Studies on phospholipid turnover argue against sloughing of tegumental membranes in adult *Schistosoma mansoni*

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SUMMARY

The tegumental membrane complex of *Schistosoma mansoni* is the site of interaction between the parasite and the host. The tegument is involved in uptake of many nutrients, but also plays a crucial role in the evasion of the actions of the host immune system. Essential for the success of this evasion is maintaining the integrity of the tegumental membranes. The rate of turnover of phospholipids was investigated by pulse-labelling worms cultured *in vitro*, followed by additional incubation in the presence of unlabelled substrates. Tegumental membranes were isolated, characterized using antibodies against specific tegumental proteins, and analysed. It was demonstrated that the most prominent fatty acid found in tegumental phospholipids, palmitate, incorporated rapidly into the phospholipid fraction during a 30 min pulse labelling. In a subsequent 20 h chase with unlabelled substrates, the incorporated radioactivity was lost again from the tegumental membrane complex. This high turnover of palmitate was found to be limited to phosphatidylcholine (PC) only. The turnover was due to deacylation/reacylation, and not to the sloughing of membranes as is the case in schistosomula. It is speculated that this rapid turnover of PC in the tegument of adult schistosomes plays a new and important role in the immune evasion by the parasite.

Key words: lipid metabolism, deacylation/reacylation, membrane sloughing, high performance liquid chromatography.

INTRODUCTION

The parasitic blood fluke *Schistosoma mansoni* is a long-term inhabitant of the mesenteric veins of its final host. Although schistosomes trigger an immune response, they are resistant to the actions of the immune system by employing multiple evasion mechanisms (Pearce & Sher, 1987; Jokiranta, Jokipii & Meri, 1995; Fishelson, 1995). Most of these mechanisms, such as antigen shedding or antigenic mimicry, are mediated through the highly specialized surface of the parasite. This site of interaction between parasite and host, the tegumental membrane complex, is composed of 2 closely apposed lipid bilayers overlaying a syncytium (McLaren & Hockley, 1977). On the interior side, the syncytium is limited by the basal lamina, a single lipid bilayer with large dilatations. The unusual tegumental membrane complex is synthesized from discoid and multi-lamellar bodies which are synthesized in cell bodies just below the basal lamina (Zhou & Podesta, 1989).

Besides having an important role in the evasion of the host immune system, the tegument is also involved in the uptake of nutritional substrates, of which glucose is quantitatively (one of) the most prominent (Bueding & Fisher, 1982; Tielens & van den Bergh, 1987). The uptake of the large amounts of glucose across the tegument is enhanced by the presence of facilitated diffusion glucose transporters in both the apical tegumental membranes and the basal tegumental membrane. Recently it was shown that there are 2 distinct schistosome glucose transporter proteins (SGTPs) involved in the uptake of glucose via the tegument. One of these glucose transporters, SGTP4, is only detectable after transformation of cercariae into schistosomula and is uniquely localized in the outer tegumental membrane complex that is formed during this transformation (Skelly & Shoemaker, 1996; Jiang *et al.* 1996). The second transporter (SGTP1) on the other hand, was shown to be present within the body of the worm and in the basal lamina and its dilatations, but not in the apical tegumental membrane complex of the schistosome (Zhong *et al.* 1995).

The tegumental membrane complex further contains a variety of proteins that can function as potent immunogens (Payares *et al.* 1985; Pearce *et al.* 1991; Xu *et al.* 1994) and can mediate – to some extent – protective immunity after immunization (Simpson, 1992). However, *in vivo*, the adult schistosome is

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resistant to the actions of the immune system directed against these antigens as long as the tegumental membranes are intact. When the integrity of these membranes is disturbed by the actions of drugs such as praziquantel or oxamniquine, a drug–antibody synergistic effect is observed, making the parasite susceptible to antisera that have no intrinsic helminthotoxic activity (Modha *et al*. 1990; Fallon & Doenhoff, 1995; Fallon, Fookes & Wharton, 1996).

The apical tegumental membranes, which are rich in cholesterol and phosphatidylcholine (PC), are continuously renewed from discoid and multilamellar bodies, supposedly sloughing old membranes into the bloodstream of the host (Samuelson, Caulfield & David, 1982; Allan, Payares & Evans, 1987; Kruger & Joubert, 1990). The rate at which this process is occurring is still unclear although this rate determines the importance of membrane renewal as an evasive strategy. Experiments on the turnover of the apical tegumental membranes have provided half-life values ranging from a few hours to several days, depending on the method of analysis and the stage of the parasite under investigation (Kusel & Mackenzie, 1975; Dean & Podesta, 1984; Saunders, Wilson & Coulson, 1987). Although it has been established that the administration of serotonin and complement to cultured schistosomes increases the incorporation of radio-isotope labelled choline into discoid and multilamellar bodies respectively (Young & Podesta, 1986; Zhou & Podesta, 1989), the turnover of the major structural components in the tegumental membranes, the phospholipids, has yet to be determined.

In this study, the fatty-acyl turnover in the phospholipids of the apical tegumental membranes is measured and compared with the turnover in the phospholipids of the worm body. Since schistosomes do not synthesize fatty acids *de novo* nor degrade them via β-oxidation (Meyer, Meyer & Bueding, 1970), the incorporation and loss of radio-isotope labelled fatty acids from schistosome membranes can be used to determine the dynamics of phospholipids in the separate membranes. SGTP1 and SGTP4 are used for the assessment of the purity of isolated membrane fractions as these glucose transporter proteins are highly specific markers for the membranes inside the worm (SGTP1) and the apical tegumental membrane complex (SGTP4) (Skelly, Tielens & Shoemaker, 1998).

MATERIALS AND METHODS

Chemicals

[1-^{14}C]-palmitic acid was obtained from Amersham (Buckinghamshire, UK). Acetonitrile was of HPLC grade and obtained from Labscan (Dublin, Ireland). Hepes was from Gibco (Paisley, UK). Phenacylbromide (Fluka AG, Buchs, Switzerland) was purified by recrystallization from hexane before use. All other chemicals were from Sigma Chemical Co. (St Louis, MO, USA) or Baker (Deventer, The Netherlands) and were of analytical grade.

Parasites

Schistosoma mansoni worms were isolated from ether-anaesthetized hamsters, 45–48 days after infection, by perfusion of the heart at 37 °C with an isolation buffer containing 20 mM Hepes, 85 mM NaCl, 5·4 mM KCl, 0·7 mM Na_2HPO_4, 1 mM $MgSO_4$, 1·5 mM $CaCl_2$, 25 mM $NaHCO_3$ and 100 mM glucose (pH 7·4) (Tielens & van den Bergh, 1987; Brouwers *et al*. 1997). The worms were washed twice in isolation buffer and finally once in incubation buffer before they were transformed to incubation vials.

Incubations

Worms were incubated for 30 min in 5 ml of minimum essential medium (MEM, Gibco) supplemented with 100 mM glucose, 1 % (w/v) bovine serum albumin, 35 μM stearic acid, 65 μM oleic acid, 65 μM linoleic acid and 85 μM of palmitic acid (all BSA-bound, concentrations including radio-isotope labelled fatty acids when appropriate). Worms were incubated in a gyratory shaker at 37 °C under an atmosphere of 95 % air and 5 % carbon dioxide in the presence of the radio-isotope labelled fatty acid (11·8 Ci/mol [1-^{14}C]-palmitic acid). Schistosomes were subsequently washed with excess (unlabelled) incubation buffer (37 °C) and transferred to 50 ml of fresh (unlabelled) incubation buffer for the duration of the chase. At the appropriate time-points, aliquots of schistosomes were taken from the vial. From these parasites, membrane fractions were prepared as described below.

Membrane preparations

Tegument surface membranes of adult *S. mansoni* were isolated essentially according to the method of Roberts *et al*. (1983). In brief, schistosomes were washed with Hanks balanced salt solution and plunged into liquid nitrogen. After thawing on ice, schistosomes were extensively washed with ice-cold Tris-buffered saline (TBS) containing 20 mM Tris–HCl (pH 7·4), 0·85 % (w/v) NaCl, 20 μg/ml L-1-tosylamide-2-phenyl-ethylchloromethyl ketone (TPCK), 5 μg/ml tosyl-l-lysinechloromethyl ketone (TLCK) and 1 mM phenylmethylsulphonyl fluoride (PMSF). The outer tegumental membrane complex was subsequently removed by applying ten 1 s vortex pulses to the parasites. The supernatant was passed over a fine stainless-steel mesh and the filtrate centrifuged at 5000 ***g*** for 30 min. The resulting

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pellet was resuspended in TBS or sample buffer (3 % SDS, 4 M urea, 10 % glycerol, 0·1 % bromophenol blue, 100 mM Tris–HCl, pH 6·8) and considered the final preparation of apical membranes.

From the resulting 'stripped' worms, membranes were isolated as described by Zhong *et al*. (1995). Stripped worms were thoroughly homogenized in ice-cold TBS containing the protease inhibitors described above using a Teflon-glass homogenizer. After addition of 10 volumes of 20 % (w/v) sucrose, the suspension was centrifuged at 4 °C for 10 min at 1000 ***g***. The supernatant was subsequently centrifuged at 105000 ***g*** for 1 h at 4 °C. The resulting pellet was resuspended in TBS or sample buffer and referred to as 'stripped worm membranes'.

Proteins were measured according to the Lowry method of Bensadoun and Weinstein using bovine serum albumin as a standard (Bensadoun & Weinstein, 1976). Alkaline phosphatase was measured according to the method of Roberts *et al*. (1983), with the incubation time adapted to 1 min because of observed inhibition of phosphatase activity by Triton X-100 during prolonged assays.

Analysis of lipids

Lipids were extracted according to the method of Bligh & Dyer (1959). Organic extracts were dried under nitrogen and dissolved in chloroform: methanol (1:1, v/v) for further analysis. Lipid classes were separated into triacylglycerols, free fatty acids, diacylglycerols and phospholipids by thin-layer chromatography on silica-G plates using petroleum ether: diethylether: acetic acid (75:25:2, v/v/v) as a mobile phase. For analysis of acyl chains, lipids were visualized by spraying the plates with rhodamine-6G in methanol: water (1:1, v/v). Spots were scraped off and transferred to screw-capped tubes. Lipids were hydrolysed in 3·5 ml of 0·3 M NaOH in methanol for 2 h at 73 °C. Non-saponified material was extracted with 2 × 1 vol. of petroleum ether. Fatty acids were extracted after acidification of the methanolic phase with 250 μl of 6 M HCl, with 3 × 1 vol. of petroleum ether. The isolated fatty acids were dried under nitrogen and converted to their phenacyl derivatives, dried under nitrogen and redissolved in acetonitrile (Wood & Lee, 1983).

SDS–PAGE and immunoblotting

SDS–PAGE was performed as described by Laemmli (1970) on 10 % SDS polyacrylamide gels. For immunoblotting, proteins were transferred to nitrocellulose membranes as described (Burnette, 1981). Nitrocellulose blots were incubated overnight in a buffer containing 20 mM Tris–HCl (pH 7·5), 0·5 M NaCl (TBS) and 1 % gelatin containing a 1:400 dilution of a rabbit anti-SGTP1, or a 1:100 dilution of a rabbit anti-SGTP4 antibody (Zhong *et al*. 1995; Jiang *et al*. 1996; Skelly & Shoemaker, 1996). After extensive washing with 20 mM Tris–HCl (pH 7·5), 0·5 M NaCl and 0·05 % Tween 20 (TTBS), the blot was incubated for 1 h in a 1:3000 dilution of goat anti-rabbit IgG peroxidase conjugate. After washing the nitrocellulose blot twice in TTBS and once in TBS for 10 min, schistosome glucose transporter proteins were visualized by incubation of the blot in the presence of 3,3′ diaminobenzidine and hydrogen peroxide.

High performance liquid chromatography (HPLC) procedures

Fatty acid phenacyl esters (FAPEs) were eluted isocratically from a 250 × 4 mm ODS-column (Merck, Darmstadt, Germany) with acetonitrile at a flow rate of 1 ml/min. Detection was at 242 nm using Perkin Elmer Nelson analytical software (Cupertino, Ca, USA) for data analysis. The detector response was linear with the molar amount of FAPE and independent of the FAPE-species (Wood & Lee, 1983). Peaks were identified by comparing retention time of samples, before and after saturation of double bonds with hydrogen and platinum, with known standards (Christie, 1989; Brouwers *et al*. 1998). When radioactive samples were used, fractions were collected every 20 s from the outlet of the UV-detector in scintillation vials, evaporated to dryness and radioactivity was counted after addition of PPO/POPOP-based scintillation fluid (Pande, 1976).

RESULTS

Purity of membrane fractions

Several methods have been published for the purification of the outer tegumental membrane complex (Kusel, 1972; Simpson *et al*. 1981; Oaks *et al*. 1981; McDiarmid, Dean & Podesta, 1983; Roberts *et al*. 1983). Of these, the freeze–thaw method developed by Roberts *et al*. (1983) is most widely used because it is relatively simple, removes the apical tegumental membranes quantitatively and yields membranes of adequate purity. In our experiments, we omitted the final step, purification of membranes over a sucrose gradient, because of the low recovery of alkaline phosphatase activity and phospholipids that we obtained. Instead, we introduced a filtration directly after removal of the apical tegumental membranes from the parasite. In this step, small worm fragments arising from the vortex pulses were removed. This procedure resulted in an apical tegumental membrane fraction in which the specific activity of alkaline phosphatase was increased 20-fold to 7·8 ± 0·1 U/mg protein ($n = 5$). Detection of cross-contamination between basal lamina and apical tegumental membranes was performed with the use of antibodies directed against

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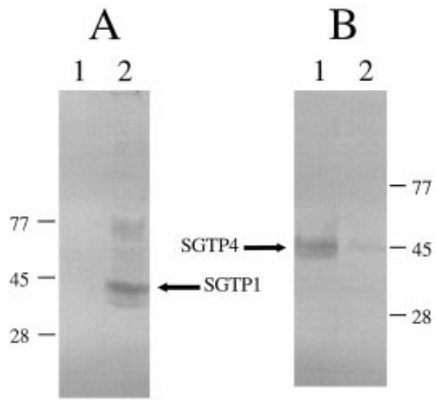

Fig. 1. Western analysis of purified membrane fractions from *Schistosoma mansoni* using antibodies directed against (A) the glucose transporter in the basal lamina and membranes inside the worm (SGTP1) and (B) the glucose transporter in the apical tegumental membranes (SGTP4). In (A) as well as in (B) lane 1 was loaded with isolated apical tegumental membranes, lane 2 contained membranes isolated from stripped worms. The amounts of sample loaded in lanes number 1 and 2 (A and B) correspond to an equal number of parasites.

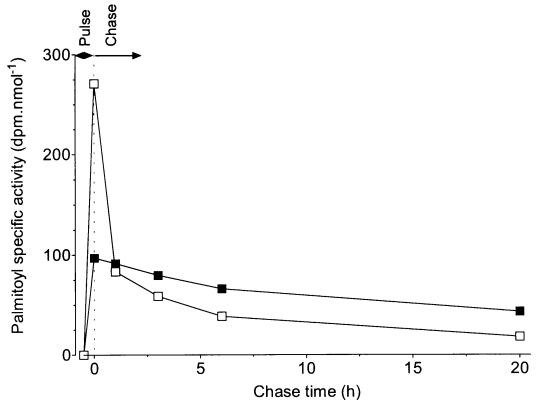

Fig. 2. Turnover of palmitic acid in the phospholipid fractions of *Schistosoma mansoni*. Schistosomes were pulse-labelled with [1-^{14}C] labelled palmitic acid for 30 min before they were transferred for varying time-periods into a medium containing only unlabelled substrates. After the appropriate chase period, the specific activity of the label was determined in the aliphatic chains of the phospholipids of the apical tegumental membrane complex (□) and the stripped worms (■) respectively. Shown is a representative experiment which was performed 3 times with similar results (twice with palmitate, once with oleate).

SGTP1 and SGTP4. In Fig. 1, Western blots were shown of the apical tegument membrane sample and the stripped-worm membrane sample, probed with antibodies against SGTP1 (Fig. 1A) and SGTP4 (Fig. 1B), respectively. The lanes containing the apical tegumental membranes in Fig. 1A and B were loaded with an equal amount of membrane proteins (5 μg). Also the amount of stripped-worm membranes loaded was equal in Fig. 1A and B (180 μg protein) and corresponded to an equal number of parasites as were used for the preparation of the apical tegumental membranes in the adjacent lanes. SGTP1 was recognized in the preparation of the membranes of the stripped worm, but not in the apical tegumental membrane fraction (Fig. 1A). SGTP4 on the other hand, was mainly detected in the apical tegumental membranes, although a faint band is seen in the lane containing membranes from stripped worms (Fig. 1B).

High turnover of fatty-acyl chains in tegumental phospholipids

To investigate the turnover of the fatty acyl chains in the phospholipids of the apical tegumental membranes in comparison to membranes in the stripped worms, schistosomes were incubated in the presence of [1-^{14}C]palmitate for 30 min. During this pulse period, radio-isotope labelled palmitate was incorporated rapidly into the phospholipids of both membrane systems, resulting in an increase of the specific activity of the palmitoyl chains linked to phospholipids of both membranes (Fig. 2). Shortly after the end of the pulse period, all label had been incorporated into complex lipids, and the amount of labelled free fatty acids in the medium as well as in the schistosomes was minimal (data not shown). The apical tegumental membranes had incorporated palmitate faster into their phospholipids than the membranes of the stripped worms, as the increase of palmitoyl specific activity was greater in the tegumental phospholipids than in the phospholipids of the stripped worm (Fig. 2). During the subsequent chase period, in which schistosomes were incubated for an additional 1–20 h in medium containing only unlabelled substrates, the specific activity of the phospholipid acyl chains in both membrane fractions decreased. The decrease in specific activity occurred more rapidly in the apical tegumental membranes. After the 20 h chase period nearly all radio-isotope labelled palmitoyl chains had disappeared from these tegumental membranes, this in contrast to the membranes of the stripped worms where still relatively large amounts of labelled palmitoyl chains were present (Fig. 2). The same pattern of rapid labelling and disappearance of the label in the apical tegumental membrane complex was observed after labelling with oleate (18:1) (not shown).

Metabolic activity of individual lipid classes in the tegumental membranes

The most predominant phospholipids found in the apical tegumental membranes were PC and phosphatidylethanolamine (PE), which is in accordance with previous characterizations of these tegumental membranes (Rogers & McLaren, 1987; Allan *et al.* 1987). The fatty acyl turnover in these 2 individual phospholipid classes was determined, to detect any difference in acyl turnover between these phos-

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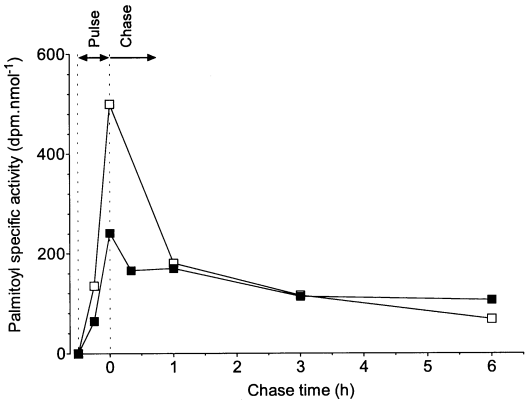

Fig. 3. Decrease in specific activity of palmitate esterified to phosphatidycholine (PC) in the tegument membranes of adult *Schistosoma mansoni* (□) and in the stripped worms (■). Worms were pulse-labelled and incubated as in Fig. 2. Shown is a representative experiment which was performed twice with similar results.

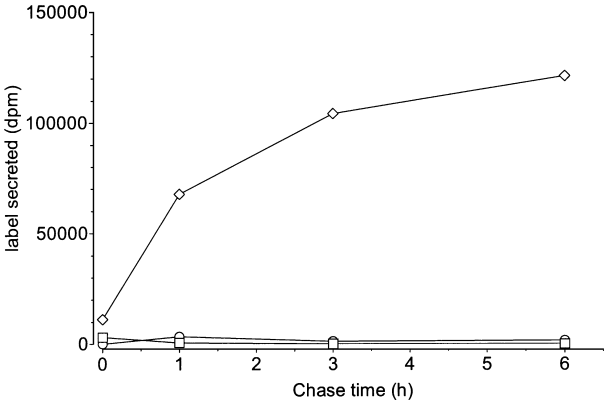

Fig. 4. Label secreted by adult schistosomes after a 30 min pulse labelling with [1-^{14}C] palmitate. After labelling, schistosomes were transferred to unlabelled medium and cultured for an additional 6 h. Samples of the culture medium were taken, extracted and the lipid classes were separated on TLC as described previously (Brouwers *et al.* 1997). Radio-isotope labelled free fatty acids (◇) were recovered in significant amounts from the medium. Only trace amounts of label associated with triacylglycerols (○) and phospholipids (□) were detectable. Shown is 1 example of a triplicate experiment.

pholipids. After analysis, significant amounts of radio-isotope labelled palmitate could only be recovered from the PC fraction, and not from the PE (or in fact, any other phospholipid) fraction. The specific activity of the fatty acyl chains of PC decreased rapidly during the subsequent chase (Fig. 3) in accordance with the observations on the total phospholipid fraction (Fig. 2).

Membrane sloughing versus acyl turnover

Analysis of the culture medium was performed to determine the molecular form of the secreted label. Free fatty acid was the only isotope-labelled lipid fraction that could be recovered from the medium, there was only a trace amount of radioactivity associated with (secreted) phospholipid or triacylglycerol (Fig. 4). It is unlikely that the tegumental membranes were the only source of the label that was recovered from the medium, as there was a larger quantity of label recovered from the medium than was incorporated into these membranes (data not shown). More important, however, is the fact that there were no labelled membrane constituents, i.e. phospholipids, recovered from the medium. This strongly suggests that there were no sloughed membranes present in the medium, as this would have led to the presence of radio-isotope labelled phospholipids.

DISCUSSION

Isolation of apical tegumental membranes

The specific activity of the enzyme alkaline phosphatase has frequently been used to assess the purity of isolated membrane fractions (Cesari, Torpier & Capron, 1983; Roberts *et al.* 1983; Payares, Smithers & Evans, 1984). Although this is a rapid and convenient method, a disadvantage is that the enzyme is not localized uniquely to the apical tegumental membrane complex. Therefore, the purity of the isolated membranes has to be calculated from the ratio of enzyme activities in the stripped worm and the isolate. The specific activity of alkaline phosphatase in the tegument fraction was highly increased to $7{\cdot}8 \pm 0{\cdot}1$ U/mg protein, which was considerably higher than the specific activity reported when the filtration step was omitted (4·5 U/mg protein), but less than after the sucrose gradient described in the original manuscript (10·3 U/mg protein) (Roberts *et al.* 1983). The filtration step proved to be a simple but valuable addition to the existing procedure, as it increased the specific activity of alkaline phosphatase with very little loss of phospholipid. Immunoblotting was used for the control of membrane purity. The use of antibodies directed against proteins uniquely located in the apical tegumental membranes (SGTP4) and in the membranes inside the worm including the underlying basal lamina (SGTP1), allowed evaluation of cross-contamination between stripped worms and the isolated apical tegumental membranes. The apical tegumental membranes appeared to be not completely removed from the schistosomes, as judged from the small amount of SGTP4 present in the stripped worm membrane fraction. It should be realized, however, that SGTP4 is also present in the precursors of the apical membrane complex (the multilamellar and discoid bodies) which are formed within the tegumental cytons. This could mean that the outer membrane complex was removed

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quantitatively. No SGTP1 staining occurred in the fraction with the apical tegumental membranes, demonstrating that there was no contamination of this fraction with basal lamina.

Turnover of the apical tegumental membranes

Palmitate is the major fatty acid found in the phospholipids of the tegument (Allan *et al*. 1987). Since schistosomes are not capable of synthesizing fatty acids *de novo*, the turnover of exogenously supplied palmitate can give reliable information about the dynamics of (palmitate containing) phospholipids. In the stripped worms, the decrease in palmitoyl specific activity was much slower than in the apical tegumental phospholipids. Incorporated label remained for a longer period of time in the stripped worm phospholipid fraction before it was replaced with unlabelled palmitate. The phospholipids of the apical tegumental membranes in contrast, were metabolically very active, as can be derived from both the high rate of incorporation of radio-isotope labelled fatty acids during the pulse period, as well as from the rapid loss of the incorporated acyl chains during the subsequent chase period. At the end of the chase period nearly all radioactivity incorporated during the pulse period had disappeared from the apical tegumental membranes, indicating that nearly all palmitoyl chains in these membranes had been replaced during the chase period.

Membrane sloughing versus acyl turnover

In newly transformed schistosomula, it has been demonstrated that a variety of surface molecules are lost from the tegumental membranes at a high rate. This has been demonstrated for glycosylphosphatidylinositol-anchored antigens, fluorescent analogues of PE, concanavalin A, and radio-isotope labelled glycoproteins and glycolipids (Caulfield *et al*. 1991; Samuelson & Caulfield, 1982; Samuelson *et al*. 1982; Pearce & Sher, 1989). Concerning the turnover of the tegumental membrane complex in adult parasites, results have been less consistent. Where some experiments indicate a rapid secretion of proteins bound to the tegument (Kusel, Mackenzie & McLaren, 1975; Dean & Podesta, 1984), studies on the loss of erythrocyte antigens and antibodies against surface-bound molecules demonstrate a very slow turnover of tegumental membranes (Saunders *et al*. 1987; Ruppel & McLaren, 1986). Our studies show a high turnover of fatty acyl chains in the tegumental phospholipid fraction of adult schistosomes. The rapid loss of radio-isotope labelled fatty acids from tegumental phospholipids can be caused by either sloughing of membrane sheets or by hydrolysis of the labelled acyl chains. To discriminate between these 2 mechanisms, the medium in which the schistosomes were cultured during the chase period was analysed. Although there was a clear secretion of label into the medium, none of the radioactivity was associated with phospholipids. From this it can be concluded that under our culture conditions there was no significant sloughing of tegumental membranes, as this would have led to the presence of radio-isotope labelled phospholipids in the medium. To exclude the possibility that label was secreted in phospholipids as part of a sloughed membrane, and was subsequently converted to free fatty acid by hydrolysis of the phospholipids by (secreted) phospholipases, we incubated the schistosomes in the presence of vesicles containing the synthetic phospholipid (17:0–17:0) PC. No heptadecanoate (17:0 as free fatty acid) was formed in the medium during the chase (data not shown), indicating that degradation by phospholipases of released PC can be excluded and is not the origin of the observed labelled fatty acids in the incubation medium. Therefore, deacylation/reacylation and not membrane sloughing caused the observed high metabolic rate of palmitate in the tegumental membranes.

Metabolic activity of individual phospholipid classes

The metabolic activity of the 2 major phospholipids (PC and PE) of the tegumental membranes was investigated and compared. The turnover of palmitate in the PC fraction of the apical tegumental membranes was very high as can be concluded from the rapid incorporation of label during the pulse, and the subsequent replacement of the labelled fatty acyl chains during the chase. In the PE fraction, however, the incorporation of palmitate into the apical membranes was below threshold limits, indicating that the turnover rate of PE was some orders of magnitude lower than the turnover rate of PC. The high deacylation/reacylation of fatty acyl chains therefore proved to be limited to the PC fraction only. This further supports the idea that membrane sloughing, if occurring at all, is a very slow process under these conditions, as the rate of turnover of PE was found to be very low and sloughing of a single phospholipid class (PC in this case) from a membrane would be highly unlikely.

Possible physiological importance of deacylation/reacylation in the tegument

The high rate of deacylation of tegumental PC described here, could be indicative of a defence mechanism applied by the schistosome against immune cells. It is attractive to speculate that contact between a host cell and the tegumental membrane complex of the schistosome would expose the host

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cell membranes to lysophosphatidylcholine (LPC) and free fatty acid, both known to be membrane destabilizing components. Assuming that the host cell is not adjusted to a similar rate of reacylation as the schistosome, one could speculate that the destabilized membrane of the host cell could interfere with the normal function of this cell. It has indeed been reported that schistosomula use LPC to lyse host erythrocytes and are able to fuse with host cell membranes (Caulfield *et al*. 1980; Golan *et al*. 1986). Our studies demonstrate that also adult worms deacylate phospholipids, forming free fatty acids and hence also LPC which could *in vivo* be used for the lysis of erythrocytes or immune cells. The observed inadequacy of immune cells may, at least partially, find its origin in the lipid metabolism of the schistosome.

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