Supplemental Table 2. Summary of 25 independent gene edited lines. Eight T1 seeds with GFP fluorescence were pooled for each line for DNA isolation, PCR amplification, and Sanger sequencing. Highlighted indicate lines with gene editing event.

ICE Score, which represents the CRISPR editing efficiency (also known as indel frequency, or the percentage of the cell population that has insertions or deletions).**Knockout Score** (KO Score). The Knockout Score represents the proportion of cells that have either a frameshift-inducing indel or a large indel in a protein-coding region that is 21 bp or greater in length that are likely to generate a complete loss-of-function mutation.

The KO score is a useful metric for researchers that are interested in understanding how many of the contributing indels are likely to result in a functional KO of the targeted gene.

The random nature of the non-homologous end joining (NHEJ) repair pathway that is most commonly used to repair double-strand breaks (DSB) induced by Cas9 generates many different types of indels. Therefore, generation of an indel in the target gene being edited does not necessarily mean that expression of the encoded protein will be knocked out, so measurement of editing efficiency or indel frequency are not the best metrics to evaluate success in CRISPR knockout experiments.

If the goal of your genome engineering work is to generate loss-of-function mutations by knocking out a gene with CRISPR-Cas9, indel analysis using the ICE Knockout Score is the most useful approach.

For a detailed explanation of the CRISPR Data Analysis Scores. Please visit the website

https://www.synthego.com/blog/crispr-knockout-score#ice-score-shows-overall-editing-efficiency

No.	Label	ICE-Score	KO-Score	gRNA1 edited event	Grna2 edited event
1	14-VAR-1-F_R	20	14	yes	yes
2	3-VAR-1-F_R	0	0		
3	1-VAR-1-F	0	0		
4	9-VAR-1-F	0	0		
5	10-VAR-1-F	0	0		
6	20-VAR-1-F_R	26	21	yes	yes
7	11-VAR-1-F_R	0	0		
8	5-VAR-1-F_R	0	0		
9	18-VAR-1-F_R	0	0		
10	19-VAR-1-F	0	0		
11	8-VAR-1-F	0	0		
12	15-VAR-1-F_R	0	0		
13	2-VAR-1-F_R	50	50	yes	yes
14	4-VAR-1-F_R	67	49		
15	12-VAR-1-F	0	0		
16	17-VAR-1-F	0	0		

17	23-VAR-1-F_R	20	16	yes	yes
18	7-VAR-1-F_R	0	0		
19	6-VAR-1-F_R	0	0		
20	16-VAR-1-F	44	41	yes	yes
21	21-VAR-1-F	73	73	yes	yes
22	26-VAR-1-F_R	23	17	yes	yes
23	25-VAR-1-F	26	20	yes	yes
24	24-VAR-1-F_R	57	57	yes	yes
25	22-VAR-1-F_R	0	0		