

# Influence of Twendee X (TwX) on Gut Microbiota and Daily Life

Questionnaire survey by Eyez, Inc. (Tokyo, Japan)

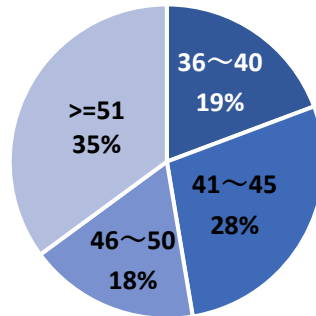
Stool microbiota analysis by Takara Bio Inc. (Shiga, Japan)

## Participant Attributes

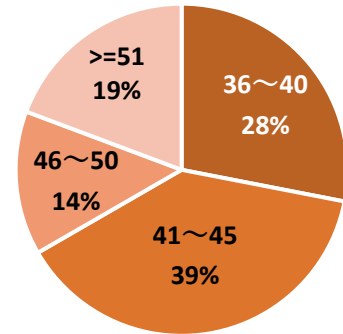
### Age distribution

Age	Male	Female
36~40	11	16
41~45	16	22
46~50	10	8
>=51	20	11

**Age distribution  
Male**



**Age distribution  
Female**



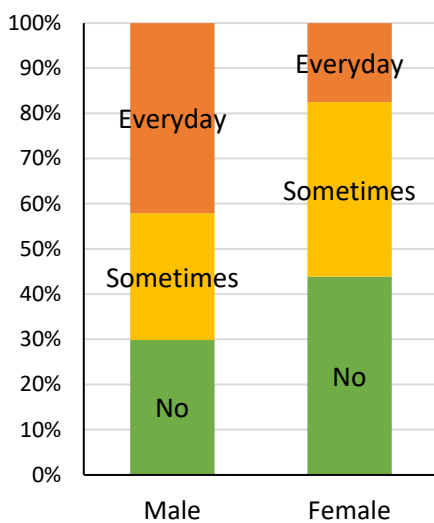
### Q. Do you drink alcohol?

	Male	Female
Everyday	24	10
Sometimes	16	22
No	17	25

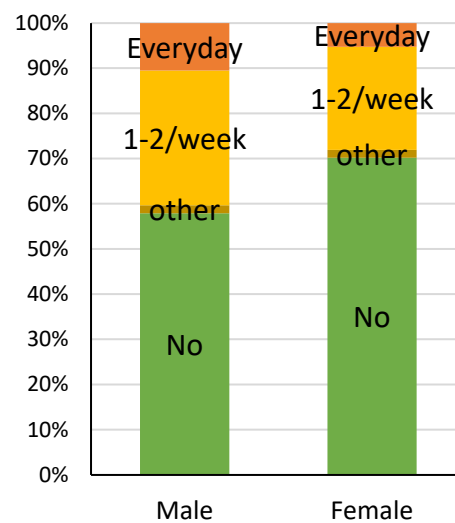
### Q. Do you exercise regularly?

	Male	Female
Everyday	6	3
1-2/week	17	13
No	33	40
Other	1	1

### Drinking



### Exercise



Comment(s) on "Other":

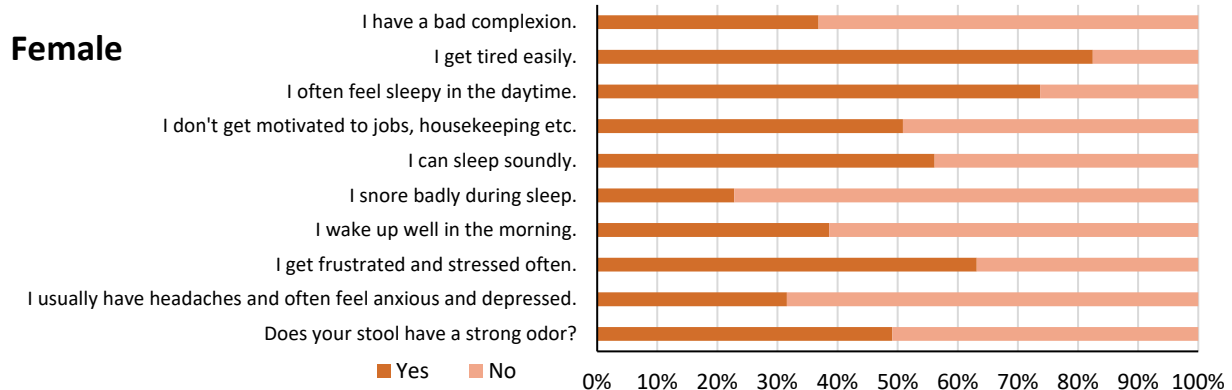
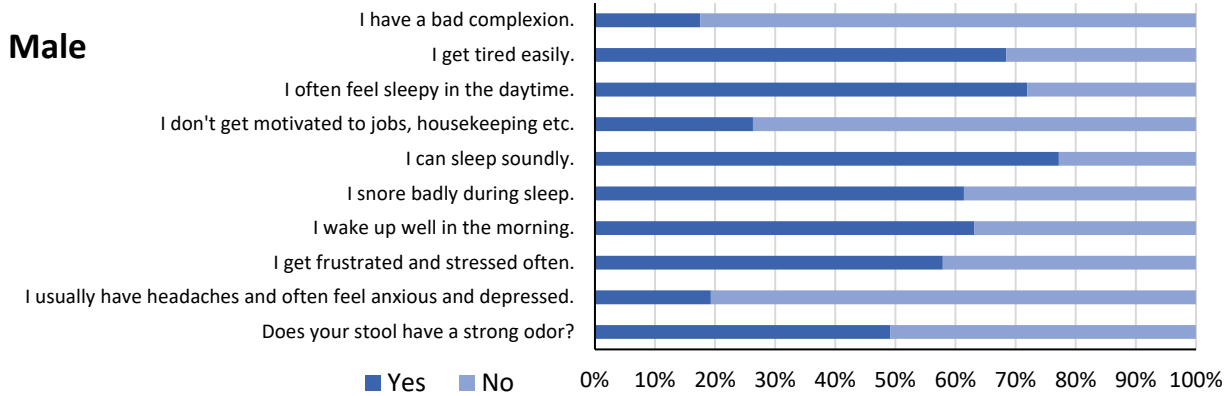
(Male) "I bike to work."

(Female) "3 per week"

**Q. Please answer the following questions**

Male	Yes		No	
	Answer	Ratio	Answer	Ratio
I have a bad complexion.	10	18%	47	82%
I get tired easily.	39	68%	18	32%
I often feel sleepy in the daytime.	41	72%	16	28%
I don't get motivated to jobs, housekeeping etc.	15	26%	42	74%
I can sleep soundly.	44	77%	13	23%
I snore badly during sleep.	35	61%	22	39%
I wake up well in the morning.	36	63%	21	37%
I get frustrated and stressed often.	33	58%	24	42%
I usually have headaches and often feel anxious and depressed.	11	19%	46	81%
Does your stool have a strong odor?	28	49%	29	51%

Female	Yes		No	
	Answer	Ratio	Answer	Ratio
I have a bad complexion.	21	37%	36	63%
I get tired easily.	47	82%	10	18%
I often feel sleepy in the daytime.	42	74%	15	26%
I don't get motivated to jobs, housekeeping etc.	29	51%	28	49%
I can sleep soundly.	32	56%	25	44%
I snore badly during sleep.	13	23%	44	77%
I wake up well in the morning.	22	39%	35	61%
I get frustrated and stressed often.	36	63%	21	37%
I usually have headaches and often feel anxious and depressed.	18	32%	39	68%
Does your stool have a strong odor?	28	49%	29	51%



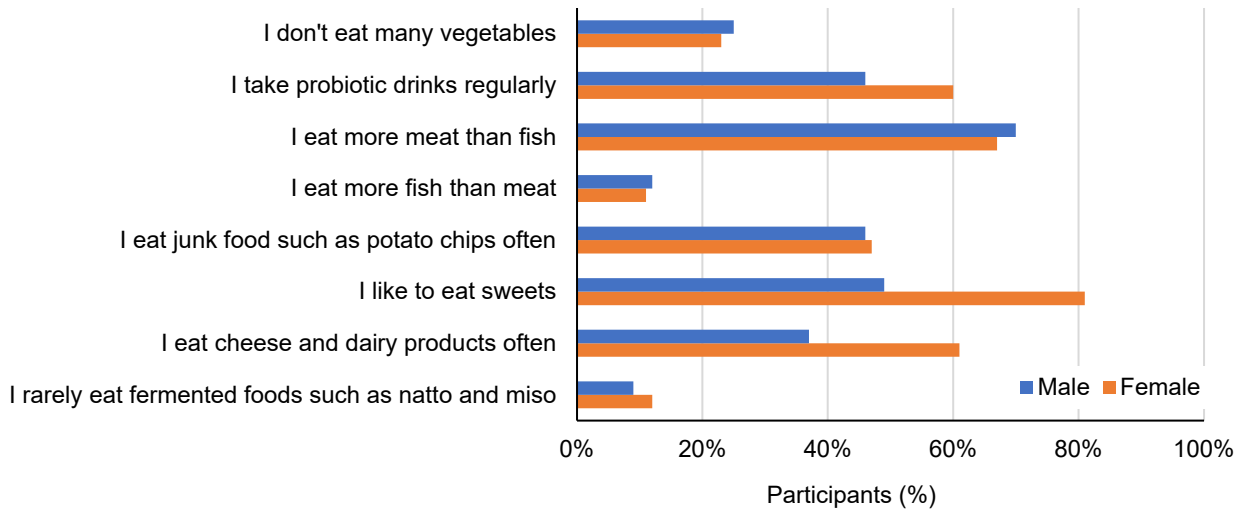
**Q. Please answer the following questions about your eating habits.**

**Male**

	Answer	Ratio
I don't eat many vegetables	14	25%
I take probiotic drinks regularly	26	46%
I eat more meat than fish	40	70%
I eat more fish than meat	7	12%
I eat junk food such as potato chips often	26	46%
I like to eat sweets	28	49%
I eat cheese and dairy products often	21	37%
I rarely eat fermented foods such as natto and miso	5	9%

**Female**

	Answer	Ratio
I don't eat many vegetables	13	23%
I take probiotic drinks regularly	34	60%
I eat more meat than fish	38	67%
I eat more fish than meat	6	11%
I eat junk food such as potato chips often	27	47%
I like to eat sweets	46	81%
I eat cheese and dairy products often	35	61%
I rarely eat fermented foods such as natto and miso	7	12%



## Results

Changes before and after 4 weeks of taking TwX

### Defecation Status (1/2)

Male	Before		After	
	Answer	Ratio	Answer	Ratio
Smooth defecation (everyday)	41	72%	43	76%
Smooth defecation (once in 2 days)	6	10%	8	14%
Smooth Defecation (other)	1	2%	3	5%
Constipated ( $\geq 3$ days)	9	16%	3	5%
Constipated ( $\geq 7$ days)	0	0%	0	0%
Constipation (defecating with laxatives)	0	0%	0	0%
Constipation (defecation with enema) (no effect of laxatives)	0	0%	0	0%

"Other" comment(s):

Due to lactose intolerance, consuming dairy products leads to having bowel movements multiple times throughout the day.

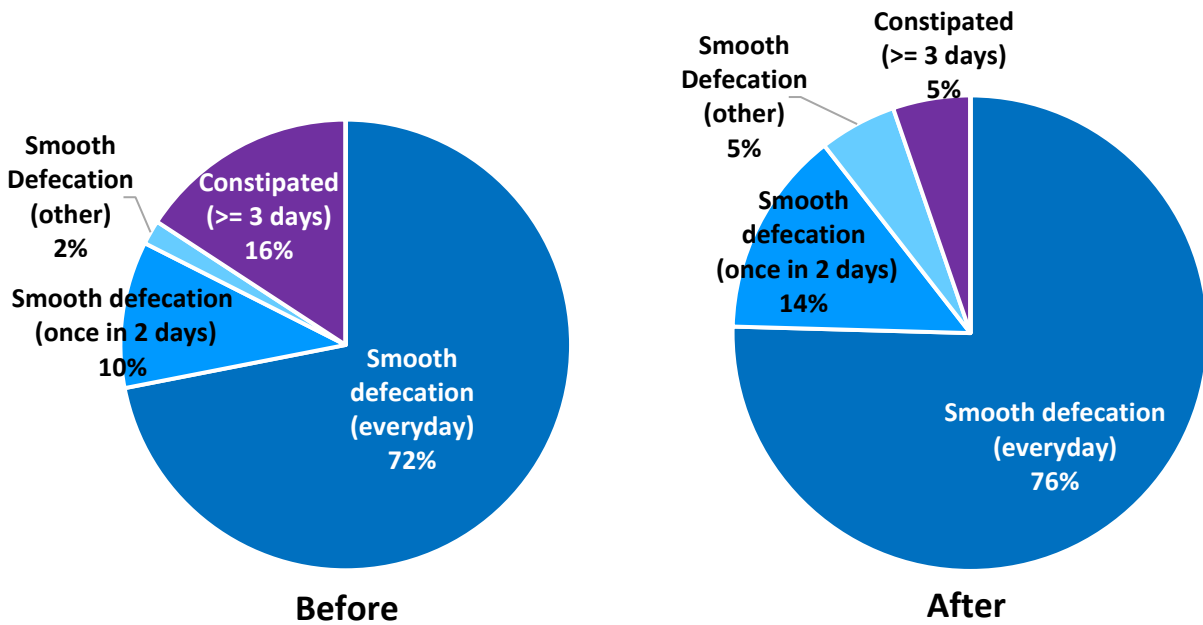
Female	Before		After	
	Answer	Ratio	Answer	Ratio
Smooth defecation (everyday)	24	42%	38	67%
Smooth defecation (once in 2 days)	19	33%	10	18%
Smooth Defecation (other)	2	4%	2	3%
Constipated ( $\geq 3$ days)	11	19%	4	7%
Constipated ( $\geq 7$ days)	0	0%	0	0%
Constipation (defecating with laxatives)	1	2%	3	5%
Constipation (defecation with enema) (no effect of laxatives))	0	0%	0	0%

"Other" comment(s):

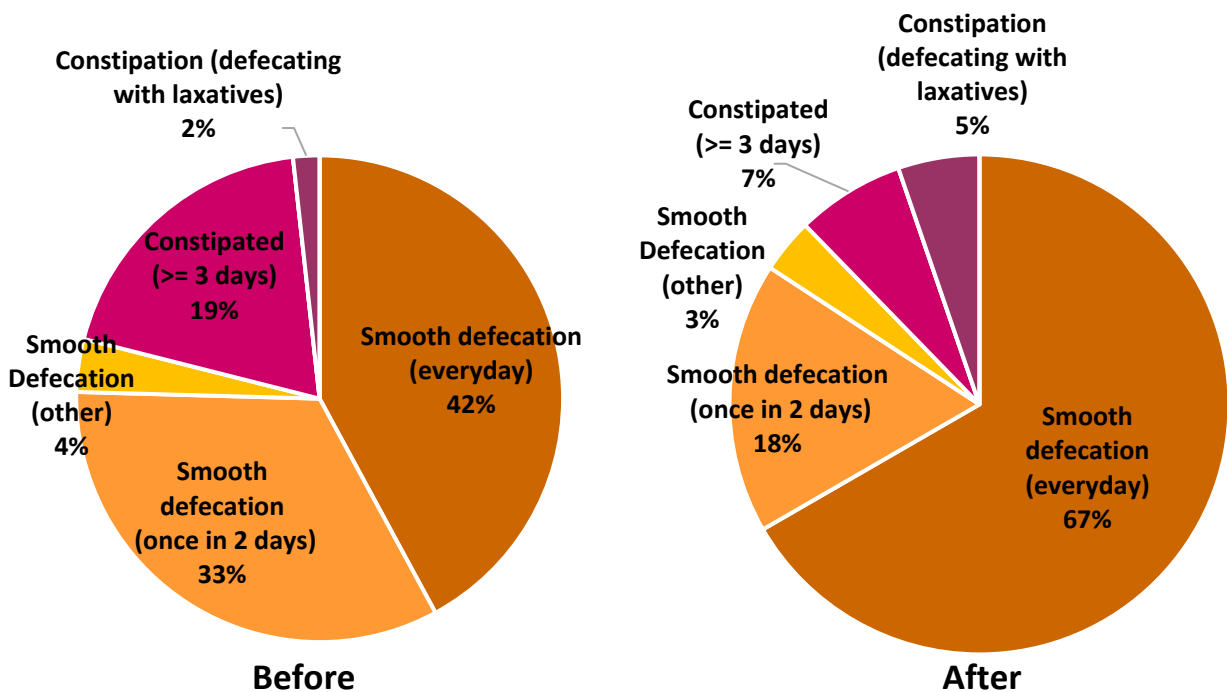
Almost every day, but sometimes there is a little or no stool for a few days.

I get constipated due to physical condition, stress, etc.

## Defecation Status (2/2)



**A. Bowel movement before/after taking Twendee X in male**



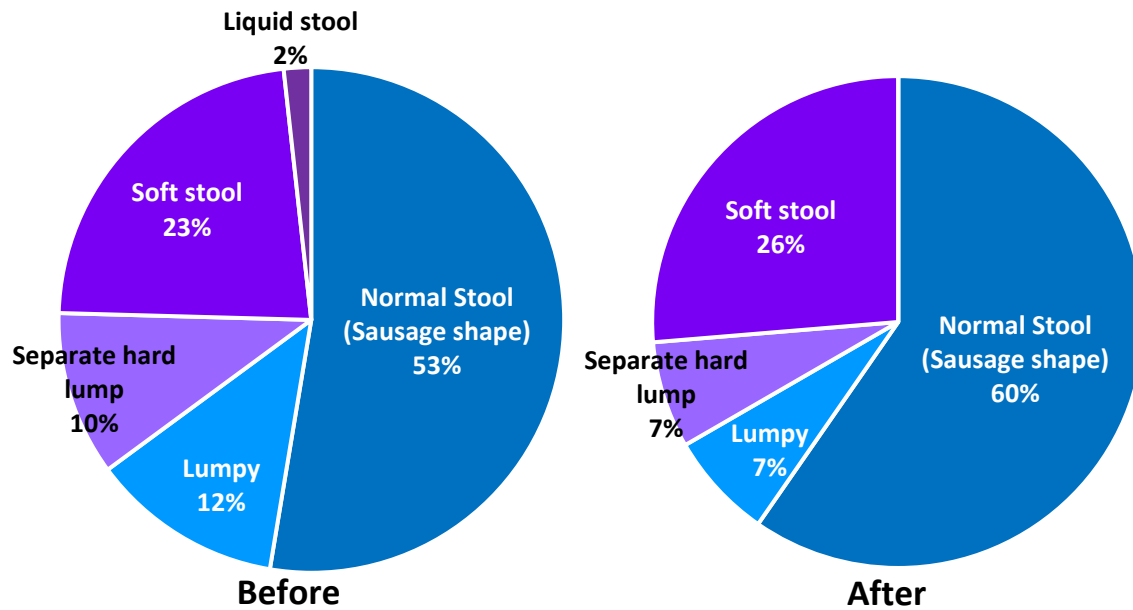
**B. Bowel movement before/after taking Twendee X in female**

## Stool shapes (1/2)

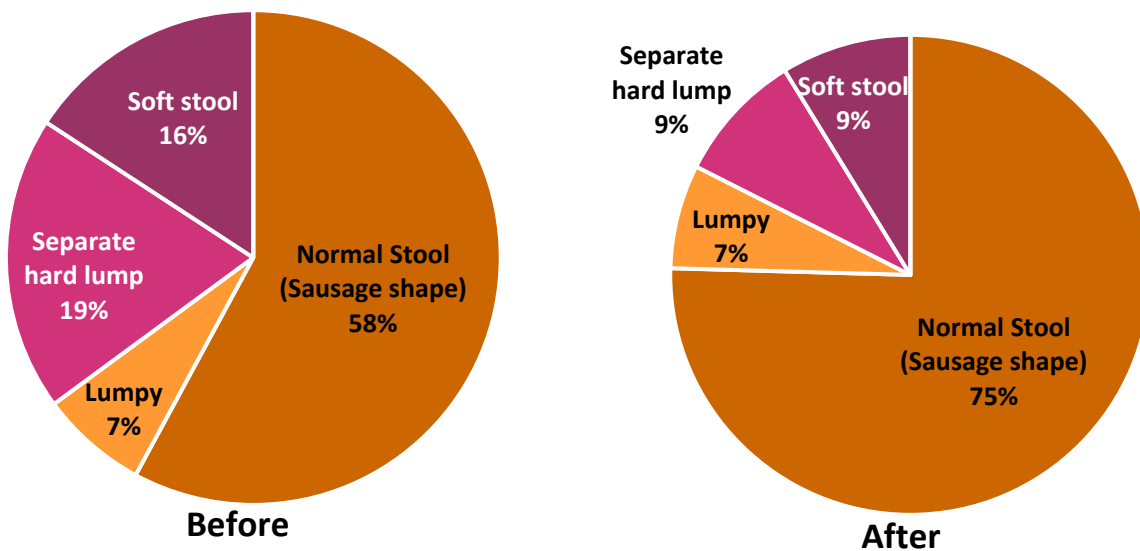
<b>Male</b>	Before		After	
	Answer	Ratio	Answer	Ratio
Normal Stool (Sausage shape)	30	53%	34	60%
Lumpy	7	12%	4	7%
Separate hard lump	6	10%	4	7%
Soft stool	13	23%	15	26%
Liquid stool	1	2%	0	0%

<b>Female</b>	Before		After	
	Answer	Ratio	Answer	Ratio
Normal Stool (Sausage shape)	33	58%	43	75%
Lumpy	4	7%	4	7%
Separate hard lump	11	19%	5	9%
Soft stool	9	16%	5	9%
Liquid stool	0	0%	0	0%

## Stool shapes (2/2)



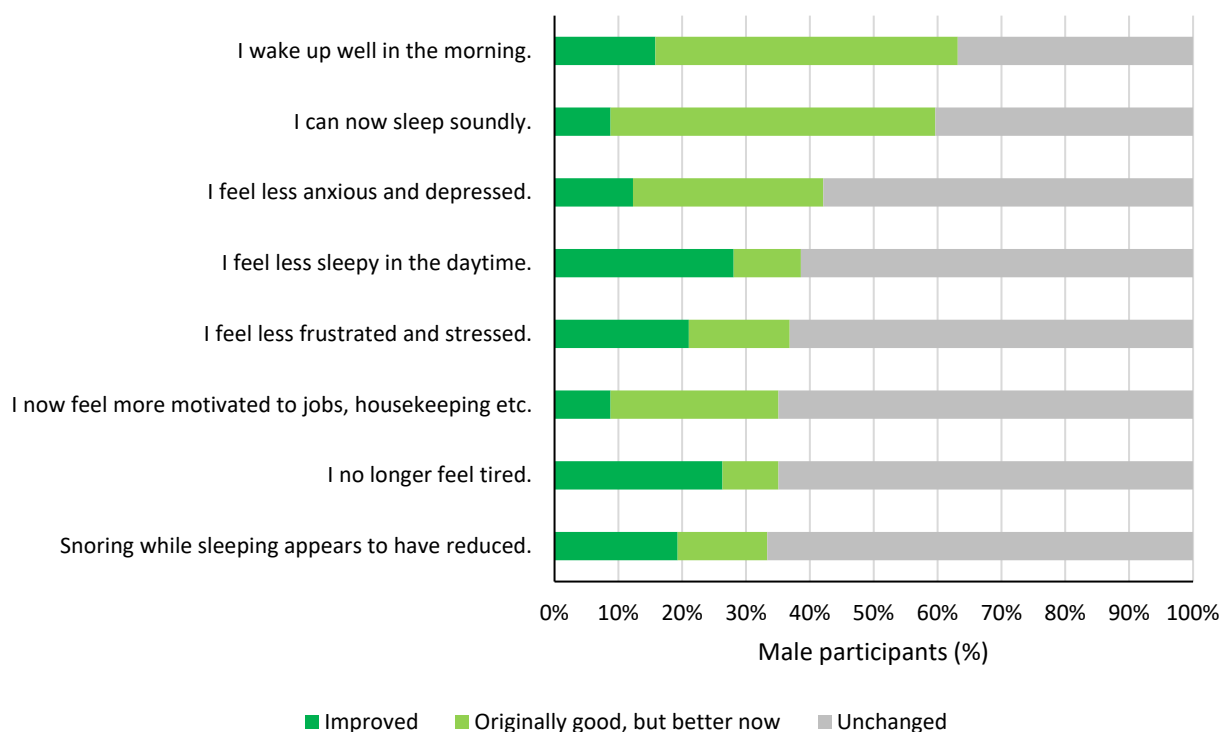
**A. Stool shapes before/after taking Twendee X in male**



**B. Stool shapes before/after taking Twendee X in female**

## Other subjective physical changes (1/2)

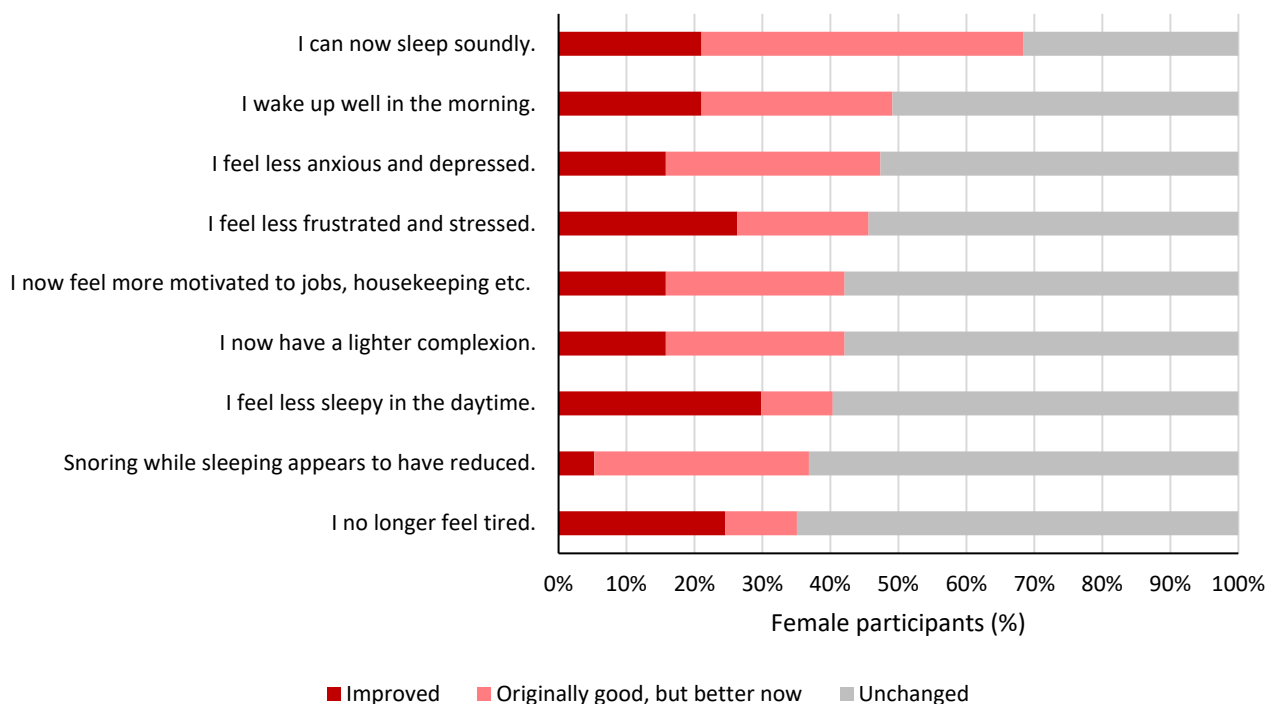
Male	Improved	Originally good, but better now	Unchanged
I wake up well in the morning.	9	27	21
I can now sleep soundly.	5	29	23
I feel less anxious and depressed.	7	17	33
I feel less sleepy in the daytime.	16	6	35
I feel less frustrated and stressed.	12	9	36
I now feel more motivated to jobs, housekeeping etc.	5	15	37
I no longer feel tired.	15	5	37
Snoring while sleeping appears to have reduced.	11	8	38





## Other subjective physical changes (2/2)

Female	Improved	Originally good, but better now	Unchanged
I can now sleep soundly.	12	27	18
I wake up well in the morning.	12	16	29
I feel less anxious and depressed.	9	18	30
I feel less frustrated and stressed.	15	11	31
I now feel more motivated to jobs, housekeeping etc.	9	15	33
I now have a lighter complexion.	9	15	33
I feel less sleepy in the daytime.	17	6	34
Snoring while sleeping appears to have reduced.	3	18	36
I no longer feel tired.	14	6	37



## Improvements after taking TwX as indicated in the comments of the survey

### ● Male

- Taking the supplement regularly helped me wake up early in the morning without an alarm.
- Before taking the supplement, my stools were soft in most case. However, a few days after starting it, my stools became normal.
- I think my physical condition is better than before.
- Somehow I feel a little healthier.
- My partner told me that I am snoring less frequently lately.
- I was able to sleep better at night and wake up better in the morning, feeling less tired.

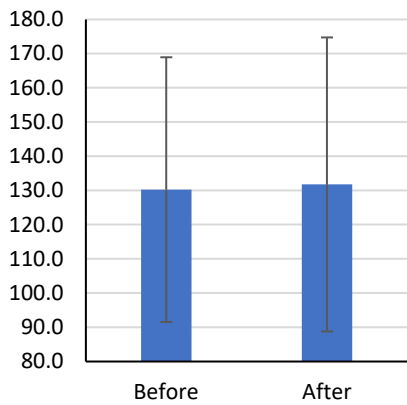
### ● Female

- The smell of stools is no longer harsh. My fart no longer smells bad.
- I now have a daily bowel movement. In addition, my hay fever has improved, and I seem to be falling asleep earlier at night.
- I wake up feeling better in the morning and can get up more easily. My skin is less dry and the problems have disappeared. I believe my skin spots have lightened.
- My bowel movements have improved considerably. Gum boils caused by pus in the gums, not mouth ulcers, have stopped appearing.
- I have noticed an improvement in my skin and no longer experience as many breakouts. I can now stay awake until night without needing to take naps.
- I feel healthy. People now frequently comment on how much better my complexion looks.

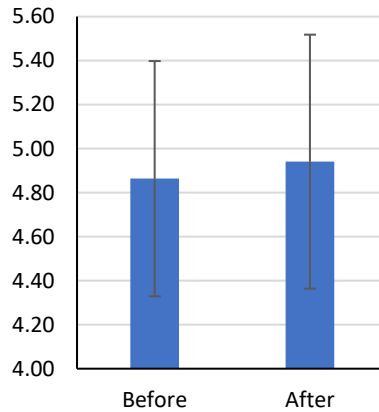
# Change in gut microbiota indices before and after taking TwX

			SD	
	Before	After	Before	After
Number of bacterial species detected	130.2	131.8	38.7	43.0
Diversity Index	4.86	4.94	0.53	0.58
Firmicutes/Bacteroidetes ratio	8.6	4.3	1.35	2.92

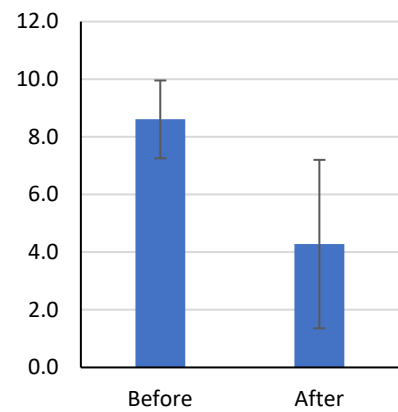
Number of bacterial species detected



Diversity Index



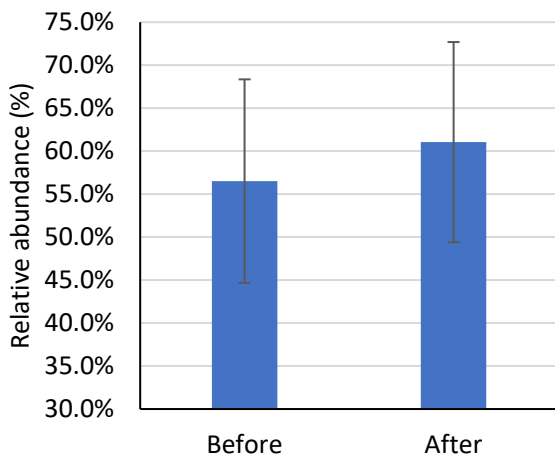
Firmicutes/Bacteroidetes ratio



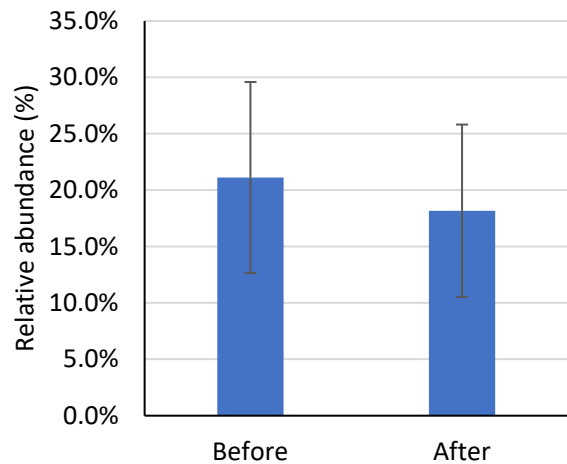
	Before	After
Firmicutes	56.5%	61.0%
SD	11.8%	11.6%

	Before	After
Bacteroidetes	21.1%	18.2%
SD	8.47%	7.65%

Firmicutes



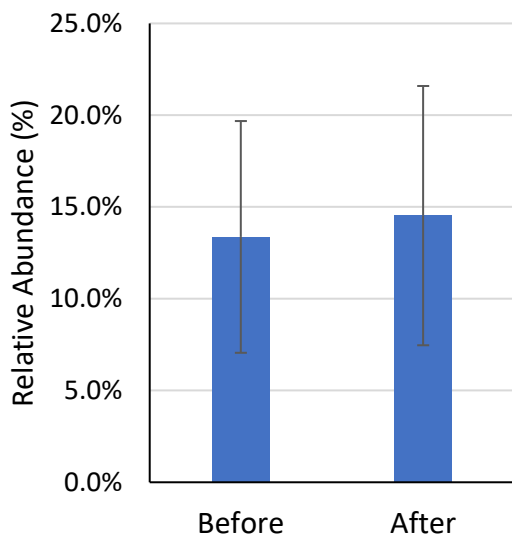
Bacteroidetes



**Butyrate-producing bacteria  
abundant in long-lived individuals\***

	Before	After
All	13.4%	14.5%
SD	6.31%	7.06%

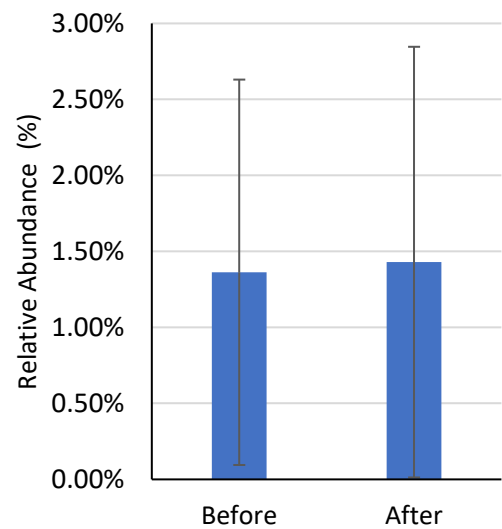
Roseburia	4.3%	4.4%
Coprococcus	2.3%	2.7%
Unclassified Lachnospiraceae	5.7%	6.3%
Lachnospira	1.1%	1.2%



**Butyrate-producing bacteria  
Major intestinal bacterial genus\*\***

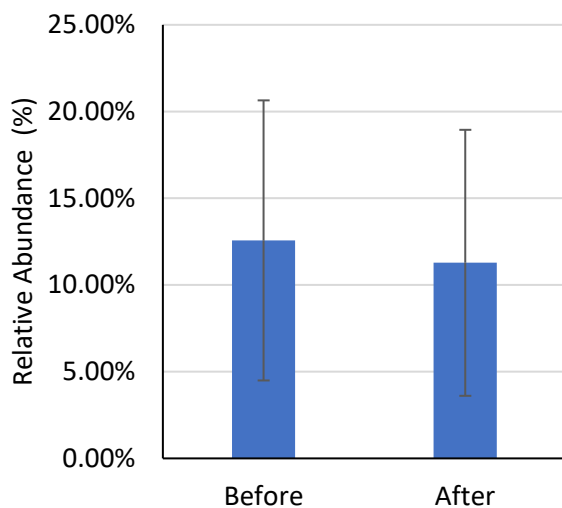
	Before	After
All	1.36%	1.43%
SD	1.27%	1.42%

Eubacterium	0.59%	0.57%
Clostridium	0.77%	0.86%



**Propionic acid-producing bacteria\*\***

	Before	After
Bacteroides	12.6%	11.3%
SD	8.08%	7.67%



\*: Naito et al., 2019

\*\* : Watabe, 2005

# Major beneficial and harmful bacteria detected

## Target beneficial bacteria

Akkermansia muciniphila
Adlercreutzia equolifaciens
Eggerthella lenta
Slackia isoflavoniconvertens
Serratia marcescens
Hafnia alvei
Bifidobacterium
Faecalibacterium
Bacteroides fragilis
Proteus vulgaris
Morganella morganii
Eubacterium
Lactococcus lactis
Ruminococcus
Roseburia
Christensenellaceae
Lactobacillus

## Target harmful bacteria

Clostridium perfringens
Campylobacter jejuni
Prevotella copri
Bacillus cereus
Klebsiella pneumoniae
Haemophilus parainfluenzae
Pseudomonas aeruginosa

The amount of beneficial and harmful bacteria present in the above was measured for each participant.

There were 5 participants with a higher presence of the top five bad bacteria than the top five good bacteria.

For those who had a predominance of bad bacteria, the percent change after taking TwX was as follows:

Beneficial bacteria change rate	1.243028
Harmful bacteria change rate	0.648176