control biscuit as 100, and the letter refers to the sugar used being S= sucrose, E= erythritol and M=maltitol.

The ingredients were: a) commercial wheat flour suitable for biscuits, IJsvogel from Meneba Meel BV (Rotterdam, The Netherlands) (composition data provided by the supplier: 15.0% moisture, 10.5% protein, 0.58% ash) b) fat (Trio Wals) provided by Unipro, Professional Bakery, (Bergen op Zoom, The Netherlands) c) granulated sucrose, (c) granulated 204 sucrose d) sweeteners: erythritol, maltitol, same supplier as for the model systems, e) sodium chloride f) sodium bicarbonate g) ammonium bicarbonate and e) deionized water was used in all the experiments.

Preparation of biscuits

First, sucrose syrup was prepared for each of the formulations by adding the sugar to distilled water and stirring for 1 hr in a water bath.

Secondly, the syrup, fat and salt were mixed in a mixer (Fit Hobart mixer, USA) for 30s at low speed (no.1), the bowl was scraped down and mixing was continued for 3min at a higher speed (no.2). Next, the flour was added and mixed in for 20s at speed no.1 and, after scraping down the bowl once more, for a further 40s at the same speed. After mixing, the dough was allowed to stand for 10 minutes during which starch and proteins absorbed the water (Pareyt et al. 2008) and in order to reduce significant differences in dough quality (Manley 1991). Then, the dough was sheeted to 4.8 mm thickness. To avoid gluten development and the subsequent deformation of shape and dimensions, the dough was turned 90° during the sheeting process, after each sheeting over the roller.

Biscuits were shaped by a biscuit molder of a circular shape with a diameter of 6.5 cm and 24 holes of 1 mm. Sixteen biscuits were placed on a baking tray and baked for 16 min in a standard electric oven with 190 °C bottom- and 210 °C top-temperatures. After baking and reaching ambient temperature, the biscuits were packed and stored in plastic bags. The biscuit properties were analyzed after 1 day of storage.

Table 2. Biscuit formulation prepared with different sucrose concentration and different sugar replacers.

Ingredients (g)	0S	25S	50S	100S	25E	50E	25M	50M	100M
Flour	55.05	58.00	60.90	66.90	57.25	61.12	55.40	55.40	55.35
Fat	17.87	18.73	19.47	21.41	18.32	19.56	17.73	17.73	17.71
Sucr	17.14	12.85	8.57	0.00	12.85	8.57	12.85	8.57	0.00
Water	9.04	9.47	10.09	10.70	9.16	9.78	8.86	8.86	8.86
Salt	0.60	0.63	0.64	0.70	0.60	0.64	0.58	0.58	0.58
sodium bicarbonate	0.19	0.20	0.21	0.23	0.20	0.21	0.19	0.19	0.19
amonium bicarbonate	0.11	0.12	0.12	0.13	0.11	0.12	0.11	0.11	0.11
Sweetener (Erythritol/Maltitol)	-	-	-	-	1.53	3.05	4.30	8.60	17.20
Total weight	100.00	100.00	100.00	100.08	100.03	100.00	100.03	100.05	100.00
ratio water/flour	0.16	0.16	0.16	0.16	0.16	0.16	0.16	0.16	0.16
ratio fat/flour	0.32	0.32	0.32	0.32	0.32	0.32	0.32	0.32	0.32
ratio salt/flour	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01

100S= no sucrose replacement, full sucrose biscuits; 75S=25% of sucrose deleted, 50S=50% of sucrose deleted, 0S=100% of sucrose deleted. 75E=25% of sucrose replaced by erythritol, 50E=50% of sucrose replaced by erythritol. 75M=25% of sucrose replaced by maltitol, 50M=50% of sucrose replaced by maltitol, 0M=100% sucrose replaced by maltitol.

2.3. Dough and biscuit analysis Starch gelatinization

The starch gelatinization was determined according to a standard procedure described by Biliaderis et al. (1980), using a DSC Q2000 (TA Instruments, New Castle, USA). Samples were heated from 20 °C to 160 °C at a rate of 10 °C/min in Perkin-Elmer stainless steel hermetically sealed pans. Both, biscuit dough and baked biscuit, were analyzed in excess of water (1:3).

Water content and water activity

The moisture content of the model system, biscuit dough and biscuits were determined according to the Approved Method 44-15.02 (AACC International 2009).

Water activity (a_w) was determined using a Decagon Aqua Lab meter (Pullman, WA, USA) calibrated with a 8.57 molal lithium chloride solution ($a_w=0.500$).

Rheological experiments

A controlled stress rheometer (AR 2000, TA Instruments, Crawley, UK), equipped with the Environmental Test Chamber (ETC) and serrated parallel plates (25 mm geometry) with a gap of 3 mm, was used for rheological characterization. Strain sweep tests (0.0001-0.1) were performed to measure the linear viscoelastic properties at a constant frequency of 1 Hz. A critical shear strain (γ_c), the strain at which a deviation from linear behavior occurs, was estimated from the normalized plot of G' and G'' (data not shown). In order to simulate the effect of the baking process, temperature sweeps at a constant deformation amplitude within the linear viscoelastic region, ($\gamma=0.0001$ at 1 Hz) were carried out by increasing the temperature from 25 °C up to 160 °C, at a constant rate of 5 °C/min. The storage modulus (G'), loss modulus (G'') and loss tangent ($\tan\delta = G''/G'$) which is consider the ratio of viscous to elastic properties were measured. Both modulus (G' and G'') are derived from the complex shear modulus G^* which represents the total resistance of dough to imposed deformation and are related with G^* as the following equation shows:

$$G^* = \sqrt{(G')^2 + (G'')^2}$$

G^* represents the total resistance of dough to imposed deformation.

Biscuit texture analysis

The texture of the biscuit was measured using a TA.TX.plus Texture Analyzer (Stable Micro Systems, Goldaming, UK). Texture analysis of the biscuits was performed by two tests:

- *Fracture Strength*. The biscuits were fractured using a three point bending probe and support (A/3PB). The experimental conditions were: test speed 0.5 mm/s; distance between supports 20 mm apart; probe travel distance 3 mm; trigger force 50 g. The force at fracture (N), the distance at break (mm) and the gradient of the curve (N/sec) were recorded

- *Puncture test*. Penetration tests were conducted with a semispherical probe (5 mm diameter), placing the sample upside down and penetrating at 0.5 mm/s (whole biscuit) to a distance of 10 mm with a trigger force of 50 g. Five holes were made in each biscuit. The parameters measured were the area under the curve (N.mm) as the resistance to penetration and the number of peaks as an index of crunchiness.

Biscuit dimensions

Biscuit thickness was measured by stacking 10 biscuits vertically against a biscuit thickness ruler, sliding a gauge to rest on top of the pile and calculate the average thickness from the height determined. Biscuit 'length' was measured by arranging 10 biscuits along the length ruler (with the stamped word parallel to its long edge) and recording the average length. Next, the biscuits were turned 90° and the average 'width' was determined. These measurements were expressed in mm as the average value/10 by duplicates.

2.4. Statistical analysis

Analysis of variance (one way-ANOVA) was applied to study the differences between formulations; least significant differences were calculated by the Tukey test and the significance at $p < 0.05$ was determined. These analyses were performed using SPSS for Windows Version 12 (SPSS Inc., USA).

3. RESULTS

3.1 Modulated Differential Scanning Calorimetry of the model systems

In MDSC, the conventional linear temperature increase/decrease is overlaid by a sinusoidal oscillation. The resulting modulated heat-flow signal can be separated into reversible and non-reversible signals, which aids in the data interpretation (Auh et al. 2003). The glass transitions (T_g) can be observed in the reversible heat flow signal, while melting/crystallization transitions can be observed in the non-reversible signals

Figure 1 shows the T_g 's of the gluten-sucrose model systems as a function of moisture content, as determined by MDSC. For S0, S0.5 and S1 the T_g of gluten was depressed with increasing sucrose content in the range of moisture contents studied. This result is in accordance with Kalichevsky et al. 1992.

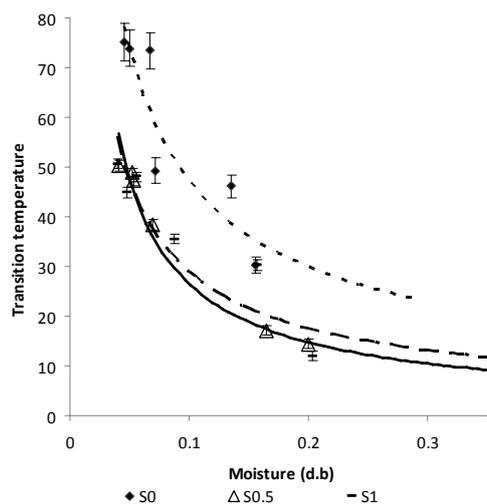


Figure 1. Effect of sucrose level in gluten the model systems gluten:water:sucrose; S0(1:1:0), S50 (1:1:0.5) and S100(1:1:1).

The polyol-containing model systems studied (table 1) did not show a glass transition in the MDSC signals. However, another phenomenon appeared in the MDSC curves for the gluten-polyol systems.

For the gluten, in presence of erythritol (example in figure 2a), the heating cycle showed a melting peak at 113°C. This peak moved to lower temperatures with increasing moisture content. During the cooling cycle at temperatures below zero, a crystallization effect occurred. As sucrose melting occurs at 179 °C (Kaizawa et al 2008) this melting transition was not observed in the gluten-sucrose samples scanned to 120 °C.

As in erythritol samples, in the systems formed by maltitol and gluten (figure 2b), no transition was found with the MDSC technique. A change in the slope of the curves was found at 41 °C approximately in all the samples, and a melting peak at 126.24 °C.

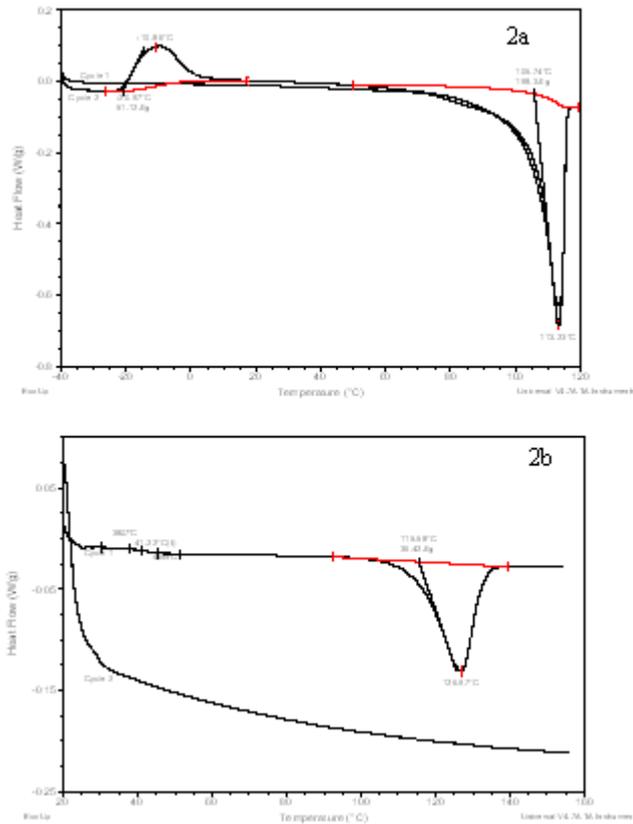


Figure 2. Thermogram of model system. Figure 2a. E100 at low moisture content (1gluten:1water:1erythritol) Figure 2b. Thermogram of model system M100 at low moisture content (1gluten:1water:1maltitol.)

The study of the model systems by MDSC revealed that the gluten in presence of plasticizers such as water, erythritol and maltitol does not go to the glassy state. In contrast to erythritol and maltitol, with the addition of sucrose the glass to rubber transition decreases to lower temperatures. As the water content is nearly constant the changes observed by sucrose addition were not caused by different amounts of water.

3.2. Calorimetric measurement in dough and biscuits.

In order to evaluate the degree of starch gelatinization in presence/absence of sugar DSC analysis was carried out on dough and biscuit samples in excess of water (1:3). A summary of the DSC results is presented in table 3. In all the thermograms (biscuit and dough) an endothermic peak between 66.53 and 75.63 °C appeared which corresponds to the starch gelatinization, in accordance with other authors (Baltsavias et al. 1999). The presence of this peak in the biscuit implies that the extent of starch gelatinization during baking was very low. In the dough samples the peak appeared at lower temperature than in the biscuit samples. This increase in the gelatinization temperature after baking can be attributed to structural changes within the starch granules occurring as a result of the heat-moisture treatment (baking), which involves mainly amylose-amylose and amylose-lipid interactions (Hoover et al. 1994).

Furthermore, the comparison of the melting enthalpies (table 3) reflects a decrease in the transition energy in the biscuits in comparison to the corresponding dough (same starch content). This decrease is associated to the fact that during the dough baking process some starch gelatinization occurs, resulting in a lower melting enthalpy after baking. Table 3 showed no significant difference among biscuits dough formulations neither in onset temperature nor peak temperature. In baked biscuits however the sucrose decrement samples (50S and 100S biscuits) presented slightly higher peak gelatinization temperatures compared to the 0S. Moreover, no effect of polyol addition on gelatinization temperatures was observed. As occurred in the onset and peak temperature, no relation between sugar content and enthalpy was found. Eliasson et al. (1992) described a shift to higher gelatinization temperatures in starch-sugar-water systems compared to starch-water systems, and they attributed this stabilizing effect of sugar to sugar molecules forming bridges between neighbouring polysaccharides located within the amorphous region of the granules thereby restricting movement of these areas. In the biscuit systems used in the current study however, this phenomenon was not found.

Table 3. Enthalpy and temperature of starch gelatinization in doughs and biscuits.

	DOUGH		BISCUIT		DOUGH	BISCUIT
	Onset T(°C)	Peak T(°C)	Onset T(°C)	Peak T(°C)	ΔH dry dough/dry starch(J/g)	ΔH dry biscuit/dry starch(J/g)
0S	63.50 ^a (0.93)	71.64 ^a (1.52)	64.93 ^{ab} (0.21)	71.52 ^{ab} (0.21)	8.83 ^a (0.99)	7.54 ^{ab} (0.57)
25S	61.55 ^a (0.13)	68.66 ^a (1.46)	67.12 ^{ab} (1.52)	73.75 ^{ab} (1.80)	8.28 ^a (0.01)	6.61 ^{ab} (0.78)
50S	61.24 ^a (0.04)	69.08 ^a (0.72)	67.48 ^{ab} (0.07)	74.40 ^b (0.54)	10.83 ^{ab} (0.79)	7.92 ^{ab} (1.92)
100S	60.94 ^a (0.08)	67.95 ^a (0.04)	69.09 ^b (0.98)	75.73 ^b (1.41)	9.36 ^{ab} (1.05)	6.44 ^a (0.31)
25E	62.30 ^a (2.60)	66.53 ^a (0.49)	64.96 ^{ab} (2.56)	71.75 ^{ab} (2.19)	12.11 ^b (1.19)	8.74 ^b (0.65)
50E	61.85 ^a (1.15)	68.53 ^a (1.48)	64.50 ^{ab} (1.42)	71.67 ^{ab} (0.76)	9.67 ^{ab} (0.04)	6.92 ^{ab} (0.04)
25M	62.39 ^a (1.86)	69.16 ^a (2.93)	65.27 ^{ab} (0.64)	73.62 ^{ab} (0.37)	10.19 ^{ab} (0.51)	6.61 ^b (0.78)
50M	61.00 ^a (0.13)	67.50 ^a (0.13)	63.81 ^a (0.27)	69.46 ^a (0.31)	11.17 ^{ab} (0.10)	8.47 ^b (0.86)
100M	61.34 ^a (0.04)	67.45 ^a (0.13)	65.52 ^{ab} (0.64)	72.89 ^{ab} (0.11)	9.32 ^{ab} (0.70)	6.92 ^b (0.86)

Values in parentheses are standard deviations. Means (in the same column with the same letter do not differ significantly ($p < 0.05$) according to the Tukey test.

3.3. Rheological experiments

Rheological measurements

The rheological properties of the biscuit dough were studied by small amplitude oscillatory shear tests. For all the dough systems the elastic component (G') was higher than the viscous component (G''). The general trend is an increase in G^* when sucrose is replaced or lowered comparing with 0S (100S>50E>50S>25E>50M>100M>25M>25S>0S), indicating dough stiffening. The G^* of the maltitol-containing doughs (25M, 50M, 100M) resembled the 0S

dough the most, while erythritol addition did not make up for the increased stiffness caused by the sucrose reduction.

In order to evaluate the effect of heating on the linear viscoelastic properties of the different dough systems, the evolution of G' (storage modulus) and G'' (loss modulus) with temperature was studied. Figure 3 shows G' and G'' versus temperature of the 0S dough as representative example where five different zones were observed.

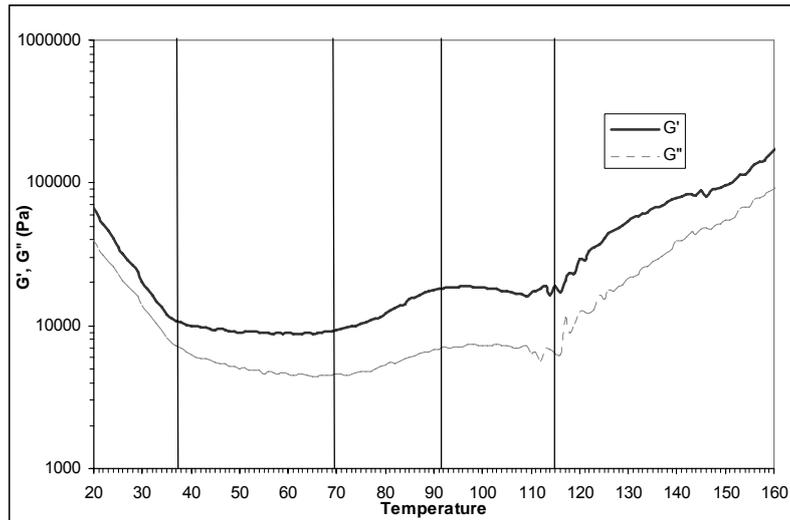


Figure 3. Temperature sweep of control dough.

- 1) The first zone, from 20 °C to 37.5 °C, where a decrease in G' was observed. This viscosity fall may be due to fat melting that occurs between 15-40 °C, depending on the fat polymorph and proportion of fat (Roos 1995).
- 2) A stable zone between 37.5 and 69.5°C, in which G' and G'' were almost independent of temperature indicating that continues heating of the dough did not cause structural changes.
- 3) Between 69.5 and 91 °C, G' increased, reflecting an increase in consistency which is related to the onset of starch gelatinization. The starch gelatinization

occurs at 60-75 °C depending upon the type of starch and the presence of other ingredient (Singh et al. 2005). It is well known that sugar delays the starch gelatinization (Chevallier et al. 2000). However, it should be kept in mind that most of the starch granules do not gelatinize during baking as observed by DSC analysis.

4) A second shorter constant range from 91.5 to 115°C.

5) Finally, from 115 °C up to the end of the test at 160 °C an increase in G' and G'' occurred. At this temperature the dough reached the water boiling point, contributing to dough stiffening (Singh et al.2005). The dough has turned into a cellular solid. Several authors previously studied the change in rheological properties in wheat dough. Bloksma (1980), Dreese et al.(1998) and Singh et al. (2005), observed three different stages during heating. Singh et al. (2005) defined a first part from 25 to 60 °C associated with bubble growth and fat melting. In the second phase, between 60 and 75 °C, a rapid bubble expansion, starch gelatinization and glutenin polymerization occurred. Between 87 and 100 °C final curing and water evaporation took part. Compared to the model described by Singh, the changes of G' and G'' with temperature were less pronounced. This may be due to differences in dough composition, with fat melting depending on the amount and polymorphism of the fat, and starch gelatinization as well as glutenin polymerization depending on the amount and type of sugar (Pareyt et al. 2009b).

The specific effects of sucrose reduction and sucrose replacement by maltitol and erythritol on the moduli are shown in Figure 4a to 4c. The graphs were grouped according to the sucrose level and sucrose replacer.

Figure 4a shows different sucrose levels from full sucrose dough (0S) to sugar free dough (100S). With the progressive sucrose decrement the transitions mentioned (fat melting, starch gelatinization, gluten polymerization) occurred at lower temperature. Both moduli (G' , G'') were higher for the sugar free biscuit dough than for the 0S dough, meaning more dough resistance. $\tan \delta$, (data not

shown), was the lowest for the sugar-free doughs, confirming the G' , G'' values shown (figure 4a).

This finding is generally in line with earlier observations, e.g. Olewnik et al. 1984 found that the dough becomes softer with sucrose increment inducing a fall in the viscosity (Maache-Rezzoug et al. 1998). Moreover, Sai Manohar et al. (1997) observed that with sucrose addition in biscuit dough, less consistency and elasticity are obtained.

Increased elasticity of the sucrose-reduced samples can be explained by more pronounced gluten development in these samples, in agreement with Pareyt et al. (2009b) who stated that gluten entanglement is restricted by the presence of sucrose. The restricted entanglement could be explained by a competition of the gluten and sucrose for water (Yamazaki 1971).

The temperature sweep results of the maltitol formulations are shown in figure 4b. The rheological behaviour of the maltitol doughs (25M, 50M and 100M) was the most similar to that of the 0S dough of all the formulations studied. In Table 4 it can be observed that maltitol has the same carbon numbers and hygroscopicity and similar molecular weight and solubility as sucrose. Therefore, from a rheological point of view, maltitol interacts with all the dough ingredients similar to sucrose.

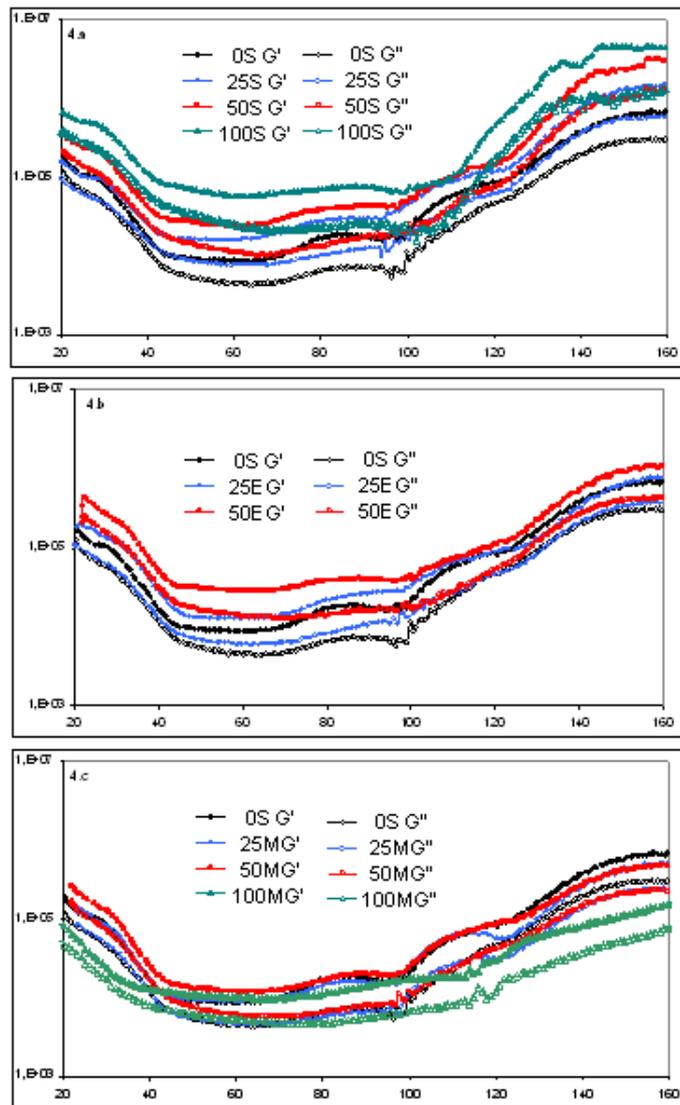


Figure 4. Evolution of the viscoelastic functions upon increasing the temperatures in short dough biscuit. 4a. Different sucrose level. 4b Different erythritol and sucrose level 4c Different maltitol and sucrose level.

Erythritol doughs (25E, 50E) showed increased moduli compared to the 0S (Figure 4c) indicating strengthening of the dough. In fact, the G' and G'' curves of the erythritol-containing doughs resembled those of the 25S and 50S formulations, indicating that erythritol behaved differently from sucrose in the dough.

Table 3. Physical and chemical properties of erythritol, maltitol and sucrose by Mitchell et al. 2008

	Erythritol	Maltitol	Sucrose
Carbon (n°)	4	12	12
Molecular weight	122	344	342
Viscosity	Very low	Medium	Low
Hygroscopicity	Very low	Medium	Medium
Solubility (% w/w at 25°C)	37	60	67

It seemed that erythritol did not affect the gluten elasticity. This could be due to the lower hygroscopicity and water solubility of erythritol than sucrose and maltitol (Table 4), which would cause it to extract less water from the gluten.

From a technological point of view, the appropriate dough elasticity is relevant to avoid dough shrinkage before cutting. When the dough showed little elasticity, shrinkage before cutting was no problem (0S, 25S, 25M); while with more gluten development (for example 100S) the difficulty increases.

3.4. Biscuit texture

The baking process transforms the dough into a cellular solid (Pareyt et al 2008). This transformation implies complex biochemical and physicochemical reactions such as protein denaturation, starch gelatinization, fat melting, Maillard reactions, water evaporation and production and thermal expansion of gases (Chevallier et al. 2002).

Fracture Strength. The force vs. distance curves for the low-sucrose and erythritol and maltitol-containing biscuits are shown in Figures 5a, 5b and 5c,

respectively. In all the curves it can be seen that the force increased up to a point where it drops instantaneously. The distance at the point of break is the resistance of the sample to bend and is related with the fragility of the sample. All the biscuits fractured under tension close to the central zone where the maximum stress occurred.

Figure 5a shows that for biscuits with reduced sucrose content (50S and 100SF biscuits) the distance until fracture increased, meaning more resistance to fracture and higher elastic response. 0S biscuits showed the highest force while the sucrose-reduced biscuits showed decreased fracture strength. 25S biscuits had similar resistance to break as the 0S biscuits but, but with lower fracture force. These findings are in agreement with previous studies (Sai Manohar et al. 1997, Pareyt et al. 2009b) who presented decreased break strength with decreased sugar content.

The present authors demonstrated the competition of sucrose with gluten for the water in earlier experiments with mixtures of gluten and sugar. A gluten network was developed in a mixograph with water. Once sugar was added, the water came out (sugar preference) and the gluten network created was broken, which was also reported by other authors (e.g. Pareyt et al. 2008). One impact of sucrose in the final biscuit is that a weak network is formed and biscuits break easily. As the sucrose quantity decreases, the gluten network becomes stronger resulting in an increasing breaking strength. Also, Maache-Rezzoug et al. (1988) affirms that sucrose disperses protein and also starch molecules, making the biscuit a fragile product. Furthermore, the decrease in breaking force was less abrupt in the sucrose-free biscuits. In agreement with Baltsavias et al. (1999), this indicates that the crack was propagated at smaller velocity caused by more energy dissipation due to plastic deformation for the sucrose-reduced biscuits. Also the study of the gradient reflects this behaviour (figure 5a), with the 0S having the highest gradient followed by 25S > 50S and 100S.

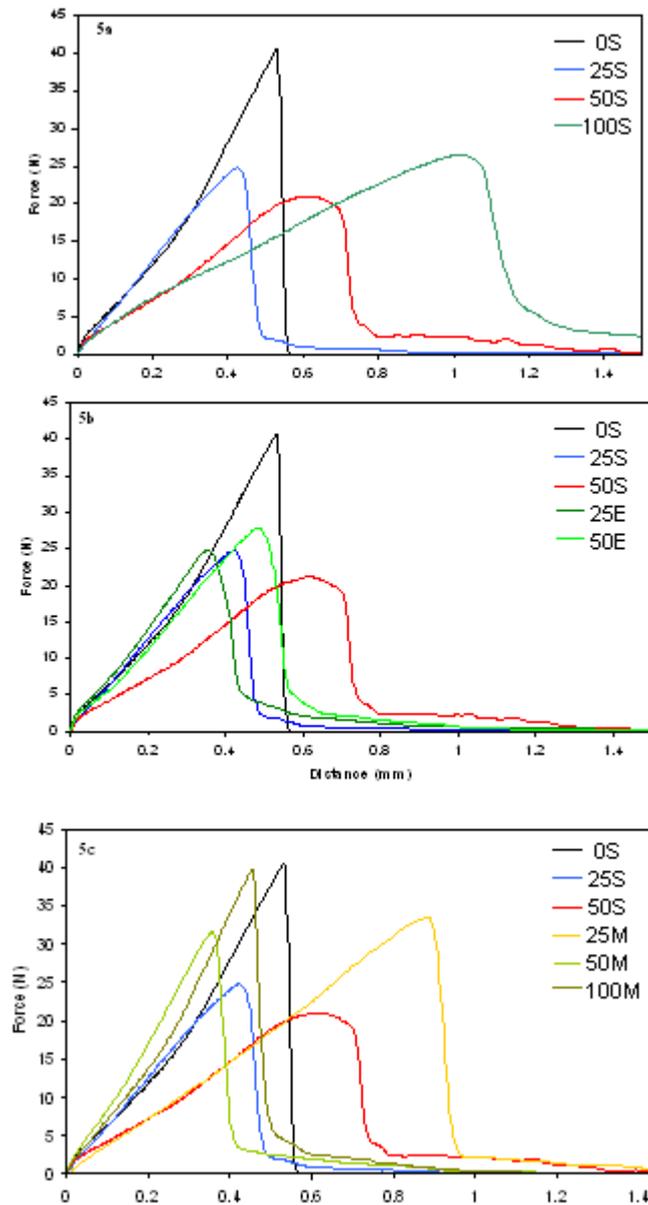


Figure 5. Effect of different sugar levels and replacers on the breaking strength in biscuits. 5a) Different sucrose concentration, 5b) different sucrose concentration and different erythritol levels, 5c) different sucrose concentration and different maltitol levels.

The effect of sucrose replacement by erythritol on biscuit fracture properties is shown in figure 5b. The fracture forces were lower for erythritol-containing samples compared to the 0S. However, the distance at break was not significantly different, implying that the erythritol biscuits (25E and 50E) were as fragile compared to the 0S but without the elastic properties that the absence of sucrose (50S) produced.

Figure 5c presents the force-distance curves of biscuit with maltitol as sucrose replacer. The resulting parameters for these formulas (25M, 50M and 100M) resembled the 0S biscuit parameters more than those of the sugar-reduced biscuit. Zoulias et al. (2000) who used maltitol as sugar replacer, obtained no difference between breaking properties of sucrose biscuits and maltitol biscuits using a cutting blade. In the present work, the 100M biscuits showed similar breaking properties to the 0S, implying that in terms of texture, maltitol was a suitable sucrose mimetic.

Puncture test. In order to obtain biscuit matrix information a puncture test was carried out. The resistance behaviour to the penetration is dependent on the puncturing location (Mandala et al. 2006). Therefore, a total of five penetrations were performed. The number of peaks and the area are presented in figure 6. The highest peak number corresponded to the 0S biscuits while the 100S biscuits showed the lowest peak number. The curves showed irregular profiles with numerous peaks as a result of local fractures of small structures or the layers while the probe passed through the product. Products 100S and 25E having more compact structures with less layering and aeration showed significant less peaks.

The area under the puncture curves (figure 6) is an indication of the resistance of the sample to the semi-spherical penetration. The sucrose addition tends to decrease the biscuit resistance. However when polyols were added, the penetration curve area increased again compared to the sucrose-reduced biscuits. In agreement with previous results, the difference between the 0S and

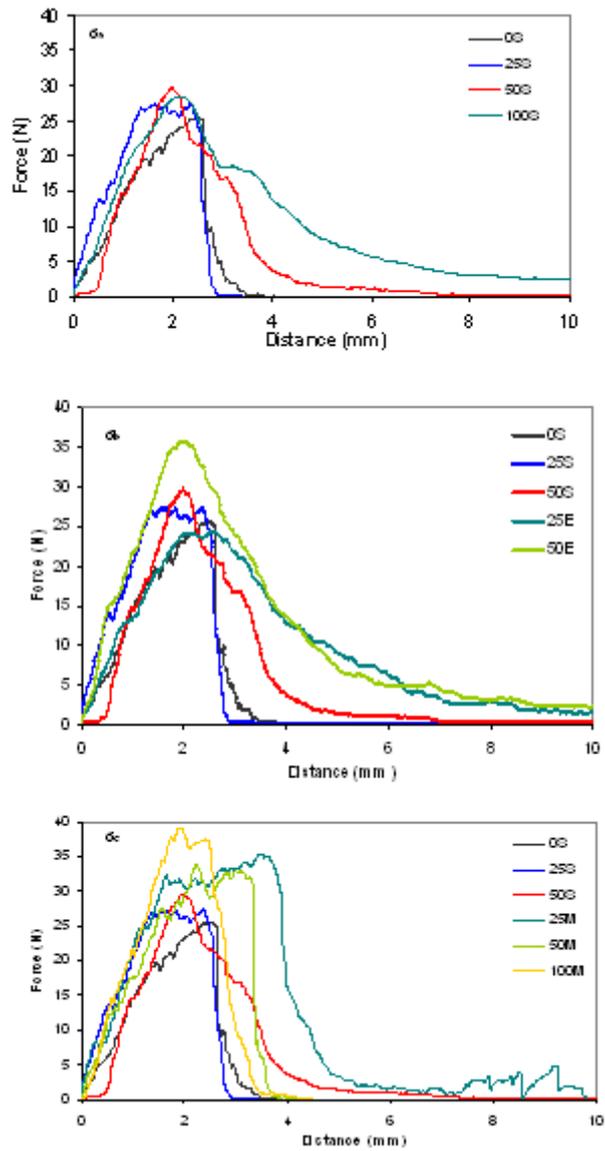


Figure 6. Effect of different sugar levels and replacers on the puncture test in biscuits. 6a) Different sucrose concentration, 6b) different sucrose concentration and different erythritol levels, 6c) different sucrose concentration and different maltitol levels.

the polyol-containing biscuits was the less accentuated when sucrose was replaced by maltitol (25M, 50M, 100M) compared to erythritol (25E, 50E)

3.5. *Biscuit dimensions*

Table 5 presents the biscuit dimensions. Sucrose reduction tended to decrease biscuit diameter (length and width) while increasing the thickness. However, no significant effects of sucrose reduction and sucrose replacement were found on biscuit dimensions. Several authors have previously observed an increase in biscuit diameter (Zoulias 2000, Sai Manohar 1997) and decrease in height (Pareyt 2009b) with increasing sucrose content. Zoulias et al. (2000) also found no changes when using maltitol as sucrose replacement. Billaux et al. 1991 related the baking spread with the viscosity. As the solubility of maltitol in water is similar to that of sucrose it is expected that the dough will flow similarly resulting in equal dimensions.

Table 5. Effect of sucrose reduce and replacement in biscuit dimensions

	Length (mm)	Width (mm)	Thickness (mm)
0S	6.89 ^a (0.05)	6.85 ^a (0.02)	8.43 ^{ab} (0.32)
25S	6.63 ^a (0.09)	6.63 ^a (0.09)	8.80 ^{ab} (0.14)
50S	6.42 ^a (0.01)	6.52 ^a (0.1)	8.45 ^{ab} (0.07)
100S	6.11 ^a (0.02)	6.09 ^a (0.01)	9.45 ^b (0.49)
25E	6.60 ^a (0.10)	6.62 ^a (0.17)	8.55 ^{ab} (0.07)
50E	6.53 ^a (0.09)	6.55 ^a (0.07)	7.80 ^a (0.42)
25M	6.89 ^a (0.07)	6.87 ^a (0.04)	8.45 ^{ab} (0.21)
50M	6.86 ^a (0.01)	6.80 ^a (0.09)	8.50 ^{ab} (0.14)
100M	6.77 ^a (0.02)	6.76 ^a (0.01)	8.45 ^{ab} (0.21)

Values in parentheses are standard deviations.

Means (N= 20) in the same column with the same letter do not differ significantly ($p < 0.05$) according to the Tukey test.

4. Conclusion

The results described in previous sections indicated that sucrose plays an important role in biscuit production and final biscuit characteristics. The amount and type of sugar replacer used have been shown to be very important factors and the effect of the sugar replacer on the gluten development and its interaction with water seem very important factors to take into account in reformulation with regard to sugar replacement.

The total absence of sucrose increases the gluten network providing elasticity to the dough and the biscuit, which is an undesirable effect in short dough biscuit. Biscuit formulas with high sucrose replacement by erythritol showed higher elasticity than the ones with sucrose or maltitol.

Only complete sucrose replacement was achieved with maltitol (100M), whilst the erythritol can be used up to 50% of sucrose replacement (50E)

Compared to the reference biscuit with about 17% (sugar/flour percentage) sucrose (0S) the complete replacement of sucrose by maltitol (100M) was not significantly different in breaking strength. Comparing 50S, 50M and 50E, the biscuit with 50% sucrose replacement with erythritol (50E) showed the largest reduction in texture quality, i.e fracture behaviour.

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RESUMEN Y DISCUSIÓN DE LOS RESULTADOS

RESUMEN Y DISCUSIÓN DE RESULTADOS

La presente tesis doctoral se enmarca dentro del proyecto de la Comisión Interministerial de Ciencia y Tecnología titulado "Reformulación de alimentos por adición de nuevos ingredientes comerciales para disminuir los contenidos en azúcar o grasas. Efectos sobre la reología, microestructura, propiedades sensoriales y aceptación". El proyecto se centra en el estudio de las interacciones entre ingredientes y los cambios estructurales que introducen los nuevos ingredientes, así como su efecto sobre la calidad y la aceptación del producto final por parte del consumidor. El proyecto aborda el estudio de dos modelos de matrices alimentarias: uno de baja humedad y otro de alta humedad.

En particular, la presente tesis doctoral se centra en el estudio de galletas de masa corta, seleccionadas como el modelo de baja humedad. Se estudia el efecto de la incorporación de fibra, el reemplazo de azúcar y el reemplazo de grasa utilizando nuevos ingredientes.

A pesar de que, actualmente en el mercado, el precio de los sustitutos utilizados elevaría el precio del producto, se esperaría compensar el incremento del precio con el beneficio obtenido al disminuir el contenido calórico y el aumento del contenido en fibra.

Los dos primeros objetivos que se abordaron se basaron en la sustitución de la harina presente en la formulación de las galletas por concentraciones crecientes de almidón resistente (AR) como fuente de fibra. El estudio de la masa reveló una mayor densidad de la misma con la adición de AR y una mayor resistencia al corte. La dureza de la masa también aumentó con la adición de AR como se mostró mediante ensayos de penetración y compresión.

El estudio reológico de las masas mostró la estructura típica de un gel débil, con valores mayores de G' respecto a G'' . Una sustitución del 20% de harina por AR mostró un comportamiento estructural similar a la galleta control según los

ensayos oscilatorios de viscoelasticidad lineal, mientras que sustituciones del 40 y 60% de harina por AR presentaron un comportamiento diferenciado con valores de G' y G'' significativamente mayores, aunque en ningún caso se afectó $\tan\delta$, es decir la relación entre la componente viscosa y la elástica. Los resultados de los ensayos de fluencia-relajación mostraron cómo la adición de AR aumentó la elasticidad y disminuyó la deformabilidad de la masa. Al igual que $\tan\delta$, el % de recuperación no se alteró por la adición de AR lo que implica que la incorporación del AR no provocó un cambio en el tipo de estructura, sino un efecto concentrador de los elementos estructurales. La deformabilidad de la masa se correlacionó positivamente con el aumento en las dimensiones de la galleta durante el horneado, es decir las masas menos elásticas fueron más deformables y dieron lugar a galletas de mayores dimensiones.

En la galleta, la adición de AR proporcionó galletas menos deformables y menos rígidas que la galleta control (sin reemplazo de harina por AR). También se observó cambios en el color al adicionar AR aumentando el valor de luminosidad. La aceptabilidad de las galletas con AR por los consumidores fue buena, así para niveles de reemplazo de la harina de hasta el 40% no se encontraron diferencias significativas con el control. Para niveles superiores de reemplazo (60%), disminuyó la aceptabilidad y se afectó negativamente la decisión de compra.

La cantidad diaria recomendada de fibra se encuentra entre 25-30g/día. La galleta consumida comúnmente en España aporta 2,1g fibra/100g de producto, mientras que la galleta más alta con almidón resistente contiene 15,4g fibra/100g de producto. Evidentemente, el consumo de galletas puede no ser diario y puede ser puntual, además, no se esperaría alcanzar los 100g de producto al día, únicamente, sino unos 30-50g; por lo que con el consumo de galletas con almidón resistente se aumentaría unos 7g de fibra a la cantidad diaria (23% de la CDR) para aquellos consumidores habituales.

Por otro lado, se estudió la sustitución de harina en un 5 y un 10% por fibra de manzana y por fibra de trigo de dos longitudes.

El estudio reológico de la masa mostró que la adición de todas las fibras a la concentración del 10% aumentó significativamente los módulos de elasticidad (G') y viscosidad (G").

En las galletas se observó que las fibras de trigo proporcionaron un olor y sabor neutro a la galleta, sin embargo, al hidratarse y crear una matriz más compacta (como mostraron los ensayos de penetración e imagen), la resistencia a la rotura frente a la galleta control aumentó y disminuyó la cantidad de picos de sonido. La resistencia a la rotura y el sonido producido con la adición de fibra de manzana resultó ser similar a la galleta control aunque esta fibra confirió un aroma y sabor afrutado.

La trayectoria oral de las galletas con fibra de manzana a dos niveles diferentes de grasa se estudió mediante la técnica sensorial dinámica llamada "Predominio Temporal de las Sensaciones", y se relacionó con las preferencias de los consumidores. Como resultado se concluyó que la primera sensación dominante al comer una galleta fue la dureza seguida de los términos crujiente/crocante, arenoso, seco en boca, pastoso y sensación grasa. Por otra parte, los consumidores penalizaron las galletas excesivamente duras y las que proporcionaron mayor sensación de sequedad bucal, que resultaron ser las altas en fibra y bajas en grasa; además, estas galletas se percibieron como las de menor carácter crujiente.

Otro de los objetivos planteados fue la sustitución de grasa por tres ingredientes diferentes: ingrediente mezcla de dextrinas (N-Dulge), inulina de alto peso molecular e hidroxipropilmetilcelulosa (HPMC).

En todas las sustituciones se observó un aumento de la fuerza de rotura de las galletas. Este efecto se vio mermado, en el caso de sustitución de la grasa por dextrinas, con la adición de AR como sustituto parcial de la harina.

La fuerza máxima de penetración aumentó con la sustitución de grasa, presentando las galletas con inulina una mayor dureza respecto a la galleta control, así como mayor intensidad del sonido emitido. Por otra parte, aunque

instrumentalmente las galletas con un 15% de inulina fueron más sonoras que la galleta control, este hecho no se vio reflejado en el análisis sensorial descriptivo, donde si que se registraron cambios en el aroma, sabor y color de las diferentes formulaciones con dextrinas y HPMC. Cuando se utilizó dextrinas y AR, el panel de entrenados halló diferencias significativas para la dureza, carácter crujiente y friabilidad de las galletas bajas en grasa respecto a la galleta control.

Los consumidores encontraron aceptables las galletas en las que la grasa había sido reemplazada por inulina o HPMC en un 15%, no resultando aceptables porcentajes mayores de sustitución (30%). Sin embargo, en las galletas con reemplazo de grasa por dextrinas y harina por AR (a niveles de 10 y 20% respectivamente) no se encontraron diferencias significativas con la galleta control.

Finalmente, la sustitución de sacarosa por inulina y eritritol también fue estudiada desde un punto de vista sensorial e instrumental. Un panel de jueces entrenados encontró diez atributos significativamente diferentes respecto a la apariencia externa/interna, la textura, el sonido al masticar, la sensación de humedad y el sabor. Ambos sustitutos produjeron galletas menos crujientes y con menor aceptabilidad por parte del consumidor, aunque la aceptación global de galletas con concentraciones de inulina del 25% no difirió significativamente de la galleta control. El atributo sensorial más correlacionado con la aceptabilidad global de los consumidores fue la aireación de la matriz. Los datos instrumentales reflejaron que el reemplazo parcial de sacarosa por eritritol disminuyó el carácter crujiente de las galletas y aumentó la dureza respecto a la galleta control, mientras que las galletas con reemplazo de sacarosa por inulina dieron galletas más blandas y con características sonoras más similares a la galleta control. Las características de textura instrumental se correlacionaron bien con el análisis sensorial descriptivo, en concreto, se vio que conforme aumentaban los eventos de sonido instrumentales los jueces clasificaban las galletas con más sonido al morder/masticar.

Para obtener un mayor conocimiento de la función de la sacarosa en las galletas de masa corta se realizó un estudio de las propiedades térmicas, reológicas y de textura tanto de la masa como de la galleta.

De los diferentes componentes de las galletas, el gluten fue uno de los más afectados por la presencia de diferentes azúcares (sacarosa, eritritol y maltitol). El maltitol y la sacarosa confirieron a la masa y a la galleta propiedades físicas similares, mientras que los resultados obtenidos con el eritritol fueron similares a los obtenidos en ausencia de azúcar, dando lugar a galletas más compactas, elásticas y con mayor fuerza de rotura en comparación con las galletas control con azúcar o con las galletas con maltitol como sustituto de sacarosa.

CONCLUSIONES

Conclusiones

Las conclusiones principales que se extraen de la presente tesis son:

- Las propiedades reológicas de la masa confieren información estructural de utilidad para la predicción del comportamiento de la masa durante las distintas etapas de la fabricación de las galletas.
- Las propiedades mecánicas y el sonido emitido durante la fractura de la galleta son parámetros fundamentales determinantes de la calidad de la galleta y están relacionados con la aceptabilidad por parte del consumidor.
- El uso de técnicas de calorimetría diferencial proporcionan información de la estructura molecular de los ingredientes de la galleta y explican su funcionalidad.
- La utilización del análisis sensorial descriptivo-cuantitativo es de gran utilidad en la reformulación de galletas, ya que permite un conocimiento global y completo de los atributos que determinan la calidad de las mismas.
- Las nuevas técnicas de análisis sensorial dinámico como el predominio temporal de las sensaciones muestran como el orden e intensidad de aparición de los atributos característicos de las galletas durante el proceso de masticación.
- El aumento de la fuerza de rotura instrumental de las galletas, es decir su dureza, es inversamente proporcional a la aceptación de las mismas.
- El conocimiento de la funcionalidad de los ingredientes de las galletas permite la elección de sustitutos que disminuyen el aporte energético o aumentan su valor nutricional con el

mínimo efecto sobre la calidad sensorial del producto y su aceptabilidad.

- El almidón resistente es un ingrediente que permite incrementar muy significativamente el contenido en fibra de las galletas, ya que se puede incorporar a altas concentraciones sin devaluación de la calidad.

- El almidón resistente confiere rigidez y mayor resistencia a la deformación de la masa, sin embargo, tras el horneado de la masa las galletas obtenidas son más blandas debido a un mayor contenido en humedad y a una menor estructuración de la matriz de la galleta.

- Las propiedades de la fibra utilizada en la sustitución de la harina influye en las características finales de la galleta.

- La adición de fibra de manzana no modifica las características texturales de la galleta confiriéndoles un sabor y aroma afrutado. Por el contrario, las fibras de trigo no modifican el sabor y el aroma pero si las características de textura y sonido de las galletas.

- El reemplazo de grasa por un ingrediente alto en dextrinas aumenta la dureza de la galleta provocando una disminución en la aceptabilidad sensorial, sin embargo, la combinación de las dextrinas con almidón resistente disminuye la dureza y mejora la aceptabilidad que se iguala a la de la galleta control con toda la grasa.

- La inulina utilizada como reemplazante del 15% de grasa aumenta el carácter crujiente de la galleta siendo más apreciada que la galleta control por los consumidores.

- El uso de la hidroxipropilmetilcelulosa para reemplazar grasa afecta a las propiedades mecánicas y da lugar a galletas más duras y con mayor emisión de sonido, que son aceptables hasta un nivel de reemplazo del 15%.
- El estudio de la funcionalidad de los azúcares en galletas muestra que azúcares similares en estructura química como son la sacarosa y el maltitol, proporcionan galletas con características similares.
- El eritritol es un sustituto de sacarosa conveniente porque no tiene efecto fermentativo en el intestino humano pero no resulta óptimo en la formulación de galletas ya que proporciona galletas más elásticas, compactas y resistentes a la fractura respecto a la galleta control.
- El uso de inulina como sustituto de sacarosa en la formulación de galletas resulta óptimo en porcentajes iguales o inferiores al 25%.