

## Ingestion of swimming pool water by recreational swimmers

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### ABSTRACT

The volume of water ingested by swimmers while swimming is of great interest to individuals who develop risk assessments using quantitative microbial risk assessment or epidemiological approaches. We have used chloroisocyanurate disinfected swimming pool waters to determine the amount of water swallowed by swimmers during swimming activity. The chloroisocyanurate, which is in equilibrium with chlorine and cyanuric acid in the pool water, provides a biomarker, cyanuric acid, that once swallowed passes through the body into the urine unchanged. The concentration of cyanuric acid in a 24 hour urine specimen and the concentration in pool water can be used to calculate the amount of water swallowed. Our study population of 549 participants, which was about evenly divided by gender, and young and adult swimmers, indicated that swimmers ingest about 32 mL per hour (arithmetic mean) and that children swallowed about four times as much water as adults during swimming activities. It was also observed that males had a tendency to swallow more water than females during swimming activity and that children spent about twice as much time in the water than adults.

**Key words** | cyanuric acid, swimming, water, water ingestion

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### INTRODUCTION

Swimming-associated gastrointestinal illness in recreational swimmers has been well documented (Pruss 1998; Wade *et al.* 2003; Zmirou *et al.* 2003). Epidemiological studies have shown that between 2 and 5% of swimmers become ill after exposure to bathing beach waters (Cabelli *et al.* 1979; Dufour 1984; Wade *et al.* 2006, 2008, 2010). The high cost of epidemiological studies, however, limits conducting such studies over long periods of time in various water environments. Quantitative microbial risk assessment (QMRA) has been suggested as an approach for estimating the health risk for swimmers who may be exposed to contaminated bathing beach waters (Ashbolt *et al.* 2010; Soller *et al.* 2010; Schets *et al.* 2011). QMRA is a process wherein various factors associated with swimming are used to estimate risk. These factors include: water quality, dose-response

associated with pathogens, fate and transport of water quality indicators and pathogens, and behavior patterns of swimmers while swimming.

The characteristics of water quality indicators and pathogens, such as dose-response, source and persistence in the environment, as well as behavioral characteristics of swimmers, such as wading, immersion of the body and immersing the head are significant factors that are commonly used to develop a risk assessment. Other behavioral factors, such as duration of body immersion and volume of water ingested have less frequently been included in the QMRA process because of the paucity of good data in this area.

The amount of water ingested while swimming has been of interest for many years. Several investigators have made

estimates about how much water might be swallowed by swimmers through a variety of techniques not utilizing empirical data related to ingestion. Table 1 summarizes such estimates that have been suggested by researchers over the last 70 years. The estimates ranged from 10 mL to 50 mL.

Very few studies directly related to swimmer water ingestion have been conducted. However, in the latter half of the 20th century chlorinated derivatives of cyanurate were increasingly used to disinfect swimming pool waters. In the presence of water, chlorinated isocyanurates dissociate to yield cyanuric acid and hypochlorous acid. The latter moiety acts as a disinfectant. The chlorinated isocyanurate acts as a stabilizer of the chlorine moiety, extending the disinfectant properties for a much longer period of time. This use led to the first empirical study of swimmer ingestion of water that was conducted by *Briggle et al.* (1981) and *Allen et al.* (1982). Their interest was not in how much water was swallowed during swimming activities, but rather in developing a new method for measuring cyanuric acid in swimming pool waters. Their studies were conducted to determine if there were health effects associated with exposure to cyanuric acid that had been added to a swimming pool in the form of a chlorinated isocyanurate. Although their studies were small, only five competitive swimmers, their investigation produced information indicating that competitive swimmers ingested, on average, about 322 mL (range 78 mL–793 mL) of pool water per hour. They also showed that cyanuric acid passed through the body unmetabolized, that only negligible amounts were absorbed through the skin, and that almost all of the cyanuric acid ingested could be recovered in a 24 h

urine sample. Furthermore, they noted, ‘Two volunteers in our study drank solutions of known CYA concentration. Subsequent analysis of their 24-h cumulative excretion indicated greater than 98% recovery of orally ingested CYA.’ *Dufour et al.* (2006) used the same approach to measure the ingestion volume of water by a small group of recreational swimmers in a pool environment. This pilot study was enabled by the widespread practice in warm climates of using chloroisocyanurate to disinfect swimming pool waters. The chlorinated compound, which is in equilibrium with the free chlorine and cyanuric acid in the pool water, provides a biomarker, cyanuric acid, that once swallowed passes through the body into the urine unchanged. The amount of ingested water can be calculated if the concentration of cyanuric acid in the pool and in a 24 h urine sample, as well as the volume of urine sample, is known. The results of this small pilot study showed that, on average, adults (greater than 18 years old) swallowed about 21 mL per hour and that children (18 years old or less) swallowed about 49 mL per hour. This small pilot study also showed that cyanuric acid in swimming pools could be used to estimate the amount of water ingested by recreational swimmers simply by measuring a swimmer’s urine for this chemical after a swimming event in a pool containing cyanuric acid.

Although the results of the pilot study demonstrated that the study design was appropriate, the small population size, especially the small number of adults, made translation of results to other and larger populations less straight forward. Therefore, a second follow-up study with a much larger population, representing different ages and genders of swimmers, was conducted. The purpose of this paper is to present the results of this large-scale study.

**Table 1** | Historical non-empirical estimates of water ingested by swimmers during swimming activity

| Source/Year                      | Ingestion estimate (mL per event) |
|----------------------------------|-----------------------------------|
| <i>Streeter</i> (1951)           | 10                                |
| <i>Steiniger</i> (1954)          | 50                                |
| <i>Shuval</i> (1975)             | 10                                |
| <i>Borneff</i> (1979)            | 50                                |
| <i>Rees</i> (1993)               | 10–15                             |
| WHO (2003)                       | 30–50                             |
| <i>Steyn &amp; Genthe</i> (2004) | 50                                |

## METHODS

### Study approach

The large-scale study was conducted using the same approach used in the pilot study (*Dufour et al.* 2006) that successfully measured the volume of water ingested by 57 swimmers while swimming. In the pilot study, swimmers

were exposed to pool water disinfected with chloroisocyanurate. The approach was as follows:

- All trials were conducted at pools where the water was disinfected with chloroisocyanurate.
- Swimmers were recruited a few days prior to the swim event.
- Swimmers were directed not to swim the day before the swim event.
- Swimmers were informed of how and why the study would be conducted. All swimmers or their parents were requested to sign consent or assent forms prior to being admitted to the study. Pool water samples were taken prior to and on the day of the swim event.
- On the day of the swim event, swimmers were directed to perform normal swimming activities for approximately 1 hour.
- On leaving the pool, swimmers were directed not to swim on the following day.
- On leaving the pool area, swimmers were instructed to pick up a urine collection container and to collect their urine over the next 24 hours.
- Swimmers were instructed to return the collected urine samples to a designated collection site and to collect their compensation for participating in the study.

### Study population

The demographic make-up of the study population is shown in Table 2. The total number of participants in the study was 549. The participants were about equally represented by males and females. The age groups, which were selected using tree analysis (that will be described later), included children aged 6–10, who comprised 12% ( $n = 66$ ) of the study population; the group labeled teens comprised individuals aged 11–15 and included 22% ( $n = 121$ ) of the study

**Table 2** | Study population by age and gender

| Age group | Male | Female |
|-----------|------|--------|
| 6–10      | 25   | 41     |
| 11–15     | 82   | 39     |
| 16+       | 170  | 192    |
| Total     | 277  | 272    |

population; and a group labeled adults comprised individuals 16 years old and older and made up 66% ( $n = 192$ ) of the study population. The labels children, teens, and adults will be used in the remainder of this paper to describe these three age groups.

### Study sites

The study was conducted at nine public swimming pools in the Columbus, Ohio area. The pools were selected based on their use of chloroisocyanurate to disinfect the pool water. The pool concentration of the chloroisocyanurate was maintained at a level of 30 to 50 mg/L, as suggested by the National Spa and Pool Institute (Tice 1997).

### Participation consent and pool safety

Swimmers were approached 2 to 3 days prior to each swim event to solicit their participation in the study. Informed consent of the participants or their legal representatives was obtained prior to the initiation of the study. Consent forms and the study design were reviewed and approved by the Battelle Human Studies Committee, and the US Environmental Protection Agency's Human Subjects Research Review Official, to ensure compliance with federal regulations governing human subjects' research. During the swimming activities a trained lifeguard was on duty.

### Sample collection

Pool water samples were collected from four locations around the pool prior to the start of the swimming activities. The sampling locations were on each side of the pool midway between the corners of the pool. 250 mL samples were collected at a depth of 25 cm in amber colored plastic bottles, which were transported to the laboratory on ice. At the laboratory, samples were transferred to a 4 °C refrigerator and analyzed within 1 week.

Urine samples were collected over a 24 hour period by each participant following the swimming event. Each participant was given a sterile 1 gallon, wide-mouthed container. The participants were directed to collect their urine over the next 24 hours and return the specimen containers to the study representative after that period of time. All

who agreed to participate returned urine samples. Urine sample data from two male and two female participants (0.7% of the total number) were discarded as outliers, corresponding to implied water ingestion in the range of 500–1,500 mL each.

### Assay methodology

The assay methods for measuring cyanuric acid in pool water have been described previously (Cantú *et al.* 2000, 2001a, 2001b; Magnuson *et al.* 2001). The pool water samples were collected, stored refrigerated, and analyzed within 1 week by HPLC methodology. The sample extraction procedure of cyanuric acid in urine samples is described elsewhere (Cantú *et al.* 2017). Briefly, to minimize biological and chemical degradation, the 24 hour urine sample was immediately preserved after collection by the addition of 1 mL of a reagent composed of 10% (v/v) perchloric acid and 1% (w/v) metaphosphoric acid. Initially, the urine was cleaned by centrifugation and solid phase extraction cartridges and eluted in aqueous form. The mixture was vortexed, and the phases were allowed to separate. An aliquot of the aqueous phase was transferred to the HPLC vial. In the vial, the pH of the sample adjusted to be compatible with the mobile phase and the sample was immediately injected into a porous graphitic column under operation of the assay methodology. The recovery efficiency for the determination of cyanuric acid from urine was calculated from urine spiked with 0.3 and 3.0 mg/L of cyanuric acid. The percent recoveries and standard deviations were respectively  $86\% \pm 7\%$  and  $93\% \pm 3\%$  for triplicate low and high level spikes, respectively (Cantú *et al.* 2017). Swimming pool samples spiked with cyanuric acid had a  $99.8\% \pm 3.0\%$  mean recovery for triplicate analysis of spike values ranging from 5 to 50 mg/L (Cantú *et al.* 2001b).

The volume of water ingested was calculated from the amount of cyanuric acid measured in the pool water and in the urine sample using the formula:

$$V^*_{\text{water ingested}} = V_{\text{urine collected}} \times \frac{U_{\text{CAconc}}}{P_{\text{CAconc}}}$$

where,  $V^*_{\text{water ingested}}$  = volume of pool water ingested;  
 $P_{\text{CAconc}}$  = cyanuric acid (CYA) concentration in pool water;

$V_{\text{urine collected}}$  = volume of urine collected over a 24 hour period; and  $U_{\text{CAconc}}$  = cyanuric acid (CA) concentration in urine.

### Data analysis

Statistical analysis of differences among age groups and between males and female swimmers was performed via SAS version 9 analysis of variance procedure (GLM, SAS Institute, NC) on log transformed mL of water ingested. Given that we did not have a firmly preconceived notion of what the relationship between ingestion and age should be or where cut-points might fall, a simple regression tree analysis (the recursive partitioning procedure from R package Rpart) was used to determine where reasonable cut-point values of age and age/gender might lie based on greatest reduction in within group standard deviation (R Core Team 2016). For illustrative purpose, empirical cumulative distributions of ingestion were developed by kernel density estimation using SAS version 9 separately for children, teens, and adults.

## RESULTS

Tree analysis was performed to determine natural breaks or cut-points in the amount of water ingested by the study population rather than making arbitrary groupings. The single greatest determination of where reasonable cut-points could be established was the age of the participants. Participants between the ages of 6 and 10 years of age (children) formed a natural cohort, as did the participants between the ages of 11 and 15 years old (teens), and participants 16 years old or older (adults). The only other natural grouping based on volume of water ingested was between gender in the greater than 16-year-old group. This result was used to define relevant age and gender groupings, which are summarized in Table 2.

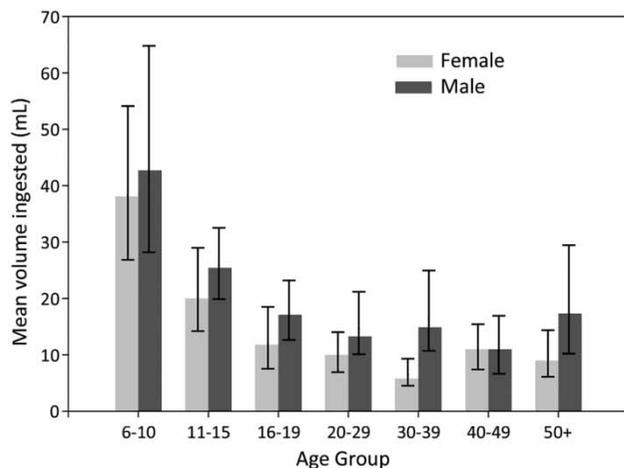
Although the data in this paper are described in terms of geometric means and confidence limits because the data are logarithmically distributed, it is appropriate to state the overall mean as an arithmetic mean in order to compare it to contemporary published data. In this case, the arithmetic mean of water ingested by all swimmers was 32 mL. The

**Table 3** | Geometric means and associated 95% confidence intervals for amount of water (mL) ingested by age group and gender

|                     | N              |                                | Geometric mean (mL) |                               | 95% CI   |  |
|---------------------|----------------|--------------------------------|---------------------|-------------------------------|----------|--|
| <b>All swimmers</b> | 549            |                                | 14                  |                               | 13 to 16 |  |
|                     | <b>Females</b> |                                | <b>Males</b>        |                               |          |  |
| <b>Age group</b>    | <b>N</b>       | <b>Geo. mean (mL) (95% CI)</b> | <b>N</b>            | <b>Geo. mean (mL) (95 CI)</b> |          |  |
| 6 to 10             | 41             | 38 (27–54)                     | 25                  | 43 (28–65)                    |          |  |
| 11 to 15            | 39             | 20 (14–29)                     | 82                  | 25 (20–33)                    |          |  |
| 16 to 19            | 24             | 12 (8–18)                      | 46                  | 17 (13–23)                    |          |  |
| 20 to 29            | 47             | 10 (7–14)                      | 39                  | 15 (10–21)                    |          |  |
| 30 to 39            | 36             | 6 (5–9)                        | 25                  | 16 (11–25)                    |          |  |
| 40 to 49            | 45             | 11 (7–15)                      | 30                  | 11 (7–17)                     |          |  |
| 50+                 | 40             | 9 (6–14)                       | 30                  | 17 (10–29)                    |          |  |

geometric mean ingested volume for all swimmers was 14 mL with 95% confidence limits of 13 to 16 mL (Table 3). In Table 3 the ingestion volumes are shown by age group and gender. The tree analysis indicated that a natural cohort was formed by swimmers over the age of 15 years. We segregated this group by 10 year age intervals and also separated all swimmers by gender. These ingestion results show that the two younger age groups swallow significantly more water than the adult group; this is uniformly true for males and females. The data in the Table 3 are further illustrated in Figure 1.

Study subjects were requested to spend an hour in the pool. However, the actual time spent in the pool, which

**Figure 1** | Geometric means and associated 95% confidence intervals for amount of water ingested by age and gender.

was self-reported, varied considerably, with the range being between 47 minutes and 104 minutes. Table 4 shows the geometric mean values of the duration of time spent in the water and their associated 95% confidence intervals of the self-reported time spent in the pool. The rate of ingestion was calculated, based on the four comparisons of primary interests, mainly among the three age groups, plus between adult men and women. Statistical significance of individual comparisons was determined by the Holms test to achieve an overall critical value  $p = 0.05$ , Bonferroni adjusted for multiple comparisons. Table 4 shows results from analysis of variance on log values with respect to ingestion, time spent in the water, and the rates of ingestion that were determined to be significant by this criterion. Among age groups, all are significantly different from each other with respect to the amount of water ingested ( $p < 0.001$  for adults versus either younger group). The volumes ingested by the two younger groups were also statistically different from each other ( $p < 0.007$ ). The amount of water ingested among males and females also differed statistically ( $p < 0.001$ ), with males ingesting a geometric mean of 13.7 mL while females ingested a geometric mean of 8.0 mL. Only the ingestion rates between children and teens and the time in the pool between females and males were not statistically significant.

The observed differences in the amount of water ingested may be influenced by which age group is reporting. Children spent nearly twice as long in the pool as teens or adults (Table 4). During the time they are in the water, the

**Table 4** | Geometric means and associated 95% confidence intervals for amount of water ingested, time spent in the pool, and implied ingestion rates by swimmer group

| Geometric means and 95% confidence intervals |        |                      |                               |                           |
|--|--------|----------------------|-------------------------------|---------------------------|
| Age group                                    | Gender | Amount ingested (mL) | Time in the pool (mL per min) | Ingestion rate (mL per h) |
| Children                                     | All    | 38.2 (28–52)         | 95.9 (88–104)                 | 23.9 (17–33)              |
| Teens  | All    | 22.1 (17–28)         | 55.8 (52–59)                  | 23.7 (19–30)              |
| Adults                                       | All    | 10.4 (9–12)          | 50.3 (49–52)                  | 12.4 (11–14)              |
|  | Female | 8 (7–10)             | 51.1 (49–54)                  | 9.4 (8–11)                |
|  | Male   | 13.7 (11–17)         | 49.5 (47–52)                  | 16.4 (13–20)              |

Differences between italic values are not significant. All other differences among age groups and gender are statistically significant at  $p < 0.01$ .

rate of ingestion implied by the combination of amount ingested and time in the water indicates that adults ingest less water per minute than those under age 16, while teens are seen to ingest water at the same as rate as children.

With respect to the adults alone, the difference in total ingestion between men and women is evidently due to their respective rates of ingestion, not time spent in the water (Table 4). Caution must be taken in interpreting these derived numbers too strictly, given they are based on self-reported data (minutes in the water); thus, they could be less reliable because of the anecdotal propensity of children, and even adults, to under- or overestimate how much time is spent in the water.

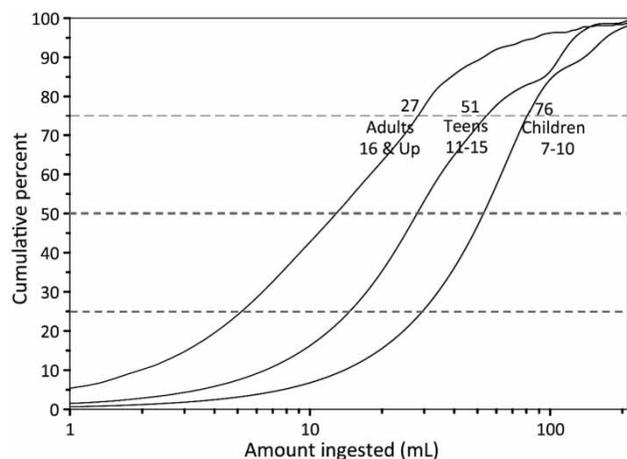
Figure 2 shows the cumulative distribution of individual ingested volumes plotted by age group. These plots measured amount ingested from least to greatest by the percent of subjects who ingested that amount or less for each of the three

age groups. The 75th percentile is shown for each age group. This indicates that 25% of the adult study participants ingested 27 mL of pool water or more, while for teens and children the corresponding amounts are 51 and 76 mL. The greatest volume of water ingested by children, teens, and adults, respectively, was 245 mL, 267 mL, and 279 mL.

## DISCUSSION

Behavioral activities of swimmers while in the water is an important aspect of determining their health risk. The major behaviors include the type of body immersion, whether it is partial or total, the duration of exposure and the amount of water ingested. During the conduct of epidemiological studies associated with swimmer health, it is critical to know if the body or head of the swimmer has been totally submerged or how much time the swimmer spent in the water. Knowledge of these behavioral characteristics allows investigators to more accurately interpret their findings (Wade et al. 2006, 2008, 2010). Similarly, investigators developing quantitative microbial risk assessments will be able to make better estimates of swimmer risk if they have data describing the amount of water an individual has swallowed while swimming. This need for ancillary information, especially water ingestion while swimming, is what has driven investigators in the past to make non-empirical estimates of the amount of water a swimmer might ingest while swimming.

Data developed during the large-scale study have, for the most part, confirmed and expanded the findings of the pilot study (Dufour et al. 2006). Results from the large-scale study show that children swallowed more water than adults



**Figure 2** | Cumulative percent of swimmers who ingested water in an amount less than the indicated quantity in mL.

while swimming. In the pilot study children (< or = to 18 years old) swallowed 2.3 times the volume swallowed by adults (>18 years old), while in the large-scale study, children (< or = to 10 years old) swallowed more than three times as much water as adults (>16 years old), and teens (11–15 years old) swallowed twice as much water as adults. In the large-scale study males swallowed 1.4 times as much water as females, whereas in the pilot study the ratio was 1.2 times as much. Interestingly, the arithmetic mean of water ingested by all participants was 32 mL (geometric mean 14 mL) in the large-scale study and 34 mL in the pilot study. The only data that were not similar in the large-scale study when compared to the pilot study were the ratio of water swallowed by men and women. In the pilot study, the ratio was 0.5, while in the large-scale study, the ratio was 1.7. This discrepancy was perhaps due to the small number of adults that participated in the pilot study.

It is of some interest that the historic non-empirical estimates that have been made were very similar to the estimates generated through experimentation. The historical estimates of water ingestion while swimming range from 10 mL to 50 mL, while the empirically produced average estimates reported by Dufour *et al.* (2006), Dorevitch *et al.* (2011), and Suppes *et al.* (2014), respectively, were 34 mL, 10 mL, and 14 mL. One might suggest that these studies have validated the non-empirical suggestions made over the last 65 years.

The overall results from the large-scale study are similar to other recently conducted studies. Dorevitch *et al.* (2011) estimated that pool swimmers swallowed an average of about 10 mL of water (arithmetic mean) with an upper confidence limit of 35 mL of water. The large-scale study arithmetic mean was 32 mL, which is within the confidence limits of the Dorevitch mean. The number of children in the Dorevitch study was too small to compare with the children in the large-scale study. Suppes *et al.* (2014) conducted a study of swimmer behavior regarding pool water ingestion. They found that the average volume of water swallowed by swimmers was 14 mL and that children swallowed about 7.3 times as much water as adults (27.5 mL versus 3.5 mL), whereas in the large-scale study the overall arithmetic mean of water ingested was 32 mL and the ratio of water swallowed in children versus adults was 2.0 (47 mL versus 24 mL).

The duration of time spent in the water may also contribute to the risk of contracting illness while swimming. Children on average spent about 1 hour and 15 minutes in the water, whereas adults on average spent about 50 minutes in the water. This greater time spent in the water could account, in part, for the larger volume of water swallowed by children. However, children and teens also had a higher rate of ingestion, implying swimming behaviors likely also play a role (anecdotally, children and teens are observed to be more active than adults). The greater ingestion of water is probably a combination of these behaviors. It should be kept in mind that duration of time spent in the water is a subjective measure, dependent on the self-reported estimate of each swimmer.

Among the upper quartiles of their respective groups, Figure 2 shows that children ingested almost twice as much water as adults 16 and older and 50% more than teens. Children, while comprising only 12% of the entire study population, made up 30% of those in the upper quartile, ingesting between 37 and 280 mL of water with an average of about 87 mL. It is interesting to speculate that children, since they swallow the most water, stay in the water the longest, and because their immune systems are not fully developed, would include most of the individuals in the 1 to 5% of all swimmers who suffer excess illness associated with swimming activities.

## CONCLUSIONS

- Swimmers on average ingest 32 mL/h (geometric mean is 14 mL) of water while swimming, with a range of 0 to 280 mL per hour.
- Children swallow almost four times as much water (38 mL/h) as adults (10 mL/h).
- Males have a tendency to swallow more water than females during swimming activities.
- Children and teens spend almost twice as much time in the water than adults.

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