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ORIGINAL PAPER

Viral dynamics and patterns of lysogeny in saline Antarctic lakes

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Abstract Antarctic lakes are extreme ecosystems with microbially dominated food webs, in which viruses may be important in controlling community dynamics. A year long investigation of two Antarctic saline lakes (Ace and Pendant Lakes) revealed high concentrations of virus like particles (VLP) (0.20–1.26 $\times$ 10^8 ml^{-1}), high VLP: bacteria ratios (maximum 70.6) and a seasonal pattern of lysogeny differing from that seen at lower latitudes. Highest rates of lysogeny (up to 32% in Pendant Lake and 71% in Ace Lake) occurred in winter and spring, with low or no lysogeny in summer. Rates of virus production (range 0.176–0.823 $\times$ 10^6 viruses ml^{-1} h^{-1}) were comparable to lower latitude freshwater lakes. In Ace Lake VLP did not correlate with bacterial cell concentration or bacterial production but correlated positively with primary production, while in Pendant Lake VLP abundance correlated positively with both bacterial cell numbers and bacterial production but not with primary production. In terms of virus and bacterial dynamics the two saline Antarctic lakes studied appear distinct from other aquatic ecosystems investigated so far, in having very high viral to bacterial ratios (VBR) and a very high occurrence of lysogeny in winter.

Keywords Antarctic lakes · Bacteria · Lysogeny · Primary production · Production · Viruses

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Introduction

The concept of the microbial loop established by Azam et al. (1983) led to a recognition of the important role of bacteria and their protozoan predators in aquatic ecosystems. Later the discovery of high concentrations of viruses (bacteriophage) in the sea (Bergh et al., 1989), suggested that viral attack and lysis of bacteria might be short-circuiting the microbial loop, by returning carbon to the organic carbon pool before it could be grazed by protozoa. Subsequent evidence showed that heterotrophic bacteria and cyanobacteria were infected by viruses, with up to 7% of bacterioplankton containing mature phage (Procter and Fuhrman 1990).

Viruses are now recognised as an important component in the microbial dynamics of planktonic environments worldwide (Fuhrman and Suttle 1993, Fuhrman 1999; Hewson et al. 2001; Pina et al. 1998; Thingstad et al. 1993, Womack and Colwell 2000; Weinbauer 2004). The indications are that viruses can contribute significantly to bacterial mortality. Estimates range from undetectable to 100%, but methods for measuring viral induced bacterial mortality have not progressed much over the last decade (Suttle 2005). Phytoplankton is also prone to infection, which can in some circumstances result in up to a 50% reduction of primary production (Suttle et al. 1990; Suttle 1992).

The role of viruses is more complex than simply causing the mortality of bacteria and phytoplankters and the recycling of carbon. Viruses play a role in maintaining clonal diversity of host communities directly, through gene transmission (transduction), and indirectly by eliminating numerically dominant host species (Jiang and Paul 1994; Wommack et al. 1999). Another potentially important role for viruses is as a source of food for flagellates (Suttle and Chen 1992).

The relationship between bacteriophage and their hosts is complex. Viruses may exist in a lysogenic condition in their hosts, replicating without the destruction of the host. This state may continue for many host generations until a factor, or factors trigger, the lytic cycle. Early studies suggested that the majority of marine viruses were lytic (Weinbauer and Suttle 1996; Wilcox and Fuhrman 1994), but more recently lysogeny has been found in both marine and freshwater systems ranging up 43% (Cochran and Paul 1998; Jiang and Paul 1998; Tapper and Hick 1998).

Antarctic lakes are characterised by plankton communities dominated by microbial plankton with few invertebrate metazoans and no fish (Laybourn-Parry 1997). While their microbial plankton has been investigated extensively, there have been few studies of viruses in these systems. Several of the earlier studies were short term (Kepner et al. 1998; Laybourn-Parry et al. 2001), but in recent years several studies spanning a year have been published (Madan et al. 2005; Säwström et al. 2006) providing a much more detailed insight into the functional dynamics of viruses in Antarctic lake ecosystems. The current study of two contrasting oligotrophic, saline lakes in the Vestfold Hills was undertaken over a year and aimed to understand the role of viruses in the annual cycle of these lakes and their relationship to bacterial production and primary production.

Methods

Sampling

Ace Lake and Pendant Lake are situated in the Vestfold Hills (68°S 78°E). Meromictic Ace Lake has an area of 0.16 km^2 and a maximum depth of 20 m with a chemocline at around 10 m. The mixolimnion has a salinity of 18 ‰ while the lower monimolimnion has a salinity of 34‰. Neighbouring Pendant Lake is unstratified and has a salinity of 16‰. It has a similar area to Ace Lake and a maximum depth of 12 m. Both lakes were sampled with a Perspex Kemmerer bottle at their deepest point between January and December 2003, through a hole drilled in the ice with a Jiffy Drill. Both lakes retained most of their ice-cover during the study: thickness ranged between 0.51 and 1.76 m in Pendant Lake and 0.35–1.73 m in Ace Lake. Access to the lakes was not possible between February and June because helicopters were unavailable. Single water samples were collected from the water column of Pendant Lake at 2 m (immediately under the ice), 4, 6 and 8 m while in Ace Lake the mixolimnion was sampled at 2, 4, 6, 8 m and just above the chemocline at 10 m. For bacterial and viral counts 50 ml of water from each depth sample was fixed in glutaradehyde to a final concentration of 2%. On each sampling occasion depth profiles of water temperature (with a field temperature probe) and photosynthetically active radiation (with a LI-COR L1-189 quantum radiometer/photometer) were made to recreate field conditions in the laboratory incubations.

Analysis of samples

Aliquots of glutaradehyde fixed material were stained with DAPI (4′,6-diamidino-2-phenylindole, Sigma) and filtered onto 0.2 μm black polycarbonate filters (Millipore) (made up to a stock solution of 100 μg ml^{-1}) and bacteria numbers enumerated under epifluorescence microscopy with UV excitation (365 nm) at 1,600×. On each preparation 50 cells were measured with a Patterson graticule and cell volumes converted to carbon using a conversion factor of 0.20 pg C μm^3 (Bratbak and Dundas 1984). Virus like particles (VLP) were enumerated from 2 ml aliquots of glutaradehyde fixed material filtered onto 0.02 μm Whatman Anodisc filters and stained with SYBR green 1 (Molecular Probes) following the protocol of Noble and Fuhrman (1997). Viruses were enumerated under epifluorescence microscopy using a blue filter (450–490 nm) as above. All preparations were made within 24 h of sample collection.

Bacterial production and primary production

Samples for bacterial and primary production were collected from 2 m (just underneath the ice), 4, 6 and 8 m in both lakes in a black Perspex Kemmerer bottle to eliminate light shock. Samples were returned to the laboratory by helicopter in summer and overland vehicle in winter and experiments set up within 2 h of collection. Bacterial production was measured by following the incorporation of [^{14}C] leucine (Kirchman 1993). For each depth, four experimental 20 ml and two control incubations with 10 nM [^{14}C] leucine were run in Whirlpaks for 90 min at field light and temperature regimen. The reaction was terminated by the addition of 0.6 ml formalin to a final concentration of 4% and ice-cold trichloroacetic acid (TCA) to a final

concentration of 10%. Samples were filtered through 0.22 μm cellulose acetate filters (Millipore) and washed with two volumes (5 ml) of 5% ice-cold TCA. The filters were dissolved with 1 ml of ethyl acetate, 10 ml of scintillation fluid added and counts conducted in a Beckman LS6500 scintillation counter. A conversion factor of 1.42×10^{17} cells mol^{-1} for the incorporation of leucine into protein was applied (Chin-Leo and Kirchman 1988). Bacterial carbon values were determined as described as above.

Primary production was measured by the incorporation of ^{14}C using the modified protocol of Steeman-Nielsen (1951; 1952). A 100 ml water sample from each depth was dispensed into a plastic bottle and 25 μCi ^{14}C–HCO_3 was added (specific activity 54 mCi $mmol^{-1}$ sodium bicarbonate, Amersham). Control samples (7 ml aliquots) were immediately removed to scintillation vials and acid (6 N HCl) killed to check isotope addition (stabilised with NaOH). From the remaining sample five 10 mL replicate aliquots were removed and each incubated in Whirlpaks for 8 h under light and temperature conditions reflecting those prevalent in the lakes. The incubations were terminated by the addition of 200 ml of 6N HCl and 7 ml aliquots transferred to scintillation vials and shaken for 3 h to drive off unincorporated CO_2. Scintillation fluid (10 ml) was then added and the samples counted. Total inorganic carbon in the water at each depth was determined by Gran titration.

Determination of lysogeny

Integrated samples were collected from 2, 4, 6 and 8 m in Pendant Lake and 2, 4, 6, 8 and 10 m in Ace Lake. The samples were divided into six replicate aliquots. Mitomycin C was added at a concentration of 1 μg ml^{-1} to three of the replicates while the other three acted as controls. Experiments were incubated in the laboratory under field temperatures for 3 days and were terminated by the addition of glutaradehyde (final concentration 2%). Long incubation times are necessary at low the low temperatures that prevail in Antarctic lakes. Samples were processed for counts of VLP and bacteria as outlined in Analysis of samples above. In calculating the percent of lysogenic bacteriophage we applied a burst size of 24, which is an average value derived from a number of studies across a range of environments (Wommack and Colwell 2000).

Viral production

During the course of the year we attempted the measurement of viral production using TdR incorporation into TCA-insoluble, deoxyribonuclease (DNase)-resistant material < 0.2 μm in size, using the method of Steward et al. (1992), modified by Noble and Fuhrman (1997). We failed to obtain consistent reliable data throughout the study period. Consequently during the summer of 2003 (December) we adopted a different approach and used a dilution technique developed by Wilhelm et al. (1998) modified by Hewson et al. (2001). Virus free water was obtained by filtering lake water (integrated from the water column) through 0.02 μm Whatman Anodisc filters. This water had been prefiltered through 1.0 μm polycarbonate filters (Millipore) to facilitate filtration through 0.02 μm filters. One litre of water from integrated depth samples was passed through a 0.22 μm Duropore filter (GV, low protein-binding, Millipore) to remove viruses. One litre of virus free water was added to the retentate and bacteria and phytoplankton were suspended from the filter by gentle washing with a pipette. This water was then divided into five 200 ml replicates contained in 500 ml acid washed Duran bottles. The bottles were incubated under field temperature and light conditions. Ten 13 ml subsamples were removed at 0, 3, 6, 9, 12, 22 and 25 h intervals and analysed for viruses and bacteria concentrations as outlined in Analysis of samples above.

Results

Bacterial production rates were higher in Ace Lake than in Pendant Lake in November and December but similar at other times during the study (Fig. 1). However, overall bacteria cell concentrations were lower in Ace Lake (Fig. 2). Bacterial production was measurable in winter, but the highest rates in both lakes occurred in spring and summer and did not correlate with highest bacterial concentrations (Figs. 1, 2). This anomaly can be explained by considering mean cell volume and biomass, highest biomass occurred in Ace Lake in December and in Pendant Lake in January in concomitant analyses (Madan et al., 2005). Generally highest production coincided with largest biomass.

Primary production showed no clear correlation with bacterial production (Pendant Lake $r = 0.30$ not significant; Ace Lake $r = 0.50$ not significant), although in these lakes it is the sole source of carbon input. There are no significant allochthonous inputs from the catchment; all drainage is derived from snow melt. On average primary production was higher in Pendant Lake, though maximum levels, which occurred in spring and summer, were of the same order of magnitude (Ace Lake 8.15 μg l^{-1} h^{-1} ± 4.0 and Pendant Lake 6.34 μg l^{-1} h^{-1} ± 3.1) (Fig. 1).

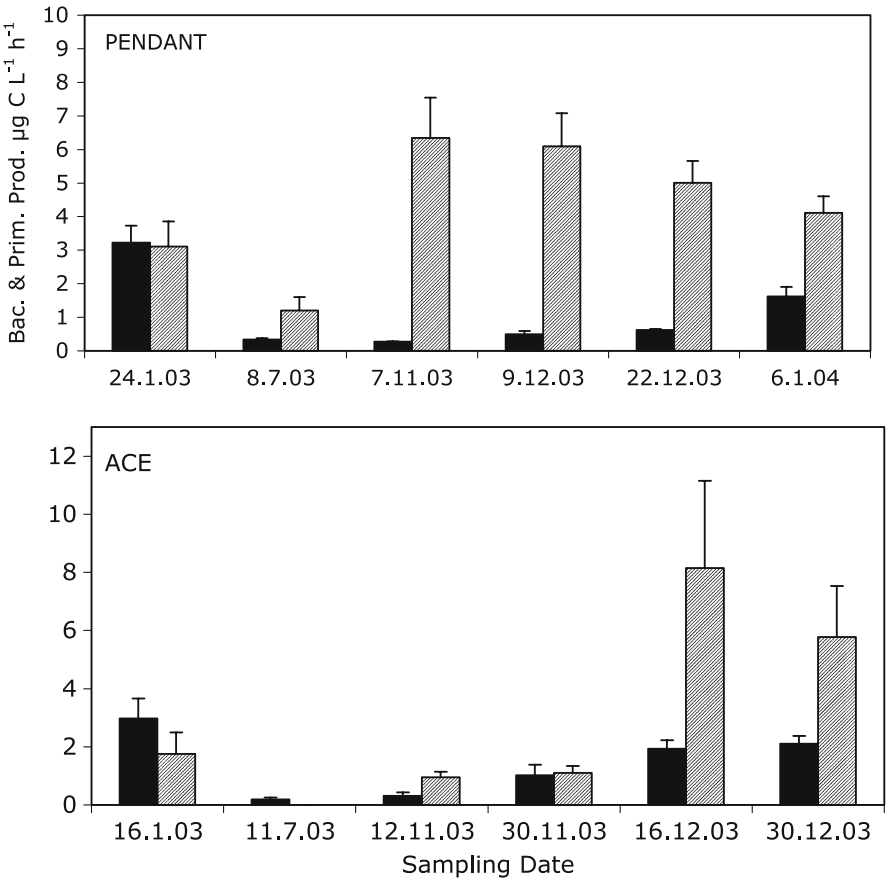

Fig. 1 Bacterial and primary production in Pendant Lake and Ace Lake during 2003 with standard errors. *Solid columns* bacterial production, *hatched columns* primary production. In Ace Lake the mean is derived from five depths in the water column; in Pendant Lake it is derived from four depths

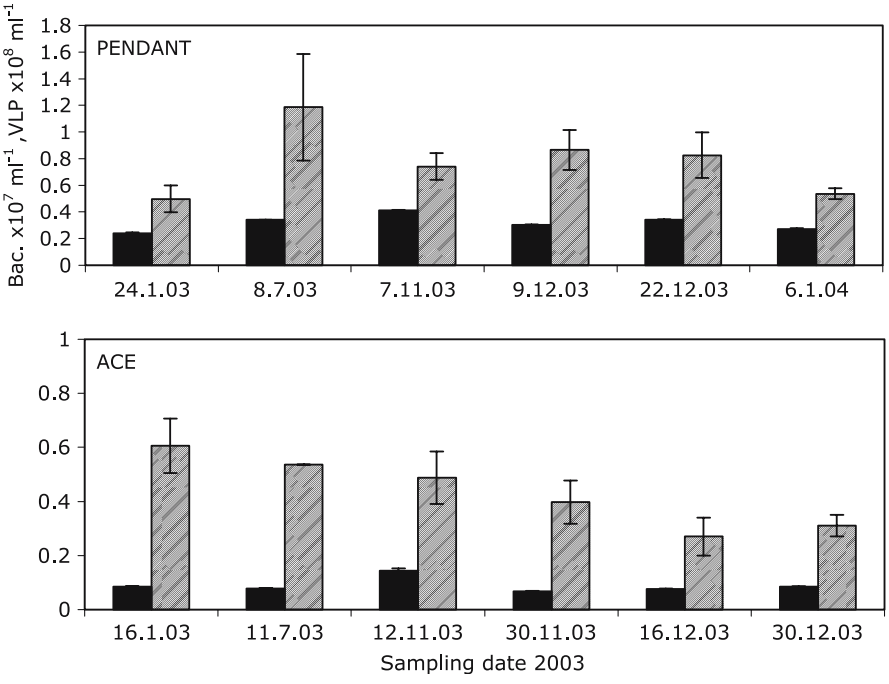

Fig. 2 Virus like particle (*VLP*) and bacterial abundances over 2003 in Pendant Lake and Ace Lake with standard errors. *Solid columns* bacteria, *hatched columns* VLP

Even in winter there was measurable photosynthesis in Pendant Lake.

VLPs ranged between 0.20 and 1.26×10^8 mL^{-1} in Pendant Lake and 0.27–1.00×10^8 ml^{-1} in Ace Lake (Fig. 2). Viral numbers exhibited no correlation with bacterial cell concentrations or bacteria production in Ace Lake but did show a positive correlate with primary production ($r = 0.73$, $P < 0.001$, $n = 28$). In

contrast in Pendant Lake viral abundance correlated with bacterial production and bacterial cell concentrations ($r = 0.75$, $P < 0.001$ and 0.570, $P < 0.001$, respectively) but not with primary production ($r = 0.30$).

In both lakes viral production declined during December towards mid-summer by 24.5% in Ace Lake and by 41.5% in Pendant Lake (Table 1). Viral production normalised to a bacterial cell indicated significantly higher production in Ace Lake (Table 1). This probably relates to higher VLP: bacterial ratios in this lake compared to Pendant Lake (Fig. 3).

The percentage of lysogenic bacteriophage determined from the induction of lysis with Mitomycin C, showed a clear seasonal pattern (Fig. 4). Highest levels of lysogeny occurred in winter and spring and declined towards summer. The percent of lysogeny reached a high level 71% in Ace Lake whereas in Pendent Lake the maximum level was 32%. In late summer (January and February) no lysogeny was detected. High levels of VLP in the water column corresponding to times of high lysogeny suggest that during winter, both the lytic cycle and lysogenic cycle were operating within the bacterioplankton community that was maintaining growth, albeit at reduced levels compared to summer (Fig. 1). The reduction in lysogeny may have been a response to increasing temperatures and photosynthetically active radiation (PAR) as the ice cover was lost, or elevated bacterial production, or a combination of factors. Late summer is typified by break up and melt of the lake ice covers and increasing temperatures from around −0.9°C in winter to 2.5°C in February (Madan et al., 2005).

Table 1 Viral production during December 2003 in Ace Lake and Pendant Lake

Lake	Date	Viral production $\times 10^6$ ml^{-1} h^{-1}	Viral production bacterium^{-1} h^{-1}
Ace lake	1 December	0.823	4.88
	16 December	0.202	3.20
Pendant	10 December	0.424	3.92
lake	23 December	0.176	2.16

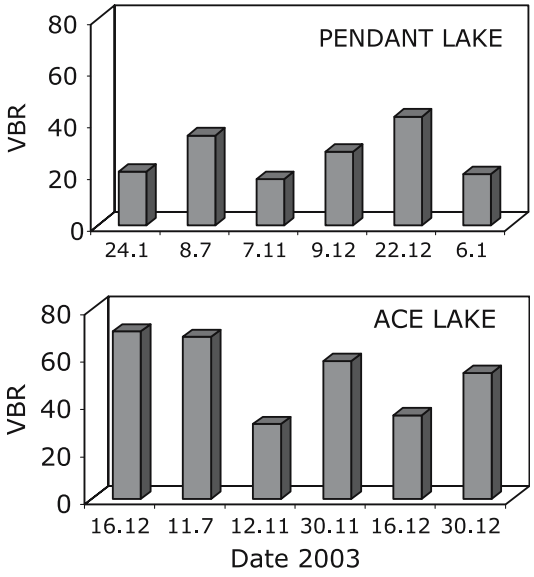

Fig. 3 Virus: bacterial ratios (*VBR*) in Pendant Lake and Ace Lake during 2003

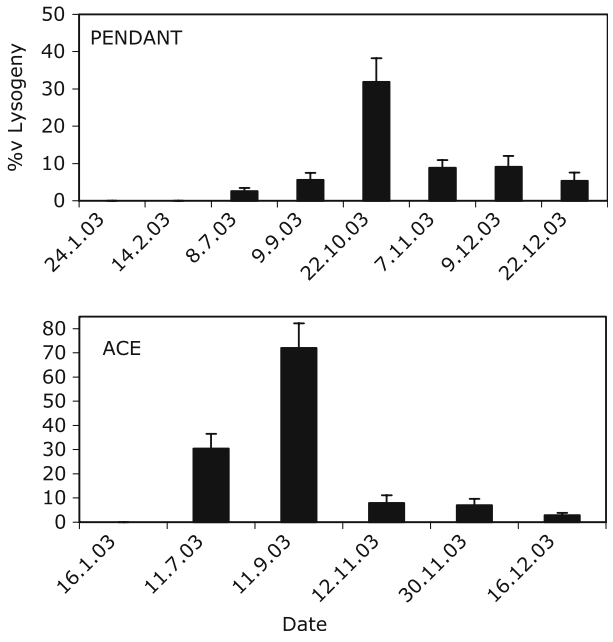

Fig. 4 Percent of lysogenic phage in Pendant Lake and Ace Lake during 2003 with standard errors

Discussion

Salinity appears to be one of the factors controlling VLP abundances. A negative correlation between salinity and viral direct counts was demonstrated along the Brisbane River and estuary, which did not necessarily correlate with bacterial cell abundance (Hewson et al., 2001) and in Tampa Bay, where seasonal salinity changes in relation virus abundances were followed (Jiang and Paul 1994). A review of VLP abundances in Quebec freshwater lakes and marine environments supports this pattern, as freshwaters had higher VLP numbers (range 4.1×10^7–2.5×10^8 ml^{-1}) compared to marine waters (range 6.7×10^4–7.1×10^7 ml^{-1}) (Maranger and Bird 1995). Saline lakes are rather different environments to estuaries, even though they may, as in the case of Ace Lake and Pendant Lake, be around half the salt concentration of sea water (16–18 ‰). In these lakes the range of VLP was

0.27×10^8–1.26×10^8 ml^{-1}, whereas at comparable salinities in the Brisbane and Noosa estuaries the maximum concentrations of VLP were 0.1×10^8 ml^{-1} and 0.24×10^8 ml^{-1}, respectively (Hewson et al., 2001) and in Tampa Bay the range was 0.02×10^8 ml^{-1}–0.32×10^8 ml^{-1} (Jiang and Paul 1994). Ace Lake and Pendant Lake can be classified as oligotrophic (Bell and Laybourn-Parry 1999). Thus while salinity may be a factor that plays a part in determining VLP abundances, there is clearly something about saline lakes, and particularly Antarctic lakes, that sets them apart irrespective of trophy or salinity.

Most data on viral production rates derive from marine environments and range from 0–9.58×10^6 ml^{-1} h^{-1} across high to low latitudes (Fuhrman and Noble 1995; Hewson et al. 2001; Steward et al. 1992, 1996). There are few data on oligotrophic lakes, either saline or freshwater, to enable comparisons. Viral production measured during the summer in Ace and Pendant Lakes declined over December (Table 1). Kepner et al. (1998) found rates of 2.04×10^6 viruses ml^{-1} h^{-1} in Lake Hoare (Dry Valleys, Antarctica) using a radiochemical method, but they did caution that their data may not be entirely reliable. A recent detailed investigation of ultra-oligotrophic Antarctic freshwater lakes revealed lytic viral production rates of 0.0033–0.029 viruses ml^{-1} h^{-1} in Crooked Lake and 0.0019–0.0098 viruses ml^{-1} h^{-1} in Lake Druzhby (Säwström et al. 2006). These rates are many times lower than those found either in the current study or Lake Hoare by Kepner et al. (1998), however Crooked Lake and Lake Druzhby are extremely oligotrophic with chlorophyll *a* concentrations usually well below 0.5 μg l^{-1} (Säwström et al. 2006). In freshwater temperate lakes in the French Massif Central oligotrophic/mesotrophic Lake Pavin had a estimated mean viral production rate of 0.079×10^6 viruses ml^{-1} h^{-1}, while in eutrophic Lake Aydat the rate was 0.424×10^6 viruses ml^{-1} h^{-1} between March and December 2000 (Bettarel et al., 2004). The limited current data suggest that in eutrophic aquatic environments lytic viruses have short turnover times compared with oligotrophic waters. Our data and that of Kepner et al. (1998) only cover a short time in summer, so temporal impacts cannot be assessed, however, in ultra-oligotrophic Crooked Lake and Lake Druzhby highest lytic viral production rates occurred in spring, but these lakes differed from the saline lakes in the current study in that lysogenic production contributed a maximum of only 1% of total viral production (Säwström et al. 2006).

The percentage of lysogenic bacteriophage displayed a clear seasonal pattern in both lakes, with low levels in early to mid summer and undetectable lysogeny in late summer. Highest levels of lysogeny occurred in winter and spring. The levels of lysogeny were at the higher end of values reported in the literature. In marine systems lysogenic bacteriophage ranged from undetectable to 100% (Weinbauer 2004), in oligotrophic Lake Superior between 0.1–7.4% (Tapper and Hick 1998) and in a Canadian fjord up to 80% (Ortman et al. 2002).

There are conflicting data on the seasonal occurrence of lysogeny. A seasonal pattern over 13 months was observed in a sub-tropical estuary, where prophage induction was recorded in the summer months at temperatures above 19°C with up to 41% of bacteria lysogenic (Cochran and Paul 1998). In Lake Superior a slightly larger proportion of the bacterial community contained lysogenic prophage in July and August compared with October (Tapper and Hick 1998). However, studies in the Gulf of Mexico found that the percentage of lysogenic bacteria was not correlated with temperature (Weinbauer and Suttle 1996, 1999) and a investigations in the Baltic and Mediterranean Seas demonstrated levels of lysogeny in excess of 50% at temperatures below 15°C (Weinbauer et al. 2003). Of particular interest is the apparent lack of any significant lysogeny in ultra-oligotrophic freshwater Antarctic Lakes where temperatures rarely rise above 4°C (Säwström et al. 2006). Thus temperature and its impact on productivity, does not appear to be a critical factor in determining the degree of lysogenic viral production in Antarctic lakes. A recent review has argued that the relationship between lysogeny and the trophic status of habitats remains to be properly elucidated (Weinbauer 2004). The evidence is conflicting, however higher levels of lysogeny are more usual in less productive environments, as was the case in the current study.

The VBR in Ace Lake and Pendant Lake were within the range 17.9–70.6 with the highest values in Ace Lake. A survey of marine and freshwater systems revealed significantly higher VBR in the latter (Maranger and Bird 1995). However, when they conducted their survey there were few data for oligotrophic freshwater systems. Recent work on two French Massif Central oligotrophic lakes revealed mean VBRs of 7 and 9 Bettaral et al. 2003). Our values are higher than those for marine systems (mean 1–5, maximum 53.8) and overlap values for freshwater systems (mean 20–25, maximum 77.5). Maranger and Bird (1995) quite reasonably predicted that VBR in saline lakes would be low, but that view is not supported by our data nor data from a short term summer survey of freshwater and saline lakes in the Vestfold Hills

(Laybourn-Parry et al. 2001). In common with Antarctic saline lakes, the freshwater lakes fall outside the predicted norm for VBR. In Crooked Lake and Lake Druzhby VBR over an annual cycle ranged from 1.2 to 8.4 (Säwström et al. 2006).

The two saline lakes in the current study differ from other lacustrine systems studied to date in having high VBR and high lysogeny during winter. Placed in the context of lakes worldwide the saline lakes in this study and the freshwater lakes in the Vestfold Hills (Säwström et al. 2006) have marked differences. These groups of saline and freshwater lakes in the Vestfold Hills are systems dominated by microbial food webs where viral dynamics probably play a role in biogeochemical cycling and microbial community dynamics. Viral mediated processes demand further study.

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References

Azam F, Fenchel T, Field JG, Gray JS, Meyer-Reil RA, Thingstad F (1983) The ecological role of water column microbes in the sea. Mar Ecol Prog Ser 10:257–263

Bell EM, Laybourn-Parry J (1999) Annual Plankton dynamics in an Antarctic saline lake. Freshw Biol 41:507–519

Bergh Ø, Børsheim KY, Bratbak G, Heldal M (1989) High abundance of viruses found in aquatic environments. Nature 340:467–468

Bettarel Y, Sime-Ngando T, Amblard C, Carrias JF, Portelli C (2003) Virioplankton and microbial communities in aquatic systems: a seasonal study in two lakes of differing trophy. Freshw Biol 48:810–822

Bettarel Y, Sime-Ngando T, Amblard TC, Dolan J (2004) Viral activity in two contasting lake ecosystems. Appl Environ Microbiol 70:2941–2951

Bratbak G, Dundas I (1984) Bacterial dry matter content and biomass estimations. Appl Environ Microbiol 48:755–757

Chin-Leo G, Kirchman DL (1988) Estimating bacterial production in marine waters from the simultaneous incorporation of thymidine and leucine. Appl Environ Microbiol 54:1934–1939

Cochran PK, Paul JH (1998) Seasonal abundance of lysogenic bacteria in a subtropical estuary. Appl Environ Microbiol 64:2308–2312

Fuhrman JA (1999) Marine viruses and their biogeochemical and ecological effects. Nature 399:541–548

Fuhrman JA, Suttle CA (1993) Viruses in marine planktonic systems. Oceanography 6:51–63

Fuhrman JA, Noble RT (1995) Viruses and protists cause similar bacterial mortality in coastal seawater. Limnol Oceanogr 40:1236–1242

Hewson I, O'Neil JM, Fuhrman JA, Dennison WC (2001) Virus-like particle distribution and abundance in sediments and overlying waters along eutrophication gradients in two subtropical estuaries. Limnol Oceanogr 46:1734–1746

Jiang SC, Paul JH (1994) Seasonal and diel abundances of viruses and occurrence of lysogeny/bacteriocinogeny in the marine environment. Mar Ecol Prog Ser 104:163–172

Jiang SC, Paul JH (1998) Gene transfer by transduction in the marine environment. Appl Environ Microbiol 64:2780–2787

Kepner RL, Wharton RA, Suttle CA (1998) Viruses in Antarctic lakes. Limnol Oceanogr 43:1754–1761

Kirchman D J (1993) Leucine incorporation as a measure of biomass production by heterotrophic bacteria. In: Kemp BF, Sherr EB, Cole JJ (eds) Handbook of methods in aquatic microbial ecology. Lewis Publishing, Ann Arbor, pp 509–512

Laybourn-Parry J (1997) The microbial loop in Antarctic lakes. In: Howard-Williams C, Lyons W, Hawes I (eds) Ecosystem processes in Antarctic ice-free landscapes. A.A. Balkema/Rotterdam/Brookfield, Rotterdam, pp 231–240

Laybourn-Parry J, Hofer JA, Sommaruga R (2001) Viruses in the plankton of freshwater and saline Antarctic lakes. Freshw Biol 46:1279–1287

Madan NJ, Marshall WA, Laybourn-Parry J (2005) Virus and microbial loop dynamics over an annual cycle in three contrasting Antarctic lakes. Freshw Biol 50:1291–1300

Maranger R, Bird DF (1995) Viral abundance in aquatic systems: a comparison between marine and fresh waters. Mar Ecol Prog Ser 121:217–226

Noble RT, Fuhrman JA (1997) Virus decay and its causes in coastal waters. Appl Environ Microbiol 63:77–83

Noble RT, Fuhrman JA (1998) Use of SYBR Green 1 for rapid epifluorescence counts of marine viruses and bacteria. Aquat Microbial Ecol 14:113–18

Ortman A, Lawrence J, Suttle C (2002) Lysogeny and lytic viral production during a bloom of the cyanobacterium *Synechococcus* spp. Microbial Ecol 43:225–231

Pina S, Creus A, Gonzalez N, Girones R, Felip M, Sommaruga R (1998) Abundance, morphology and distribution of planktonic virus-like particles in two high mountain lakes. J Plankt Res 20:2413–2421

Procter LM, Fuhrman JA (1990) Viral mortality of marine bacteria and cyanobacteria. Nature 343:60–62

Säwström C, Anesio A M, Granéli W, Laybourn-Parry J (2006) Seasonal viral loop dynamics in two large ultra-oligotrophic Antarctic freshwater lakes. Microbial Ecol (in press)

Steeman-Nielsen E (1951) Measurement of the production of organic matter in the sea. Nature 167:684

Steeman-Nielsen E (1952) The use of radioactive carbon (^{14}C) for measuring organic matter in the sea. J Cons Perm Int Explor Mer 18:117–140

Steward GF, Wikner J, Cochlan WP, Azam F (1992) Estimation of virus production in the sea, I. Method development. Mar Microb Food Webs 6:57–78

Steward GF, Smith DC, Azam F (1996) Abundance and production of bacteria and viruses in the Bering and Chukchi seas. Mar Ecol Prog Ser 131:287–300

Suttle CA (1992) Inhibition of photosynthesis in phytoplankton by the submicron size fraction concentrated from seawater. Mar Ecol Prog Ser 87:105–112

Suttle CA (2005) Viruses in the sea. Nature 437:356–361

Suttle CA, Chen F (1992) Mechanisms and rates of decay of marine viruses. Appl Environ Microbiol 58:3721–3729

Suttle CA ,Chan AM, Cottrell MT (1990) Infection of phytoplankton by viruses and reduction of primary productivity. Nature 387:467–469

Tapper MA, Hick RE (1998) Temperate viruses and lysogeny in Lake Superior bacterioplankton. Limnol Oceanogr 43:95–103

Thingstad TF, Heldal M, Bratbak G, Dundas I (1993) Are viruses important partners in microbial food webs? Trends Evol Ecol 8:209–212

Weinbauer MG (2004) Ecology of prokaryotic viruses. FEMS Microbiol Rev 28:127–181

Weinbauer MG, Suttle CA (1996) Potential significance of lysogeny to bacteriophage production and bacterial mortality in coastal water of the Gulf of Mexico. Appl Environ Microbiol 62:4374–4380

Weinbauer MG, Suttle CA (1999) Lysogeny and prophage induction in coastal and offshore bacterial communities. Aquat Microbial Ecol 18:217–225

Weinbauer MG, Brettar I, Höfle M (2003) Lysogeny and virus-induced mortality of bacterioplankton in surface, deep and anoxic waters. Limnol Oceanogr 48:1457–1465

Wilcox RM, Fuhrman JA (1994) Bacterial viruses in coastal seawater: lytic rather than lysogenic production. Mar Ecol Prog Ser 114:35–45

Wilhelm SW, Brigden S, Suttle CA (1998) The role of viruses in organic carbon cycling in the sea. Eos 79;OS168

Wommack KE, Ravel J, Hill RT, Colwell RR (1999) Population dynamics of Chesapeake Bay virioplankton: total community analysis by pulsed-field gel electrophoresis. Appl Environ Microbiol 65:231–240

Wommack KE, Colwell RR (2000) Virioplankton: viruses in aquatic ecosystems. Microb Mol Biol Rev 64:69–114