APPLIED AND ENVIRONMENTAL MICROBIOLOGY, Oct. 1987, p. 2534–2538
0099-2240/87/102534-05$02.00/0
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Vol. 53, No. 10

Plasmid-Associated Bacteriocin Production and Sucrose Fermentation in *Pediococcus acidilactici*

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Received 4 May 1987/Accepted 31 July 1987

Production of bacteriocin activity designated pediocin PA-1 was associated with the presence of a 6.2-megadalton plasmid in *Pediococcus acidilactici* PAC1.0. The bacteriocin exhibited activity against *P. acidilactici*, *P. pentosaceus*, *Lactobacillus plantarum*, *L. casei*, *L. bifermentans*, and *Leuconostoc mesenteroides* subsp. *dextranicum*. Partial characterization of pediocin PA-1 is described. The molecular weight of pediocin PA-1 was ca. 16,500. Additionally, strain PAC1.0 was found to contain a 23-megadalton plasmid associated with sucrose-fermenting ability.

The pediococci are a group of gram-positive homofermentative lactic acid bacteria. These organisms may be found as saprophytes on vegetable material (12). Commercially, pediococci are important in the fermentation of vegetables (13) and meats (4, 14). Reports from our laboratory (8, 9) and others (3, 10) have shown the presence of plasmid DNA in *Pediococcus* spp. Daeschel and Klaenhammer (3) and Graham and McKay (10) have shown an association between bacteriocin and plasmid DNA in strains of *Pediococcus pentosaceus* and *P. cerevisiae*, respectively. Recently, we reported the association of raffinose fermentation and plasmid DNA in strains of *P. pentosaceus* (9). We report here evidence for the association of plasmid DNA and sucrose-fermenting ability and bacteriocin production in *P. acidilactici* PAC1.0.

MATERIALS AND METHODS

Bacterial strains and media. The bacterial strains used are listed in Table 1. Carbohydrate fermentation was determined as described previously, with medium BM (8). Stock culture and cultures for routine use were stored as previously described (9).

Plasmid isolation and purification. Plasmid DNA was isolated, and DNA samples were subjected to agarose gel electrophoresis as previously described (8). Reference plasmid DNA was prepared as previously described (8).

Curing studies. Stability of plasmid-encoded traits and elimination of plasmid DNA by growth at elevated temperatures were accomplished by methods previously described (9). PAC1.0 cells to be used in curing experiments were streaked on BM-sucrose. A single acid-producing colony was then transferred for use in curing experiments. In experiments involving curing of the bacteriocin-producing phenotype (Bac$^+$), single isolated colonies were streaked onto duplicate plates. Isolated colonies streaked on assay plates were screened for the Bac$^+$ phenotype by methods described below. The duplicate plate was used as an inoculum source for curing experiments. Curing was accomplished by growing cells for 18 h at 45, 47, and 50°C as previously described (9).

Bacteriocin assay. Production of bacteriocin was assayed by spotting cells on MRS agar (Difco Laboratories, Detroit, Mich.) and incubating them at 35°C for 18 h. Assay plates were exposed to chloroform vapor for 30 min and overlaid with soft agar (0.75%) seeded with indicator cells. Plates were incubated at 32°C for 18 h. Isolates producing a clear zone of inhibition were considered as producing bacteriocin.

Preparation of cell extracts and enzyme assays. Conditions for cell growth, preparation of cell extracts, determination of protein concentrations, and enzyme assays were accomplished by methods previously described (9).

Partial purification of bacteriocin. Two liters of MRS broth (Difco), inoculated at 1% with an 8-h-old culture of strain PAC1.0 (optical density at 600 nm, 0.60) grown in the same medium, was incubated statically at 35°C for 18 h. After 18 h, the pH of the culture was determined and adjusted to pH 6.0. Cells were removed by centrifugation at 16,300 × *g* for 15 min at 5°C. The supernatant was filtered through a 0.45-μm (pore size) filter (Millipore Corp., Bedford, Mass.). The supernatant was assayed for bacteriocin activity by spotting 5 μl of a serial twofold dilution series onto MRS plates overlaid with soft agar seeded with indicator cells. Assay plates were incubated at 32°C. The indicator strain was *P. cerevisiae* FBB63 (10), reidentified as *P. pentosaceus* by using carbohydrate utilization patterns and temperature growth characteristics as described by Garvie (7). One arbitrary unit (AU) of bacteriocin was defined as 5 μl of the highest dilution of culture supernatant yielding a definite zone of growth inhibition on the indicator lawn. The titer was expressed as the reciprocal of the highest dilution showing inhibition.

Ammonium sulfate (Sigma Chemical Co., St. Louis, Mo.) was added to the filtered supernatant to 30% (wt/vol) saturation at 5°C. After gentle stirring for 18 h, the mixture was centrifuged at 16,300 × *g* for 20 min at 5°C. The supernatant was brought to 60% (wt/vol) ammonium sulfate under the above conditions. After 18 h, a precipitate was collected by centrifugation. The precipitate was reconstituted in 40 ml of 0.05 M Tris-maleate (Sigma) buffer (pH 6.5; TM buffer). The reconstituted precipitate was dialyzed against TM buffer at 5°C by using Spectrophor no. 1 membrane tubing (Spectrum Medical Industries, Inc., Los Angeles, Calif.), and the titer of its activity was determined. The reconstituted dialyzed precipitate was then subjected to two ion-exchange procedures for further purification. (i) For DEAE-Sephadex batchwise separation, the bacteriocin preparation (40 ml) was added to 3 volumes of DEAE-Sephadex A-25 (Sigma) equilibrated with TM buffer at 5°C and stirred slowly for 1 h.

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TABLE 1. Strains used in this study

Strain	Resident plasmid size(s) (MDa)	Relevant phenotype(s)[a]	Comment or reference[a]
P. acidilactici			
PAC1.0	23, 6.2	Suc^+ Bac^+	8
PAC1.14	23	Suc^+ Bac^-	Bac^- PAC1.0; this study
PAC1.17	6.2	Suc^- Bac^+	Suc^- PAC1.0; this study
PAC1.19	None	Suc^- Bac^-	Suc^- Bac^- PAC1.0; this study
P. pentosaceus PPE1.0	30	Raf^+	9

[a] Abbreviations: Suc^+, ability to ferment sucrose; Bac^+, ability to produce bacteriocin PA-1; Raf^+, ability to ferment raffinose; −, loss of indicated phenotype.

The slurry was then filtered through a Büchner funnel, and its titer was determined. (ii) For CM-Sephadex batchwise separation, the filtrate was added to 3 volumes of CM-Sephadex C-25 (Sigma) equilibrated with TM buffer at 5°C and stirred slowly for 1 h. The slurry was then filtered through a Büchner funnel and washed with 3 volumes each of TM buffer and TM buffer containing 0.4 M NaCl. Activity was eluted with 1 volume (40 ml) of TM buffer containing 0.6 M NaCl. The active fraction was dialyzed against TM buffer and then concentrated 10-fold in the dialysis tubing by removal of water with Carbowax 20 (Fisher Scientific Co., Pittsburgh, Pa.). The titer of the partially purified bacteriocin was determined, and it was used in studies to characterize the bacteriocin.

Effects of heat treatment and enzymes. A partially purified sample of bacteriocin PA-1 (6,400 AU/ml) was assessed for thermostability and enzymatic effects on activity. Bacteriocin was incubated with each enzyme at a final concentration of 500 μg/ml for 60 min. Incubation in the presence of α-chymotrypsin and trypsin was at 25°C, and all other enzyme-bacteriocin mixtures were incubated at 37°C. Inactivation of the enzymes was achieved by boiling them for 3 min.

Temperature stability of the bacteriocin was assessed by heating a solution of bacteriocin to 80°C for 60 min, 100°C for 3 and 10 min, and 121°C for 15 min. After each treatment, bacteriocin samples were assayed for bacteriocin titer.

Enzymes. All enzymes were obtained from Sigma. α-chymotrypsin (type II; 47 U/mg) and lipase (type 1; 8.6 U/mg) were dissolved in 0.05 M Tris hydrochloride (pH 8.0) containing 0.01 M $CaCl_2$; protease (type V; 1 U/mg), lysozyme (grade I; 41,400 U/mg), papain (type III; 10 U/mg), and trypsin (type IX; 15,000 U/mg) were dissolved in 0.05 M Tris hydrochloride (pH 8.0); pepsin (3,200 U/mg) was dissolved in 0.2 M citrate buffer (pH 6.0); phospholipase C (type I; 10 U/mg) was dissolved in 0.05 M Tris hydrochloride (pH 7.0) containing 0.01 M $CaCl_2$.

pH stability of activity. Partially purified PA-1 (1 ml) was dialyzed against buffers of various pHs. The bacteriocin solution (6,400 AU/ml) was dialyzed for 18 h with two changes against 0.05 M glycine hydrochloride buffer (pH 2.0), 0.05 M citrate buffer (pH 3 to 6), 0.05 M Tris hydrochloride (pH 7 to 9), and 0.05 M carbonate-bicarbonate buffer (pH 10 to 11). After dialysis, the contents of the tubing were assayed for bacteriocin activity.

Adsorption studies. Adsorption of bacteriocin PA-1 to sensitive and resistant cells was accomplished by procedures similar to those described by Barefoot and Klaenhammer (2). Cells from overnight MRS broth cultures were subcultured in 25 ml of fresh broth and grown to a concentration of 10^8 CFU/ml. Cells were harvested by centrifugation, washed twice in 0.05 M Tris-maleate (pH 6.5), and suspended in 0.5 ml of the same buffer containing bacteriocin at 200 AU/ml. The mixture was incubated on ice for 1 h. Cells were removed by centrifugation followed by filtration through a 0.22-μm-pore-size filter (Millipore). The titer of activity in cell filtrate was determined. Controls included incubation of bacteriocin PA-1 without cells and cells with no PA-1 added.

Molecular weight determination. The molecular weight of bacteriocin PA-1 was determined by gel filtration. Three milliliters (1,600 AU/ml) of the concentrated preparation was applied to an ascending Sephacryl S-200 column (2.6 by 75 cm; Pharmacia Fine Chemicals, Piscataway, N.J.) in 0.05 M Tris-maleate buffer (pH 6.5). The elution volume of the bioactive PA-1 fraction was compared with the elution volumes of standard proteins. Protein in the eluate was determined by A_{280}. Bacteriocin activity was assayed as described above. The protein standards and their molecular weights included the following: lysozyme, 14,300; carbonic anhydrase, 29,000; bovine albumin, 66,000; β-amylase, 200,000 (Sigma). The following formula was used: $K_{AV} = (V_e - V_o)/(V_t - V_o)$, where V_e is the elution volume for the protein, V_o is the elution volume for blue dextran 2000, and V_t is the total bed volume.

RESULTS

Plasmid-associated phenotypes. *P. acidilactici* PAC1.0 had previously been reported (8) to contain two plasmids of 23 and 6.2 (reported as 4.7) megadaltons (MDa), designated as pSRQ10 and pSRQ11, respectively (Fig. 1, lane B). Strain PAC1.0 expresses a sucrose-fermenting (Suc^+) phenotype and produces a bacteriocin (Fig. 2A). Previous studies with three strains of *P. pentosaceus* (9) had associated the ability to ferment raffinose with the presence of plasmid DNA. It was therefore of interest to determine whether one of the resident plasmids observed in strain PAC1.0 might code for the Suc^+ phenotype. The effect of growth at elevated temperature on the stability of the Suc^+ phenotype was evaluated by growing PAC1.0 at 45, 47, and 50°C. Individual colonies obtained from each of the treatments were evaluated for the Suc^+ phenotype. Segregants that did not ferment sucrose (Suc^-) were detected at frequencies of 1.1 ± 0.1, 1.4 ± 0.01, and 4.7 ± 0.02% from growth at 45, 47, and 50°C, respectively. Representative Suc^+ and Suc^- segregants from each of the treatments were surveyed for their plasmid content. Suc^+, bacteriocin-producing (Bac^+) segregants had both the 23- and 6.2-MDa plasmids (Fig. 1, lane B). Isolates expressing a Suc^- Bac^+ (Fig. 2B) phenotype lacked the 23-MDa plasmid (Fig. 1, lane D). A representative Suc^- Bac^+ strain was designated PAC1.17.

To establish whether the remaining plasmid codes for the Bac^+ phenotype, strain PAC1.17 was grown at 45°C. Non-bacteriocin-producing (Bac^-) segregants of PAC1.17 were obtained at a frequency of 3.7%. A representative Bac^- segregant of PAC1.17 was designated PAC1.19 (Fig. 2D). Strain PAC1.19 showed no detectable plasmid DNA. To further confirm possible association of the Bac^- phenotype with plasmid pSRQ11, strain PAC1.0 was grown at 45, 47, and 50°C. Suc^+ Bac^- segregants of PAC1.0 were obtained at frequencies of 2.4, 2.1, and 2.7%, respectively. Survey lysis of representative Suc^+ Bac^- isolates showed the presence of

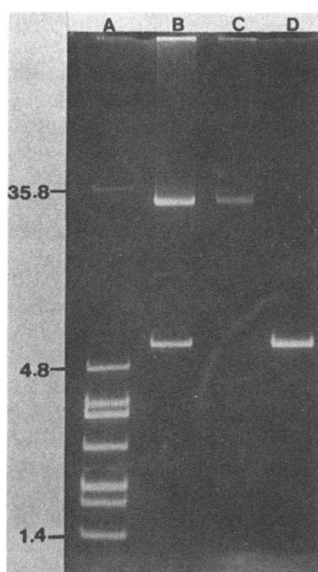

FIG. 1. Agarose gel electrophoresis of CsCl-ethidium bromide-purified plasmid DNA from *P. acidilactici* PAC1.0 and derivative strains. Electrophoresis of DNA was in 0.7% agarose at 100 V for 2 h. Lanes: A, *Escherichia coli* V517 (bands identified top to bottom) 35.8-, 4.8-, 3.7-, 3.4-, 2.6-, 2.0-, 1.8-, and 1.4-MDa covalently closed circular DNA; B, PAC1.0 Suc^+ Bac^+; C, PAC1.14 Suc^+ Bac^-; D, PAC1.17 Suc^- Bac^+. The molecular masses of *P. acidilactici* plasmid DNAs are shown in Table 1. The molecular mass of standard plasmid DNA is indicated.

plasmid pSRQ10 (23 Mda) and the absence of pSRQ11 (Fig. 1, lane C). A representative Suc^+ Bac^- isolate was designated PAC1.14 (Fig. 2C).

Plasmid-associated enzyme activity. The association between the presence of plasmid pSRQ10 and the Suc^+ phenotype was further investigated by determining plasmid-associated enzyme activity in PAC1.0 and its Suc^- derivative PAC1.17. Activities of 0.12, <0.01, and 1.26 nmol of reducing sugar (expressed as glucose) per min per mg of protein (U) were observed in cell extracts of PAC1.0 grown in the presence of glucose, fructose, and sucrose, respectively. Strain PAC1.17, grown in the presence of glucose and fructose, showed no sucrose hydrolase activity (<0.01 U).

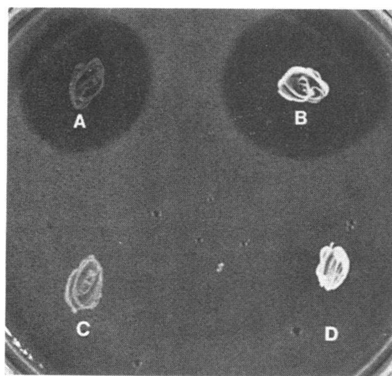

FIG. 2. Antagonistic properties of PAC1.0 and derivatives toward *P. pentosaceus* PPE1.0. (A) PAC1.0 Suc^+ Bac^+. (B) PAC1.17 Suc^- Bac^+. (C) PAC1.14 Suc^+ Bac^-. (D) PAC1.19 Suc^- Bac^-. The method used to detect bacteriocin production is described in Materials and Methods.

TABLE 2. Antagonistic spectrum of pediocin PA-1 produced by *P. acidilactici* PAC1.0[a]

Strain	Sensitivity	Source or reference[b]
Pediococcus pentosaceus		
PPE1.0	+	9
PPE1.2	+	9
ATCC 25744	+	ATCC
PPE5.0	+	9
ATCC 25745	+	ATCC
ATCC 33316	+	ATCC
Pediococcus acidilactici		
PAC1.0	−	8
PAC1.14	−	This study
ATCC 8081	+	ATCC
ATCC 25740	+	ATCC
ATCC 25741	+	ATCC
Pediococcus cerevisiae FBB63[c]	+	L. McKay, UM
Lactobacillus plantarum		
WSO	+	H. Fleming, NCSU
SD	+	MLT collection
Lactobacillus casei		
842	+	MLT collection
EV	+	MLT collection
Lactobacillus bifermentans 35409	+	ATCC
Leuconostoc mesenteroides subsp. *dextranicum* LDE1.0	+	MLT collection

[a] The conditions used for the plate assay are described in Materials and Methods.
[b] Abbreviations: ATCC, American Type Culture Collection (Rockville, Md); NCSU, North Carolina State University (Raleigh); UM, University of Minnesota (St. Paul); MLT, Microlife Technics (Sarasota, Fla).
[c] Reidentified as *P. pentosaceus* (see Materials and Methods).

The α-galactosidase and α-glucosidase activities of PAC1.0 extracts were also tested by using *o*-nitrophenyl-α-D-galactopyranoside and *p*-nitrophenyl-α-D-glucoside, respectively, as substrates. Neither α-galactosidase nor α-glucosidase activity was observed in PAC1.0 extracts.

Inhibitory spectrum of PA-1. The plate assay system, with PAC1.0 as a Bac^+ strain and PAC1.14 as the control Bac^- strain, was used to evaluate the spectrum of bacteriocin activity. Activity of the inhibitory product produced by *P. acidilactici* PAC1.0 was designated as pediocin PA-1. Strain PAC1.14 was included to detect any inhibitory effects on indicator strains from by-products of metabolism, such as lactic acid. Strain PAC1.0 showed activity against different strains of *P. acidilactici*, *P. pentosaceus*, *Lactobacillus plantarum*, *L. casei*, *L. bifermentans*, and *Leuconostoc mesenteroides* subsp. *dextranicum* (Table 2). No activity was observed against *Streptococcus lactis*, *S. lactis* subsp. *diacetylactis*, *S. cremoris*, or *S. thermophilus*. The Bac^- strain PAC1.14 showed no sensitivity to pediocin PA-1. Strains of *Staphylococcus aureus* showed sensitivity equal to that of the parent and cured strains, indicating that inhibition was probably due to the suppressive effects of lactic acid. With partially purified pediocin PA-1 (32,000 AU/ml), strains of *Micrococcus varians*, *M. sodonensis*, *Staphylococcus xylosus*, *S. epidermidis*, *S. carnosus*, *Lactobacillus acidophilus*, *L. lactis*, and *L. bulgaricus* were tested for sensitivity to the bacteriocin. None of the strains showed sensitivity to pediocin PA-1.

It was of interest to determine the relative sensitivities of

TABLE 3. Effects of various treatments on pediocin PA-1 activity

Treatment	Residual activity (AU)[a]
Protease	0
Papain	0
Pepsin	0
α-Chymotrypsin	0
Lysozyme	32
Phospholipase C	32
Lipase	32
80°C for 60 min	32
100°C for 3 min	32
100°C for 10 min	32
121°C for 15 min	2
RNase	32
DNase	32
None	32

[a] AU, Arbitrary units, defined in Materials and Methods.

various strains to the bacteriocin. With a high-titer (128×10^4 AU/ml) preparation of pediocin PA-1, the relative sensitivities of various susceptible strains were determined. Strains PPE1.0 and *P. acidilactici* ATCC 8081 showed sensitivity to 5 AU, while *L. plantarum* WSO was sensitive to 2 AU. PAC1.0 and its derivative strains PAC1.14 and PAC1.19 showed no sensitivity to the high-titer preparation.

Adsorption studies. Plate assay methods and spotting of purified bacteriocin had shown that strain PAC1.0 and its Bac^- derivative strains showed no apparent sensitivity to pediocin PA-1. Since adsorption of a bacteriocin to sensitive cells is an initial step in the action of a bacteriocin (16), it was of interest to determine whether there were quantitative differences in the adsorption of pediocin PA-1 to sensitive and insensitive strains. *P. acidilactici* PAC1.0, PAC1.14, and PAC1.19, *P. pentosaceus* PPE1.0 and FBB63, and *L. plantarum* WSO all reduced an initial pediocin PA-1 concentration of 200 AU/ml to <20 AU/ml, which was the lower limit of detection. The sensitive and insensitive strains adsorbed the bacteriocin to equivalent degrees.

Effects of enzymes, heat treatments, and pH. Partially purified bacteriocin was tested for sensitivity to various enzymes, heat, and pH. Pediocin PA-1 was sensitive to protease, papain, and α-chymotrypsin (Table 3). Bacteriocin activity was not affected by lipase, phospholipase C, lysozyme, DNase, and RNase, or heating to 80 and 100°C. Exposure to 121°C did partially destroy activity (Table 3). Bacteriocin activity was found to be most stable between pHs 4 and 7, with some loss of activity at pHs 2, 3, 9, and 10. Most of the activity was lost at pH 11.

Molecular weight determination. The molecular weight of pediocin PA-1 was estimated by ascending gel filtration chromotography with Sephacryl S-200. Comparison of the elution volume of the bioactive fraction with those of standard proteins resulted in a peak of activity corresponding to a molecular weight of ca. 16,500 (Fig. 3).

DISCUSSION

The results presented provide evidence for plasmid-encoded sucrose utilization and bacteriocin production in *P. acidilactici* PAC1.0. Sucrose hydrolase activity was associated with the presence of pSRQ10, a 23-MDa plasmid, while bacteriocin production in the same strain was associated with pSRQ11, a 6.2-MDa plasmid. Loss of the 23-MDa plasmid resulted in simultaneous loss of sucrose-fermenting ability and associated sucrose hydrolase activity. Strains cured of plasmid pSRQ10 expressed a Suc^- Bac^+ phenotype, indicating possible involvement of plasmid pSRQ11 in bacteriocin production. Strains cured of both plasmids expressed a Suc^- Bac^- phenotype, while those cured only of plasmid pSRQ11 expressed a Suc^+ Bac^- phenotype, further indicating involvement of pSRQ11 in bacteriocin production. Unlike the previously reported (9) sucrose-fermenting *P. pentosaceus* strains, *P. acidilactici* PAC1.0 showed no ability to utilize α-galactosides such as melibiose or raffinose. No α-galactosidase or α-glucosidase activity was observed in extracts of strain PAC1.0. Evidence of inducible sucrose hydrolase activity was obtained with extracts of glucose- and sucrose-grown cells. The lack of α-D-glucosidase and α-galactosidase activity suggests that the sucrose hydrolase activity may be due to a β-D-fructofuranoside fructohydrolase. Further characterization is necessary to determine the exact specificity of the enzyme and possible permease activity associated with the presence of plasmid pSRQ10.

Graham and McKay (10) have reported the association of production of a bacteriocinlike substance in *P. cerevisiae* with the presence of a 10.5-MDa plasmid but have reported no evidence that immunity or resistance to the bacteriocin is associated with the same plasmid. Daeschel and Klaenhammer (3) reported that association of bacteriocin production and immunity is encoded on a 13.6-MDa plasmid in *P. pentosaceus*. The bacteriocin described by Daeschel and Klaenhammer (3) showed activity not only against *Pediococcus* spp. but also against *Clostridium*, *Staphylococcus*, and *Streptococcus* spp. The present study demonstrated that the antagonistic substance produced by *P. acidilactici* PAC1.0

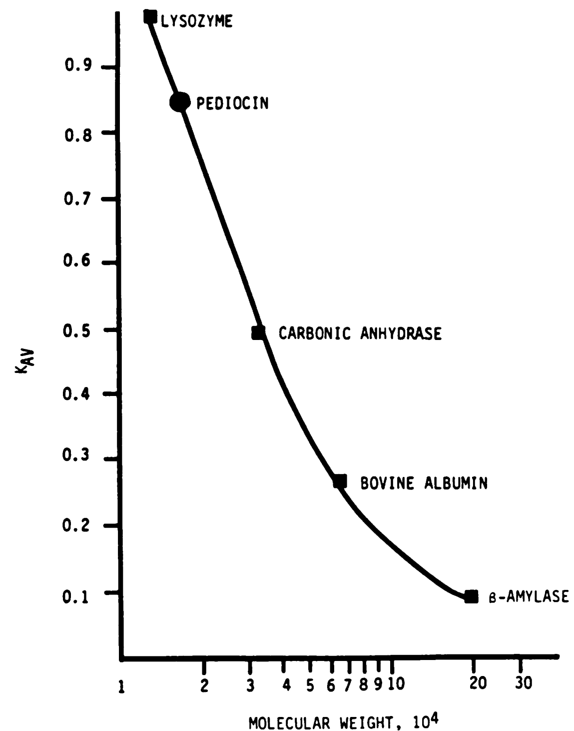

FIG. 3. Molecular weight determination of pediocin PA-1 from *P. acidilactici* eluted from a Sephacryl S-200 column in 0.05 M Tris-maleate buffer (pH 6.5).

has properties which are similar to the bacteriocinlike activity in gram-positive bacteria (16). The molecular weight of ca. 16,500 (Fig. 3), as determined by gel filtration, for pediocin PA-1 is comparable to the molecular weight of bacteriocins described in *Streptococcus* (16), *Bacillus* (5), and *Lactobacillus* (17) spp. Pediocin PA-1 showed activity against related species (Table 2) and is bactericidal in its mode of action. Pediocin PA-1 is proteinaceous in nature (Table 3), as indicated by sensitivity to various proteolytic enzymes. Additionally, production of the bacteriocinlike material is plasmid associated. No evidence for plasmid-encoded bacteriocin immunity was obtained, since the cured Bac^- isolates did not become sensitive to the bacteriocin. The mechanism of resistance, which is usually determined by loss of a specific receptor for a particular bacteriocin or immunity (16), of strain PAC1.0 cells to the bacteriocinlike action of pediocin PA-1 is unknown. However, strains cured of plasmid pSRQ11 that expressed a Bac^- phenotype and the parental strain showed no sensitivity to pediocin PA-1 by either the plate assay method or exposure to high-titer, partially purified pediocin PA-1. Sensitive and insensitive cells adsorbed pediocin PA-1 at comparable levels, suggesting that the specificity of PA-1 may not be entirely dependent on the presence of specific receptor sites on sensitive cells. These results are similar to those reported by Barefoot and Klaenhammer (2) for lactacin B, which demonstrated adsorption to both sensitive and insensitive *Lactobacillus* spp. The nonlethal binding observed with pediocin PA-1 has also been reported for other bacteriocins, such as staphylococcins (6, 11), lactocin LP27 (17, 18), and streptococcin B-74628 (15). As noted by Tagg et al. (16), the nonlethal binding may be a reflection of the high surface activity of some bacteriocins that makes them capable of adsorbing nonspecifically to various bacteria. Further studies are needed to determine the mechanisms for pediocin PA-1 specificity, resistance, and mode of action. The associations of sucrose fermentation and production of pediocin PA-1 with plasmids pSRQ10 and pSRQ11, respectively, are, to our knowledge, the first reported plasmid-linked properties of *P. acidilactici*.

The natural role of these plasmid-linked properties can only be speculative and may give strains containing them a selective advantage in nature. In the case of PAC1.0, the ability to utilize sucrose may be related to its natural ecological environment. *P. acidilactici* and *P. pentosaceus* are known to reside on plants naturally (12) and to be involved in the fermentation of a variety of vegetables (13). Sucrose is the most common disaccharide found in plants (1), and thus, strains containing genetic information allowing them to utilize this sugar may have a selective advantage in vegetable fermentation. The action of the bacteriocin against related organisms may allow one strain to predominate over competing related microorganisms. Strain PAC1.0 represents a strain which, possibly through a selective process, has retained or obtained information which may give it a selective advantage in its natural environment. Further characterization of these and other plasmids found in the pediococci will enable industry to tailor the genetic composition of a strain(s) and control such characteristics as strain dominance and fermentation capability.

ACKNOWLEDGMENTS

We gratefully acknowledge the assistance of Jennifer R. Dusza in the preparation of the manuscript. We thank Ebenezer R. Vedamuthu for critical reading of the manuscript.

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