Legume crops offer many benefits to agricultural sustainability. Legumes capture atmospheric nitrogen through a symbiotic relationship with soil bacteria in a process called “biological nitrogen fixation.” This process reduces the reliance on synthetic nitrogen fertilizers.

Among legumes, soybeans are a major source of protein and oil. Soybeans are grown in a range of latitudes and environments. One of the challenges to improving soybean productivity is the high demand of nitrogen in comparison to cereals and oilseed crops.

Biological nitrogen fixation can be improved by breeding and selection that focuses on the plant, the nitrogen-fixing bacteria, and better matching plant and bacteria (Photo 1).

**Soybean Nitrogen Fixation and Yield**

Soybean production depends heavily on the total nitrogen uptake supplied by the biological nitrogen fixation process. A strong connection has been documented between plant nitrogen demand and final yield for soybeans. For example, a soybean crop yielding 70 bushels per acre needs a total nitrogen uptake (above ground only) of approximately 320 pounds per acre (Figure 1). Plant nitrogen demand increases with yield at the rate of 100 pounds nitrogen per acre for each 20 bushels per acre. Approximately 50% of the total nitrogen uptake is coming after full-pod stage (R4 stage), increasing the nitrogen need of the crop during the later crop growth stages. From the end-season nitrogen uptake, on average 50–60% of this amount is met by biological nitrogen fixation.

The primary nitrogen sources are: 1) biological nitrogen fixation and 2) inorganic nitrogen coming from the soil reservoir. The nitrogen fixed is assimilated and exported in the xylem as three nitrogen sources: 1) ureides (allantoin and allantoic acids), 2) amino-nitrogen, and 3) nitrate-nitrogen. As nodulation activity increases, the ureide content of the xylem

![Figure 2. Map of the United States referencing all the experimental locations. Colors represent different soybean maturity groups (Tamagno et al., 2018, Scientific Reports Journal).](image)

![Photo 1. a) Soybean plant at V3 (three-leaf) stage with 43 centimeter total length, growing without inorganic nitrogen supply under greenhouse conditions. The seed was inoculated with an inoculant containing a minimum (3.0 x 10^9 colony forming unit mL^-1) of Bradyrhizobium japonicum strain. b) and d) showing roots (and nodules). c) shows the nodule starting its activity, based on its internal coloration.](image)
The presence of a non-fixing crop (check plant). The ureide method is an indirect and point-measurement of biological nitrogen fixation, it is less labor intensive and expensive but needs calibration with isotopic labeled method (15N, for example natural abundance).

**Study Description**

In collaboration with WinField United this study established trials in 23 locations across the Midwest in the 2016 growing season (Figure 2). This study investigated the effect of nitrogen fertilizer application on biological nitrogen fixation and its implications for soybean productivity and protein concentration.

The study characterized the seasonal dynamics of biological nitrogen fixation (via ureide method, Figure 3) and its consequences for soybean productivity with an emphasis on growth and shoot biomass allocation mechanisms.

**Result**

Application of nitrogen at different growth stages affected neither yield nor protein concentration. Overall, the biological nitrogen fixation process increased until the beginning of seed formation and then decreased until harvest (Figure 4).

Nitrogen fertilizer reduced the peak (up to 16%) of biological nitrogen fixation. Biomass allocation to seeds was reduced with increasing biological nitrogen fixation.

**Conclusions**

Application of nitrogen fertilizer at different growth stages affect neither yield nor protein, but reduced the peak (up to 16%) of biological nitrogen fixation. This study highlights the importance of improving biological nitrogen fixation by breeding and selection targeting the plant, the nitrogen-fixing bacteria, and better matching plant and bacteria. This is particularly relevant in areas of the US Midwest where selection for maintenance of biological nitrogen fixation in dry soils is needed to improve soybean productivity under drought and heat stress conditions.

Based on: Tamagno, S., Sadras, V. O., Haegele, J. W., Armstrong, P. R., & Ciampitti, I. A. (2018). Interplay between nitrogen fertilizer and biological nitrogen fixation in soybean: implications on seed yield and biomass allocation. Scientific Reports, 8(1), 17502. [https://doi.org/10.1038/s41598-018-35672-1](https://doi.org/10.1038/s41598-018-35672-1)