Magnetic Resonance Imaging 33 (2015) 336–345

Contents lists available at ScienceDirect

Magnetic Resonance Imaging

journal homepage: www.mrijournal.com

MR microscopy for noninvasive detection of water distribution during soaking and cooking in the common bean

Urša Mikac [a,*], Ana Sepe [a], Igor Serša [a,b]

[a] Jožef Stefan Institute, Jamova 39, 1000 Ljubljana, Slovenia
[b] Department of Biomedical Engineering, Kyung Hee University, 1 Seocheon-dong, Giheung-gu, Yongin-si, Gyeonggi-do 446–701, Korea

ARTICLE INFO

Article history:
Received 10 April 2013
Revised 17 September 2014
Accepted 8 December 2014

Keywords:
Common bean (*Phaseolus vulgaris*)
Soaking
Cooking
Magnetic resonance microscopy

ABSTRACT

Magnetic resonance microscopy (MRM) was used to study water distribution and mobility in common bean (*Phaseolus vulgaris*) seed during soaking at room temperature (20 °C) and during the cooking of presoaked and dry bean seed in near-boiling water (98 °C). Two complementary MRI methods were used to determine the total water uptake into the seed: the T_2-weighted 3D RARE method, which yielded an increased signal from regions of highly mobile (bulk) water and a suppressed signal from regions of poorly mobile (bound) water; and the 3D SPI method, which yielded an increased signal from regions of water restricted in motion and a suppressed signal from the bulk water regions owing to the short repetition time of the method. Based on these results, it can be concluded that during soaking water enters the bean through the micropyle, migrating below the seed coat. The raphe and hypocotyl are hydrated first, while the cotyledon tissue is hydrated next. It was also observed that the imbibition rate increases with an increasing soaking temperature.

1. Introduction

A variety of legumes play an important role in the human diet. Of legumes, common beans are widely grown and consumed in various regions of the world. They are high in starch, protein and dietary fiber and are an excellent source of minerals and vitamins [1]. Beans can be dried and stored for a very long time if kept in a cool and dry environment.

Most of the legumes used for human food require hydration before cooking to cut down on the amount of cooking time and to reduce the amount of oligosaccharides, which cause flatulence [2]. The hydration of seeds before or during cooking is essential for protein denaturation, starch gelatinization and seed softening. A common procedure for assessing the processing quality of dry beans is to measure their total water uptake, which can be efficiently followed by measuring their weight increase at different hydration times. Different models of hydration introducing various hydration parameters have been proposed based on weight increase [3,4].

The microstructural characteristics of beans during soaking and cooking have been studied by way of a scanning electron microscope [5]. Soaking under different conditions, such as water temperature, relative humidity, and salt concentration, has been studied for a number of bean varieties [6–10]. The effects of atmospheric pressure cooking and high-pressure cooking on the physiochemical and nutritional properties of different bean varieties have been investigated as well [11–13]. In addition, the impact of different storage conditions on bean quality has been examined [14,15].

The various parts of seeds have different water absorption properties. Measuring water distribution in seeds during imbibition and cooking *in situ* is a demanding task. Several methods have been employed to trace water imbibition in seeds, such as tracer measurement using dyes [16], scanning electron microscopy [17,18], neutron beam [19] and X-ray CT [20]. Magnetic resonance imaging (MRI) provides a very efficient and noninvasive way to follow water distribution in seeds. The method can detect both molecular mobility and localization. In addition, it is fast enough to follow changes during imbibition. MRI has been employed to study water uptake in seeds [21–25] and to follow changes during cooking and baking [26–30].

In our study, MRI was used to measure water distribution inside beans during soaking (hydration) and cooking. Two sets of experiments were performed. In the first set, beans were soaked before cooking, while in the second set they were cooked without presoaking. The water distribution in the beans was followed by MRI using two complementary pulse sequences: the T_2-weighted 3D RARE (rapid acquisition with relaxation enhancement) sequence and the 3D SPI (single point imaging) sequence. The T_2-weighted 3D RARE sequence enabled the detection of highly mobile water molecules, i.e., those in

* Corresponding author at: Jožef Stefan Institute, Jamova 39, 1000 Ljubljana, Slovenia. Tel.: +386 1 477 3314; fax: +386 1 477 31 91.
E-mail address: urska.mikac@ijs.si (U. Mikac).

http://dx.doi.org/10.1016/j.mri.2014.12.001
0730-725X/

regions with unrestricted water mobility and therefore relatively long spin–spin (T_2) relaxation time. The 3D SPI sequence enabled the detection of bound water regions, i.e., the regions with restricted water mobility and therefore relatively short T_2 relaxation time. The combination of both imaging methods enabled an efficient monitoring of water distribution and mobility in the bean seeds during soaking and cooking. The SPI method enabled the detection of bound water at low concentrations at the beginning of imbibition, while the RARE method enabled a more precise determination of water distribution in the seed at later stages, when the amount of water was higher and the T_2 relaxation time was longer.

2. Materials and methods

2.1. Beans

Fig. 1 shows a schematic presentation of the bean seed structure and a representative T_2-weighted MR image in an identical slice. The relevant seed structures are: seed coat, plumule, cotyledon, hilum, micropyle and hypocotyl-radicle axis. A seed coat envelops and protects the embryo, which consists of two cotyledons (food storing structures) that are connected with each other along the radicle (primary root), hypocotyl (shoot system) and plumule (first leaves). The point where the bean is attached to its pod is called the hilum (a scar visible at the bean's exterior). At one end of the hilum is a small pore called the micropyle.

Dry common bean seeds (*Phaseolus vulgaris*), a Slovenian autochthon variety "Savinjski sivček" with weight of 0.35 ± 0.03 g and water contents of 8.5 ± 0.5 %, were used in the study. The water content in the seeds was determined by the gravimetric method. The seeds were weighed first, then dried in an oven at 70 °C until completely dry and finally weighed again. The water content was then determined as the ratio of the water mass loss (the difference between the initial and the final mass of the seed) divided by the mass of the completely dry seed.

2.2. MR imaging of beans during soaking and cooking

MRI experiments were performed with an Apollo (TecMag, Houston TX, USA) MRI spectrometer with a superconducting 2.35 T horizontal bore magnet (Oxford Instruments, Abingdon, UK) equipped with micro-imaging accessories (Bruker, Ettlingen, Germany) including radiofrequency (RF) coil inserts of various sizes and micro-imaging gradients with a maximum strength of 250 mT/m. In the MRI experiments, a bean seed was embedded in cotton wool and inserted into a glass tube filled with water. Thus, any eventual bean motion during soaking and cooking was prevented. The tube with the bean seed was then inserted into a 20 mm diameter RF coil, which was in turn inserted into an MR microscopy probe inside the magnet. The probe enabled sample thermoregulation by an air stream of controlled temperature that was blowing on the sample, thus maintaining the sample temperature constant throughout the imaging experiment. Air temperature was controlled by directing an air stream at a constant flow rate into in a glass Dewar tube containing a heater. The heater power was adjusted dynamically by a temperature controller that received temperature readings from a copper-constantan thermocouple sensor inside the RF coil. Such an experimental scheme allowed for a dynamic following of bean soaking and/or cooking and cooling without removing the bean from the magnet.

Two sets of experiments were performed. In the first set, bean seeds were first soaked for 15 h at room temperature, then cooked for 1.5 h and finally cooled in the RF probe for another 3 h. In the second, the seeds were cooked for 3 h (without presoaking) and then cooled for another 3 h. Each type of experiment was repeated three times. The imbibition of water in the bean seed during soaking and cooking was followed by way of sequential MR imaging using the 3D SPI sequence [31,32] and the 3D RARE sequence [33]. The images were acquired every 60 min during the soaking in cold water (approximately 20 °C) and every 20 min during cooking in near-boiling water at 98 ± 1 °C.

The 3D RARE images were acquired with an isotropic resolution of 266 μm, imaging matrix 64 × 64 × 64, inter-echo time $TE = 1.64$ ms, repetition time $TR = 2$ s, turbo factor 64 and scan time 140 s. The ordering of *k*-space lines in imaging using the RARE sequence was sequential, thus the corresponding images were T_2-weighted. The signal intensity S of the RARE sequence is intricate [33], however, it can be estimated by the expression

$$S_{RARE} = \rho \exp\left(-\frac{TE_{eff}}{T_2}\right)\left[1-\exp\left(-\frac{TR}{T_1}\right)\right]. \quad (1)$$

Here ρ is the spin density, TR the repetition time, T_1 and T_2 are the spin–lattice and spin–spin relaxation times, respectively, and TE_{eff} is the effective echo time of the RARE sequence, i.e., the time between the excitation pulse and the echo tagged with a zero phase encode value. The TE_{eff} was 54.12 ms, yielding to T_2-weighting for the sample's regions with short T_2 values. The signal intensity of the SPI sequence is on the other hand given by [32]

$$S_{SPI} = \rho \exp\left(-\frac{t_p}{T_2}\right)\left[\frac{1-\exp\left(-\frac{TR}{T_1}\right)}{1-cos\varphi\cdot\exp\left(-\frac{TR}{T_1}\right)}\right]\sin\varphi, \quad (2)$$

where t_p is the phase encoding time and φ is the RF excitation pulse flip angle. The spatial resolution of the 3D SPI images was equal to 500 μm isotropic. This is almost half of the resolution obtained with the 3D RARE method. The lower resolution of the SPI method was due to the hardware limitations associated with a limited gradient strength and a need for a short encoding time t_p, which was only 0.15 ms. Other parameters of the 3D SPI sequence were: imaging matrix 64 × 64 × 16, RF excitation pulse flip angle $\varphi = 20°$, repetition time $TR = 5$ ms and scan time 330 s. To save imaging

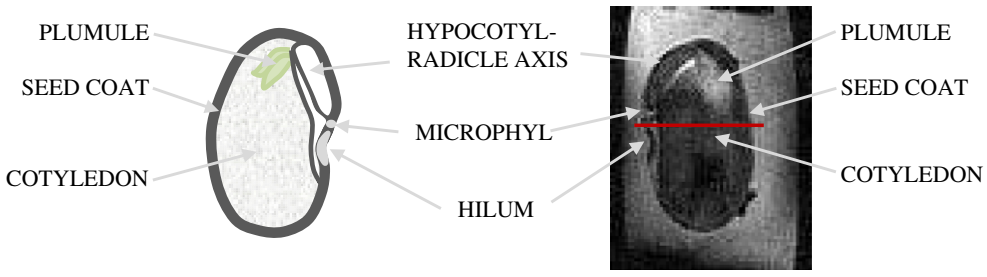

Fig. 1. Bean seed anatomical structures: seed coat, plumule, cotyledon, hilum, micropyle, and hypocotyl-radicle axis. The structures are pointed out by arrows in a drawing (left) and in a representative T_2-weighted image in a transverse slice across the bean seed (right).