

.I

*

BALB/c
 Echinococcus PAO Spm CSF PAO
 . granulosus
 Spm-PAO Spm Spm-PAO
 .Spm 200 PAO 1200
 PAO-Spm Spm
 CSF PAO
 :
 macrophages
 PAO
 (Morgan et al., 1981)
Plasmodium chaubaudi
 C75B1 Polyamine (PA)
 parasitemia super cations
Plasmodium yoelii (202- 88)
 pH
 .(Bachrach,1973; Wallace, 2003)
Babesia rodhaini (PAO) Polyamine Oxidases
Dirofilaria immitis Spd Spm
Schistosoma mansoni PA
 (PA-PAO) .(Abraham et al.,1979; Binda et al.1998)
 PAO Spd Spm
 PA .(Ferrante et al., 1986a) PAO
 (1,3) *

.2008/3/4

2007/5/17

(1-0.5) CSF 15 (Ferrante et al., 1990)
 (Ferrante et al., 1988) (Storer et al., 1988)
 al., 1986a)
 (10 5) (Ferrante et al., 1986b)
 () Echinococcosis
 . hydatid disease
 (Jumaa et al., 2001)
 CSF (Eckert and Deplazes, 2004) Cyclo-Zoonotic disease
 (Scharcterle and) Larval stage
 Pollack,1973; Lowrey et al.,1951)
 .CSF (Metacestode
 CSF PAO *Echinococcus*
 PAO
 .(Flayeh,1988; Dahel,1995) *Echinococcus granulosus*
 AL-) CSF PAO Cystic Echinococcosis
 CSF PAO Cystic hydatid disease
 Hames and) *Echinococcus multilocularis*
 (Clark and (Hooper,2000 Alveolar
 DEAE Switzer, 1976) Alveolar hydatid Echinococcosis
 PAO . - .(Vuitton, 2004) disease
 . Chemical Surgical
 Radiological
 BALB/c Immunological modulation Plant extracts
 . /
 /6-5 Immunostimulators
 4-3 Immunomodulators
 . Specific
 non-specific
 . /
 (Smyth, 1985)
 .(Smyth and Baret, 1980)
 Intraperitoneal Cavity
 .(Wangoo et al., 1989) 2000
 PAO
 10/ 200 25

عدد البلاعم المختزلة

$$100 \times \left(\frac{\text{عدد البلاعم المختزلة}}{\text{عدد البلاعم الكلي}} \right) =$$

(1600,1200,800,400,200)
PAO (10/

/ 2000
5 . 24

10/ 200
/ 5 (

(Dottorini et al., 1985)
(Ali-khan,1978) . 24 / 2000
5 .

.t 5 .(+ ve Control)
-ve) PBS
F . (Control

Randomly Complete Block Design (RCBD)

Dunnett

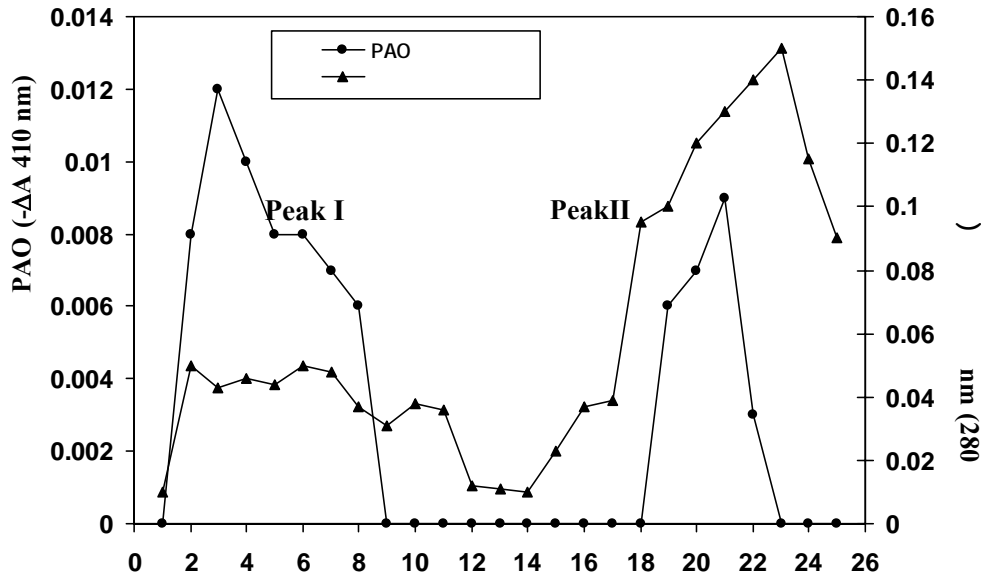
PAO 5
(Bruning and Kintz, 1977)
200
10/

CSF PAO
PAO (1)
8.18 ± 66.12
/ 147.53
/ PAO 5
200
PAO 10/
DEAE- Cellulose 72
(peak I) .(1) PAO 6
1201.92 (40-10)
II /
(110 -95) (peak II)
/ 1157.22
(Al-Katib, (II,I) 2000) Tetrazolium
CSF PAO Formazan granules
PAO : (Park et al.,1968)
()

(Cervelli et al., 2001)

(Murray et al., 2002)

PAO



CSF PAO : (1)

(1)
CSF PAO

%		/) ()	** ()	* /) ()			CSF
100	-	66.12	374.94	41.66	5.67	9.0	
165.26	2.23	147.53	619.65	72.90	4.20	8.5	
							DEAE-Cellulose
291.71	18.17	1201.92	1093.75	31.25	0.91	35	Peak I
222.22	17.50	1157.22	833.20	41.66	0.72	20	Peak II

*

()

U

**

Spm-PAO

PAO (%)	()	
	Spm	PAO
24		
5.41±*37.61	200	200
4.18±32.0	200	400
7.97±****61.92	200	800
3.26±****66.34	200	1200
6.16±***47.14	200	1600
3.80±*35.0	200	-
0.98±27.72	C ⁺	

PAO (1600-200) (200) Spm
 24 Spm
 Spm-PAO
 (P < 400 1200 %66.34
 %32.0 0.001)
 Spm .%27.72
 (P < 0.05)
 %35
 Phagocytic .% 27.72

C⁻ activity
 C⁺ Spm-PAO (3)

(P < 0.05) *
 (P < 0.01) **
 (P < 0.005) ***
 (P < 0.001) ****

± ()

)
 (Seiler) PAO
 .(et al.,1995

(Bachrach, 1981) Antiviral Antimicrobial
 (Levitz et al., 1990)
 .(Rzepczyk et al., 1984; Ferrante et al.,1984) Antiparasitic
 (Ali and Salih, 2001; Ali and Abdulla, 2004)

()
 (Garrett and Grisham, Spm .2005)

Spm
 Spm
 Innate immunity
 Spm

Lysozyme
 .(Powroznik et al., 2004)
 Spm

(Ali and Abdulla,2004)

(Jenkins et al.,1990)

Pseudomonas. aeruginosa

PAO

(3)

(Ryu and Kim, 2000)

PAO

(1600-200)

Pasteurella multocida (P-2383)

(200) Spm

18

24 Spm

3

1.574

(Czuprinski et al., 1984)

(P < 0.001)

1200

1600

0.484

3

0.570

Spm

Spm (3)

()

0.510

Spm-PAO

(3)

PAO

()

24

Spm

Spm

()			()	
48	24	3	Spm	PAO
0.16±****0.98	0.07±****1.02	0.08±****0.920	200	200
0.03±***0.448	0.05±***0.622	0.03±***0.806	200	400
0.02±0.314	0.04±0.530	0.15±0.842	200	800
0.04±****0.810	0.17±****1.080	0.35±****1.574	200	1200
0.02±0.308	0.03±0.416	0.11±0.484	200	1600
0.13±0.480	0.09±0.506	0.08±0.570	200	-
0.02±0.320	0.03±0.420	0.09±0.51	C ⁺	

(P < 0.05)

*

C⁺

(P < 0.005)

(P < 0.01)

**

(P < 0.001)

± ()

(Ali and Abdulla, 2004)

PAO

PAO

(Morgan et al., 1980)

PAO

(3) (2)

PAO

200

1200

PAO

Spm

()

(Allison et al.,

(Bachrach, 1981)

.1978)

PAO

PA

(4)

P.falciparum

PAO

H₂O₂

(Chaturvedi et al.,2004)

72

(Spm-PAO)

Apoptosis

PA

%63.12

.%42.6

(5)

Mesocestoides corti

72

(Spm-PAO)

(Jenkins et al.,1990)

(5)

%67

(P < 0.001)

(%)

.%55.4

()

72)

Spm

PAO

(4)

(%)

()

%	()	
	Spm	PAO
1.19±****67.0	200	1200
3.23±55.4	C ⁺	

Spm

PAO

72

(%)	()	
	Spm	PAO
0.90±****63.12	200	1200
1.67±42.60	C ⁺	

C⁻

C⁺

*

**

(P < 0.05)

(P < 0.01)

(P < 0.005)

(P < 0.001)

± ()

()

72) (7) PAO

6 (Spm- PAO) (6)

1.23 72 (Spm- PAO)

0.97 3 1.28

1.20 48

0.90 3 (P < 0.001)

1.15 0.97

3 1.70

48

1.0 0.90

(P < 0.001) (P < 0.001)

(6) ()

PAO 72 Spm

()			()	()	
48	24	3		Spm	PAO
0.158±****0.60	0.015±****0.93	0.031±****1.28	1	200	1200
0.254±****0.70	0.215±****1.20	0.158±****1.70	2		
0.041±0.48	0.158±0.80	0.012±0.97	1	C ⁺	
0.145±0.35	0.155±0.71	0.158±0.90	2		

(7) () (72)

Spm PAO

()			()	()	
48	24	3		Spm	PAO
0.223±****0.75	0.198±****1.06	0.236±****1.23	1	200	1200
0.127±****0.40	0.136±****0.75	0.215±****1.20	2		
0.055±****0.62	0.065±****0.86	0.206±****1.15	3		
0.041±0.48	0.158 ±0.80	0.012 ±0.97	1	C ⁺	
0.145±0.35	0.155 ±0.71	0.158± 0.90	2		
0.048±0.52	0.097±0.77	0.203 ±1.00	3		

(P < 0.001)

C⁺

± ()

- Spm-PAO
- PAO
- Spm-PAO
- (Morgan et al., 1980; Chaturvedi et al., .2004) (Ali and Abdulla, 2004; Ali and Yaseen, 2006)
- Depressed CMI
(Ali and Salih, 2000; Ali and Yaseen, 2006)
- . (Ryu and Kim, 2000)
- inflammation. *Inflamm. Res.*, 8(1-2):27-35.
- Bachrach, U. 1973. Functions of naturally occurring polyamines. Academic Press. INC. New York. London.2.
- Bachrach, U. 1981. Metabolism of polyamines. In: Polyamine in Biology and Medicine. (Morris D.R. and Marton L.J., eds).157-168, Marcel Dekker, New York.
- Binda, C., Coda, A. Angelini, R., Federico, R., Ascenzi, P. and Mattevi, A. 1998. Crystallization and Preliminary X-ray analysis of polyamine oxidase from *Zea mays*. *Acta crystallogr.D.Biol.Crystallogr*, 54(2):1429-1431.
- Bruning, J.L. and Kintz, B.L. 1977. *Computational handbook of statistics*. 2nd ed., Scott Foresman Company, Gleriew .
- Cervelli, M., Cona, A., Angelini, R., Pdticelli, F., Federico, R. and Mariottini, P. 2001. A barley polyamine oxidase isoform with distinct structural features and subcellular localization. *Eur. J. Biochem.*, 268(13):3816-3830.
- Chaturvedi, R., Cheng, Y., Asim, M., Bussiere, F.I., Xu, H., Gobert ,A.P., Hacker, A., Casero, Jr. R.A., Wilson, K.T. 2004. Induction of polyamine oxidase 1 by *Helicobacter pylori* causes macrophage apoptosis by hydrogen peroxide release and mitochondrial membrane depolarization. *J. Biol. Chem.* 279(38):40161-40173.
- Clark, J.M. and Switzer, R.L. 1976. Experimental Biochemistry.2nd ed., W.H. Freeman and Company San Francisco.
- Czuprinski, C.J., Henson, P.E. and Campbell, P.A. 1984. Killing of *Listeria monocystogenes* by inflammatory neutrophils and mononuclear phagocystes from immune and non-immune mice. *J. Leukocyte Biol.* 35:193-208.
- Dahel, K.A.D. 1995. *A study of polyamine oxidase enzyme*
- Abraham, A.K., Olsnes, S. and Pihl, A. 1979. Fidelity of protein synthesis *in vitro* is increased in the presence of spermidine. *FEBS. Lett.* 101:93.
- Ali, A.A. and Abdulla, I.T. 2004. Specific and non-specific cellular immune response in BALB/c mice against infection with hydatid disease by the polysaccharide extracted from *Pseudomonas aeruginosa*. III. Delayed type hypersensitivity and Phagocytosis. *Riv. Parassitol.*, XXI (LXV)-1:17- 24.
- Ali, A.A. and Salih, N.E. 2001. Immunomodulation in mice against infection with unilocular hydatid disease by the polysaccharide pullulan. III. Cellular and humoral immune response. *Riv. Parassitol.*, XVIII (LXII)-2:161-170.
- Ali, A.A. and Salih, N.E. 2000. Pullulan and delayed-type hypersensitivity in mice infected with unilocular hydatid disease. *Riv. Parssitol.*, XVII (LXI)-2:175-182.
- Ali, A.A. and Yaseen, S.S. 2007. Effect of lipopolysaccharide extracted from *Escherichia coli* in immune response of BALB/c mice against infection with secondary hydatid disease. III. *Phagocytic index and delayed-type hypersensitivity*. *J.Edu.Sci.* 20(1): 145-154.
- Ali-khan, Z. 1978. *Echinococcus multilocularis*:cell mediated immune response in early and chronic alveolar murine hydatidosis. *Exp. Parasitol*, 46:157-165.
- Al-Katib, S.M.Y. 2000. *Occurrence and properties of polyamine oxidase isoenzymes in the milk and cerebrospinal fluid*. Ph.D. Thesis, Coll. Edu. Univ. Mosul. (in Arabic).
- Allison, A.C., Ferluga, J., Prydz, H. and Schorlemmer, H.U. 1978. The role of macrophage activation in chronic

- Antifungal activity of the PAO-polyamine oxidase. *Antonie van Leeuwenhoek.*, 58(2):107-114.
- Lowrey, O.H., Rosebrough, N.J., Farr, A.L. and Randall, R.J. 1951. Protein measurements with the folin phenol reagent. *J. Biol. Chem.*, 193: 265-275.
- Morgan, D.M.L., Christensen, J.R. and Allison, A.C. 1981. Polyamine oxidase and the killing of intracellular parasites. *Biochem. Soc. Trans.*, 9:563-564.
- Morgan, D.M.L., Ferluga, J. and Allison, A.C. 1980. Polyamine oxidase and macrophage function. In: Polyamines in Biomedical Research, (Gaugas, J.M., ed.), 303-308, Wiley, Chichester and New York.
- Murray-Stewart T., Wang, Y., Devereux, W. and Casero, Jr. R.A. 2002. Cloning and characterization of multiple human PAO splice variants that code for isoenzyme with different biochemical characteristics. *Biochem. J.*, 368: 673-677.
- Park, P.H., Filkring, S.M. and Smith Wick, E.M. 1968. *Lancet* 2:532-534.
- Powroznik, B., Gharbi, M., Dandrifosse, G. and Peulen, O. 2004. Enhancement of lysozyme stability and activity by polyamines. *Biochimie*, 86:651-656..
- Ryu, H. and Kim, C. 2000. Immunologic reactivity of a lipopolysaccharide-protein complex of type A *Pasteurella multocida* in mice. *J. Vet. Sci.* 1(2):87-95.
- Rzepczyk, C.M., Saul, A.J. and Ferrante, A. 1984. Polyamine oxidase-mediated intraerythrocytic killing of *Plasmodium falciparum*. *Infect. Immun.*, 43(1):238-244.
- Scharcterle, G.R. and Pollack, R.L. 1973. A simplified method for the quantitative assay of small amounts of protein in biological materials. *Anal. Biochem.*, 51:654-655.
- Seiler, N., Moulinoux, J.P., Havouis, R. and Toujas, L. 1995. Characterization of amine oxidase activities in macrophages from human peripheral blood, *Biochem. Cell Biol.* 73(5-6):275-281.
- Smyth, J.D. and Barrett, N.G. 1980. Procedures for testing the viability of human hydatid cysts following surgical removal especially after chemotherapy. *Trans.Roy. Soc. Trop.Med. Hyg.* 74:649-652.
- Smyth, J.D. 1985. *In vitro* culture of *Echinococcus spp.* Proc. 13th ed., Int. Cong. Hydit, Madrid, 84-95.
- Storer, R.J., Ferrante, A., Bates, D.J., Zola, H. and Morgan, D.M.L. 1988. Retroplacental polyamine oxidase: antimicrobial, antiinflammatory, antitumor and in the blood of normal, depressed or schizophrenic persons. M.Sc. Thesis, Univ.
- Dottorini, S., Sparovli, M., Bellucci, C. and Magnini, M. 1985. *Echinococcus granulosus*: Diagnosis of hydatid disease in man. *Ann. Trop. Med. Parasitol.*, 74:43-49.
- Eckert, J. and Deplazes, P.X. 2004. Biological, epidemiological, and clinical aspects of echinococcosis, a zoonosis of increasing concern. *Clin. Microbiol.Rev.*, 17(1):107-135.
- Ferrante, A., Ljungstrom, I., Rzepczyk, C.M. and Morgan, D.M.L. 1986a. Differences in sensitivity of *Schistosoma mansoni* Schistosomula, *Diriofilaria immitis* microfilariae and *Nematospiroides dubius* third-stage larvae to damage by the polyamine oxidase-polyamine system. *Infect. Immun.* 53(3):606-610.
- Ferrante, A., Maxwell, G.M., Rencis, V.O., Allison, A.C. and Morgan, D.M.L. 1986b. Inhibition of the respiratory burst of human neutrophils by the polyamine oxidase-polyamine system. *Int. J. Immunopharmacol.*, 8:411.
- Ferrante, A., Rzepczyk, C.M. and Saul, A.J. 1984. Polyamine oxidase- mediated trypanosome killing: the role of hydrogen peroxide and aldehydes. *J. Immunol.* 133(4):2157-2162.
- Ferrante, A., Storer, R.J. and Cleland, L.J. 1990. Polyamine oxidase activity in rheumatoid arthritis synovial fluid. *Clin. exp. Immunol.*, 80:373-375.
- Flayeh, K.A. 1988. Spermidine oxidase activity in the serum of normal and schizophrenic subjects. *Clin. Chem.*, 34: 401-403.
- Garrett, R.H. and Grisham, C.M. 2005. Mechanisms of enzyme action. In: Biochemistry, 3rd ed., Thomson Learning Academic Resource Center, Brooks/Cole, a division of Thomson Learning, Inc., 453.
- Hames, B.D. and Hooper, N.M. 2000. Instant notes on biochemistry. 2nd ed. Bios. Scientific Publishers Limited.
- Jenkins, P., Dixon, J.B., Rakha, N.K. and Carter, S.D. 1990. Regulation of macrophage-mediated larvicidal activity in *Echinococcus granulosus* and *Mesocostoides corti* (cestoda) infection in mice. *Parasitology*, 100:309-315.
- Jumaa, H.I., Al-Kennany, E.R. and Al-Hayali, F.Q. 2001. Histological and histochemical study of hydatid cyst wall of *Echinococcus granulosus* from sheep and cattle naturally infected.Iraqi. *J. Vet.Sci.*,14(1):39-56..
- Levitz, S.M., Di Benedetto, D.J. and Diamond, R.D. 1990. Inhibition and killing of fungi by the polyamine system.

- perspective of polyamine metabolism. *Biochem. J.*, 376:1-14.
- Wangoo, A., Ganguly, N.K. and Mahajan, R.C. 1989. Phagocytic activity of monocytes in murine model of *Echinococcus granulosus* of human origin. *Indian. J. Med.Res.*89:40-42.
- immunosuppressive properties further characterization. Basic and Clinical Aspects (ed. By Y. Tomoda, S. Mizutani, O. Narita and A. Klopper). 349 NSP Science Press. Utrecht.
- Vuitton, D.A. 2004. Echinococcosis and allergy. *Clin. Rev. Aller. Immunol.*, 26(2):93-104.
- Wallace, H.M., Fraser, A.V. and Hughes, A. 2003. A

Immune Effects of Partially Purified Polyamine Oxidase(PAO) from Cerebrospinal Fluid(CSF) Against Infection with Secondary Hydatid Disease in Mice.I. Phagocytosis and Delayed-Type Hypersensitivity

*Khawla A. Al-Flayeh, Asmaa Abdulaziz. and Wathba I. Ali **

ABSTRACT

The study investigated the immune system response to infection with secondary hydatid disease in BALB/c mice activated by PAO with spermine then infected with protoscoleces of *Echinococcus granulosus*. Pathological changes occurred in mice activated by PAO-Spm, and by Spm alone, were followed in comparison with the positive control group throughout one month, depending on certain criteria which included changes in non-specific and specific immune response represented by phagocytosis and delayed-type hypersensitivity, respectively. The results revealed that PAO-Spm system is better than Spm alone, and the optimum concentration of PAO-Spm is 1200 µg PAO with 200µg Spm.

Pathological changes were followed in activated mice with the optimum concentration of PAO in comparison with +ve control group throughout two and three months. The results revealed an increase in the rate of phagocytic index and foot pad thickness, respectively, in activated mice with PAO-Spm. Whereas an increase in the phagocytic index, but no change in foot pad thickness occurred in mice activated with Spm alone.

Therefore, it was concluded that PAO isolated from CSF with Spm could be considered as an effective immunomodulator against infection with secondary hydatid disease.

Keywords: Polyamine Oxidase, Hydatid Disease, Immunity.

* Department of Chemistry, Faculty of Education, Mosul University, Iraq (1&3); and Department of Biology, Faculty of Education, Mosul University, Iraq. Received on 17/5/2007 and Accepted for Publication on 4/3/2008.