



Passive DNA adjuvant efficacy in a murine model of hepatitis B virus infection

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Passive DNA (PD)
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ABSTRACT

The DNA adjuvant for hepatitis B vaccine (DNA adjuvant) was evaluated in a murine model of hepatitis B virus (HBV) infection. The DNA adjuvant was composed of a plasmid encoding the HBV surface antigen (HBsAg) and a plasmid encoding the hepatitis B core antigen (HBcAg). The DNA adjuvant was administered to mice along with the HBsAg vaccine. The DNA adjuvant significantly enhanced the cellular immune response (CD4⁺ and CD8⁺ T cells) and the antibody response (anti-HBsAg) compared with the HBsAg vaccine alone. The DNA adjuvant also significantly reduced the viral load (HBV DNA) in the liver and the serum. The DNA adjuvant was highly effective in protecting mice from HBV infection. The DNA adjuvant was a promising adjuvant for HBV vaccine.

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1. Introduction

Hepatitis B virus (HBV) is a major cause of liver disease and cancer. The HBV vaccine is highly effective in preventing HBV infection. However, the HBV vaccine is composed of recombinant HBsAg and is not able to induce a strong cellular immune response. The DNA adjuvant is a promising adjuvant for HBV vaccine. The DNA adjuvant is composed of a plasmid encoding the HBsAg and a plasmid encoding the HBcAg. The DNA adjuvant is administered to mice along with the HBsAg vaccine. The DNA adjuvant significantly enhanced the cellular immune response (CD4⁺ and CD8⁺ T cells) and the antibody response (anti-HBsAg) compared with the HBsAg vaccine alone. The DNA adjuvant also significantly reduced the viral load (HBV DNA) in the liver and the serum. The DNA adjuvant was highly effective in protecting mice from HBV infection. The DNA adjuvant was a promising adjuvant for HBV vaccine.

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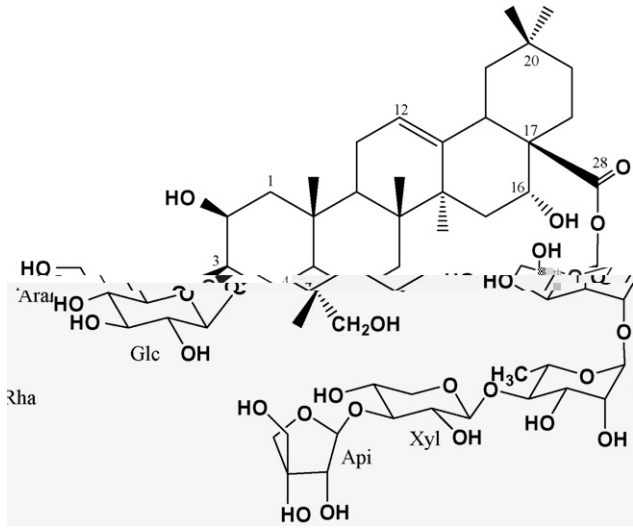


Fig. 1. Cefcazidime (PD, C₅₇H₉₂O₂₈, MW: 1224.5854) structure. The structure shows the core bicyclic system and the complex sugar side chain. The side chain is composed of several sugar units: Ara (arabinose), Glc (glucose), Rha (rhamnose), Xyl (xylose), and Api (apiose). The core structure includes a decalin-like ring system with a double bond at C12 and a methyl group at C20. The numbering of the carbon atoms in the core is 1, 3, 4, 12, 16, 17, and 20. The side chain is attached at C16 and C17. The structure is shown in a perspective view with wedged and dashed bonds indicating stereochemistry.

HB A [10]. add , A r e
cca a ca eac a e e f ec a da a

S ed c ec ed f e HB A ed ce de a e
 cc d , Ha ba ced a (HBSS, S a), a
 ced a r f c r d a ed r a e ee e
 ba a de ce d ad ee c e
 ee ed a c r de (0.8%, /). Af e ca r f
 a (380 x a 4°C fr 10), e e ed ce e e a ed
 r ee e PBS, a dr e d ded c ee ed . Ce
 be e ec ed a e c ee b r a b ed e
 e c ec le. Ce ab e ce ed 95%. S c e ee
 eed ed r ee fr e fa 96 e fla b c e
 a e (N c) a 5×10^6 ce / 100μ c ee ed , ee
 af e C A (f a c c r a 5μ /), LPS (f a c c r a
 10μ /), HB A (f a c c r a 4μ /), r ed ee
 added a f a e f 200μ . T e a e ee c ba ed a
 37°C a da ee $5\% \text{CO}_2$. Af e 44 (fr C A d
 LPS) r 68 (fr HB A), 50μ f MTT (2 /) a added
 eac e d c ba ed fr fr e 4 . T e a e ee c
 r f ed (1400×5) d e r r ed MTT a r e ed
 ca ef b e 150μ fa DMSO (S a, USA) r
 (192 μ DMSO 8 μ 1NHC) a added eac e, d e
 ab r ba ce a e a a ed ELISA eade a 570 af e 15 .
 T e a de (SI) a ca c a ed ba ed ef
 fr a: SI = e ab r ba ce a ef c r e d ded b
 e ab r ba ce a ef a ed c r e .

HB A ec fic I G, I G1, I G2a, d I G2b b de e a
 e e de ec ed d da e a e b a r ec ELISA.
 b ef, c e a e e ee ca ed 100μ HB A
 (2 μ / 50 M ca b a e b ca b a e b ffe, H 9.6) fr
 24°C a 4°C. T e e ee a ed r ee e PBS c a
 0.05% (/) T ed 20 (PBS/T ed), d b c ed 5%
 FCS/PBS a 37°C fr 2 . Af e r ee a , 100μ fa e e
 f d ed e a a e r 0.5% FCS/PBS a c r ee added
 r ca e e . T e a e ee d c ba ed fr 2 a 37°C ,
 f ed b r ee e f a . A l f 100μ f abb
 el G r e ad e da ec : a ed ed 1:20,000, a
 el G1 e da ec : a e 1:16,000, I G2a e da e
 c : a e 1:8000, d I G2b e da ec : a e 1:8000
 0.5% FCS/PBS ee added eac a e. T e a e ee fr e
 c ba ed fr 2 a 37°C . Af e a , e e da e ac
 a a a ed a f : 100μ f b r a e (10 f
 d a ed a e d 37.5μ f $30\% \text{H}_2\text{O}_2$ 25 f 0.1M
 cr a e a e b ffe, H 5.0) a added eac e. T e a e
 a c ba ed fr 10 a 37°C , d d e eac a e
 a ed b add 50μ / e f 2N H_2SO_4 . T e ca da
 a ea r ed ELISA eade a 490 , ee e f e a
 a e a e be b'c ed d be ed r c
 a , ELISA a a ee e fr ed e a ed a fr a f
 e a e .

T e NK ce ac f ed a de e ed a C
 96R N Rad ac e C c A a K (P e a). YAC 1
 ce ee ed a a e ce d eed 96 e Ub
 c e a e a 4×10^4 ce / e RPMI 1640. S c e
 r e a ed a de c bed ab e ee ed a e effec r ce d
 ee added a 2×10^6 ce / e e E/T r a 50:1. Eac e
 a r e ea ed fr e . T e a e a ca r f ed a $250 \times$ fr
 o

Table 1
 Selected primer sequences used for RT-PCR.

Gene	Primer sequence	Product (bp)	Accession
GAPDH	5' AAATGGTGAAGTCCGGTGTG 3' 5' TGAAGGGGTCGTTGATGG 3'	108	NM.001001303
IL 2	5' GCACCCACTTCAAGCTCCA 3' 5' AAATTTGAAGGTGAGCATCCTG 3'	174	NM.008366
IFN γ	5' CGGCACAGTCATTGAAAGCCTA 3' 5' GTTGCTGATGGCCTGATTGC 3'	199	NM.008337

GAPDH, housekeeping gene.

cells (ABC). After fixation for 30 min, cells were stained with DAB (TMB) at 37°C for 15 min. The reaction was stopped by adding 100 μ l of 0.1 M sodium acetate. The reaction was read by ELISA reader at 450 nm.

Selected cells were seeded in 24 well plates (Nunc) at 5×10^6 cells / 1 cm², and after 24 h (in culture at 4 μ l /) were added with 2 μ l (culture). The cells were cultured at 37°C in a humidified atmosphere with 5% CO₂.

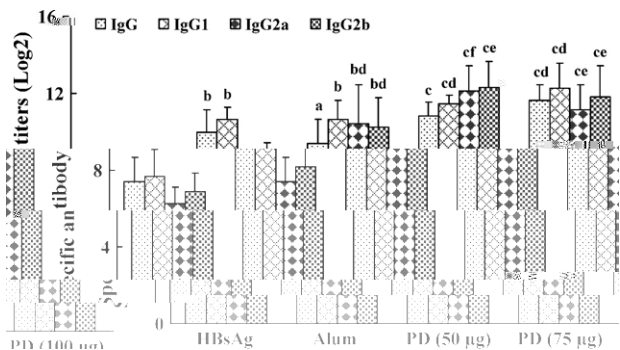


Fig. 3. Effect of adjuvant (D(PD) HB A) on the efficacy of I G, I G1, I G2a, and I G2b in eliciting HB A specific antibody response. Se a e e c e d 2 e e a f e e a b d a , d HB A e c f i c I G, I G1, I G2a, d I G2b b d e e a e e e a r e d b d e c ELISA a d e b e d e e . T e a e e e e e d a e a ± S.E. (= 5). S i c a d f f e c e HB A a e r e e d e a e d a < 0.05, b < 0.01, d c < 0.001; e HB A / A r a d < 0.05, e < 0.01, d f < 0.001.

I G1, I G2a, d I G2b b d e e e e e e e a r e d 2 e e a f e e a a a e ELISA d e r e e e F . 3. HB A a e d c e d e e f HB A e c f i c I G, I G1, I G2a, d I G2b b d . A d PD f i c a d c e d e e HB A e c f i c I G d I G1 e HB A e d c e (< 0.05, < 0.01, < 0.001). H e e , e c b a a f HB A PD e e c a a d e f 100 μ r d c e d e e a I G d I G1 e c a e d A d d HB A c b a . S i c a d a c e HB A e c f i c e I G2a d I G2b e e e b e e d PD e d c e c a e d HB A a e r (< 0.01 < 0.001). M r e e , I G2a d I G2b b d e e e c e e e d PD e e e e a e a r e a e d c e . T e e e e e e f i c a d f f e c e (> 0.05) e e I G2a d I G2b e e b e e c e r e d HB A / A d HB A a e . T e f d d c a e d a PD f i c a d a c e d e HB A e c f i c a b d r d c c e e d HB A . M r e e , a b d e d c e d b PD a d , a d e d HB A a f e e e e e e e e c e d b A a d , a d e d a c c e .

T e e f f e c f PD NK c e a c c e e d HB A e e F . 4. PD f i c a d c e d e e a c f NK c e e HB A e d c e a r e e

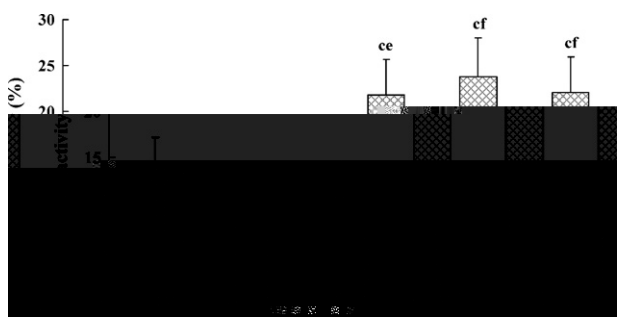


Fig. 4. Effect of adjuvant (D(PD) NK cell activity in response to HB A . S i c e e e e e a e d 2 e e a f e e a a , d a a e d f r NK c e a c b e LDH e e a e e d a d e c b e d e e . T e a e e e e e e d a e d a c < 0.001; e HB A / A r a e < 0.01 d f < 0.001.



Fig. 5. Effect of adjuvant (D(PD) CTL activity in response to HB A . S i c e e e e e a e d 2 e e a f e e a a , d a a e d f r CTL a c b e LDH e e a e e d a d e c b e d e e . T e a e e e e e e d a e d a ± S.E. (= 5). S i c a d f f e c e HB A a e d HB A / A r e e e d e a e d a c < 0.001 d f < 0.001 r e e c e .

d e (< 0.001). T e e e e , e e , f i c a d f f e c e (> 0.05) e a c f NK c e b e e d c e r e d HB A / A d HB A a e . T e f d d c a e d a PD c d r e a c a f NK c e c a c c e e d HB A .

f i

T e e f f e c f PD a d e c f i c CTL a c e HB A e d c e e e F . 5. I a f c e HB A a e d c e d e e e c f i c CTL a c 2 e e a f e e e c d a a . A d d f A HB A d d e e f r e c e a e HB A e c f i c CTL a c a b e e e d HB A a e . c l a , PD f i c a d c e d e e c f i c a c f CTL c e e d HB A a r e e d e (< 0.001). T e f d d c a e d a PD c d r e e c f i c a c f CTL c e e d HB A .

f i

r d e a e e f f e c f PD T 1 d T 2 c e r e d HB A , c e r d c f c e HB A e d c e e d e e c e d ELISA . T e c a b a c r e e f i l 2 , I F N γ , d I L 10 e e c r c e d e c e a d a d , d e c r r e a c e f f i c e e a b e a 0.9980. A F . 6. e c d f c e I L 2 , I F N γ , d I L 10 e c r e e a a f HB A a e d f i c a c e f e a c e e d HB A / PD e e f i c a e a e e HB A c r c e (< 0.01), e a PD f i c a d c e d e r d c f e T 1 d T 2 c e HB A e d c e . H e e , A f i c a c e a e d e I L 10 r d c e HB A e d c e (< 0.001).

f i

S i c e PD f i c a d c e d I G2a d I G2b b d r e e e d T 1 c e r d c , e e e a r e d I L 2 d I F N γ R N A e e e c e f c e e d HB A . T e c e f e e d c e e e a e d r HB A , d a R N A e e r a c e d . R e a e a e RT PCR f r I L 2 d I F N γ c e R N A e r e e e e f r e d . G A P D H a e d a a a c r f r e RT PCR e a c . A F . 7. e I L 2 d I F N γ R N A e r e e c e e d PD e e e

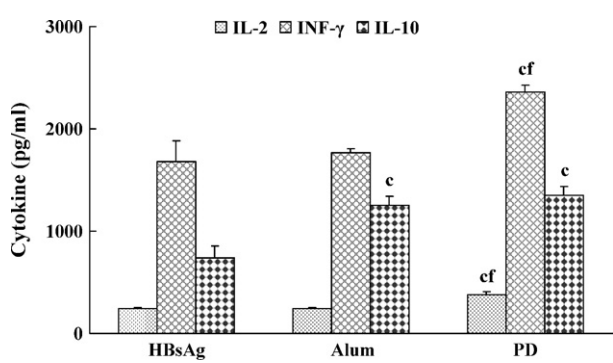


Fig. 6. Effect of adjuvant D(PD) on HBsAg-specific IL-2, INF-γ, and IL-10 production. Mice were immunized with HBsAg (4 μg) in the presence of alum or PD. Serum cytokine levels were measured 48 h after immunization by ELISA. The data represent the mean ± S.E. (n = 5). Significant differences between HBsAg/A and HBsAg/PD are indicated by letters: a, b, c, d, e, f, g, h, i, j, k, l, m, n, o, p, q, r, s, t, u, v, w, x, y, z, aa, ab, ac, ad, ae, af, ag, ah, ai, aj, ak, al, am, an, ao, ap, aq, ar, as, at, au, av, aw, ax, ay, az, ba, bb, bc, bd, be, bf, bg, bh, bi, bj, bk, bl, bm, bn, bo, bp, bq, br, bs, bt, bu, bv, bw, bx, by, bz, ca, cb, cc, cd, ce, cf, cg, ch, ci, cj, ck, cl, cm, cn, co, cp, cq, cr, cs, ct, cu, cv, cw, cx, cy, cz, da, db, dc, dd, de, df, dg, dh, di, dj, dk, dl, dm, dn, do, dp, dq, dr, ds, dt, du, dv, dw, dx, dy, dz, ea, eb, ec, ed, ee, ef, eg, eh, ei, ej, ek, el, em, en, eo, ep, eq, er, es, et, eu, ev, ew, ex, ey, ez, fa, fb, fc, fd, fe, ff, fg, fh, fi, fj, fk, fl, fm, fn, fo, fp, fq, fr, fs, ft, fu, fv, fw, fx, fy, fz, ga, gb, gc, gd, ge, gf, gh, gi, gj, gk, gl, gm, gn, go, gp, gq, gr, gs, gt, gu, gv, gw, gx, gy, gz, ha, hb, hc, hd, he, hf, hg, hh, hi, hj, hk, hl, hm, hn, ho, hp, hq, hr, hs, ht, hu, hv, hw, hx, hy, hz, ia, ib, ic, id, ie, if, ig, ih, ii, ij, ik, il, im, in, io, ip, iq, ir, is, it, iu, iv, iw, ix, iy, iz, ja, jb, jc, jd, je, jf, jg, jh, ji, jj, jk, jl, jm, jn, jo, jp, jq, jr, js, jt, ju, jv, jw, jx, jy, jz, ka, kb, kc, kd, ke, kf, kg, kh, ki, kj, kl, km, kn, ko, kp, kq, kr, ks, kt, ku, kv, kw, kx, ky, kz, la, lb, lc, ld, le, lf, lg, lh, li, lj, lk, ll, lm, ln, lo, lp, lq, lr, ls, lt, lu, lv, lw, lx, ly, lz, ma, mb, mc, md, me, mf, mg, mh, mi, mj, mk, ml, mm, mn, mo, mp, mq, mr, ms, mt, mu, mv, mw, mx, my, mz, na, nb, nc, nd, ne, nf, ng, nh, ni, nj, nk, nl, nm, nn, no, np, nq, nr, ns, nt, nu, nv, nw, nx, ny, nz, oa, ob, oc, od, oe, of, og, oh, oi, oj, ok, ol, om, on, oo, op, oq, or, os, ot, ou, ov, ow, ox, oy, oz, pa, pb, pc, pd, pe, pf, pg, ph, pi, pj, pk, pl, pm, pn, po, pp, pq, pr, ps, pt, pu, pv, pw, px, py, pz, qa, qb, qc, qd, qe, qf, qg, qh, qi, qj, qk, ql, qm, qn, qo, qp, qq, qr, qs, qt, qu, qv, qw, qx, qy, qz, ra, rb, rc, rd, re, rf, rg, rh, ri, rj, rk, rl, rm, rn, ro, rp, rq, rr, rs, rt, ru, rv, rw, rx, ry, rz, sa, sb, sc, sd, se, sf, sg, sh, si, sj, sk, sl, sm, sn, so, sp, sq, sr, ss, st, su, sv, sw, sx, sy, sz, ta, tb, tc, td, te, tf, tg, th, ti, tj, tk, tl, tm, tn, to, tp, tq, tr, ts, tt, tu, tv, tw, tx, ty, tz, ua, ub, uc, ud, ue, uf, ug, uh, ui, uj, uk, ul, um, un, uo, up, uq, ur, us, ut, uu, uv, uw, ux, uy, uz, va, vb, vc, vd, ve, vf, vg, vh, vi, vj, vk, vl, vm, vn, vo, vp, vq, vr, vs, vt, vu, vv, vw, vx, vy, vz, wa, wb, wc, wd, we, wf, wg, wh, wi, wj, wk, wl, wm, wn, wo, wp, wq, wr, ws, wt, wu, wv, ww, wx, wy, wz, xa, xb, xc, xd, xe, xf, xg, xh, xi, xj, xk, xl, xm, xn, xo, xp, xq, xr, xs, xt, xu, xv, xw, xx, xy, xz, ya, yb, yc, yd, ye, yf, yg, yh, yi, yj, yk, yl, ym, yn, yo, yp, yq, yr, ys, yt, yu, yv, yw, yx, yy, yz, za, zb, zc, zd, ze, zf, zg, zh, zi, zj, zk, zl, zm, zn, zo, zp, zq, zr, zs, zt, zu, zv, zw, zx, zy, zz.

5, IL 10, IL 13. For example, the effect of adjuvant on the production of cytokines is dependent on the type of antigen and the adjuvant used. In this study, we found that the use of PD as an adjuvant significantly increased the production of IL-2, INF-γ, and IL-10 in mice immunized with HBsAg. This is in contrast to alum, which did not significantly affect cytokine production. The results suggest that PD is a more effective adjuvant for enhancing the immune response to HBsAg. The mechanism of action of PD is not fully understood, but it is thought to be related to its ability to stimulate the innate immune system. PD is a complex of liposomes and a protein, which may act as a TLR ligand and activate signaling pathways that lead to the production of cytokines. The use of PD as an adjuvant may be a promising strategy for the development of more effective vaccines.

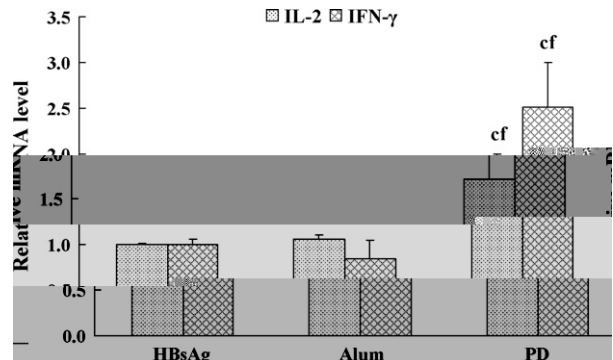


Fig. 7. Effect of adjuvant D(PD) on HBsAg-specific IL-2 and IFN-γ mRNA levels. Mice were immunized with HBsAg (4 μg) in the presence of alum or PD. The relative mRNA levels of IL-2 and IFN-γ were measured 24 h after immunization by RT-PCR. The data represent the mean ± S.E. (n = 5). Significant differences between HBsAg/A and HBsAg/PD are indicated by letters: a, b, c, d, e, f, g, h, i, j, k, l, m, n, o, p, q, r, s, t, u, v, w, x, y, z, aa, ab, ac, ad, ae, af, ag, ah, ai, aj, ak, al, am, an, ao, ap, aq, ar, as, at, au, av, aw, ax, ay, az, ba, bb, bc, bd, be, bf, bg, bh, bi, bj, bk, bl, bm, bn, bo, bp, bq, br, bs, bt, bu, bv, bw, bx, by, bz, ca, cb, cc, cd, ce, cf, cg, ch, ci, cj, ck, cl, cm, cn, co, cp, cq, cr, cs, ct, cu, cv, cw, cx, cy, cz, da, db, dc, dd, de, df, dg, dh, di, dj, dk, dl, dm, dn, do, dp, dq, dr, ds, dt, du, dv, dw, dx, dy, dz, ea, eb, ec, ed, ee, ef, eg, eh, ei, ej, ek, el, em, en, eo, ep, eq, er, es, et, eu, ev, ew, ex, ey, ez, fa, fb, fc, fd, fe, ff, fg, fh, fi, fj, fk, fl, fm, fn, fo, fp, fq, fr, fs, ft, fu, fv, fw, fx, fy, fz, ga, gb, gc, gd, ge, gf, gh, gi, gj, gk, gl, gm, gn, go, gp, gq, gr, gs, gt, gu, gv, gw, gx, gy, gz, ha, hb, hc, hd, he, hf, hg, hh, hi, hj, hk, hl, hm, hn, ho, hp, hq, hr, hs, ht, hu, hv, hw, hx, hy, hz, ia, ib, ic, id, ie, if, ig, ih, ii, ij, ik, il, im, in, io, ip, iq, ir, is, it, iu, iv, iw, ix, iy, iz, ja, jb, jc, jd, je, jf, jg, jh, ji, jj, jk, jl, jm, jn, jo, jp, jq, jr, js, jt, ju, jv, jw, jx, jy, jz, ka, kb, kc, kd, ke, kf, kg, kh, ki, kj, kl, km, kn, ko, kp, kq, kr, ks, kt, ku, kv, kw, kx, ky, kz, la, lb, lc, ld, le, lf, lg, lh, li, lj, lk, ll, lm, ln, lo, lp, lq, lr, ls, lt, lu, lv, lw, lx, ly, lz, ma, mb, mc, md, me, mf, mg, mh, mi, mj, mk, ml, mm, mn, mo, mp, mq, mr, ms, mt, mu, mv, mw, mx, my, mz, na, nb, nc, nd, ne, nf, ng, nh, ni, nj, nk, nl, nm, nn, no, np, nq, nr, ns, nt, nu, nv, nw, nx, ny, nz, oa, ob, oc, od, oe, of, og, oh, oi, oj, ok, ol, om, on, oo, op, oq, or, os, ot, ou, ov, ow, ox, oy, oz, pa, pb, pc, pd, pe, pf, pg, ph, pi, pj, pk, pl, pm, pn, po, pp, pq, pr, ps, pt, pu, pv, pw, px, py, pz, qa, qb, qc, qd, qe, qf, qg, qh, qi, qj, qk, ql, qm, qn, qo, qp, qq, qr, qs, qt, qu, qv, qw, qx, qy, qz, ra, rb, rc, rd, re, rf, rg, rh, ri, rj, rk, rl, rm, rn, ro, rp, rq, rr, rs, rt, ru, rv, rw, rx, ry, rz, sa, sb, sc, sd, se, sf, sg, sh, si, sj, sk, sl, sm, sn, so, sp, sq, sr, ss, st, su, sv, sw, sx, sy, sz, ta, tb, tc, td, te, tf, tg, th, ti, tj, tk, tl, tm, tn, to, tp, tq, tr, ts, tt, tu, tv, tw, tx, ty, tz, ua, ub, uc, ud, ue, uf, ug, uh, ui, uj, uk, ul, um, un, uo, up, uq, ur, us, ut, uu, uv, uw, ux, uy, uz, va, vb, vc, vd, ve, vf, vg, vh, vi, vj, vk, vl, vm, vn, vo, vp, vq, vr, vs, vt, vu, vv, vw, vx, vy, vz, wa, wb, wc, wd, we, wf, wg, wh, wi, wj, wk, wl, wm, wn, wo, wp, wq, wr, ws, wt, wu, wv, ww, wx, wy, wz, xa, xb, xc, xd, xe, xf, xg, xh, xi, xj, xk, xl, xm, xn, xo, xp, xq, xr, xs, xt, xu, xv, xw, xx, xy, xz, ya, yb, yc, yd, ye, yf, yg, yh, yi, yj, yk, yl, ym, yn, yo, yp, yq, yr, ys, yt, yu, yv, yw, yx, yy, yz, za, zb, zc, zd, ze, zf, zg, zh, zi, zj, zk, zl, zm, zn, zo, zp, zq, zr, zs, zt, zu, zv, zw, zx, zy, zz.

The effect of adjuvant on the production of cytokines is dependent on the type of antigen and the adjuvant used. In this study, we found that the use of PD as an adjuvant significantly increased the production of IL-2 and IFN-γ mRNA levels in mice immunized with HBsAg. This is in contrast to alum, which did not significantly affect mRNA levels. The results suggest that PD is a more effective adjuvant for enhancing the immune response to HBsAg. The mechanism of action of PD is not fully understood, but it is thought to be related to its ability to stimulate the innate immune system. PD is a complex of liposomes and a protein, which may act as a TLR ligand and activate signaling pathways that lead to the production of cytokines. The use of PD as an adjuvant may be a promising strategy for the development of more effective vaccines.

4. Discussion

The results of this study show that the use of PD as an adjuvant significantly increased the production of IL-2, INF-γ, and IL-10 in mice immunized with HBsAg. This is in contrast to alum, which did not significantly affect cytokine production. The results suggest that PD is a more effective adjuvant for enhancing the immune response to HBsAg. The mechanism of action of PD is not fully understood, but it is thought to be related to its ability to stimulate the innate immune system. PD is a complex of liposomes and a protein, which may act as a TLR ligand and activate signaling pathways that lead to the production of cytokines. The use of PD as an adjuvant may be a promising strategy for the development of more effective vaccines. In addition, the use of PD as an adjuvant significantly increased the relative mRNA levels of IL-2 and IFN-γ in mice immunized with HBsAg. This is in contrast to alum, which did not significantly affect mRNA levels. The results suggest that PD is a more effective adjuvant for enhancing the immune response to HBsAg. The mechanism of action of PD is not fully understood, but it is thought to be related to its ability to stimulate the innate immune system. PD is a complex of liposomes and a protein, which may act as a TLR ligand and activate signaling pathways that lead to the production of cytokines. The use of PD as an adjuvant may be a promising strategy for the development of more effective vaccines.

de ra ed a PD d a ed e a f e e e ,
 de cedaba cedT 1/T 2 e e e HB A ce
 a a ca ed e e ce d f I G2a, I G2b d
 I G1 e e [29].

de ce a e ab a T ce de ed c e
 e e ed e ad a ac f PD, e a a ed e
 T 1/T 2 c e e e r fi e HB A ed ce
 ELISA. PD e e a ed e r d c f
 T 2 c e IL 10, b a r d a ced e r d c
 f T 1 c e IL 2 d IFN γ c e e HB A
 ed ce. H IL 2 e e c r e a ed e d c
 f a d ec fic ce a r fe a e r e e, e e
 e e f IFN γ c e e e a e f I G2a d
 I G2b b d e . S a , e ce ed A d
 PD ad e e f IL 10 d c r e ded e e e
 f I G1 e e ce. T e e, e HB A ec fic b d
 e d c e r fi e c fi a e PD r ed a
 ba ced T 1/T 2 e e e e A a a
 ca ed r ed T 2 e e e e . r de
 fr e e c da e e ec r e be fr e c a e
 e a f T 1 c e, e ed r ea e RT PCR
 a e RNA e r e A e e 42 e T823 3.1(eR)

PD 782021Tf8.62120TD(0.641)T/F11Tf1.050TD0.000145715.5(c



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