

## Occurrence of *Cryptosporidium* oocysts and *Giardia* cysts in the Nakdong River and their removal during water treatment

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### ABSTRACT

This study was conducted in preparation of a pending *Cryptosporidium* regulation in Korea. The study had two main objectives: 1) to examine the occurrence of *Cryptosporidium* oocysts in the Nakdong River; and 2) to evaluate their removal during water treatment. Occurrence of *Giardia* cysts was also examined. Average (arithmetic mean) numbers of *Cryptosporidium* oocysts and *Giardia* cysts at the treatment intake site were  $2.6\text{ l}^{-1}$  and  $4.8\text{ l}^{-1}$ , respectively. Generally, the number of *Giardia* cysts was higher than that of *Cryptosporidium* oocysts at more sites, but the difference was minimal. Comparison of tributaries indicated that livestock wastes were more serious pollutants than sewage in terms of protozoa contamination. In general, fewer oocysts and cysts were detected during winter. No correlation was found for such water quality parameters as T-N, T-P, TOC, DO, pH and temperature with the numbers of oocysts and cysts except for suspended solids, which showed the highest correlation ( $R^2 = 0.55$ ). Removal of *Cryptosporidium* oocysts was evaluated using a *Cryptosporidium* tracer, which has similar characteristics to *Cryptosporidium* oocysts. The tracer removal depended on turbidity removal. Coagulation followed by sedimentation resulted in 1.2–1.5 log removal of the tracer under optimal conditions. Filtration resulted in 1.3–1.5 log removal of the tracer. These treatability experiments showed that traditional water treatment processes could achieve 2.5–3.0 log removal of the oocysts.

**Key words** | *Cryptosporidium*, *Giardia*, occurrence, removal, tracer, treatability

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### INTRODUCTION

The Korean government has regulated *Giardia* cyst removal from drinking water through treatment since 2004 (MOE 2002). The regulation stipulates a 3-log removal for *Giardia* cysts, to be satisfied by proper operation of filtration and disinfection. A credit of 2.5-log removal is given to a water treatment plant using rapid filtration provided the plant maintains filtrate turbidity levels of less than 0.3 NTU in 95% of monthly measurements. The remaining credit is given after the plant satisfies the required CT value (the concentration of residual chlorine multiplied by the chlorine contact time). Besides the regulation of *Giardia*

cyst removal, water treatment facilities are also required to monitor *Cryptosporidium* levels in raw water as well as in treated water. Such monitoring results will then be used for the future regulation of *Cryptosporidium*. This study was conducted in preparation of the upcoming regulation of *Cryptosporidium* oocysts.

There were two objectives in this study. The first objective was to examine the occurrence of *Cryptosporidium* oocysts and *Giardia* cysts, and the second was to evaluate their removal during water treatment. The specific objectives were:

- Measuring the numbers of *Cryptosporidium* oocysts and *Giardia* cysts and their seasonal variation in raw waters of the Nakdong River
- The effect of upstream tributaries on *Cryptosporidium* levels
- Identification of water quality parameters that correlate with numbers of *Cryptosporidium* oocysts and *Giardia* cysts in the raw water
- The removal of *Cryptosporidium* oocysts during water treatment
- The effect of high turbidity levels on the removal of *Cryptosporidium* oocysts

## MATERIALS AND METHODS

### Monitoring of *Cryptosporidium* and *Giardia*

The Nakdong River was selected as the raw water source. This river, notorious for being organically contaminated, is a major raw water resource for the southeast part of Korea. Nine sampling points were selected and are shown in Figure 1. 'N' indicates the Nakdong River and 'T' its tributary. The sampling points were numbered from upstream (N1) to downstream (N6). N4 is the water treatment facility's intake site for city 'D' with 2.5 million people. Five sampling points were selected

upstream of the intake site, with three points downstream. T1 (on a tributary) was selected because it was highly contaminated by livestock wastes, while T2 was mainly contaminated by sewage. They would show the effects, on tributaries, of livestock waste and sewage contamination on numbers of *Cryptosporidium* oocysts and *Giardia* cysts. T3 (on a tributary) was selected to examine the combined effect of industrial waste and sewage contamination. Sampling was conducted every month for a 1-year period. *Cryptosporidium* oocysts were measured from January to December except at two sites (N4, N6). For N4, samples were collected from May to December, and for N6, from March to December. *Giardia* cysts were measured from May to December at all sites. *Cryptosporidium* oocysts and *Giardia* cysts were measured in accordance with the USEPA method (USEPA 1999). For this study, however, the only difference is the sample volume. It was increased from 10 litres to 20 litres for more optimal detection. The recovery rates of *Cryptosporidium* oocysts and *Giardia* cysts were 44.5% and 45% based on this test.

### Removal

Removal (treatability) of *Cryptosporidium* was evaluated using a *Cryptosporidium* tracer (Table 1), which has similar characteristics to *Cryptosporidium* oocysts. The tracer was

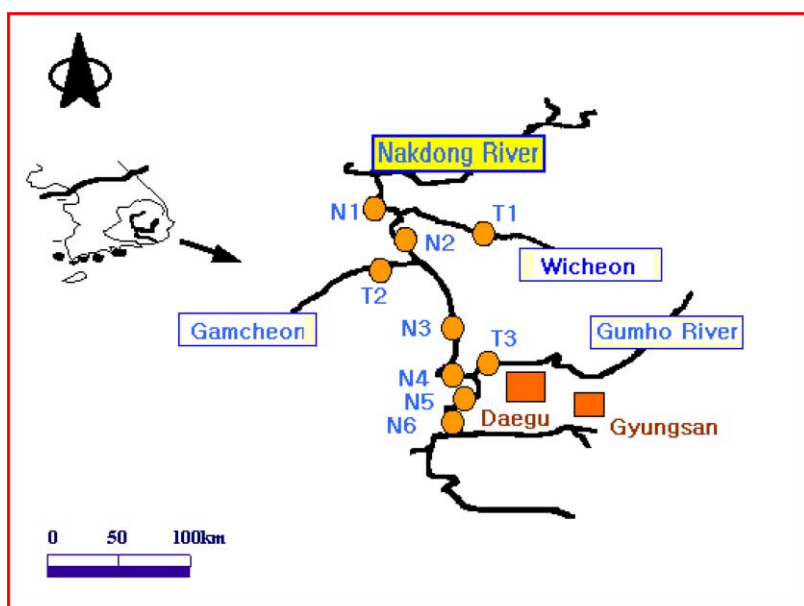


Figure 1 | Sampling sites on the Nakdong River.

**Table 1** | Comparison of *Cryptosporidium* tracer and oocyst

Description	<i>Cryptosporidium</i> tracer	<i>Cryptosporidium parvum</i> oocyst
Material	Polymethylmethacrylate (PMMA)	
Size	5 $\mu\text{m}$	4–6 $\mu\text{m}$
Specific gravity	1.19	1.05–1.10
Zeta potential	–29 mV (pH 6.6)	–25 mV (pH 6.0–6.5)

added to the raw water taken from N4. The evaluation focused on examining how conventional water treatment processes removed the tracer. Coagulation, flocculation and sedimentation were simulated with jar tests, while filtration was simulated with small filter columns. Poly-aluminium chloride (PACl) was selected as a coagulant because the water treatment facility taking Nakdong River water uses PACl. Table 2 shows the conditions of coagulation, sedimentation and filtration in this study.

**Table 2** | Conditions of coagulation, sedimentation and filtration

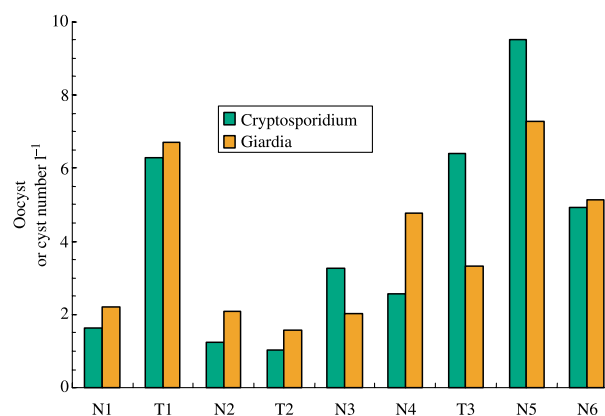
Description	Conditions
Coagulant	PACl (polyaluminium chloride)
Rapid mixing time and speed	3 minutes at 140–150 rpm
Slow mixing time and speed	30 minutes at 25–35 rpm
Sedimentation time	1 hour
Column dimension	$\phi 100 \text{ mm} \times \text{L}1,500 \text{ mm}$
Filter media material	Sand
Filter media depth	750 mm
Gravel depth	300 mm
Filtration rate	134 $\text{m day}^{-1}$
Effective size of filter media	0.6 mm
Uniformity of filter media	1.5

## RESULTS AND DISCUSSION

### Occurrence

*Cryptosporidium* oocysts and *Giardia* cysts were detected in all samples, as shown in Figure 2. Table 3 summarises the results. Average (arithmetic mean) numbers of *Cryptosporidium* oocysts and *Giardia* cysts during the study period were  $2.6 \text{ l}^{-1}$  and  $4.8 \text{ l}^{-1}$  at the intake point (N4), respectively. The number of *Cryptosporidium* oocysts at N4 was in the range of  $0.45\text{--}5.5 \text{ l}^{-1}$  and that of *Giardia* cysts was in the range of  $0.25\text{--}21.6 \text{ l}^{-1}$ . More *Giardia* cysts were detected than *Cryptosporidium* oocysts. The number of cysts was higher than that of oocysts at six sampling sites, and vice versa at three sites. Generally, upstream sites were more contaminated by *Giardia* cysts, downstream sites by *Cryptosporidium* oocysts. However, the difference was so minimal that it could not be confirmed statistically. A similar result was obtained in another study. Lee et al. (2000) measured *Cryptosporidium* oocysts and *Giardia* cysts in the Han River, which is the raw water source for Seoul City.

The oocysts and cysts at T1–T3 were examined in order to compare the effect of different sources of contamination. The examination revealed that the average numbers of oocysts ( $6.3 \text{ l}^{-1}$ ) and cysts ( $6.7 \text{ l}^{-1}$ ) at T1 were greater than those at T2 ( $1.0 \text{ l}^{-1}$  and  $1.6 \text{ l}^{-1}$ ) and T3 ( $6.4 \text{ l}^{-1}$  and  $3.3 \text{ l}^{-1}$ ). This result suggests that livestock wastes are the more likely source of protozoa contamination than sewage or industrial wastes. However, the livestock contamination was effectively diluted as there was no difference in the protozoa levels

**Figure 2** | Average numbers of *Cryptosporidium* oocysts and *Giardia* cysts at the sampling sites.

**Table 3** | Occurrence of *Cryptosporidium* oocysts and *Giardia* cysts in the Nakdong River in 2001

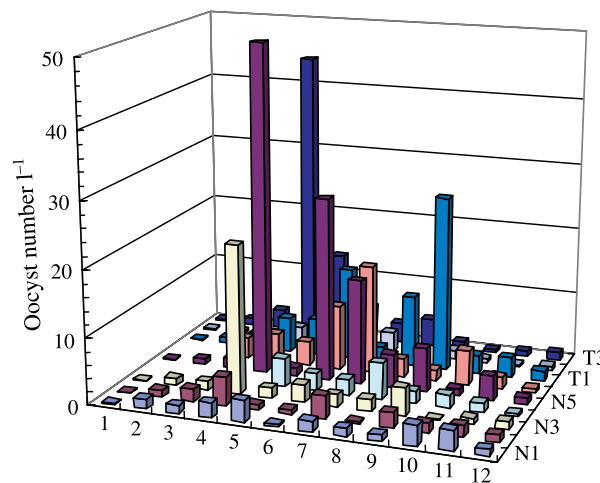
Sampling sites	<i>Cryptosporidium</i> oocysts l <sup>-1</sup>	<i>Giardia</i> cysts l <sup>-1</sup>
N1	0.15–3.30 (1.62)*	0.45–5.40 (2.22)
T1	0.10–26.50 (6.28)	1.50–13.40 (6.70)
N2	0.05–4.30 (1.60)	0.10–4.45 (2.09)
T2	0.25–3.00 (1.02)	0.20–3.35 (1.59)
N3	0.10–22.40 (3.27)	0.10–6.45 (2.04)
N4	0.45–5.50 (2.56)	0.25–21.60 (4.78)
T3	0.20–43.90 (6.40)	0.10–9.25 (3.33)
N5	0.10–49.70 (9.49)	0.80–24.20 (7.28)
N6	0.65–16.35 (4.93)	1.40–10.75 (5.14)

\*Values in parentheses indicate the average values.

between the water upstream of the livestock contamination (N1) and the water at the downstream site (N2).

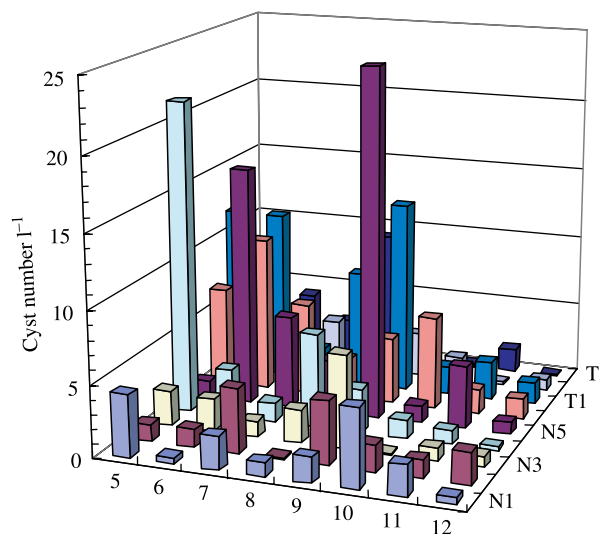
The number of *Cryptosporidium* oocysts in the Nakdong River (3.31 l<sup>-1</sup>) increased at N3 to more than twice the number from the upstream sites N1 (1.61 l<sup>-1</sup>) and N2 (1.31 l<sup>-1</sup>). The numbers of *Giardia* cysts were higher at the intake site (N4) than at the other upstream sites. The protozoa level peaked at N5, and reduced at N6. Sudden increases in the number of *Cryptosporidium* oocysts at N3 suggest that the river upstream of N3 might be contaminated by sewage or livestock wastes. The tributary of T3 and sewage from city ‘D’ possibly aggravated the protozoa contamination at N5, where after dilution reduced the contamination at N6.

Figures 3 and 4 show the seasonal variation of protozoa levels at all sampling sites during the study period. Examination of the seasonal variation revealed that, generally, fewer oocysts and cysts were detected during winter. The lowest numbers of *Cryptosporidium* oocysts were generally detected in January. The exceptions were the sampling sites of N3, N4 and N6. Although the January counts at N3 were still low, the lowest count was observed in October. The January counts were unavailable for N4 and N6. Unlike the lowest number of oocysts, there was no definite pattern for the highest number of oocysts, but more oocysts were generally found during spring



**Figure 3** | Seasonal variation of *Cryptosporidium* oocysts at all sampling sites.

and summer. The highest number was detected during April (N2, N3, N5, T3), May (T1), July (N6, T2), August (N4) and September (T1). Other studies also reported a high level of *Cryptosporidium* oocysts during spring and early summer (LeChevallier et al. 1997; States et al. 1997). The lowest numbers of *Giardia* cysts were mostly measured in December, except at three sites (N2, N3 and T2). The lowest count was detected in August (N2), October (N3) and November (T2). As with *Cryptosporidium* oocysts, there seemed to be no definite pattern for the highest number. The highest number was detected during May through October: May (N4), June (N6), July (N2, T2), August (T3), September (N3, N5, T1) and October (N1).



**Figure 4** | Seasonal variation of *Giardia* cysts at all sampling sites.

An attempt was then made to identify a water quality parameter that would correlate with the numbers of oocysts and cysts. Parameters such as T-N, T-P, TOC, DO, pH and temperature were investigated but with no success. Some correlation was evident with suspended solids (SS) ( $R^2 = 0.55$ ). This is in line with other studies (Elimelech & O'Melia 1990; LeChevallier & Norton 1992). Besides those chemical parameters, the physical parameter of flow rate was also evaluated. However, there was no correlation found between flow rate and the protozoa level.

### Treatability

*Cryptosporidium* tracer was removed effectively through the conventional water treatment processes of coagulation, sedimentation and filtration. According to researchers who investigated the treatability of *Cryptosporidium* oocysts and *Giardia* cysts during water treatment (LeChevallier & Norton 1995), *Giardia* cysts were almost completely removed during sedimentation. However, *Cryptosporidium* oocysts were slightly more difficult to remove, with a removal performance of 98% after filtration.

Coagulation was effective in removing the *Cryptosporidium* tracer (Figure 5) showing that properly practised coagulation, when followed by sedimentation, could achieve a 1.2–1.5 log removal of the tracer. The tracer removal pattern behaved similarly to the turbidity removal pattern. As coagulation approached the optimum condition with increas-

ing PACl dose, the turbidity removal improved and so did the tracer removal. Further increases in PACl dose depressed the pH, which aggravated the turbidity removal as well as the tracer removal. Figure 5 clearly shows the settled tracer level changing in line with the settled turbidity level. The maximum tracer removal (1.5 log) was obtained at pH 6.48, at which point the settled turbidity reached the lowest level. The tracer removal was sensitive to changes in pH, as for the turbidity removal. In order to examine the effect of pH, PACl was added to the raw water at 10 ppm. While the solution pH was changed to 5, 6, 7 and 8, residual water qualities were monitored. Figure 6 shows the results. According to Figure 6, the settled turbidity, tracer and SS reached their lowest levels at pH 7. As the pH was varied from 7, residual levels of SS, turbidity and tracer increased. These results indicate that the removal performance of *Cryptosporidium* oocysts is affected by coagulation conditions of dosage and pH. The optimum coagulant dosage and pH should be used in order to maximise the removal of *Cryptosporidium* oocysts.

The effect of high levels of turbidity in the raw water was then examined because high levels of turbidity cause operational problems at local water treatment plants. Operators experienced difficulties in coagulating highly turbid water and the settled turbidity subsequently became too high, especially during the flooding season. Turbidity of raw water was increased from 10 NTU to 100 NTU by adding kaolin in order to examine the effect of high turbidity. Figure 7 shows the best settled water qualities at each condition.

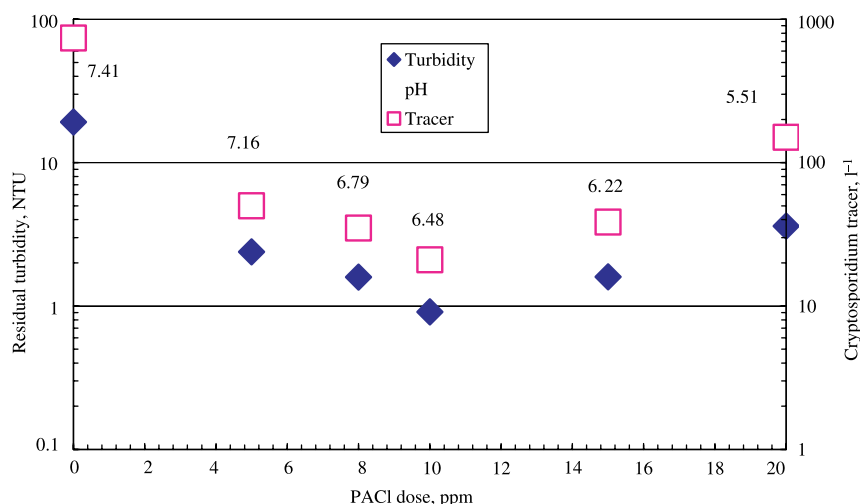
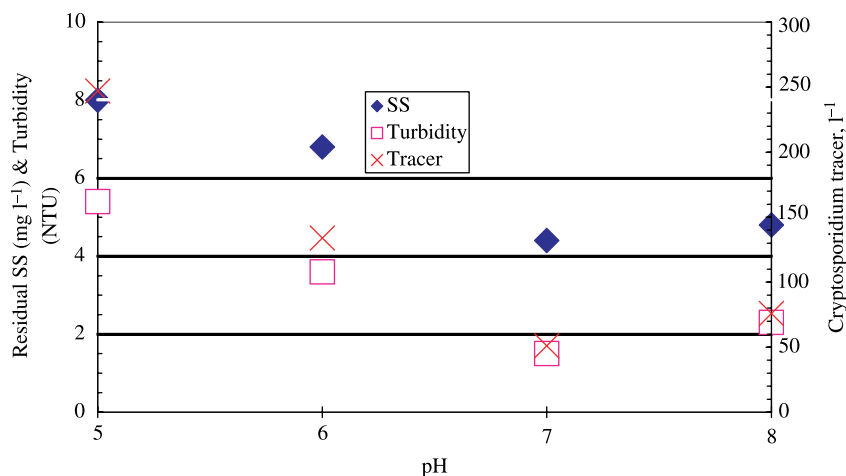


Figure 5 | The settled turbidity and *Cryptosporidium* tracer as a function of PACl dose.

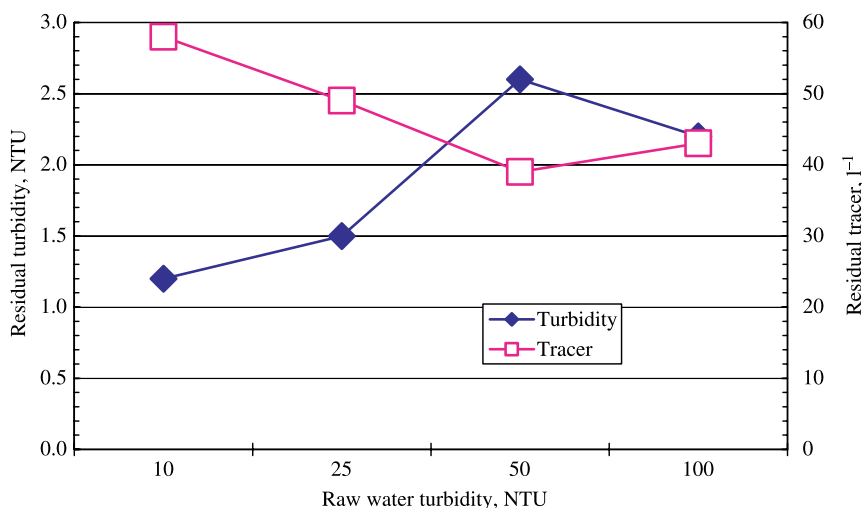


**Figure 6** | The effect of pH on the settled SS, turbidity and *Cryptosporidium* tracer.

According to **Figure 7**, the tracer removal was less sensitive to a change in the raw water turbidity than the turbidity removal. The settled turbidity increased more than twice with increasing raw water turbidity, but the settled tracer level decreased. The tracer removal performance deteriorated slightly only when the raw water turbidity increased to 100 NTU. The better removal of the tracer could be related to its shape. A sphere is superior to other shapes in terms of settling. Therefore, sedimentation of the spherical tracer produced more stable results even when the raw water turbidity was extremely high.

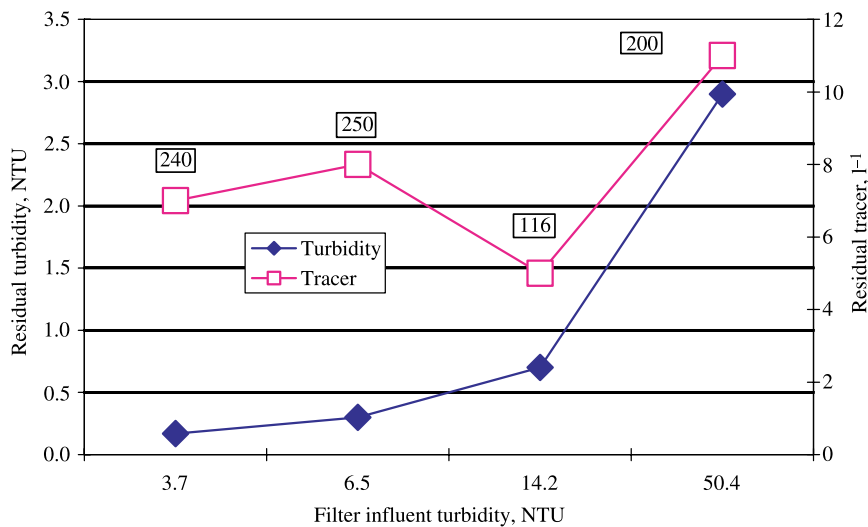
The effect of influent quality on tracer removal performance was examined in the filter column test. As shown in **Figure 8**, the filter influent turbidity was varied from 3.7 NTU

to 50.4 NTU, and the tracer number from  $116\text{ l}^{-1}$  to  $250\text{ l}^{-1}$ . The column test showed that the removal performance was not affected by the influent quality. Filtration maintained the stable removal performance of the tracer (1.3–1.5 log). The filtrate turbidity level was more sensitive to the influent turbidity change than the filtrate tracer level. The filtrate turbidity level increased with increased turbidity of the filter influent. Less change was observed for the filtrate tracer level, which exceeded  $10\text{ l}^{-1}$  only when the filter influent turbidity exceeded 50 NTU. Filtration was able to maintain 1.5-log removal of the tracer up to the influent turbidity level of 6.5 NTU. The removal performance deteriorated slightly as the filter influent turbidity increased to 10 NTU (1.4 log) and 50 NTU (1.3 log).



**Figure 7** | The effect of high turbidity of the raw water on the settled turbidity and *Cryptosporidium* tracer.





**Figure 8** | The effect of the turbidity loading on the filtrate turbidity and *Cryptosporidium* tracer (values inside the box indicate the influent tracer level).

## CONCLUSIONS

This study revealed serious protozoan contamination of the Nakdong River. The numbers of *Cryptosporidium* oocysts at the intake site were in the range of  $0.45\text{--}5.51\text{ l}^{-1}$  (average of  $2.61\text{ l}^{-1}$ ) and *Giardia* cysts were in the range of  $0.25\text{--}21.61\text{ l}^{-1}$  (average of  $4.81\text{ l}^{-1}$ ). The sites upstream of the water treatment facility intake on the Nakdong River were more contaminated by *Giardia* cysts, while the downstream sites were more contaminated by *Cryptosporidium* oocysts. The protozoa numbers were generally low during winter. However, there was no definite pattern for the high levels. Livestock wastes appeared to be a more important source of protozoa contamination than sewage or industrial wastes. Suspended solids level was the only water quality parameter showing some correlation with the occurrence of *Cryptosporidium* oocysts.

This study showed that conventional water treatment processes could be effective in removal of *Cryptosporidium* oocysts. Proper coagulation, if followed by sedimentation, could achieve 1.2–1.5 log removal of *Cryptosporidium* oocysts, and filtration could achieve another 1.3–1.5 log removal. Therefore, conventional water treatment processes could achieve 2.5–3.0 log removal of *Cryptosporidium* oocysts. Since the tracer removal closely followed the turbidity removal pattern, it is important to maintain a

good performance of turbidity removal during water treatment to minimise the possibility of protozoa breakthrough.

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