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Present and past microbial life in continental pan sediments and its response to climate variability in the southern Kalahari

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ABSTRACT

Terrestrial climate archives are rare in the arid southwestern African region, which makes paleoclimate and paleoenvironmental studies difficult. Since there are only ephemeral lacustrine systems in the area, in this study a continental pan (playa) is evaluated as a climate archive. Climate has a strong impact on the pan ecosystem, causing adaptation of indigenous microorganisms to varying temperature, precipitation and salinity conditions. Here a combined approach of inorganic and organic geochemical investigations, including lipid biomarker analyses, was carried out to examine the response of indigenous microbial communities to environmental changes and to characterize the nature, abundance and depth distribution of recent (phospholipids) and past (glycerol dialkyl glycerol tetraethers, GDGTs) microbial life within the sediments of Witpan, located in the southern Kalahari. Lipid biomarkers contain information about changes in biogeochemical processes and climate variation, therefore we tested here whether they can be used to reconstruct paleoclimatic changes such as past precipitation periods in arid terrestrial ecosystems. Despite the extreme environmental conditions with rather low TOC values, restricted availability of water and substrates in the pan system, bacterial life was observed along the depth profile of Witpan. Bacterial membrane phospholipid life markers showed their highest abundance in the surface layers, indicating that microbial life in Witpan is strongly influenced by near-surface processes. A series of saturated, branched and unsaturated phospholipid fatty acids (PLFAs) were detected and several phyla of Bacteria, such as gram-positive and gram-negative bacteria were present. Some PLFAs and intact archaeal membrane lipids point to the presence of a halophilic microbial community in the surface layers. Biomarkers for past microbial life (archaeol, branched and isoprenoid GDGTs) were absent or had very low concentrations during the dry Holocene sequence (below the surface sediments). However, during the Last Glacial Maximum (LGM), considered to represent a period with increased precipitation, an increased abundance of these biomarkers was observed. Thus, these results demonstrate the potential of microbial biomarkers in pan systems to preserve climate signals over geologic timescales. The data indicates that microbial biomarkers can be used to trace paleo-precipitation periods in semi-arid to arid environments and that pan structures can form suitable geo-archives for biomolecules in areas where other terrestrial archives are missing.

1. Introduction

In temperate and tropical terrestrial environments, deposits from lakes and wetlands are frequently used for paleoenvironmental reconstructions. Since there are almost no lakes in arid landscapes other archives have to be used for climate studies (Telfer et al., 2009). Periods of increased humidity in the Kalahari region have been inferred by investigating various carbonate deposits, such as calcretes (Nash and McLaren, 2003), stromatolites (Lancaster, 1986), speleothems (Brook et al., 1999) and tufas (Doran et al., 2015). For paleohydrological and paleoclimatic studies, fluvial systems and slack-water deposits have been considered (Heine, 2004). However, proxy data from paleoenvironmental archives in southwestern Africa are sparse, extremely heterogeneous and indicate different regional environmental reactions to climate variations (Heine, 2005).

The arid southwestern African landscape is characterized by pan depressions, especially in the eastern part of Namibia and in the north-central and western areas of the Republic of South Africa

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where evaporation exceeds precipitation. Pans are closed depressions which are representative of low relief areas in arid and semi-arid environments (Goudie and Wells, 1995). Erosional processes such as eolian deflation have been considered to shape pan systems mainly during dry periods (Shaw and Bryant, 2011). During more humid phases with higher groundwater levels sediments accumulate within pans (Lancaster, 1976, 1986). Sediments are transported and deposited via surface run off and internal drainage systems. Additionally, deposition of eolian sediments into standing shallow water during humid phases is possible (Holmgren and Shaw, 1997). In semi-arid and arid regions this can cause the development of pans (Goudie and Thomas, 1985) and over time the formation of archives for paleoclimate evolution in the area.

Precipitation in this region is controlled by the seasonal shift of the Intertropical Convergence Zone (ITCZ). The ITCZ is located at the boundary between northern and southern trade winds and is caused by the near-vertical ascent of warm and humid air from low latitudes near the equator. While during austral winter (June to September) the ITCZ above the African continent is located north of the equator, in the austral summer (December to March) the ITCZ strongly shifts to the south bringing humidity to the east and center of southern Africa (Ahrens and Samson, 2010). In the Kalahari region, precipitation occurs only occasionally by seasonal rain showers during austral summer (summer rainfall zone). Strong precipitation leads to transient runoff in ephemeral rivers and pans are filled temporally with water. Due to the high evaporation in this area, desiccation of surface waters and aridification of the southwestern Kalahari is a fast process on a seasonal scale. Along the western coastal region of southern Africa precipitation occurs between April and September. This winter rainfall zone is influenced by the annual northward migration of the Southern Hemisphere westerlies. During the last glacial–interglacial transition the seasonality and amount of precipitation has changed in southern Africa (Gasse et al., 2008). Expansion of the Antarctic sea-ice during the Last Glacial period caused northward migration of the westerlies (Stone, 2014). This led to an extension of the winter rainfall zone affecting the precipitation in southwestern Africa during the Last Glacial Maximum (LGM) (Stone, 2014).

In the southern Kalahari region two studies concerning the geological characterization and depositional age assessment of pan deposits have been conducted on pan structures in Botswana (Holmgren and Shaw, 1997) and in the northwest of South Africa (Telfer et al., 2009). Holmgren and Shaw (1997) described the evolution of a shallow endorheic basin, the Lebatse Pan, in the southeast Kalahari in Botswana and examined the environmental conditions during the formation of the pan. The sediment stratigraphy of the pan indicated its potential as an archive for geochemical and geophysical analyses, since geomorphological, physical and chemical properties indicated different depositional phases (Holmgren and Shaw, 1997). Telfer et al. (2009) were able to show that Witpan in northwest South Africa contains a Late Pleistocene sedimentary fill. They published optically stimulated luminescence (OSL) dating ages for Witpan sediments, showing that a thick sediment package within Witpan was deposited between 18 to 22 ka BP. They assumed rapid sedimentation during this period due to "wetter than present" conditions. Their study describes the potential of pan sediments in northwest South Africa as an archive for environmental changes during the Late Quaternary.

Although the local geomorphology shows evidence of changes in the southwestern African region, they sometimes can be ambiguous in terms of interpreting the actual climatic conditions (Thomas and Burrough, 2012). The use of geo-proxy data for integrated regional records of climate change in southern Africa is quite challenging and thus it is important to explore new geo-archives and to search for additional proxy indicators for the reconstruction of climatic and environmental conditions.

Due to the occasional presence of water, pans can form a habitat for a diverse ecosystem. In contrast to higher organisms, relatively little is known about the survival and adaption of microorganisms in such hot desert ecosystems. Climate has a strong impact on the pan microbial ecosystem, causing adaptation of microbial communities to varying temperatures, low water availability, salt precipitation, and salinity conditions (Makhalanyane et al., 2015). To elucidate microbial communities in pans a combined approach of microbial lipid biomarkers and geochemical analysis can be used to describe the composition, abundance and distribution of microbial communities with respect to climatic and environmental variation. Microbial membrane phospholipid (PL) esters and their side chain fatty acid (PLFA) inventory represent biomarkers that are indicative for living Bacteria (Zelles, 1999), whereas the fatty acid side chains can provide a fingerprint of the community structure on a broad taxonomic level (Kaur et al., 2005). The PL life markers are rapidly degraded after cell death (Logemann et al., 2011), thus their occurrence indicate the presence of living cells in geological samples (White et al., 1979). Intact membrane phospholipid ethers are characteristic for Archaea. These biomolecules seem to be significantly more stable due to their ether-bound moieties restricting their potential to act as life markers (Logemann et al., 2011). Main membrane constituents of halophilic archaea are archaeol phosphatidyl glycerophosphate (PGP), archaeol phosphatidyl glycerophosphate methyl ester (PGP-Me), archaeol phosphatidic acid (Ar-PA), archaeol phosphatidyl glycerol (Ar-PG) and archaeol phosphatidyl glycerosulfate (Ar-PGS) (Kates, 1993). Additionally, analog dialkyl glycerol diether (DGD) structures occur where the side chain can contain one or two sesterterpanyl side chains (25 carbon atoms) instead of the phytanyl side chain (20 carbon atoms) found in archaeol (Dawson et al., 2012). Side chains can also contain double bonds or hydroxy groups (Kates, 1993; Dawson et al., 2012).

In addition to the intact lipids, geological samples often contain microbial membrane core lipids that are already partly degraded. These compounds have lost their head groups, but their lipid cores are very stable and are well preserved in sedimentary settings (Pease et al., 1998). Compounds such as archaeol and glycerol dialkyl glycerol tetraethers (GDGTs) occur ubiquitously in water, soil, peat and sediments and represent characteristic biomarkers for past Archaea and Bacteria (Schouten et al., 2013). After the transition from biosphere to geosphere, such fossil molecules can be retraced to their biological precursors and changes in the ancient microbial ecosystems can be described (Schwark, 2013). Branched GDGTs (brGDGTs) are ubiquitous compounds in lake sediments (Blaga et al., 2009) as well as in soils (Weijers et al., 2007) and they are known to derive from Bacteria (Weijers et al., 2006), whereas isoprenoid GDGTs (iGDGTs) including dialkyl glycerol diethers such as archaeol are synthesized by Archaea (Kates, 1996).

The current study is focused on depth-related variations of the abundance and composition of present and past microbial communities in pan deposits in the southern Kalahari region with a specific focus on the climate history in this area. Former studies suggested an increased precipitation in the southwestern African region during the LGM (e.g., Chase and Brewer, 2009) and drier conditions towards and within the Holocene period (e.g., Lim et al., 2016). This information forms the climatic and environmental background for the interpretation of the past microbial biomarker data found in pan deposits in the southwestern Kalahari region. Water availability is a prerequisite for microbial life processes. Thus, we wanted to test our hypothesis that the abundance and composition of past microbial biomarkers in pan deposits can be used to trace periods of increased paleo-precipitation in arid landscapes such as the southern African region. Additionally, the study will contribute to the question whether pan sediments are

appropriate geo-archives for paleoclimatic reconstruction in the southern Kalahari area. Furthermore, we will investigate for the first time the depth distribution, and on a broad taxonomic level the composition, of modern bacterial communities in pan deposits. As the study site we selected Witpan located in the northwestern part of the Republic of South Africa, since for this pan some information on the climatic history is already available to evaluate the microbial biomarker data in a paleo-climatic context.

2. Material and methods

2.1. Study site

Witpan (Fig. 1a and b, 26°40′S, 20°09′E) is located in a broad belt of pans within the southwestern Kalahari in the northwestern part of the Republic of South Africa. It is ca. 5 km long and surrounded by linear dunes. On its southern side a well-developed lunette dune surrounds the pan. The pan floor subsurface (30 cm) is described as alkaline with an pH of 9.5 (Thomas et al., 1993). The pan consists of a northern and southern basin with different filling histories (Telfer et al., 2009). The northern area is characterized by a sporadic, channelized drainage system, which might be responsible for depositional processes (Holmgren and Shaw, 1997). The northern and southern basins are characterized by silt and clay, but the southern basin additionally shows some intervals of increased coarser material such as fine to very fine sand (Telfer et al., 2009). Both erosional processes from the lunette dune or eolian input are discussed as a reason for the coarser material in the southern basin (Telfer et al., 2009). Deflation has been considered as a main formative process leading to the formation of this lunette dune in the southern part of the pan (Thomas et al., 1993). Interactions of water and wind are important for lunette shaping processes and can describe periods of changing climatic conditions. Surface runoff (Fig. 1c) is promoted by a cycling process where sediment is wind-deposited onto the dunes and returned to the pan floor by water during storm events (Thomas et al., 1993). In contrast to the erosional processes Optically Stimulated Luminescence (OSL) data, determined for deposits from the southern basin, clearly indicate rapid deposition of sediments during the LGM, interpreted as a wetter period in this area. Thus, the sedimentary history of pans is the result of erosional and depositional phases. Although this might prevent a continuous deposition of sedimentary material, the OSL data suggests that Witpan forms a geo-archive for climatic information of the past at least since the Late Glacial (Telfer et al., 2009).

Biogenic proxies such as pollen and diatoms are sparse in Witpan sedimentary fills and only some degraded phytoliths have been found (Telfer et al., 2009). Witpan might not allow standing water over longer periods of time (Telfer et al., 2009). Today water comes with occasional rainfall and fills the pan at least for several weeks (information from native farmers).

2.2. Sampling and sample material

To take advantage of the paleo-climatic information provided by Telfer et al. (2009) for the southern basin of Witpan, sediment material for the current study was also collected in the southern part of the pan (Fig. 1b; 26°40.658′S 20°09.45′E) during a sampling campaign at the end of the dry season in November 2013. However, the current study site was closer to the southern lunette dunes. The top layers (9 cm) of the sampling site were covered by a salty crust. In the upper 50 cm, samples were taken from a trench excavated into the sediment. Within the upper 15 cm, samples were taken at 3 cm intervals, followed by 5 cm intervals down to 50 cm depth. Sampling was continued at 10 cm intervals from 50–180 cm depth by drilling a short core with an Eijkelkamp hand auger. Samples for biomarker analyses were taken from fresh and clean surfaces within the trench and from inner parts of the drill core. Samples were stored immediately in liquid nitrogen, transported to GFZ Potsdam and after arrival in our laboratories they were kept at −24 °C. Overall, 29 samples from the Witpan were collected for geochemical characterization as well as for biomarker studies.

The ground water table after drilling at the study site was at a depth of 230 cm. The collected Witpan deposits showed a large variability in grain size distribution. Silt and evaporite crystals dominated the top layers (0–14 cm), followed by a mixture of silt and sand. Between 25 and 119 cm sand made up the largest proportion. Clay and silt appeared between 119 and 180 cm and dominated the sediment composition in this depth interval. The shifting grain sizes reflected fluctuating environmental conditions during deposition and possibly changing sediment sources (Schüller et al., 2015). The Witpan sedimentary fill shows a clear change in sedimentary conditions around 20 ka BP (110 cm depth) (Schüller and Wehrmann, 2016).

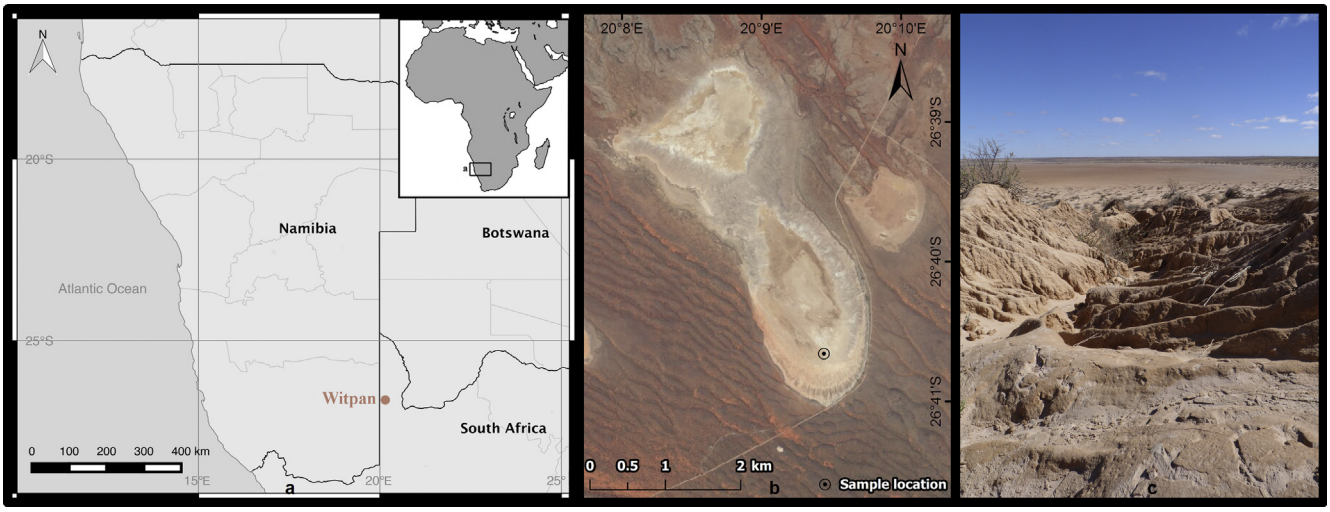

Fig. 1. (a) Map of study site at the border between Namibia and South Africa. (b) Aerial image of Witpan in northwestern South Africa (26°40′S, 20°09′E) with the sampling location. Map provided by Robert Milewski (GFZ German Research Centre for Geosciences – Helmholtz Centre Potsdam); Sources: Esri, DigitalGlobe, GeoEye, Earthstar Geographics, CNES/Airbus DS, USDA, USGS, AEX, Getmapping, Aerogrid, IGN, IGP, swisstopo, and the GIS User Community. (c) View from the southern rim of Witpan towards the sampling site (picture taken in October 2013).

For the Witpan deposits it was possible to obtain five ^{14}C-radiocarbon dates on bulk TOC. The age data in the project was provided by Schüller and Wehrmann (2016). The first two data points revealed Holocene ages with 1.7 ± 0.08 ka BP and 5.4 ± 0.1 ka BP at 10 and 25 cm depth, respectively. The TOC increase at 55 cm depth (Fig. 2a) was dated back to 19.8 ± 0.3 ka BP. Another date was determined at 110 cm depth, yielding 20.8 ± 0.3 ka BP, which was only slightly older than the sample from 55 cm depth and a last age date was provided for sediment from 205 cm depth with 20.5 ± 0.3 ka BP. Thus, the age data from 110 and 205 cm depth indicated essentially the same age and together with the age date at 55 cm a very rapid deposition of sedimentary material. This is in accordance with the observation presented in Telfer et al. (2009), who reported that sediments in the southern part of Witpan between 50 and 90 cm (deeper sediments have not been dated) accumulated around 20 ka BP. Following both age assignments, the sediments between 55 and 205 cm depth fell into the range of the LGM. Overall the radiocarbon age assessment confirms that Witpan indeed contains a Late Glacial to Holocene sediment fill. Furthermore, in the time period of the LGM the age model by Schüller and Wehrmann (2016) shows strong similarities to the age model of Telfer et al. (2009) based on OSL data. The fact that both independent methods of age determination (one based on time of organic matter production and one on time of last exposure of inorganic matrix to sunlight) show similar results, provides good indication that the organic matter (and with that the biomarkers) in the pan essentially represents organic material from the respective time period and not reworked older material from the pan catchment area. Thus, Witpan represents an excellent background to test our hypothesis whether microbial biomarkers can be used as indicators for paleo-precipitation periods in arid areas.

2.3. Total organic carbon

The total organic carbon (TOC) content was measured with a Euro EA 3000 element analyzer (EuroVector) using freeze-dried and homogenized samples (3 mg). The samples were wrapped in Ag capsules after they have been treated with HCl (20%) at 85 °C to remove carbonates (Schmidt et al., 2014).

2.4. Ion chromatography

Porewater concentrations of the sediment samples were rather small and the samples were therefore leached according to Blume et al. (2011). Five g of sample material was suspended in 25 ml of deionized water, shaken for 90 min and then centrifuged to remove solids. The supernatant was filtered through 0.45 μm cellulose acetate filter (Whatman Aqua 30/0.45 CA) and diluted 1:500 with MilliQ water. The concentrations of anions and organic acids were determined by ion chromatography (IC) in replicates with conductivity detection (ICS 3000, Dionex Corp. and Sykam IC). The following ions were determined from the leachates: fluoride, chloride, acetate, formate, sulphate and nitrate. Details of the analytical settings are provided in the supplement (Supplementary Table S1). Specifics of the method to detect organic acids were described by Vieth et al. (2008); for inorganic ions see Noah et al. (2014). Quantification standards, which contain all investigated compounds, were measured in different concentrations on every measurement day.

2.5. Lipid biomarker analysis

Approximately 80 g of freeze-dried samples was homogenized, ground and extracted using a flow blending system with a 200 ml mixture of methanol (MeOH)/dichloromethane (DCM)/ammonium

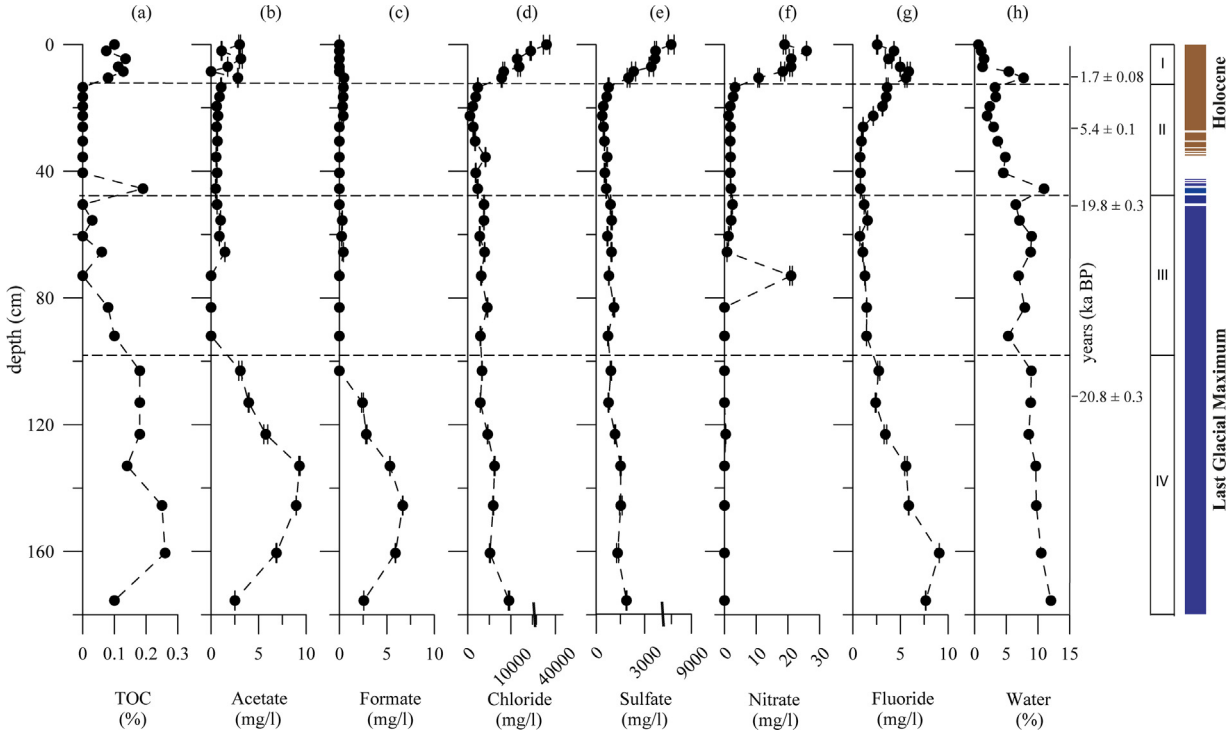

Fig. 2. Biotic and abiotic parameters of Witpan sedimentary fills: (a) total organic carbon (TOC), (b) acetate, (c) formate, (d) chloride, (e) sulfate, (f) nitrate, (g) fluoride and (h) water content. Ions were obtained by sediment leaching. Age data provided by Schüller and Wehrmann (2016). Additional age date point: 20.5 ± 0.3 ka BP at 205 cm (not shown). Note different scales of *x*-axis.

acetate buffer (2:1:0.8, v:v:v, pH = 7.6) according to the procedure of Bligh and Dyer (1959). Subsequently, the solvent extract was collected in a separation funnel and for phase separation DCM and water were added to achieve a ratio of MeOH/DCM/ammonium acetate buffer (pH 7.6) of 1:1:0.9 (v:v:v). Afterwards the organic phase was removed and the water phase was re-extracted two times with DCM. The organic phases were combined and the solvent was evaporated using a TurboVap® 500 system (Biotage) and finally a gentle stream of nitrogen. The obtained extract was separated into fractions of different polarity (low polarity lipids, free FAs, glycolipids and phospholipids) using a pure silica column (1 g silica gel 63–200 μm) and a Florisil® column (1 g magnesium silica gel, 150–250 μm) in sequence. The low polarity fraction was eluted with 20 ml of $CHCl_3$, the free FAs with 50 ml of methyl formate blended with 12.5 μl of glacial acetic acid and the glycolipid fraction with 20 ml of acetone. After removal of the Florisil® column the PLs were eluted with 25 ml of MeOH from the silica column. To improve the recovery of PLs, the silica column was rinsed with 25 ml of a MeOH/water mixture (60:40, v:v) and the extract was captured in a separation funnel. DCM and water were added for phase separation (MeOH/DCM/water, 1:1:0.9, v:v:v), the organic phase was removed, and the water phase was re-extracted two times with DCM. Finally, the organic phases were combined and evaporated to dryness and stored at −20 °C until analysis. The PL fraction was used for subsequent PLFA analysis. The method applied is described in Zink and Mangelsdorf (2004).

2.6. Detection of phospholipid fatty acids (PLFAs)

For phospholipid fatty acids (PLFA) detection half of the PL fraction was used for mild alkaline hydrolysis via ester cleavage (Müller et al., 1990). The resulting fatty acid methyl esters were measured by gas chromatography–mass spectrometry (GC–MS). Compounds were identified on a gas chromatograph (Trace GC Ultra, Thermo Electron Corporation) equipped with a cold injection system (Thermo Electron Corporation) and a 50 m × 0.22 mm × 0.25 μm BPX5 (SGE) column coupled to a DSQ MS Thermo Finnigan Quadrupole MS (Thermo Electron Corporation). The GC was run in splitless mode. The injector temperature was programmed from 50 to 300 °C at a rate of 10 °C/s. The initial oven temperature was 50 °C (1 min isothermal), heating rate 3 °C/min to 310 °C (held for 30 min). Helium was used as carrier gas at a continuous flow rate of 1 ml/min. The GC–MS was operated in the electron impact (EI) ionization mode at 70 eV. Full-scan mass spectra were recorded from m/z 50–650 at a scan rate of 1.5 scans/s.

2.7. Detection of intact phospholipid esters and ethers

The second half of the PL fraction was used for the analysis of intact phospholipid esters and ethers. Analyses were performed on a Shimadzu LC10AD HPLC coupled to a Finnigan TSQ 7000 triple quadrupole mass spectrometer with an electrospray interface. Samples were separated with a LiChrospher 100 diol column (2 × 125 mm, 5 μm; CS-Chromatographie Service) equipped with a pre-column filter. Compound separation was achieved by the following solvent gradient: 1 min 100% A, increasing over 20 min to 35% A and 65% B using a linear gradient followed by 40 min of reconditioning. Eluent A is a mixture of *n*-hexane:isopropanol:formic acid:ammonia (25% in water; 79:20:1.2:0.04, v:v:v:v), eluent B is isopropanol:water:formic acid:ammonia (25% in water; 88:10:1.2:0.04, v:v:v:v. The flow rate was set to 0.35 ml/min. The method is described in Rütters et al. (2001). ESI source conditions are as follows: spray voltage 4 kV; capillary temperature 220 °C; nitrogen sheath gas at 60 psi; without auxiliary gas. Full scan mass spectra were recorded in the negative ion mode over the range m/z 400–1800 at a scan time of 2 s.

2.8. Detection of glycerol dialkyl glycerol tetraethers (GDGTs)

The low polarity lipid fractions were dissolved in 250 μl DCM/MeOH (99:1, v:v) and a 40-fold excess of *n*-hexane was added to precipitate asphaltenes, which were then removed by filtration over sodium sulfate. The *n*-hexane soluble fraction was separated into an aliphatic/alicyclic hydrocarbon, aromatic hydrocarbon, and hetero compound (NSO-compounds containing nitrogen, sulfur, and oxygen) fraction using a medium-pressure liquid chromatography (MPLC) system (Radke et al., 1980). To study glycerol dialkyl glycerol tetraethers (GDGTs), the NSO fractions were analyzed by High Performance Liquid Chromatography–Atmospheric Pressure Chemical Ionization–Mass Spectrometry (HPLC–APCI–MS) according to a modified method after Hopmans et al. (2000) and Schouten et al. (2007). Samples were measured on a Shimadzu LC10AD HPLC coupled to a Finnigan MAT TSQ 7000 mass spectrometer. Compounds were separated on a Prevail Cyano column (2.1 × 150 mm, 3 μm; Alltech) equipped with a pre-column filter. Tetraethers were eluted isocratically with *n*-hexane (99%) and isopropanol (1%) for 5 min, followed by a linear gradient to 1.8% isopropanol in 40 min and 1 min to 10% isopropanol. This was maintained for 5 min to clean the column and set back to initial conditions and held for 16 min for equilibration. The flow rate was set to 200 μl/min. Atmospheric pressure chemical ionization (APCI) conditions were as follows: corona current of 5 mA (5 kV), a vaporizer temperature of 350 °C, a capillary temperature of 200 °C and voltage of 7.5 V; nitrogen sheath gas at 60 psi was used without auxiliary gas. Mass spectra were generated by selected ion monitoring (DeLong et al., 2008) in the positive ion mode. The following m/z values 1302, 1300, 1298, 1296, 1294 and 1292 were used for isoprenoid GDGTs, 1049, 1047, 1045, 1035, 1033, 1031, 1021, 1019 and 1017 for branched GDGTs and 654 for archaeol. For semi-quantitative determination of GDGT concentration an external synthetic archaeol standard was measured.

3. Results

3.1. Abiotic and biotic parameters

According to differences in sediment properties (Fig. 2) and microbial biomarker quantities (see Sections 3.2 and 3.3), the sediment sequence was subdivided into four intervals. The first interval, from 0 to 14 cm, represented the surface layer. The second interval from 14 to 50 cm was characterized by a very low organic input and low microbial biomarker contents. The third interval started below 50 cm and ended at 95 cm with a slight increase in total organic carbon and lipid fossil biomarkers. The fourth interval (95–180 cm) showed a further increase of TOC (up to 0.26%) and fossil biomarkers. The separation of Witpan deposits into intervals is maintained below.

Following the age assessment by Schüller and Wehrmann (2016), the surface interval I and presumably the main part of interval II were assigned to Holocene ages. The end of the Last Glacial period was not really resolved, but might be included in the lower part of interval II. Finally, the sediments of intervals III and IV represented the LGM.

In Witpan deposits the total organic carbon content (TOC) values was rather low and ranged from 0.03 to 0.26 wt% (Fig. 2a). In surface interval I, TOC contents up to 0.14 wt% were measured. In interval II, TOC contents where below the detection limit. After a spike at 55 cm depth, a gradual increase in TOC was observed in intervals III and IV and values of up to 0.23 wt% (at 160 cm) were detected.

Small organic acids such as acetate and formate were detected in varying concentration between 0.2 and ~9 mg/l (Fig. 2b and 2c).

The acetate depth profile generally resembled the TOC profile (with the exception of the sample at 55 cm) and showed higher concentrations in the surface interval I and in the deepest interval IV. Formate showed an equal depth distribution with exception of the surface layer where almost no formate was detectable.

Chloride was the predominant anion with up to 38,000 mg/l in interval I (Fig. 2d), followed by sulfate with concentrations (Fig. 2e) up to 7500 mg/l in the surface interval. Below the surface layer chloride and sulfate concentrations decreased down to 480 and 370 mg/l, respectively before starting to slightly but progressively increase again from 120 to 170 cm to 9560 and 1880 mg/l. Nitrate was mainly detected in surface interval I with values up to 26 mg/l (Fig. 2f). In intervals II and III concentrations were quite low (around 2 mg/l) with an exception at 70 cm depth (21 mg/l). Below 70 cm, nitrate could not be detected. Dissolved fluoride was found in the surface interval I and in deepest interval IV of Witpan sediment (Fig. 2g) and showed a similar profile to that of TOC (Fig. 2a). The water content was around 0.6% in the top layers and increased slowly but more or less steadily with depth to up to ~12% (Fig. 2h).

3.2. Analyses of phospholipid fatty acids (PLFA) life markers and intact archaeal lipids

In Witpan sediments saturated ($C_{14:0}$ to $C_{20:0}$), branched (*iso*/*anteiso*-$C_{15:0}$, *iso*-$C_{16:0}$, *iso*/*ai*-$C_{17:0}$, 10Me-$C_{16:0}$), unsaturated ($C_{16:1\omega9}$, $C_{16:1\omega7c}$, *cis*/*trans*-$C_{16:1\omega5c,t}$, $C_{18:1\omega7c}$ and $C_{18:1\omega9}$) and cyclo (cy-$C_{17:1}$ and cy-$C_{19:1}$) PLFAs were identified. PLFAs were detectable down to 123 cm depth (Fig. 3a). In surface interval I the highest diversity of PLFAs was observed (Fig. 3a). Overall cyclopropyl and mono-unsaturated PLFAs dominated the surface interval (Fig. 3a and Table 1). In intervals II to IV, the relative proportion of saturated PLFAs significantly increased, more or less dominating the PLFA profile (Fig. 3b). The proportion of branched PLFAs appeared to increase slightly below the surface interval. Additionally, the *iso*/*anteiso* ratio of $(iC_{15:0} + iC_{17:0})/[(iC_{15:0} + iC_{17:0}) + (aiC_{15:0} + aiC_{17:0})]$ PLFAs showed a trend to more *iso*-branched PLFAs within the surface interval (Fig. 3c). Highest concentration of bacterial life markers are identified in the top interval I with up to ca. 31,000 ng/g sed. Below a decrease down to values around 800 ng/g sed was observed (Fig. 6a).

Intact archaeal membrane lipids were also detected in Witpan deposits (Fig. 4). Only dialkyl glycerol diether (DGD) lipids were detected. Fig. 4 indicates the presence of archaeol phosphatidic acid (ArPA), hydroxyarchaeol phosphatidyl glycerol (ArOH-PG), archaeol phosphatidyl glycerol (ArPG) and archaeol phosphatidyl glycerophosphate methyl ester (Ar-PGP-Me). Additionally, their structurally related DGDs with one phytanyl (C_{20}) and one sesterterpanyl (C_{25}) ether side chain were detected in surface interval I.

3.3. Analyses of the past microbial biomarkers

Archaeol was detected throughout the entire sediment core (Fig. 5a). The archaeol profile revealed enhanced concentrations up to 1100 ng/g sed (sediment dry weight) in surface interval I. In interval II the concentration was quite low and increased slightly in interval III with a spike at 92 cm depth (3000 ng/g sed). In the deepest core section (interval IV), concentrations gradually increased up to 1400 ng/g sed at 160 cm depth. Crenarchaeol was only present in trace amounts (< 5% of total GDGTs, Fig. 5b). It was mainly detected in the surface layers with values up to 0.6 ng/g sed and additionally in the deepest core section (interval IV). Isoprenoid GDGTs (iGDGTs) were detected with 0–3 cyclopentyl rings (Fig. 5c and d). Within this compound group, iGDGT-0 was the dominant compound (Fig. 5c). While detection of iGDGT-0 was low in intervals I, II and III, in interval IV it showed a depth profile similar to that of archaeol. This can also be seen in the ratio of archaeol vs GDGT-0 which showed a sudden decrease especially in interval IV (Fig. 5e). The contents of iGDGTs 1–3 content is much lower than for iGDGT-0. iGDGT-1 and iGDGT-2 contribute 10–30% in interval III and IV and < 5% in interval I and II to the total content of iGDGTs in Witpan deposits. Archaeal biomarkers including archaeol and iGDGTs (up to ~3300 ng/g sed., Fig. 6b) significantly dominated over bacterial branched GDGTs (brGDGTs; up to ~100 ng/g sed, Fig. 6c). The depth profile of brGDGTs for the deeper core section (intervals II–IV) showed some similarities to the archaeal signal, however, there is only a small amount of these biomarkers in surface interval I. At our study site, established parameters to reconstruct pH and mean air temperature, like Cyclisation of Branched Tetraether (CBT) and Methylation of Branched Tetraether (MBT) ratios (Weijers et al., 2007) were not applicable due to the overall low abundance of brGDGTs, especially those with one or two additional cyclopentyl rings.

4. Discussion

4.1. Pan deposits as life habitat for microorganisms

Investigation of the chemical constituents in pan sediments provides valuable information on potential substrates (electron donor and acceptor) for microbial metabolisms and on the environmental living conditions of the indigenous microbial community.

Witpan is covered by a salt-rich loose layer about 9 cm thick with a firm salt crust on top. Chloride and sulfate are the predominant anions in the top layer. Solutes are concentrated in closed depressions induced by natural physical and chemical processes (Mares, 1999). In semi-arid to arid environments rain dissolves salts in the top layers and will evaporate from the soil before it can deeply infiltrate. This process leads to an accumulation of salts within or at the surface of the top soil causing the high saline conditions in the pan deposits. Long-term drying results in accumulation of chloride in top soils by an evapotranspirative enrichment of infiltrated rainfall in semi-arid regions, as reported for the southwestern United States and Australia (Scanlon et al., 2007). Chloride provides information on water movement due to a relatively simple cycle. Infiltrating precipitation causes chloride to move into or through the upper layers of the soil and accumulate upon evaporation of the water. Chloride moves conservatively in liquid water through the hydrologic cycle (Scanlon et al., 2009).

Sulfate is another major anion but with a more complex cycle, because it has several sources as well as sinks and is involved in different biochemical reactions. High concentrations of sulfate in the top layers contribute to soil salinity (Scanlon et al., 2009). Nitrate is formed in many semi-arid regions from both atmospheric deposition and nitrogen fixation (Walvoord et al., 2003; Deans et al., 2005). Nitrate also occurred mainly close to the surface layer of Witpan (Fig. 2f). Both nitrate and sulfate are excellent electron acceptors for heterotrophic metabolism (Bertrand et al., 2015).

Sulfate and fluoride are subject to ion exchange processes such as adsorption, desorption and surface complexation (Alloway, 2013). Fluoride plays an important role in semi-arid regions, where high levels are common in groundwater. Elevated dissolved fluoride concentration could be also a signal of eroded material in the pan. Higher concentrations in surface layers and interval IV (Fig. 2g) may indicate an increased supply of material into the pan during intervals I and IV. This might indicate enhanced water supply during these periods.

Buried organic matter is the most important carbon and energy source (electron donor) for organo-heterotrophic microorganisms in sedimentary systems (Schaechter, 2009) and small organic acids such as acetate and formate are preferred substrates for microor-

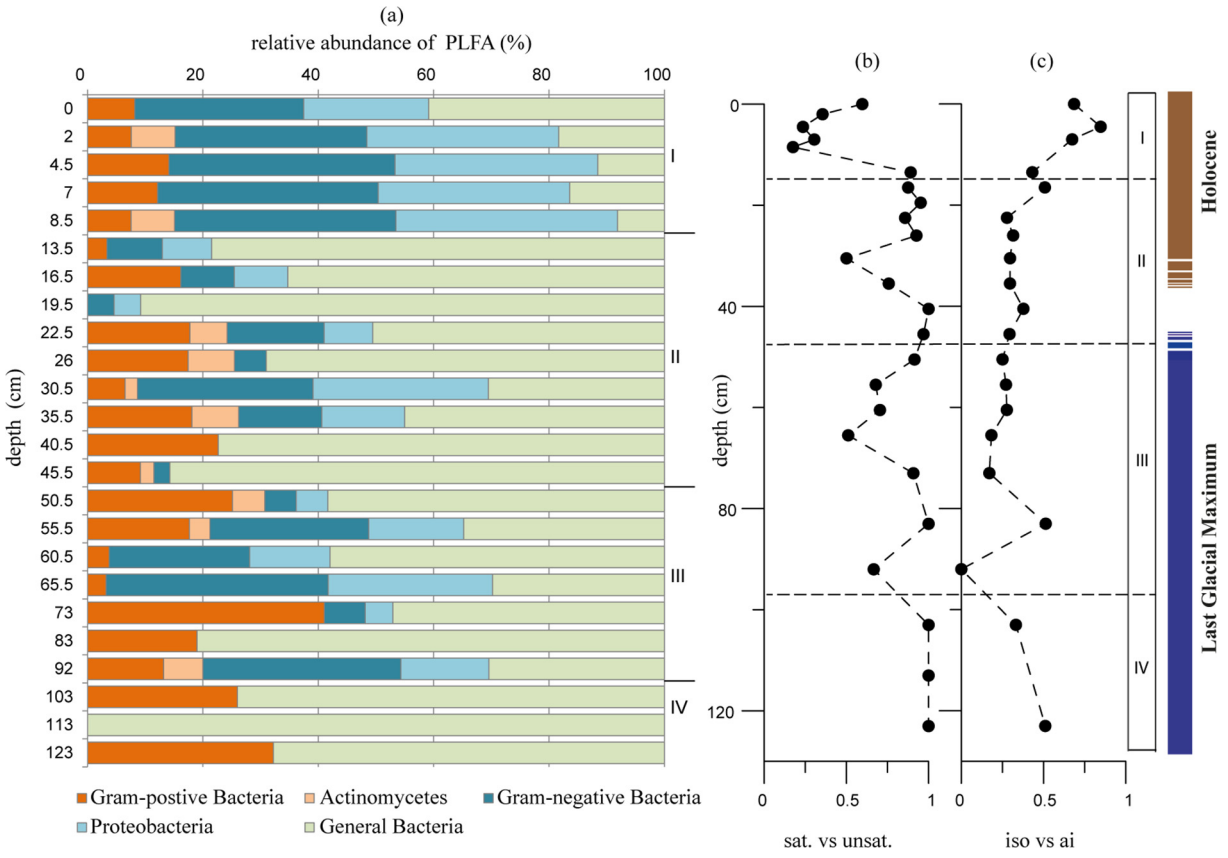

Fig. 3. (a) Relative abundance of phospholipid fatty acid (PLFA) with depth transferred into broad taxonomic information on the microbial community composition in Witpan sediments: (orange) gram-positive bacteria, (light orange) *Actinomycetes*, (blue) gram-negative bacteria, (light blue) Proteobacteria and (light green) general bacteria. Assignments and references see Table 1. (b) Ratios of total saturated/monounsaturated FAs ($C_{14:0} - C_{20:0}/(C_{14:0} - C_{20:0}) + (C_{16:1\omega9}, C_{16:1\omega5} + C_{16:1\omega7c} + C_{18:1\omega7} + C_{18:1\omega9})$ and (c) *iso*/*anteiso* FAs ($iC_{15:0} + iC_{17:0}/(iC_{15:0} + iC_{17:0}) + (aiC_{15:0} + aiC_{17:0})$ in Witpan sediments with depth. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

Table 1
Phospholipid fatty acids (PLFAs) and their origin. ω = methyl branch or double bond position counted from the tail end side of the fatty acid; *iso*/*anteiso* = methyl branch in ω2 and ω3; X:Y = number of carbon atoms and number of double bonds; c = double bond in cis-configuration; cyc = cyclopropyl ring.

Lipid marker (fatty acids)	Microorganism	References
iso/*ai*15:0, *iso*16:0, *iso*/*ai*17:0 (branched and saturated)	Gram-positive	Kaur et al. (2005)
10Me16:0	Actinomycetes	Zhang et al. (2007)
16:1ω5, 16:1ω7c, 16:1ω9c, 18:1ω7c, 18:1ω9c, cyc17:1. cyc19:1 (monoenoic and cyclopropane unsaturated)	Gram-negative	Piotrowska-Seget and Mrozik (2003) and Zelles (1999)
16:1ω7c, 18:1ω7c, 18:1ω9c	Proteobacteria	Ringelberg et al. (2008)
14:0, 15:0, 16:0, 17:0, 18:0, 19:0, 20:0 (saturated)	General bacteria	Rhead et al. (1971)

ganisms (see below). Thus, for the Witpan sedimentary fills our data indicate increased substrate potential (electron donors and acceptors) for microorganisms in surface interval I and in the deeper intervals III and especially IV (Fig. 2b, c, e and f).

4.2. Living microbial communities in pan deposits

Phospholipid-derived fatty acids (PLFAs) in sedimentary systems are characteristic markers for living bacterial communities (Zelles, 1999). Thus, the occurrence of PLFA markers indicates the presence of bacterial life in Witpan deposits. Especially surface interval I shows higher concentrations, indicating the influence of near-surface processes on the abundance and presumable activity of the bacterial community. The increased abundance of PLFAs in the surface interval I fall together with an enhanced content of substrates (TOC, acetate; Fig. 2a, b and c) and electron acceptors (sulfate, nitrate: Fig. 2e and f). Usually acetate as well as sulfate and nitrate are rapidly consumed in an active microbial environment (Bertrand et al., 2015); thus, the fact that their concentrations are enhanced in the surface layers might indicate restricted or seasonal microbial activity. The reason for this is probably the low water content in the surface sediments (Fig. 2h). Free water is important for substrate exchange and metabolic processes and therefore represents a prerequisite for microbial life and activity. The Witpan sampling campaign was at the end of the dry season (May to November), a time of presumably reduced microbial activity. However, the strong PLFA signal for microbial life in the surface layers suggests the presence of bacteria which might be active at a low level during this time and then increase their activity during the rainy season (December to April) when more water becomes available (approx. 150 mm/year in this season). In interval II bacterial life signals are low and even in the deeper intervals III and especially IV where the potential substrates (TOC, acetate and formate) increase again higher levels of life markers could not be detected, indicating that an enhanced deeper microbial community does not exist in the investigated sediments.

The PLFA side chain inventory represents a fingerprint of the community structure on a broad taxonomic level (Kaur et al., 2005). This method can be used to identify several phyla of Bacteria. The PLFAs were separated into five groups (Table 1), according

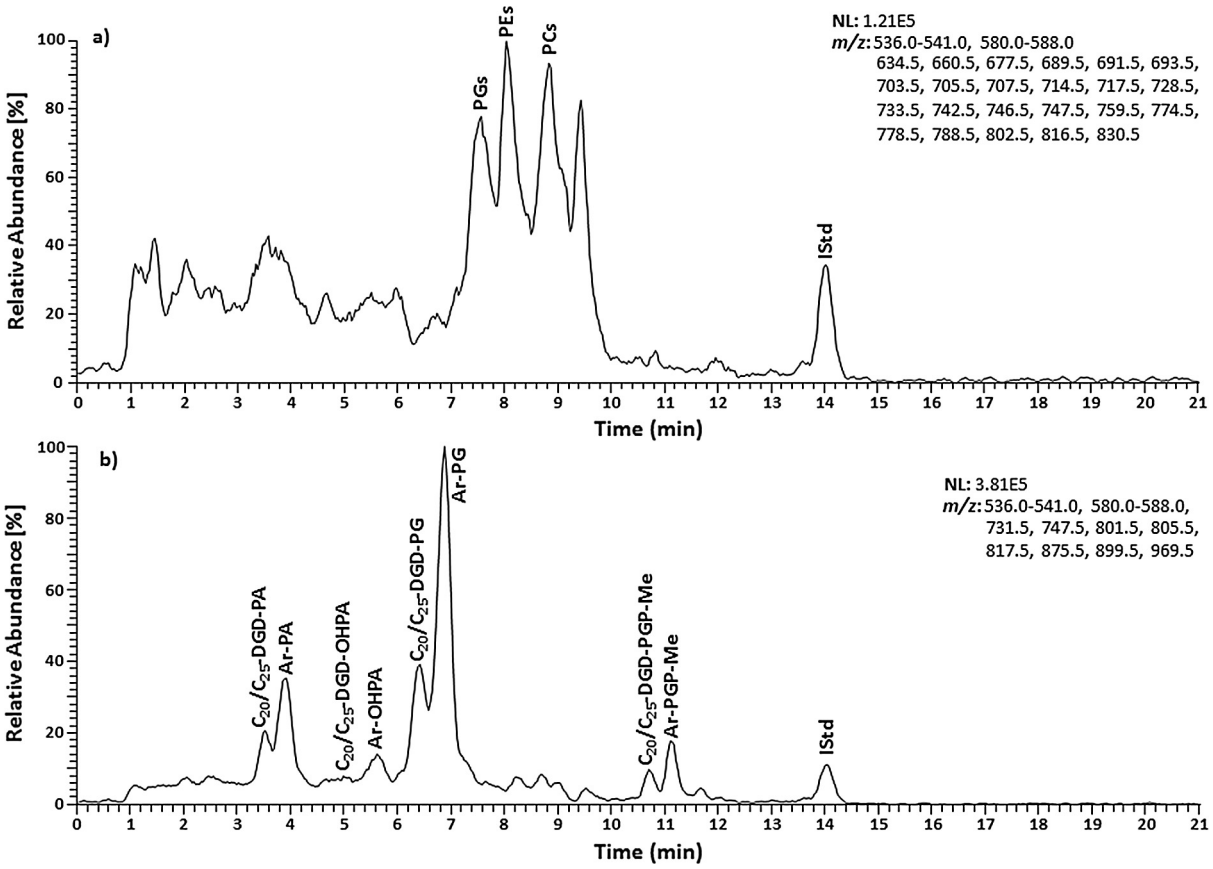

Fig. 4. HPLC–ESI-MS run of: (a) detected (bacterial) intact phospholipid esters, and (b) of detected (archaeal) intact phospholipid ethers extracted from Witpan sediment material at 1 cm depth. PGs = phosphatidyl glycerols, PEs = phosphatidyl ethanolamines and PCs = phosphatidyl cholines. Please note that each PG, PE and PC peak represent a series of these compounds (same head group) with different fatty acid side chains. Ar-PA = archaeol phosphatidic acid, ArOH-PG = hydroxyarchaeol phosphatidyl glycerol, Ar-PG = archaeol phosphatidyl glycerol and Ar-PGP-Me = archaeol phosphatidyl glycerophosphate methyl ester. Additionally, their structurally related dialkyl glycerol diethers (DGDs) with one phytanyl (C_{20}) and one sesterterpanyl (C_{25}) ether side chain were detected.

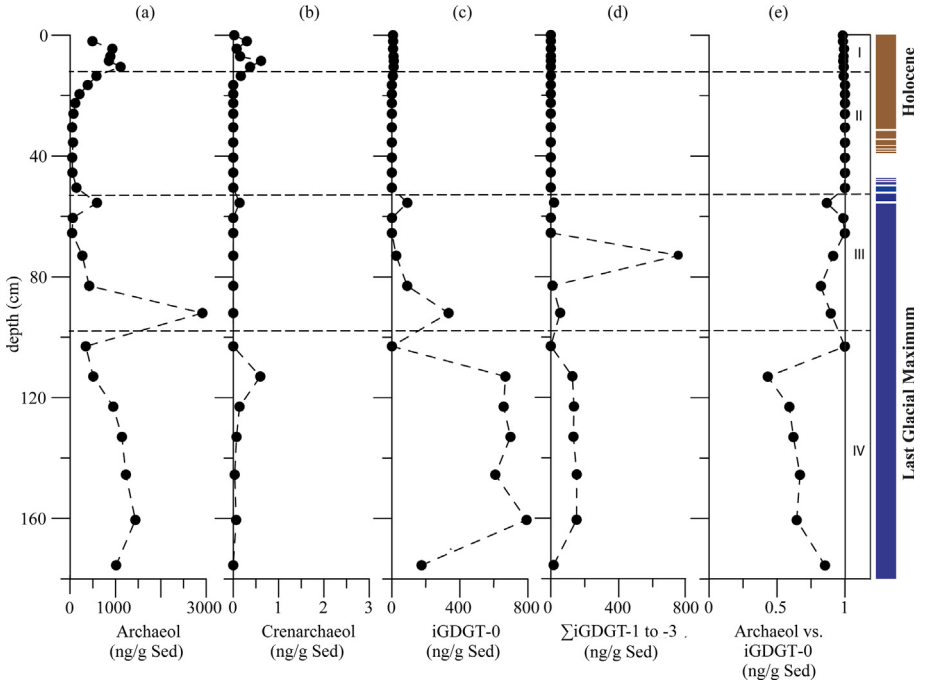

Fig. 5. Past microbial lipid biomarkers of Witpan deposits with depth: (a) archaeol, (b) crenarchaeol, (c) isoprenoid glycerol dialkyl glycerol tetraether with no cyclopentyl rings (iGDGT-0), (d) iGDGT-1 to -3 (1–3 cyclopentyl rings), and (e) ratio of relative abundance of archaeol and iGDGT-0. Note different scales of *x*-axis.

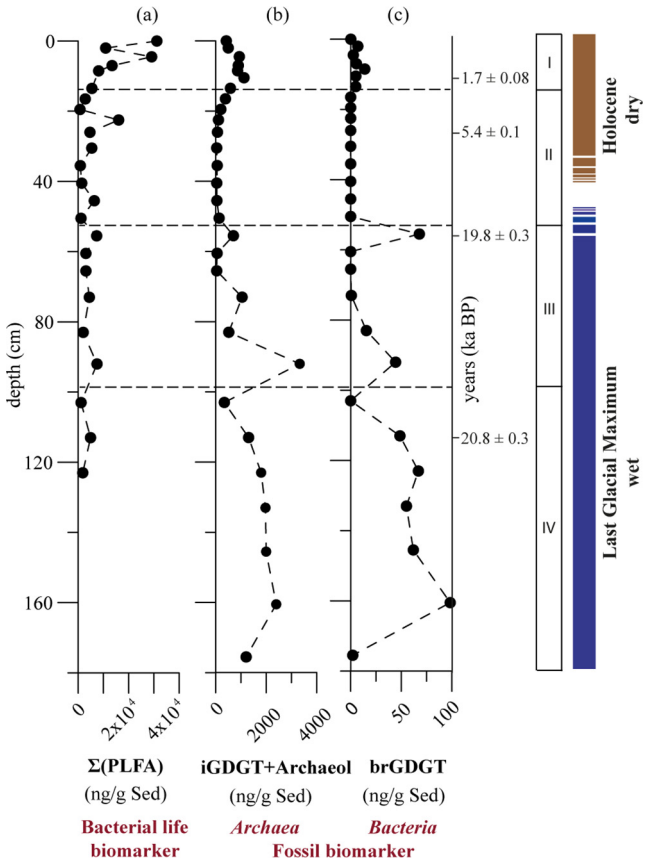

Fig. 6. Depth profiles of phospholipid fatty acid life markers and past microbial biomarkers in the Witpan sediments from Last Glacial Maximum to Holocene time. (a) phospholipid derived-fatty acid (PLFA) profile as indicator for viable bacteria, (b) isoprenoid glycerol dialkyl glycerol tetraethers (iGDGT-0 to 3) + archaeol and (c) branched glycerol dialkyl glycerol tetraethers (brGDGT) as indicators for past archaeal and past bacterial life, respectively. Age data provided by Schüller and Wehrmann (2016). Note different scales of x-axis.

to their structural similarity or biological origin. Saturated fatty acids such as C_{16} and C_{18} fatty acids are part of the lipid inventory of most living organisms (Rhead et al., 1971). Due to their ubiquitous occurrence in microorganisms they are not very specific. The saturated and branched PLFAs, e.g. *iso*/*ai*-$C_{15:0}$, *iso*/*ai*-$C_{17:0}$, are known biomarkers for gram-positive bacteria (Kaur et al., 2005; Romano et al., 2008; Villanueva et al., 2014) and they are often abundant in hypersaline environments (Ventosa et al., 1998; Ghozlan et al., 2006). Actinomycetes, indicated by 10Me-$C_{16:0}$ (Zhang et al., 2007), represent a small percentage (Fig. 3) of the microbial community in Witpan deposits, probably due to a lower tolerance to salinity than other bacteria (Zahran, 1997; Fierer et al., 2003). Actinomycetes are gram-positive bacteria and they are able to form endospores, which are resistant to desiccation. As outlined in Table 1, gram-negative bacteria contain a large proportion of cyclopropane and monounsaturated fatty acids (Zelles, 1999; Piotrowska-Seget and Mrozik, 2003). Furthermore, monoenoic FA with 16 and 18 and cyclopropyl FA with 17 and 19 carbon atoms (see Table 1 dark blue group for comparison) are discussed in the context of halophilic bacteria (Ventosa et al., 2011). It was observed that high NaCl concentrations cause increased proportion of cyclopropyl FA (Ohno et al., 1979).

In the current study, the phospholipid composition of the top interval I differ from those below. PLFAs, characteristic of gram-negative bacteria, dominate the top layers (Fig. 3a) which might indicate a hypersaline environment and good living conditions for halophiles. The content of these compounds increases with salt concentration and refers to an increase of gram-negative halophilic bacteria (Ventosa et al., 1998). Particularly in carbon-limited areas of arid environments, microbial communities are often characterized by photoautotrophic organisms, such as Proteobacteria. Especially the upper 10 cm of Witpan are characterized by PLFAs that are typical of Proteobacteria (Ringelberg et al., 2008, Table 1). Gram-negative Proteobacteria are distributed worldwide and are often found in desert soil bacterial communities (Spain et al., 2009; Lefèvre et al., 2012). They are considered to be relevant in nutrient-limited arid environments (Boldareva-Nuianzina et al., 2013). Below the surface interval I a higher proportion of non-specific saturated PLFAs dominate with an overall significantly decreasing abundance of PLFAs (Fig. 3a). However, the discussed FAs and associated microbial groups still play a role but on a much lower level.

The ratio of saturated to monounsaturated fatty acids indicates a higher proportion of unsaturated fatty acids in the near-surface sediments (Fig. 3b). This ratio often reflects temperature adaptation, where a higher proportion of saturated fatty acids indicates an adaptation towards higher temperatures (Russell, 1989). Thus, the ratio seems not to resemble temperature adaptation but rather variations in the overall microbial community in response to the stronger halophilic conditions in the surface layer. By regulating the relative proportion of *iso* to *anteiso* fatty acids in their cell membranes, microbes can adapt to extreme temperature conditions. At higher temperatures often an enhanced proportion of *iso* fatty acids is observed (Kaneda, 1991). In the present study, the *iso*/*anteiso* ratio of fatty acids indeed shows an upward directed trend to more *iso*-fatty acids, which might indicate an adaptation towards warmer surface conditions (Fig. 3c).

Additionally, surface interval I contains a series of intact archaeal membrane markers (Fig. 4). Their potential as life markers is restricted due to their higher stability compared to PL esters (Logemann et al., 2011). However, their simultaneous occurrence with the PL ester life markers suggests a living bacterial and archaeal community in the surface interval I of Witpan. The intact archaeal lipids detected are known to occur in halophilic archaea (Kates, 1993; Oren, 2002), which is in accordance with the high salt concentration in the top layers of the pan. Also the presence of archaeal lipids with a C_{25} ether side chain is characteristic for halophilic archaea (Kates, 1993; Dawson et al., 2012). Currently ongoing microbiological investigations will provide a deeper insight into the species forming the halophilic archaeal and bacterial community in Witpan.

Aridity and the associated saline and alkaline sediment conditions have a large influence on microbial ecosystems (Shen et al., 2008). The current data suggest increased microbial activity for the surface deposits of Witpan and there are indications that near-surface microbial communities show seasonal microbial activity during wet periods. Johnson et al. (2005) and Schirmack et al. (2015) also reported that microorganisms can desiccate and be inactive for most of the time but become quickly hydrated and active again when water becomes available.

In interval IV the low numbers of PLFA life markers (Fig. 6a) indicate a low abundance of living microorganisms despite an increasing feedstock potential (TOC, acetate and formate) in deeper sediments below 95 cm depth. Low water content affects transport, survival, and activity of microorganisms (Kieft et al., 1993). In contrast, in the surface layer water potential fluctuates greatly during rainy events, which might lead to occasional or seasonal stimulation of microbial activity in surface layers.

4.3. Past microbial response to climate changes in southern Kalahari pan deposits

Two types of GDGTs were preserved in Witpan deposits: iGDGTs indicating past archaeal biomass and brGDGTs represent-

ing past bacterial biomass (Figs. 5 and 6). The concentrations of brGDGTs and iGDGTs in terrestrial sediments potentially reflect the relative supply of GDGT-producing bacteria and archaea (Jia et al., 2013). Usually iGDGTs are the dominant fossil biomolecules in marine and lacustrine environments (Schouten et al., 2013), whereas brGDGTs are predominant in soils (Weijers et al., 2006, 2007). Isoprenoid GDGT concentrations of Witpan were significantly higher than those of brGDGT (Fig. 6b and c), indicating that the input of soil organic material into the pan system from the surrounding is overall only a minor process. Microbial signals within the pan seem to be mainly produced in situ within the respective time and are not only reworked material from older sediments and soils surrounding the pan (see also Section 2.2 for comparison).

Archaeol is the dominant microbial biomarker in surface interval I. Halophilic archaea synthesize only archaeol and no iGDGT-0. Therefore, the relative abundance of archaeol vs iGDGT-0 has been used as a paleosalinity proxy for hypersaline systems due to the predominance of halophilic Euryarchaeota (Turich and Freeman, 2011; Wang et al., 2013). As outlined above the salt concentration in Witpan surface interval I is much higher than in the underlying sediments and also salt crystals are found below the firm salt crust on top. The surface interval is rich in chloride, sulfate, nitrate and fluoride (Fig. 2) and hosts an abundant halophilic microbial community. Thus, the increased archaeol and low iGDGT concentrations in interval I can be explained by a pronounced halophilic archaeal community in the salt-rich surface layers of Witpan. Since a living halophilic community was detected close to the surface (see Section 3.2), the archaeol signal in the upper 20 cm likely reflects degraded remnants of a currently living and active archaeal community adapted to the high salt conditions. Compared to all other iGDGTs, crenarchaeol is mainly detected in the surface layers (Fig. 5b). Crenarchaeol is prevalent in lacustrine and marine environments and most likely originates from ammonia-oxidizing *Thaumarchaeota* (Schouten et al., 2013), whereas it is only a minor compound in terrestrial systems (Hopmans et al., 2004). Due to our observation that microbial life is more abundant in near-surface deposits, the crenarchaeol signal in the top layers most likely represents the remains of living *Thaumarchaeota* in the sediments.

While in the surface interval the interpretation of the fossil microbial biomarkers is complicated by a currently living microbial community, in the deeper sedimentary section with low or no life marker detection the archaeol, iGDGT and brGDGT signals represent archaeal and bacterial communities of the past (Figs. 5 and 6). In an environment of restricted water availability past microbial biomarkers might have the potential to indicate periods of increased precipitation. Former paleoclimate studies in the Kalahari reconstructed humid and dry intervals in the past. Telfer et al. (2009) studied the paleoenvironmental history of Witpan and postulated a relatively wet phase around 20 ka BP. In addition, studies of Chase and Meadows (2007) and Shi et al. (2001) referred to an enhanced windiness during the LGM caused by stronger westerlies in the Southern Hemisphere. This resulted in increasing precipitation in the southern Kalahari during the LGM due to a spatially extended and intensified winter rainfall zone (Chase and Meadows, 2007). Pollen records from the southern Namib Desert indicated increased water availability during the Last Glacial period compared to the Holocene (Lim et al., 2016). Also, other studies described a change from wetter conditions during the LGM to drier conditions at the transition to and within the Holocene. Thus, Gasse et al. (2008) interpreted different terrestrial and nearshore proxies such as pollen data, dust grain size data and windiness patterns that were directly related to variations in rainfall seasonality, atmospheric CO_2 concentration and temperature affecting evapotranspiration. They postulated increasing temperature with a gradual drying from 14 ka to the Holocene in this region. This basic climatic information forms the background on which the past microbial biomarkers detected in Witpan deposits over time can be interpreted.

The dry Holocene period in the southern Kalahari region (Weldeab et al., 2013) is characterized by an overall low bacterial and archaeal abundance in Witpan deposits (Figs. 5 and 6). This is accompanied by very low contents of buried organic matter within these sediments (Fig. 2a), which might indicate sparse living conditions during the Holocene interval II. In contrast, the more humid LGM period (Lancaster, 2002; Gasse et al., 2008; Stone, 2014) is characterized by increased abundance of fossil bacterial and archaeal biomarkers and a rise of the organic matter and substrate parameters (TOC, acetate and formate). This clearly indicates significant environmental changes from LGM deposits (intervals III and especially IV) to the overlying Holocene sediments. Thus, the biomarker proxies points to significantly better living conditions for microorganisms during the wetter LGM, which is reasonable since moisture is an important issue for microorganisms, stimulating microbial diversity and activity (Chen et al., 2007). Therefore, the wetter conditions during the LGM are a good explanation for the higher abundance of past microbial markers in Witpan deposits from the LGM. In contrast to intervals I and II, where only archaeol is dominant, in intervals III and IV iGDGTs significantly increase relative to archaeol (Fig. 5a and c), which can also be observed in the archaeol/iGDGT-0 ratio (Fig. 5e). This implies an archaeal community less dominated by halophilic archaea during the LGM and therefore less saline habitat conditions. Although still in low concentration, brGDGTs also show enhanced abundance during intervals III and IV pointing to a somewhat higher supply of soil material from the catchment area also supporting increased precipitation during the LGM.

It might be argued that the concentration of past microbial biomarker signals is not only the result of past microbial abundance and is overprinted by post-depositional degradation with the consequence that concentration cannot be used to trace past microbial abundances. However, as mentioned above, the past microbial biomarker core lipids used here are already the product of early degradation processes (loss of their head groups) showing that the core lipids are quite stable against degradation. Furthermore, a scenario where degradation should be stronger during the dry Holocene than during the more humid LGM, which should have stimulated microbial activity by higher water availability, is not very plausible. Thus, although degradation might be involved in the transformation from a life biomarker to a past biomarker signal, the data presented here suggest that in a geological context the past microbial biomarkers are well preserved in the arid depositional environment and that the fossil biomarker signal still carries significant information on their production in the past.

The surface interval I differs from the overall dry Holocene period. At least occasionally or seasonally wetter conditions seem to stimulate a living microbial community (see Section 3.2) and accumulation of organic matter in near-surface sediments. This microbial life might be triggered by rainfall events providing biological resources such as water and nutrient into the pan system. The past biomarker signal is also increased in interval I (Fig. 5a). The situation in the top interval is complicated by the actual living microbial community and recycling of organic matter surely plays a role. However, considering the stability of the past markers, their increase in interval I might indeed indicate that conditions during the latest Holocene are different from those of the earlier Holocene period in this area. For instance, the time period of the Little Ice Age (LIA, 15th to 19th century) is thought to represent a more humid period in this region (Ramisch et al., 2017). Thus, periods of increased rainfall events such as the LIA might have stimulated again the establishment of a larger microbial community during the latest Holocene period.

Overall, our past microbial markers reasonably support the climatic conception in the southern African region with higher precipitation during the LGM and drier conditions towards and within the Holocene period (Chase and Meadows, 2007; Telfer and Thomas, 2007). Therefore, these results show that in arid to semi-arid areas past microbial biomarkers in pan systems can be an appropriate tool to indicate paleo-climatic changes, especially variations in paleo-precipitation, temperature and the resulting paleo-aridity. Thus, even though pan deposits might not be continuous and the depositional history is often complex, these data emphasized the potential of pans to preserve paleo-climatic information encoded in deposited characteristic biomolecules.

5. Conclusions

The present study investigated sediment geochemistry and microbial biomarkers for present and past microbial life in Witpan deposits in the southern Kalahari region (southwestern Africa). We evaluated the impact of past climate variations on the abundance and composition of microorganisms during the Last Glacial Maximum to Holocene.

Despite the extreme environmental conditions with low TOC contents, restricted water availability, and high salt concentration, bacterial life is present mainly in the surface sediments of Witpan and significantly decreases with depth. Thus, present microbial life seems to be closely related to surface processes, which control water, substrate and nutrient availability. Biomarkers suggest a viable microbial community dominated by halophilic microorganisms in the salt-rich surface sediments.

Low or absent biomarkers for past bacterial and archaeal life in the Holocene interval below the surface layers indicate the lack of abundant past microbial life during the Holocene sequence, considered to represent a dry period in the study area. In contrast, during the postulated wetter LGM higher abundance of past microbial life and higher feedstock potential is displayed by past microbial biomarkers and organic matter proxies. The data suggest that water availability is a driving factor for the abundance of past microbial life in Witpan.

Thus, our results show that past microbial biomarkers, particularly GDGTs and archaeol, can be used to trace paleo-climatic and paleo-environmental changes such as the precipitation history in semi-arid to arid environments. Furthermore, this study supports the potential of pan deposits to act as appropriate geo-archives for biomolecules in dry areas, where other terrestrial records are scarce.

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Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.orggeochem.2017.04.00.

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