Fig. 4 Southern blot analysis of *Pst*I fragments of *S. coelicolor* plasmids. Lanes 1, pIJ601; lanes 2, SCP1(410 kb); lanes 3, SCP1(440 kb); lanes 4, SCP1(470 kb); lanes 5, pM138; lanes 6, pM146. Plasmids, SCP1s(410, 440 and 470 kb), pM138 and pM146 were isolated from *S. coelicolor* A3(2), M138 and M146, respectively as in Fig. 1. These plasmids and pIJ601 were digested with *Pst*I and subjected to conventional agarose gel electrophoresis and Southern blot analysis with ³²P-labelled pIJ601 as a probe.

DNA remained at the origin. Southern hybridization analysis confirmed this interpretation; ³²P-labelled pKSL hybridized to all chromosomal DNA bands but not to that of PR2-45. On the other hand, contaminating degraded chromosomal DNA in the probe hybridized weakly to all intact chromosomal DNAs remaining at the origin. These results clearly indicate that large linear DNA cannot be separated from degraded chromosomal DNA in this system.

That the presence of pKSL is related to antibiotic production in *S. lasaliensis* suggests that this plasmid is involved in the production of lasalocid and/or echinomycin. We further searched for and detected giant linear plasmids from five other strains in which antibiotic production is postulated to be plasmid determined including *S. coelicolor* A3(2) (ref. 15).

Next, we tried to test for localization of antibiotic biosynthetic genes on giant linear plasmids using the *S. coelicolor* system, because the cloned methylenomycin biosynthetic (*mmr*) and resistant (*mmr*) genes are known to form a gene cluster on a unisolatable plasmid, SCP1 (ref. 13). OFAGE analysis (Fig. 3) surprisingly revealed that the wild-type strain, *S. coelicolor* A3(2) contains a series of giant linear plasmids (410–560 kb) with a size difference of 30 kb, whereas two other SCP1⁺ strains, *S. coelicolor* M138 and M146 have only one plasmid of 350 kb. On the other hand, the two SCP1[−] strains *S. coelicolor* M124 and M130 have no plasmid: Probe pIJ601, which contains the *mmr* gene, hybridized to all plasmid bands regardless of their lengths. Furthermore, *Pst*I digestion revealed that all plasmids contained the same 2.6-kb *mmr* fragment (Fig. 4); the larger fragment in lane 5 might be formed by partial digestion of the plasmid. These results demonstrate that both a series of giant linear plasmids in *S. coelicolor* A3(2) and a 350-kb plasmid in *S. coelicolor* M138 and M146 are SCP1 itself. Variation of SCP1 in *S. coelicolor* A3(2) might be formed by multiplication of a 30-kb segment of a 350-kb plasmid, a possibility which should be studied further. We have revealed for the first time the physical identity of SCP1 and confirmed the presence of the methylenomycin biosynthetic genes on it. However, we cannot exclude the possibility that SCP1 is present at the same time in an integrated form in *S. coelicolor* A3(2), because the probe hybridized weakly to the origin of lane 9 in Fig. 3.

We found giant linear plasmids in a total of six antibiotic producing strains of *Streptomyces* by the newly developed OFAGE technique. Hayakawa *et al.*¹ reported a linear plasmid (pSLA2 of 17 kb) from *S. rochei* 7434-AN4, a producer of lankacidins, which has been the only linear plasmid isolated from *Streptomyces*. The sizes of linear plasmids so far isolated, including pSLA2, range from 5.4 to 17 kb. Therefore, the plasmids we describe here are the first examples of giant linear plasmids from prokaryotes or eukaryotes; the size of pKSL (520 kb) is one-twentieth that of chromosomal DNA (10⁴ kb)¹⁶. This new type of genetic structure is very interesting in view of the many unstable genetic characters of *Streptomyces* such as aerial mycelium formation¹⁷, pigment production¹⁸, drug resistance¹⁹ and arginine biosynthesis²⁰ in addition to antibiotic production^{21,22}. The relation between a giant linear plasmid and antibiotic production in *S. lasaliensis* is circumstantial, but it

has been confirmed in *S. coelicolor*. The giant linear plasmids may function not only in *S. coelicolor* but also in other strains in which antibiotic production is postulated to be plasmid determined.

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- Hayakawa, T., Tanaka, T., Sakaguchi, K., Otake, N. & Yonehara, H. *J. gen. appl. Microbiol.* **25**, 255–260 (1979).
- Hirochika, H., Nakamura, K. & Sakaguchi, K. *EMBO J.* **3**, 761–766 (1984).
- Francou, F. *Molec. gen. Genet.* **184**, 440–444 (1981).
- Gunge, N., Tamaru, A., Ozawa, F. & Sakaguchi, K. *J. Bact.* **145**, 382–390 (1981).
- Palmer, J. D., Shields, C. R., Cohen, D. B. & Orton, T. J. *Nature* **301**, 725–728 (1983).
- Levings, C. S. III & Sederoff, R. R. *Proc. natn. Acad. Sci. U.S.A.* **80**, 4055–4059 (1983).
- Kirby, R., Wright, L. F. & Hopwood, D. A. *Nature* **254**, 265–267 (1975).
- Wright, L. F. & Hopwood, D. A. *J. gen. Microbiol.* **95**, 96–106 (1976).
- Carle, G. F. & Olson, M. V. *Nucleic Acids Res.* **12**, 5647–5664 (1984).
- Chater, K. F. & Bruton, C. J. *Gene* **26**, 67–78 (1983).
- Chater, K. F. & Bruton, C. J. *EMBO J.* **4**, 1893–1897 (1985).
- Westley, J. W., Evans, R. H. Jr, Williams, T. & Stempel, A. *JCS Chem. Commun.* 71–72 (1970).
- Keller-Schierlein, W., Mihailovic, M. Lj. & Prelog, V. *Helv. chim. Acta* **42**, 305–322 (1959).
- Carle, G. F. & Olson, M. V. *Proc. natn. Acad. Sci. U.S.A.* **82**, 3756–3760 (1985).
- Kinashi, H. & Shimaji, M. *J. Antibiotics* **40**, 913–916 (1987).
- Bernigni, R., Petrov, P. A. & Carere, A. *Appl. Microbiol.* **30**, 324–326 (1975).
- Redshaw, P. A., McCann, P. A., Pentella, M. A. & Pogell, B. M. *J. Bact.* **137**, 891–899 (1979).
- Schremph, H. *Molec. gen. Genet.* **189**, 501–505 (1983).
- Hintermann, G., Cramer, R., Vogtli, M. & Hutter, R. *Molec. gen. Genet.* **196**, 513–520 (1984).
- Ishihara, H., Nakano, M. M. & Ogawara, H. *J. Antibiotics* **38**, 787–794 (1985).
- Ochi, K. *J. Bact.* **150**, 592–597 (1982).
- Stonesifer, J., Matsushima, P. & Baltz, R. H. *Molec. gen. Genet.* **202**, 348–355 (1986).
- Shirahama, T., Furumai, T. & Okanishi, M. *Agric. Biol. Chem.* **45**, 1271–1273 (1981).
- Hopwood, D. A. *et al. Genetic manipulation of Streptomyces, a Laboratory Manual* (John Innes Foundation, Norwich, 1985).

Corrigendum

Structure, sequence and expression of the hepatitis delta (δ) viral genome

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A GUANOSINE residue was omitted from the nucleotide sequence of the hepatitis δ viral (HDV) genomic RNA between residues 1,113 and 1,114 (Figs 4 and 5). HDV RNA therefore consists of 1,679 nucleotides and Table 1 should now read as given below.

Table 1 Large open reading frames (ORFs) within HDV RNA and the complementary anti-HDV RNA strand

Strand	ORF no.	Translational frame	Nucleotide position*	Total no. of AAs	No. of AAs beginning with first Met
HDV RNA	1	2	539	165	152
HDV RNA	2	1	786	156	121
HDV RNA	3	1	1,608	120	0
HDV RNA	4	3	1,297	115	68
HDV RNA	10	1	435	116/39†	33
HDV RNA	11	3	937	119	86
Anti-HDV RNA	5	1	1,619	221/202‡	214/195‡
Anti-HDV RNA	6	3	1,341	288	179/34‡
Anti-HDV RNA	7	1	506	148/80§	148/80§
Anti-HDV RNA	8	1	821	104/56	74/26
Anti-HDV RNA	9	2	91	101	0

Only ORFs longer than 300 nucleotides are shown. AAs, amino acids.

* The position of the first nucleotide in each ORF is indicated according to the numbering in the corrected Fig. 4.

† Ambiguity arising from clonal heterogeneity at position 553.

‡ Ambiguity arising from clonal heterogeneity at position 1,012.

§ Ambiguity arising from clonal heterogeneity at position 264.

|| Ambiguity arising from clonal heterogeneity at position 653.