

How is the safety of genome-edited foods confirmed?

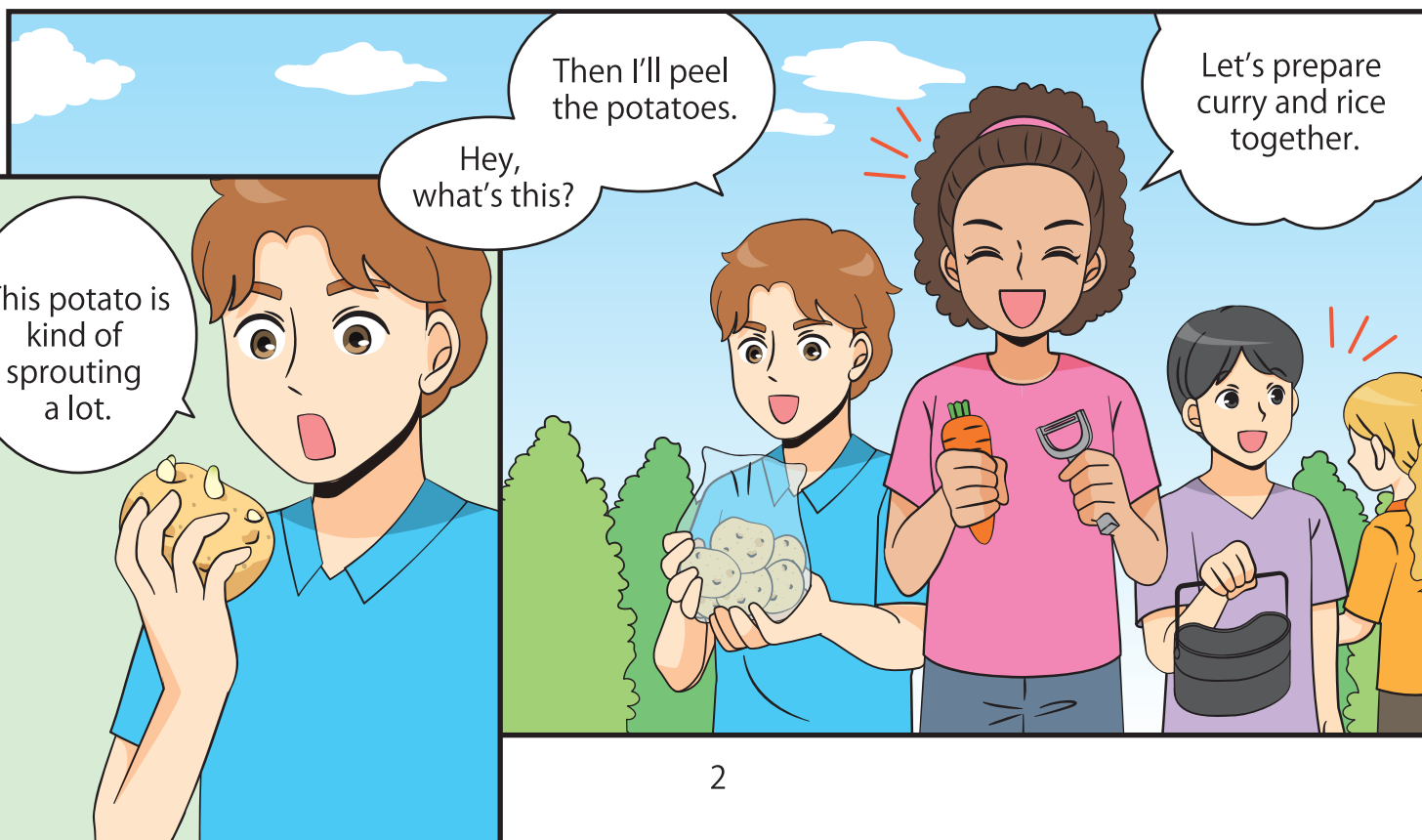
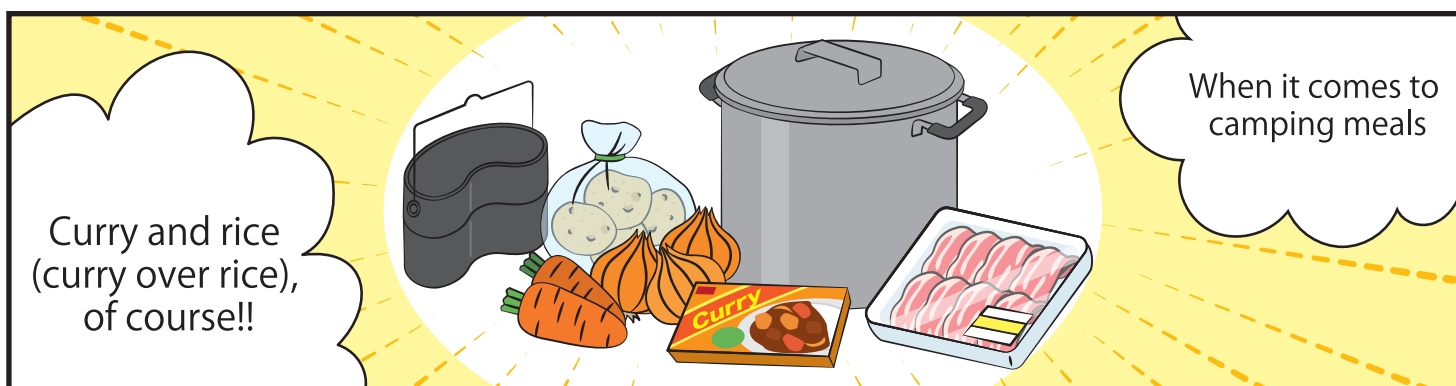
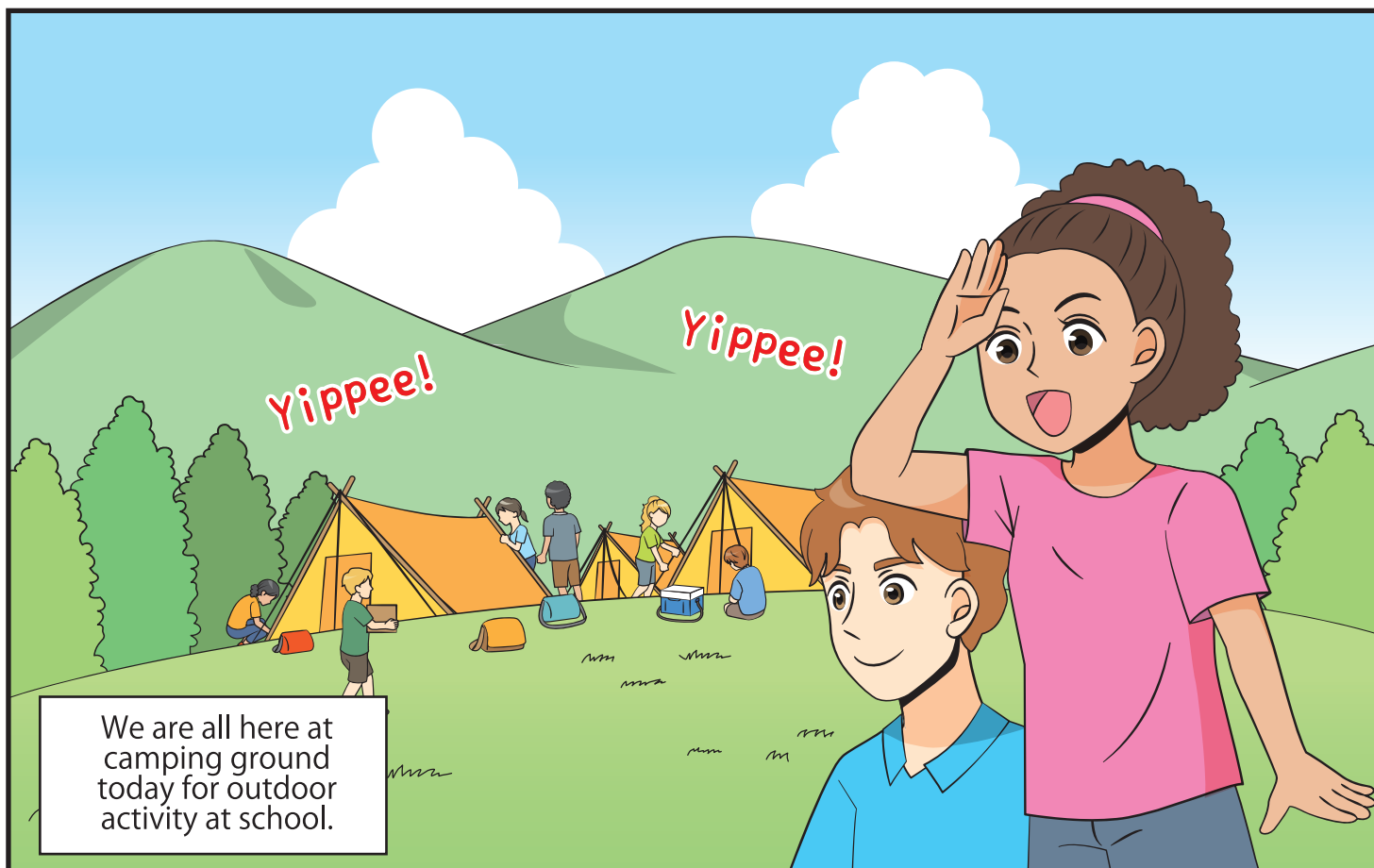
Genome-edited
solanine-free
potatoes

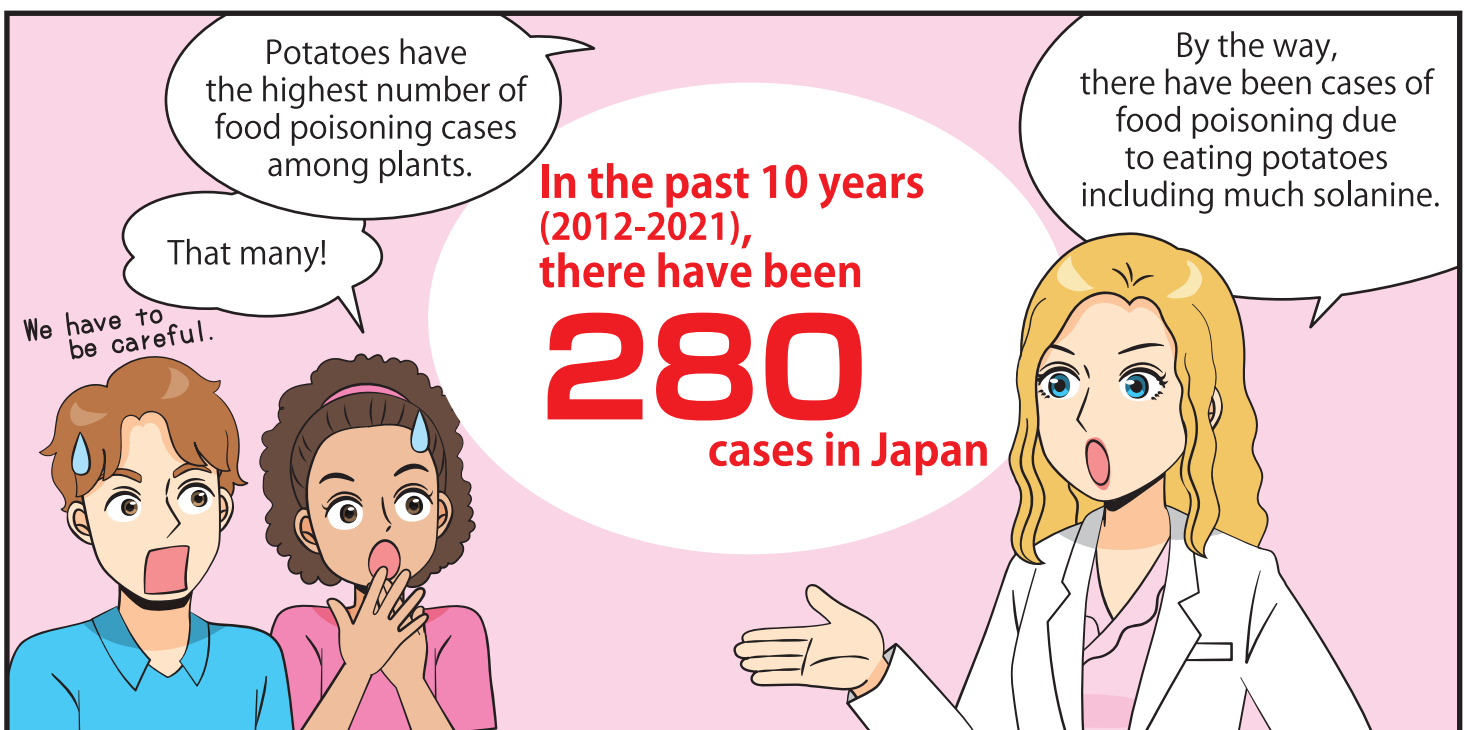
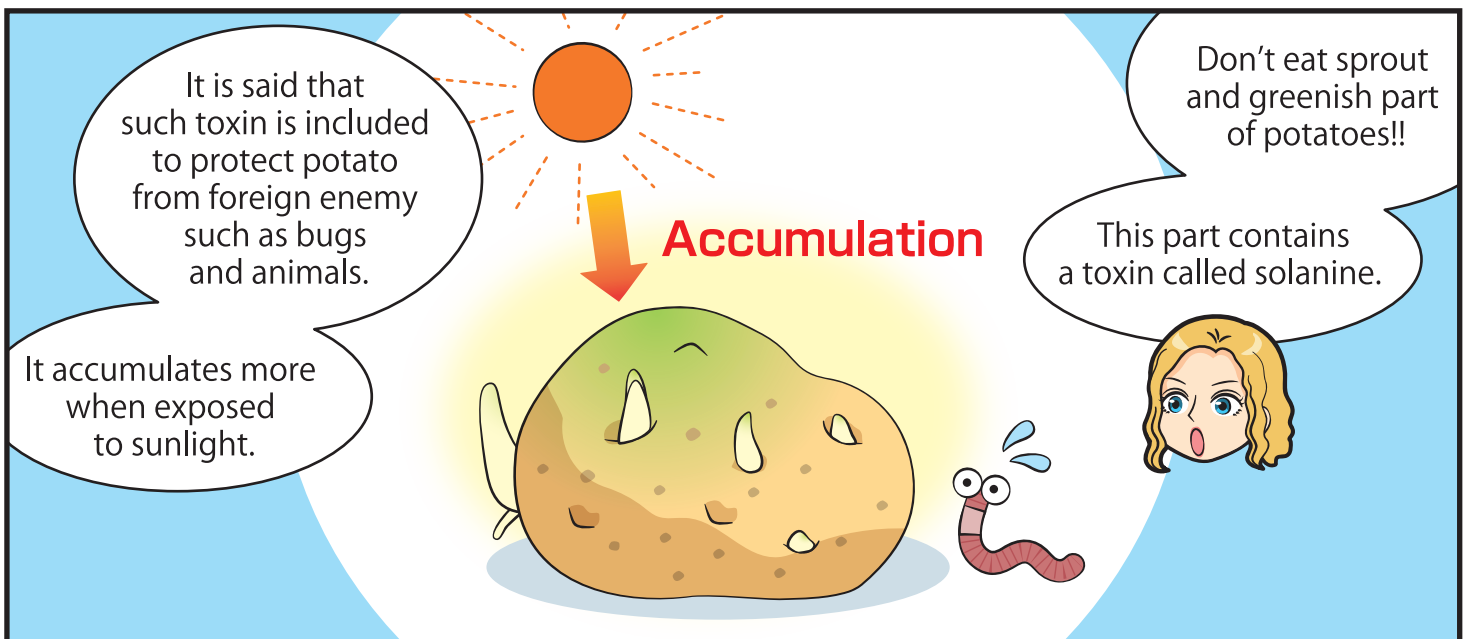
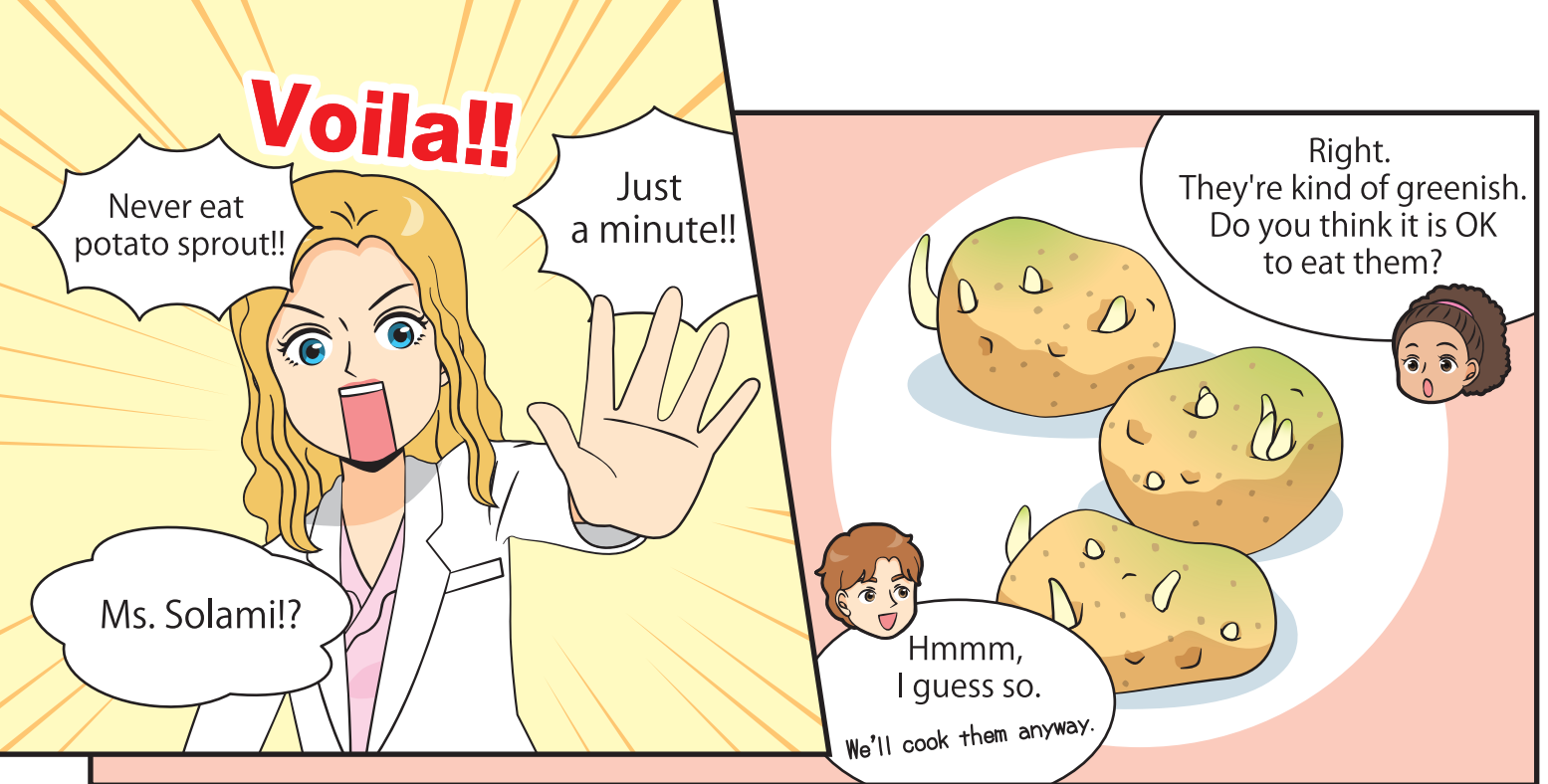


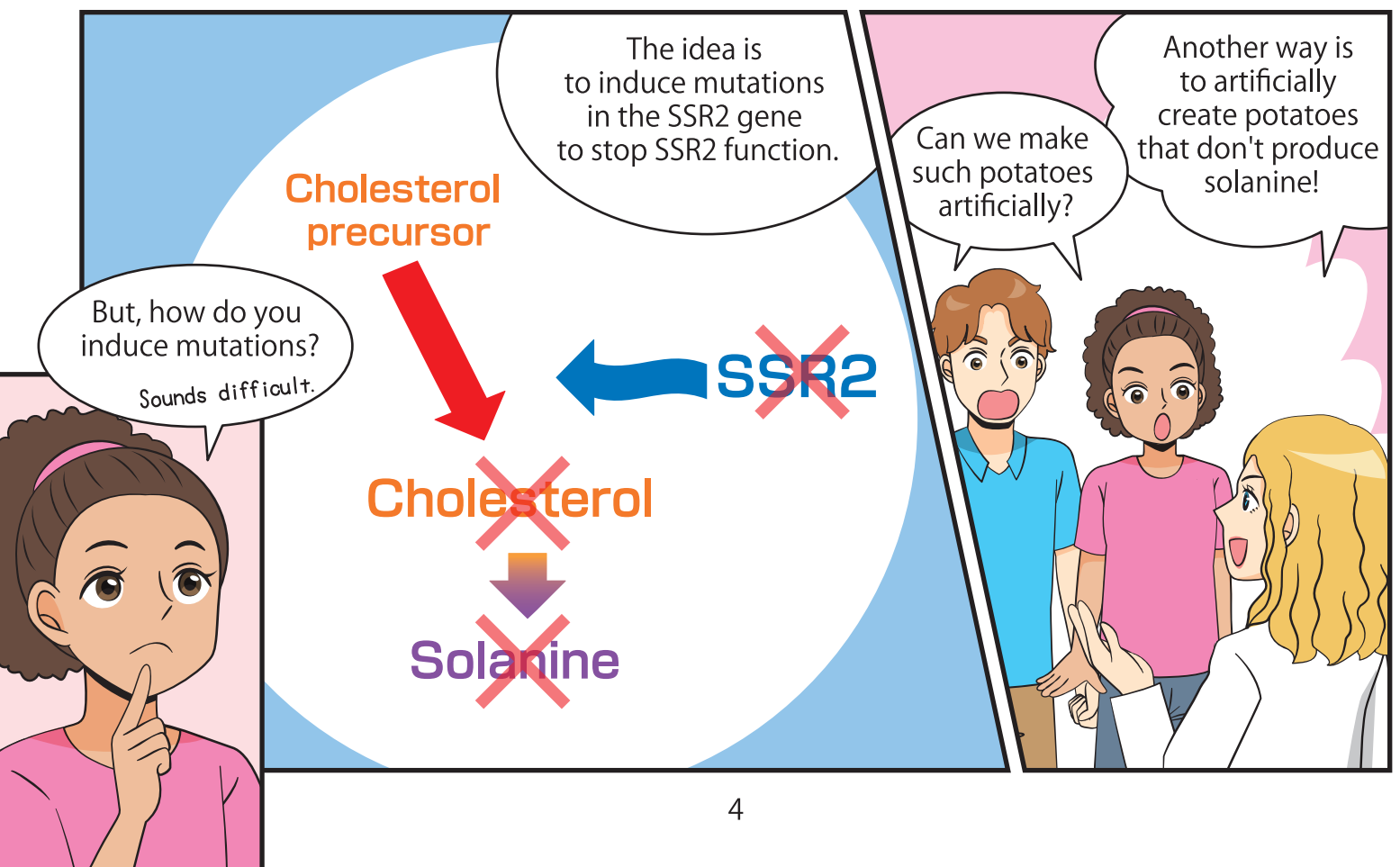
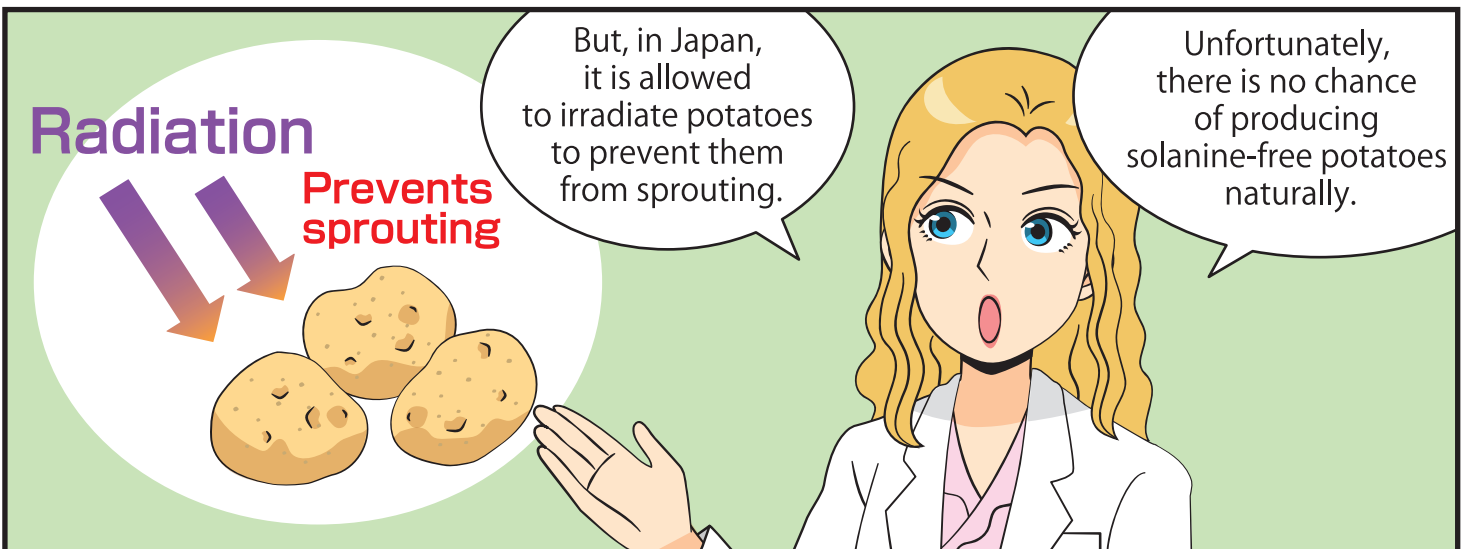
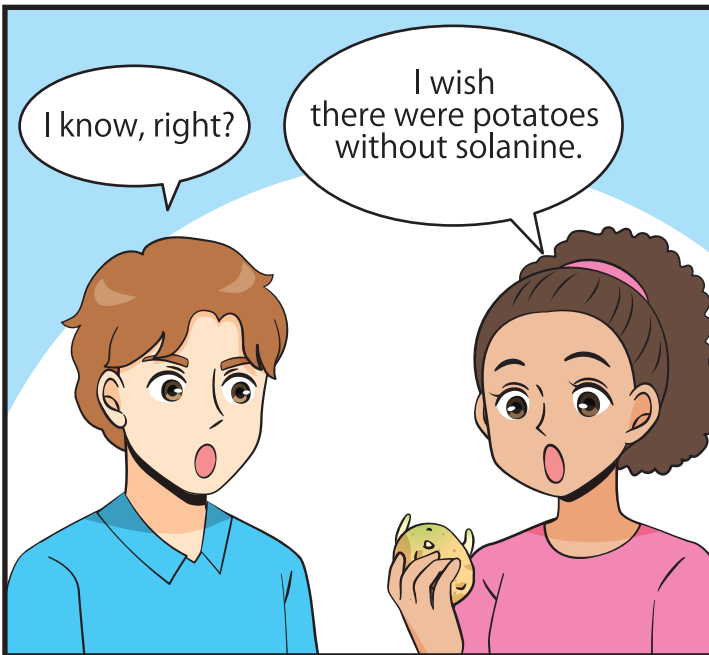
Natl. Inst. Health Sci., Japan

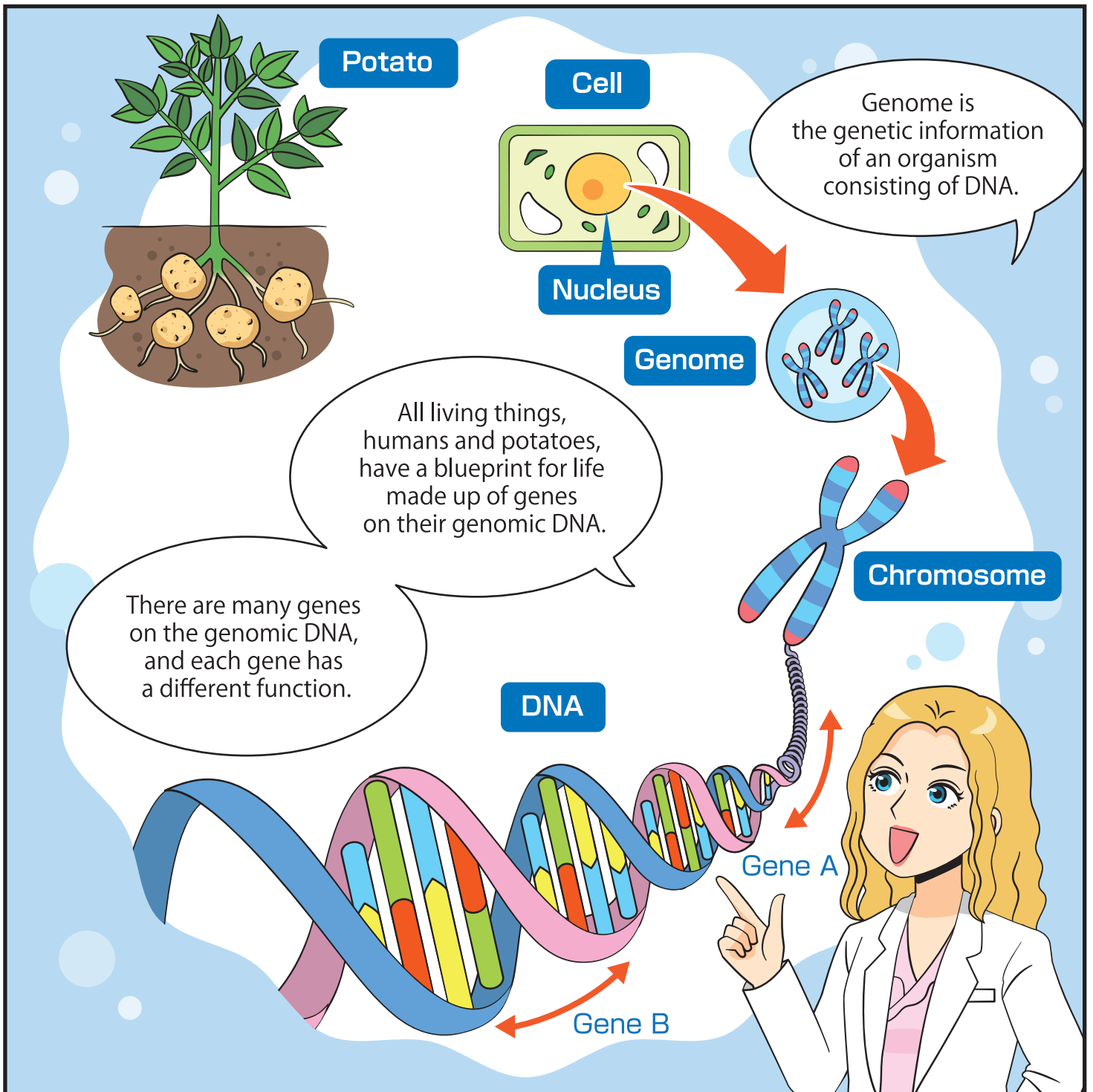
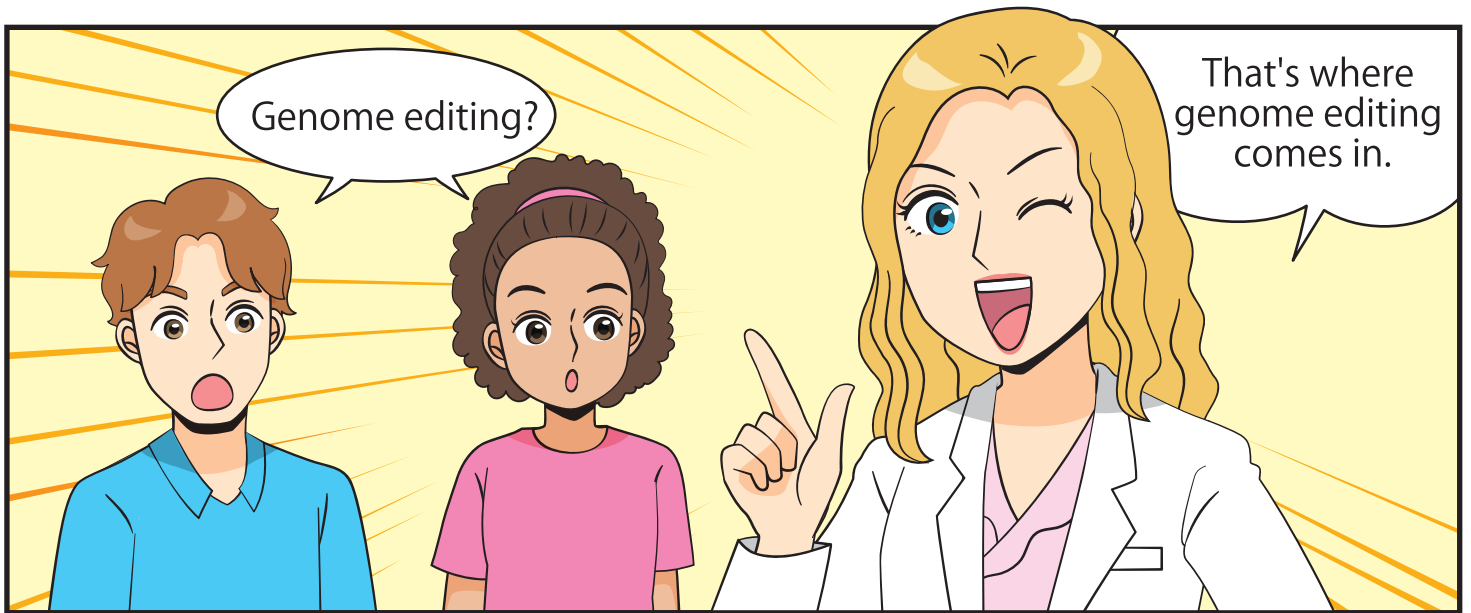
Genome-edited potatoes that do not produce solanine have not been submitted for notification yet. This cartoon was created using the example of the solanine-free potatoes, and assumes how safety is confirmed in the prior consultation phase before notification and distribution.

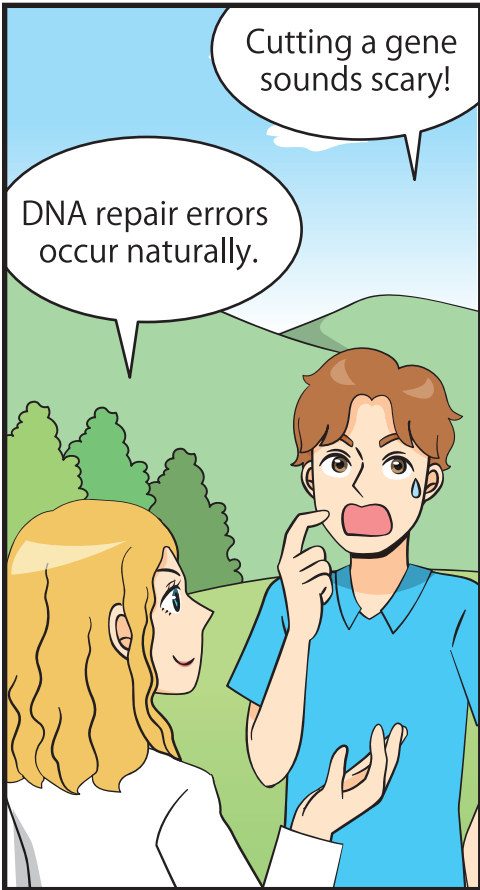
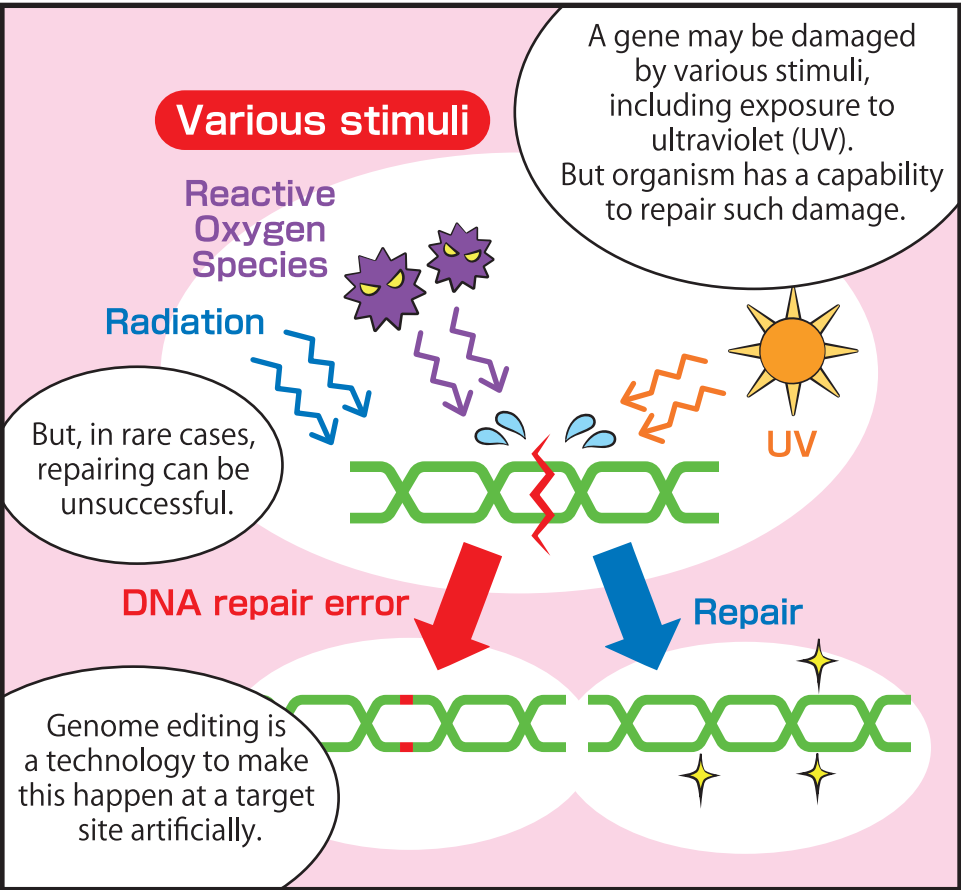
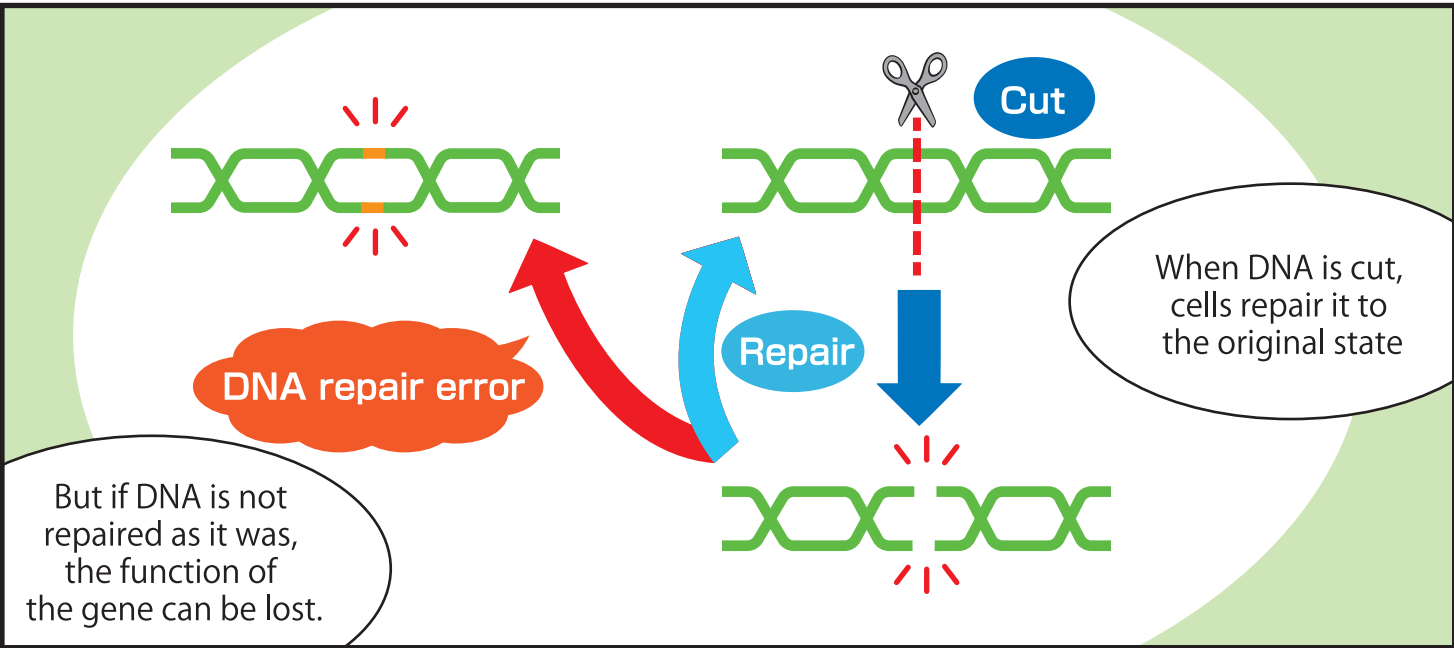
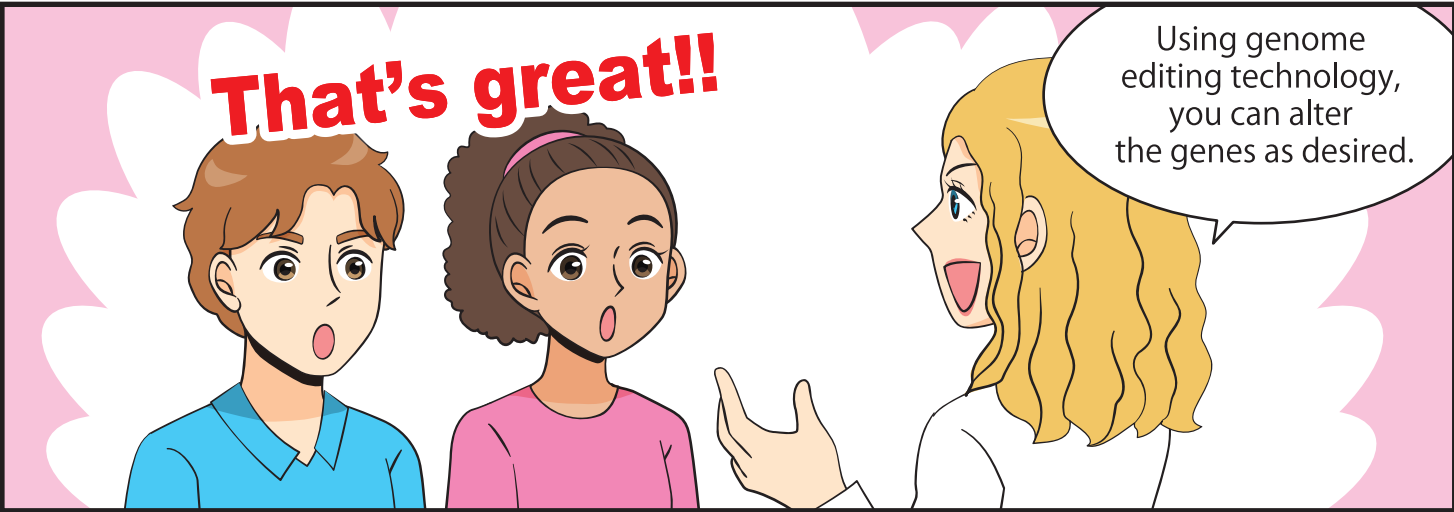
- In Japan, a notification system for genome-edited foods was launched in October 2019. The safety of genome-edited foods is confirmed during the prior consultation phase of the notification system, and a notification will be submitted to and accepted by the Consumer Affairs Agency (CAA) when it is determined that there are no safety concerns and the requirements for notification are met.
- In this MANGA (cartoon), outline of the prior consultation and notification system for the genome-edited foods is introduced.

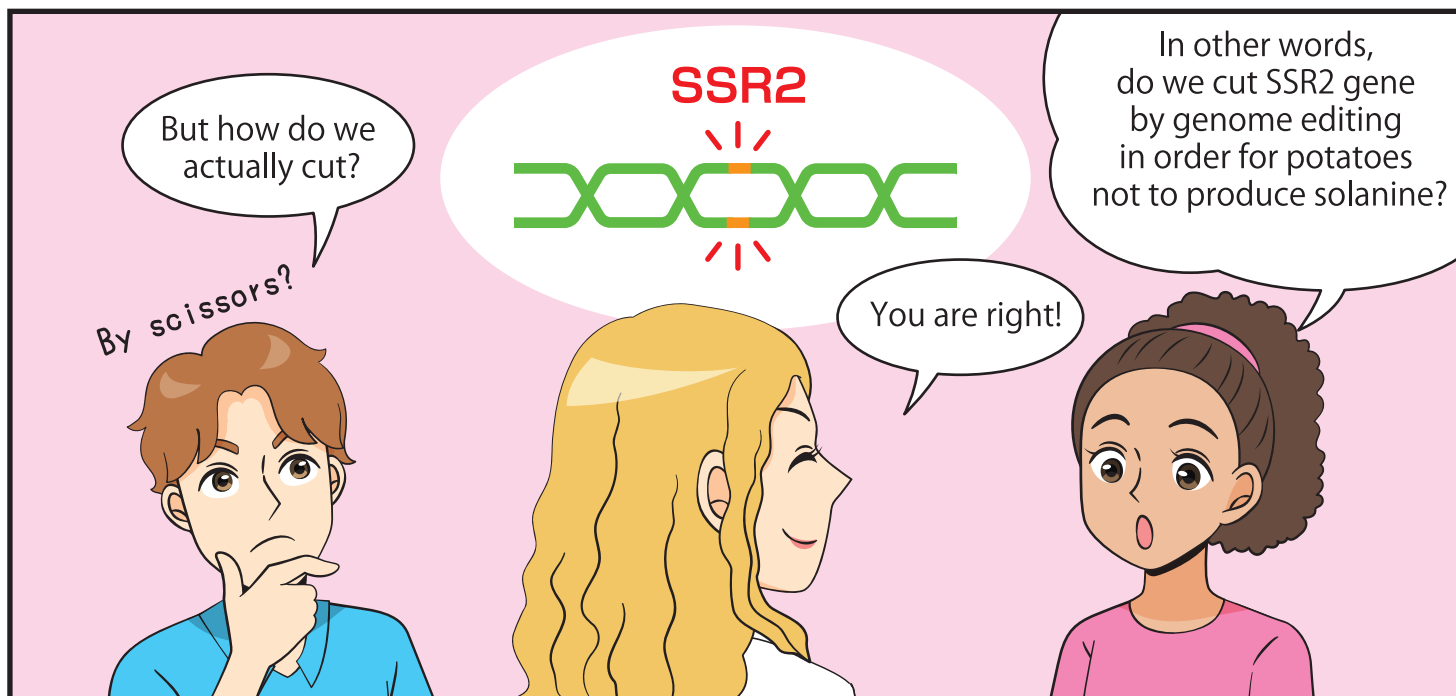






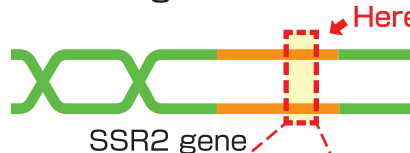






Genome editing procedure

- 1 Decide a location of SSR2 gene where to cut.



- 2 Prepare a molecule that binds to the target location.

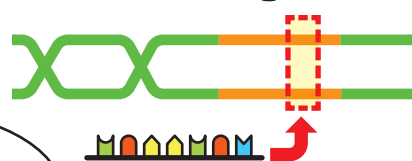


This is the outline of the process.

- 3 Deliver the molecule to the plant to be genome-edited.

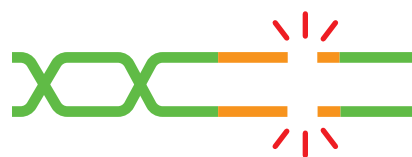


- 4 That molecule binds to the SSR2 gene.

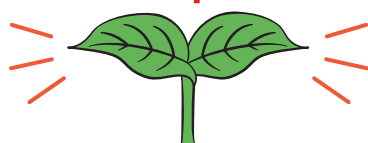


How do you deliver that molecule?

- 5 SSR2 gene is cut.



Genome editing is complete!!



When this Agrobacterium is delivered to potato nursery plants by infection, the molecule binds to the location to be cut.

Infection

Potato nursery plants

Molecule to bind to the site to be cut

Vector (transporter)

We use a soil bacterium, called Agrobacterium, which can deliver a part of DNA sequence to plants.

Agrobacterium

We connect the molecule, to be combined to the location desired to be cut, to a transporter called a vector, and have the Agrobacterium deliver.

Not yet! Now comes the main part.

Now we can harvest potatoes without solanine!!

Is the genome editing now completed?

SSR2

Then, SSR2 gene will be cut.

I see. It is cut like scissors.

Mutated No change Mutated Mutated

We grow the nursery plants after genome editing, check DNA, and select only the individuals to which mutation has occurred to SSR2 genes.

That means, we select the individuals whose gene was not repaired to its original state. Right?

Deletions

Substitution

Insertion

Repair errors

Cut

Repair

We check how the base sequence of SSR2 gene of the selected potatoes has changed.

DNA is composed of four base pairs, A, T, G, and C. The sequence is called a base sequence.

A Adenine G Guanine
T Thymine C Cytosine

DNA

What is "base sequence"?

Change in genes due to the change of base sequence.

Threonine Valine Tyrosine Alanine Arginine Arginine

ACT GTA TAT GCC CGA CGG A
TGA CAT ATA CGG GCT GCC T

Cut

Threonine Valine Tyrosine Alanine Arginine Arginine

ACT GTA TAT GCC CGA CGG A
TGA CAT ATA CGG GCT GCC T

Unsuccessful repair

Threonine Valine Tyrosine Proline Aspartic acid Glycine STOP

ACT GTA TAT CCC GAC GGA TAA
TGA CAT ATA GGG CTG CCT

Missing base pair The sequence shifts forward one by one.

The combination of the three base pairs in a sequence determines one amino acids, and the sequence of the amino acids determines the characteristics of the protein.

If one base is missing due to a repair error, it will be shifted one by one. As a result, the amino acids will be different from the original.

That results in a loss of function of the original gene. Right?

Search tools and databases for toxicity and allergenicity are available online.

Search tools and databases for toxicity and allergenicity are available online.

We will also find out whether a substance causing food allergy is generated or not due to the sequence change.

We should not create different toxins when trying to eliminate solanine. Right?

Whether other toxic substances are generated due to the increase in the amount of cholesterol precursor.

Cholesterol precursor

Phytosterol

Cholesterol

Solanine

Whether SSR2 gene is involved in generating toxic substance due to the loss of its function.

SSR2

We will also check whether toxic substances, other than solanine, are generated or not due to the sequence change.

Yes, understood.

Furthermore, we examine if any change has occurred at a location other than SSR2 gene.

At a location other than the cut location:

Are there same sequences?

Is there 1 base different sequence?

Is there 2 bases different sequence?

We examine the sequences in the potato genome that are similar to the targeted sequence in advance. If there are similar sequences, we will find out to see if they have changed before and after genome editing.

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After deletion

Environment with high nutrition is suitable for Agrobacterium to grow.

Confirm that Agrobacterium does not increase.

Environment including antibiotic is not suitable for Agrobacterium to grow.

Agrobacterium is deleted.

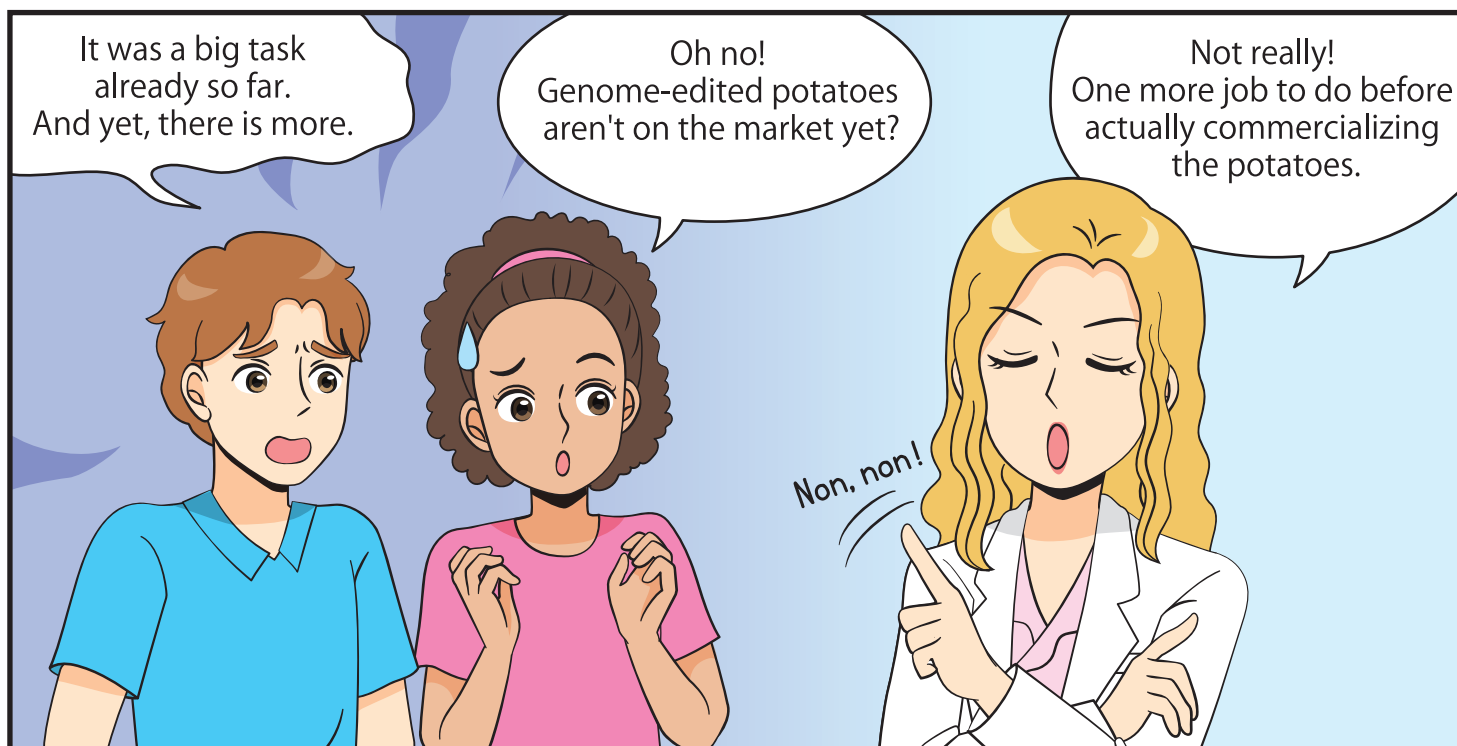
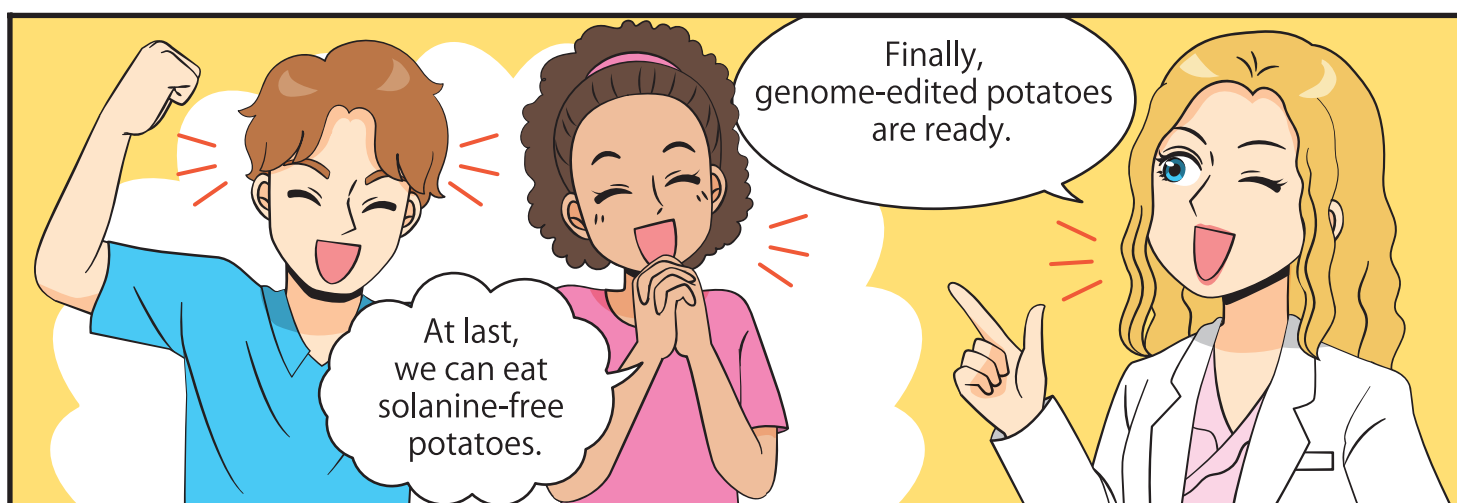
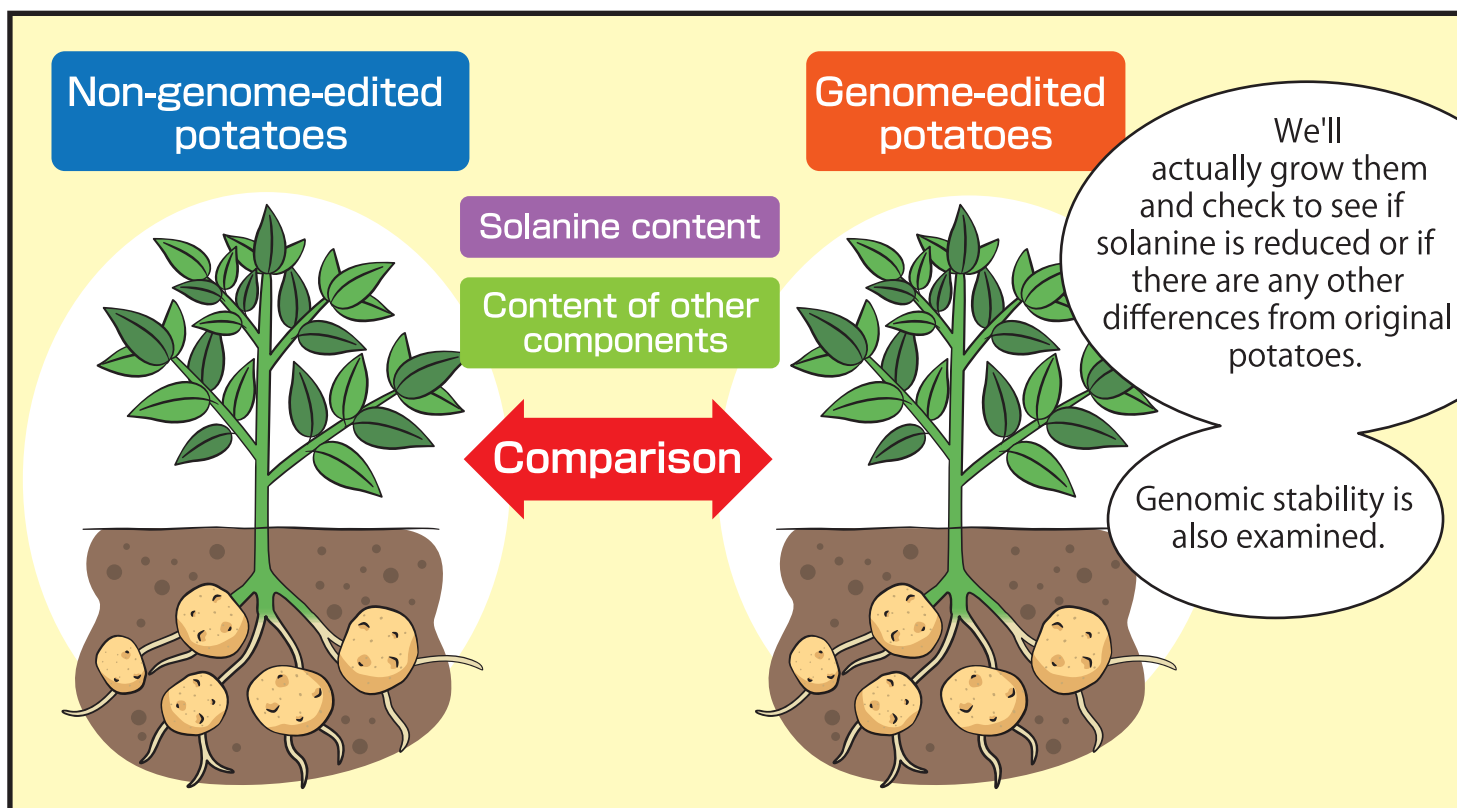
We will further check if Agrobacterium is deleted from the nursery plant.

There are so many things to be done, aren't they?

Right...

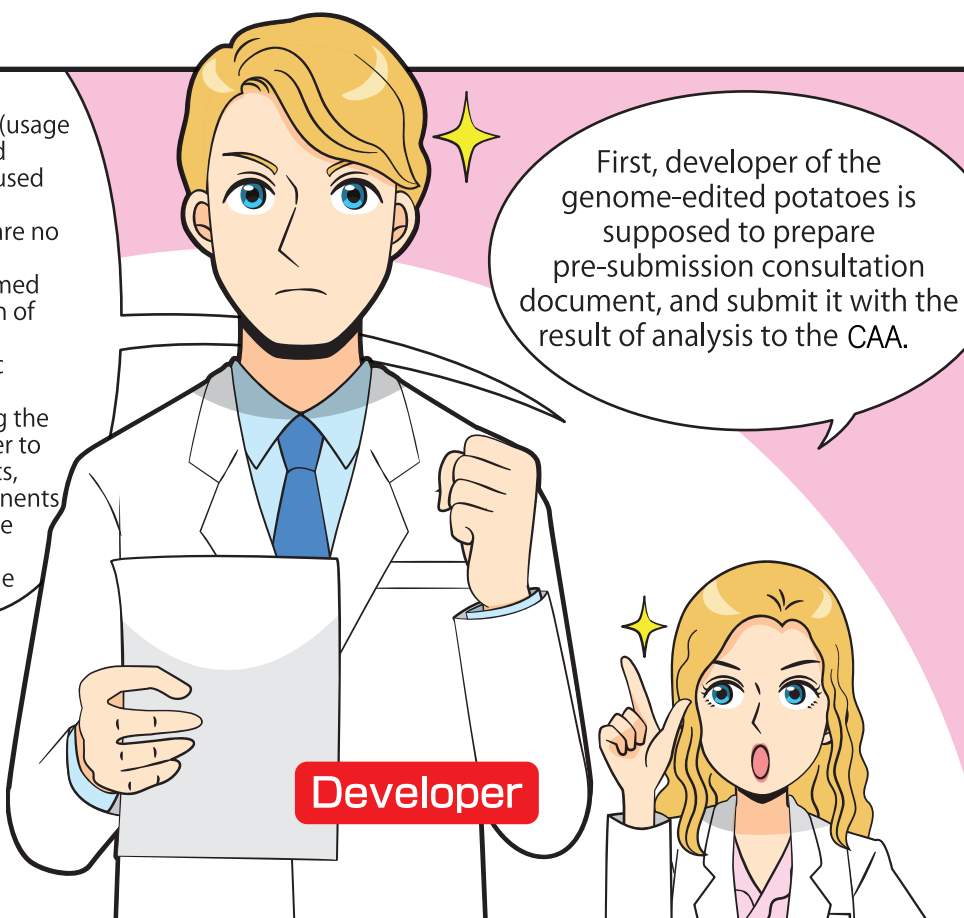
PCR method	It is checked to see if the vector sequence has not increased. (a small sequence is checked)
Southern blotting method	It is checked to see that DNA of a vector is not detected. (a large fragment is checked)
Analysis based on phenotypes.	It is checked to see that a characteristic, indicated when a vector is included, is not observed.
Next generation sequencer	It reads whole genome sequence to see if vector sequences exist in the genome.

We will find out the absence or presence of the vector sequences used.



Content;

- (1) Names of item and breed and summary (usage and intended use) of the developed food
- (2) Method of genome editing technology used and details of modification
- (3) Information on confirmation that there are no remaining foreign genes or their parts
- (4) Information on confirmation that confirmed changes in DNA do not cause production of new allergens having adverse effects on human health or increase of known toxic substances contained
- (5) For items in which modification affecting the metabolic system was performed in order to increase or decrease specific components, information on changes in major components (nutrient components only) related to the target metabolic system
- (6) Year and month of marketing (*Notify the MHLW of it after marketing)



First, developer of the genome-edited potatoes is supposed to prepare pre-submission consultation document, and submit it with the result of analysis to the CAA.

Developer

Wait a minute!

Too many!!

That's enough!

Examples of the material showing the result of analysis are;

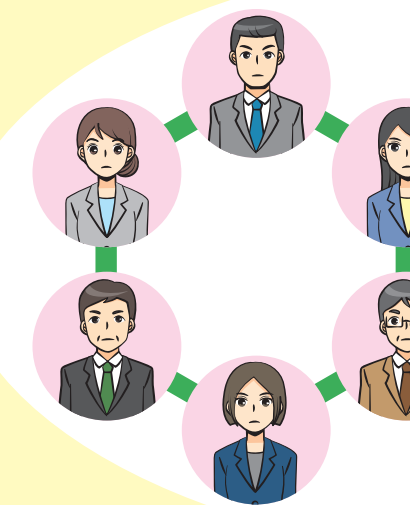
- The result of analysis investigating how SSR2 gene sequence has changed (Example: Electrophoretic image file)
- Result of a search for existence/non-existence of a sequence similar to the one at the designated location.
- Result of a search to see if a new protein is not generated.
- Result of a study to see if allergenicity and toxicity do not exist.
- Result of determination of solanine content etc.
- Details of each measurement and analytical method. And so on.

**Pharmaceutical Affairs[※]
and Food Sanitation Council**


Consumer Affairs Agency

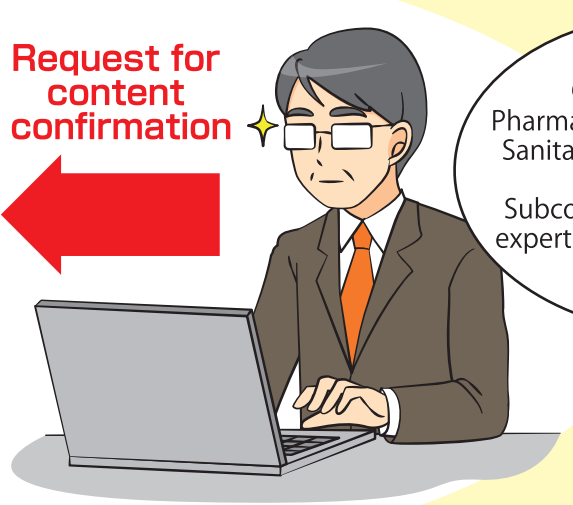
After checking the document,

CAA will request the Pharmaceutical Affairs and Food Sanitation Council (hereinafter referred to as "the Subcommittee"), consisting of experts, to check the content of the document.

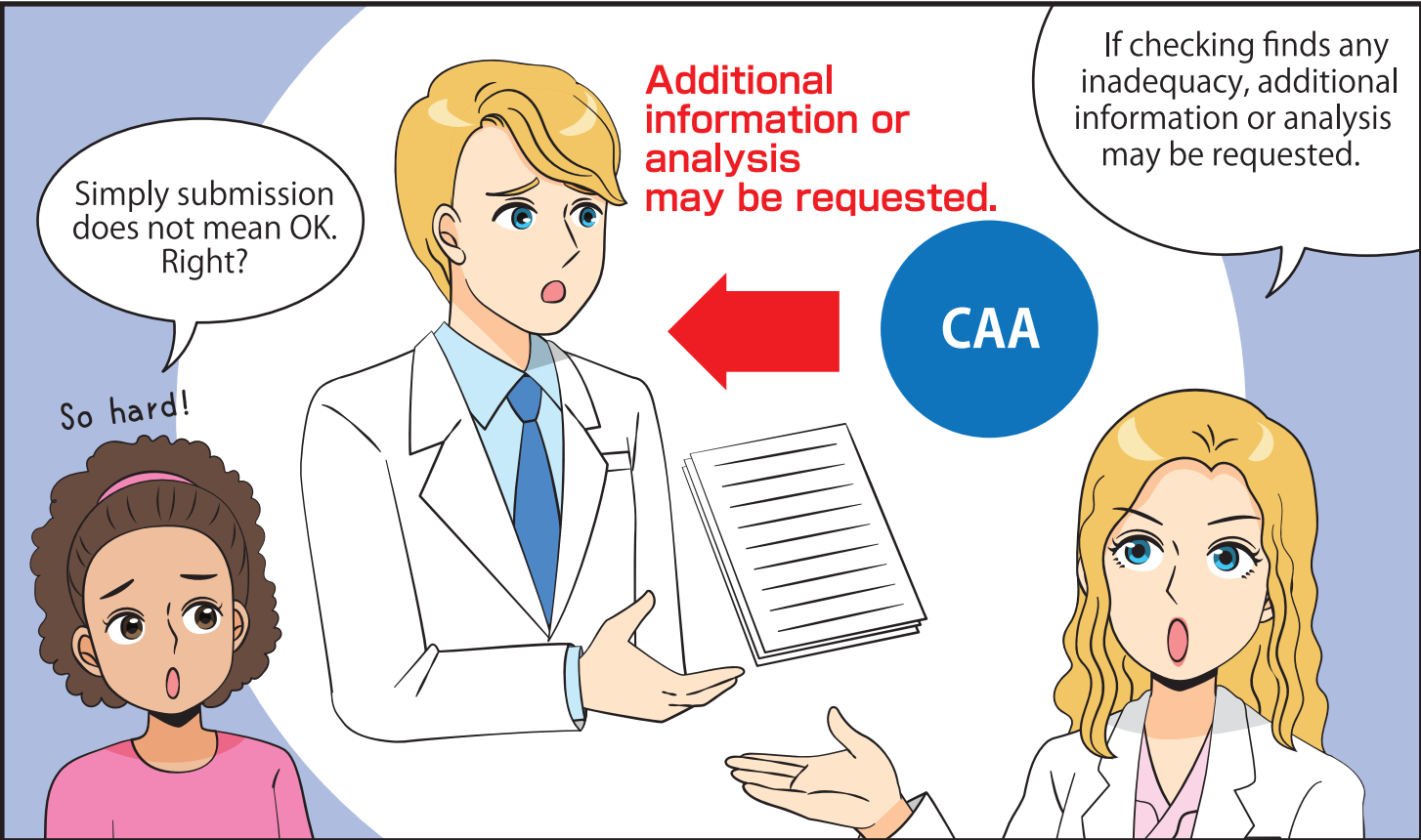
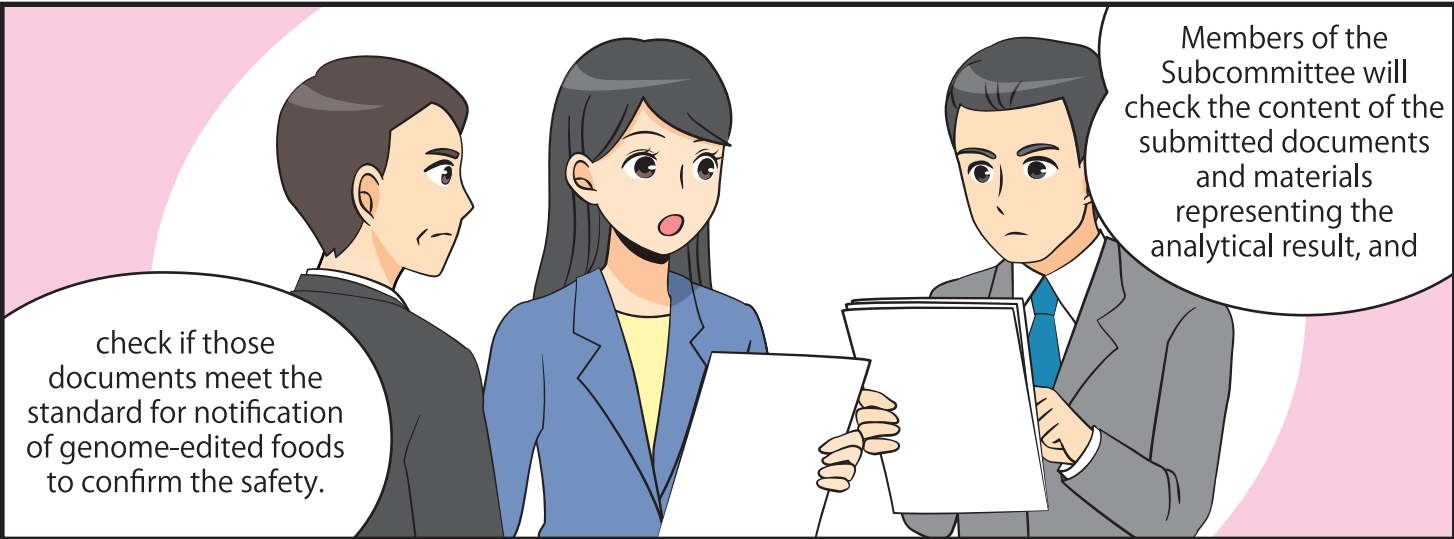


Request for content confirmation





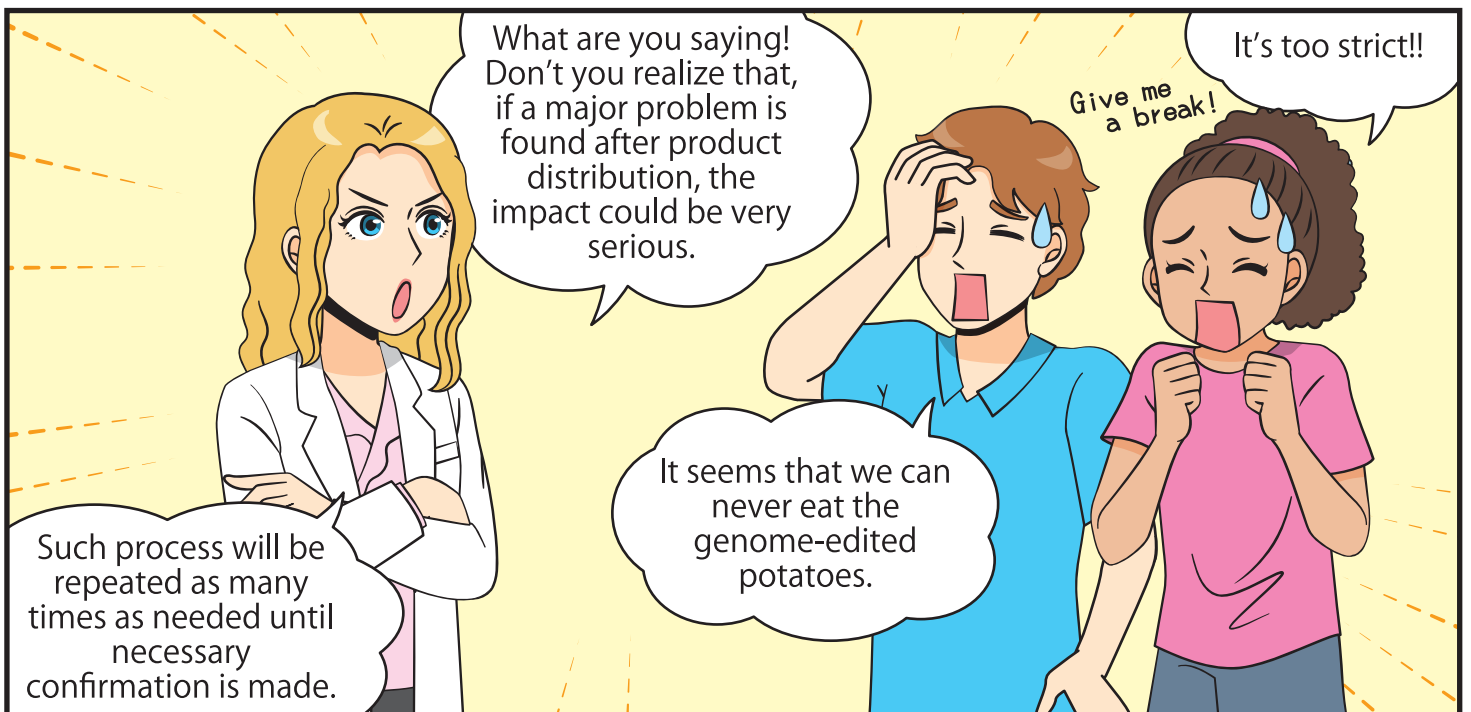
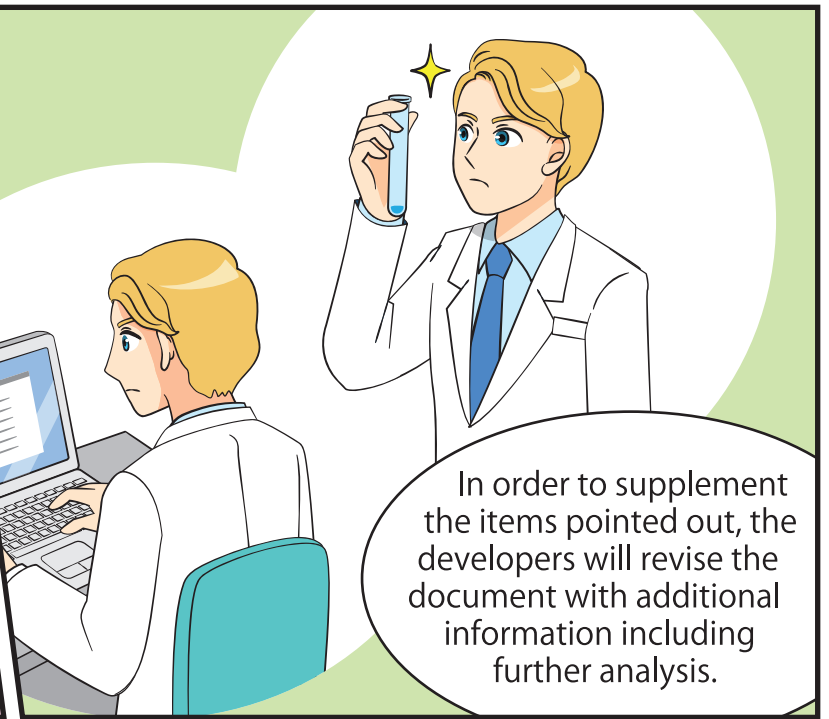
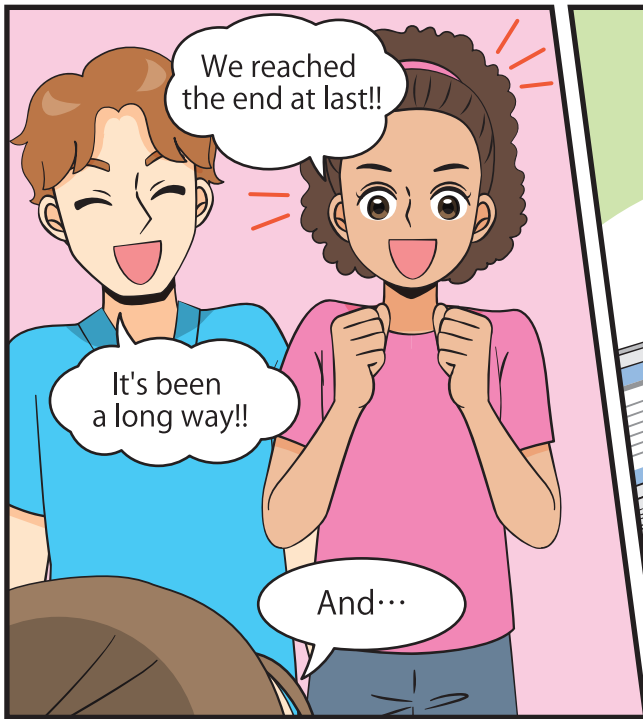
※Subcommittee on Genetically Modified Foods, Newly Developed Food Committee of the Food Sanitation Council

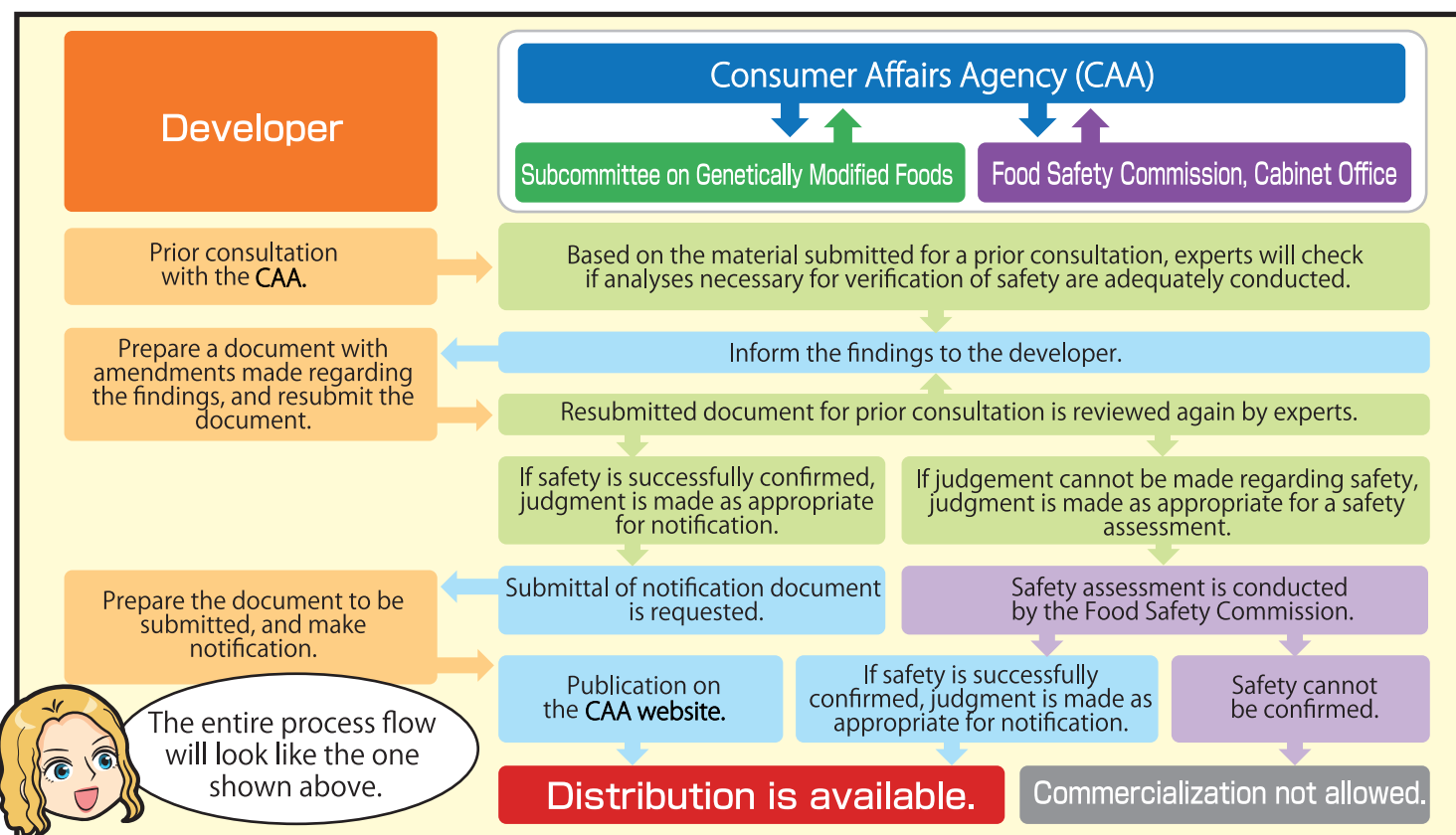


Content of the application document	Items pointed out by the Subcommittee
<p>Tool necessary for genome editing (molecule to be combined to the location desired to be cut) was inserted into the cell of ○○species potatoes by Agrobacterium method, and mutation was observed in SSR2 gene.</p> <p>Cut location is □□region of SSR2 gene. When genome editing was performed on individuals of △pieces, mutation of missing 1 base was observed on individuals of ◇pieces, and reduction of solanine content was confirmed.</p>	<p>•What kind of vector was used for genome editing?</p> <p>•Where on SSR2 gene was modified? Please show us the location in an appropriate figure?</p> <p>•How was the solanine content measured? How many individuals were investigated, and how were the measurements? Based on the database, were the changes within the variable range under the cultivation conditions?</p>
<p>Because a method to express target protein transiently by Agrobacterium is used, foreign genes are not inserted into the potato genome. The fact that foreign genes did not remain was confirmed by PCR method, Southern Blotting method and analysis based on phenotype.</p>	<p>•Was the position of the primer appropriate in PCR method? Did the primers cover the sequence to be analyzed? Could you also submit the picture of electrophoresis?</p> <p>•Where was the probe position for analysis of Southern hybridization analysis? Was the target fragment detected? Please show us the result.</p>

Hey! It is till so many!!

Table on the left shows the points to be checked.





If solanine-free potatoes are distributed in the market, concern for food poisoning can be reduced!

First, I was a little bit scared about cutting genes. But now I am comfortable about eating.

I did not know that such a severe review is made before commercialization.

It's great to know that safety is adequately confirmed, and yet good taste and nutrition stay the same as the conventional potatoes.

By the way ~

Oh no!

I completely forgot about it.

I will do it right away.

What about potatoes peeling?

We've been waiting for a long time.

Potatos in Curry Rice



END