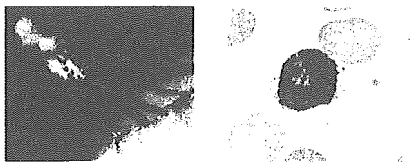
Research on Malaria Transmission in Thailand



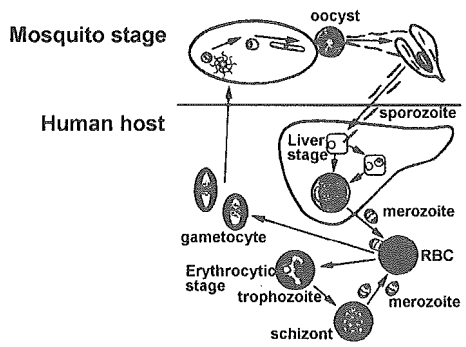
Jetsumon (Prachumsri) Sattabongkot

Entomology Department, AFRIMS
Biology Department, Faculty of Sciences, Burapha University
Pathobiology Department, Faculty of Sciences, Mahidol University

Objective

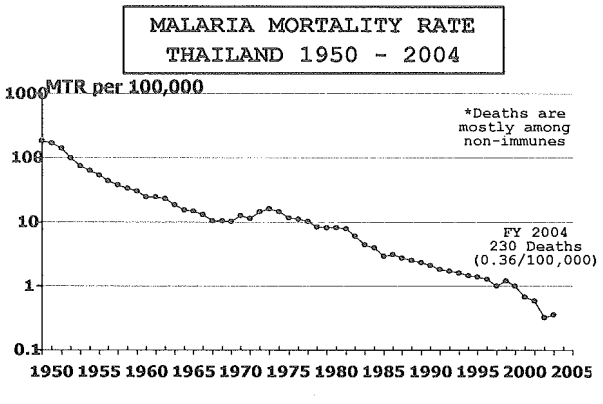
- Study the important factors to malaria transmission

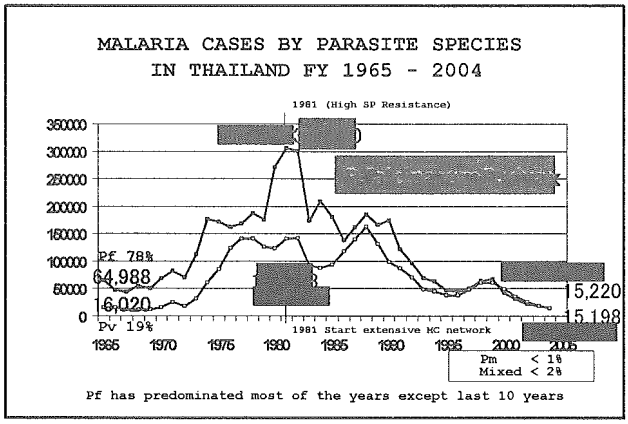
Malaria Life cycle

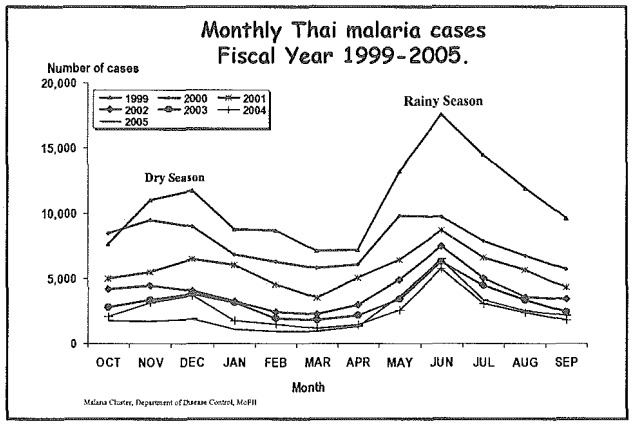


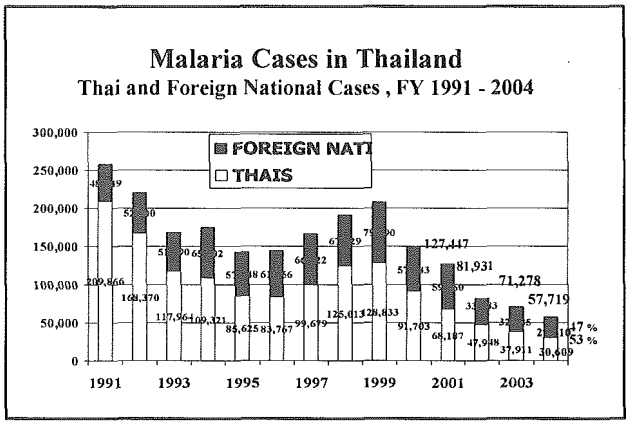
Malaria Transmission

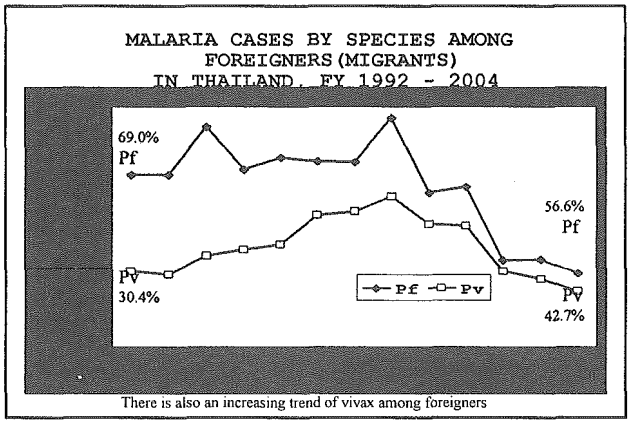
- Human
  - Immune response
- Vector species
  - Biology
  - Vectorial Capacity/Competence
- Parasites
  - Biology
  - Drug sensitivity

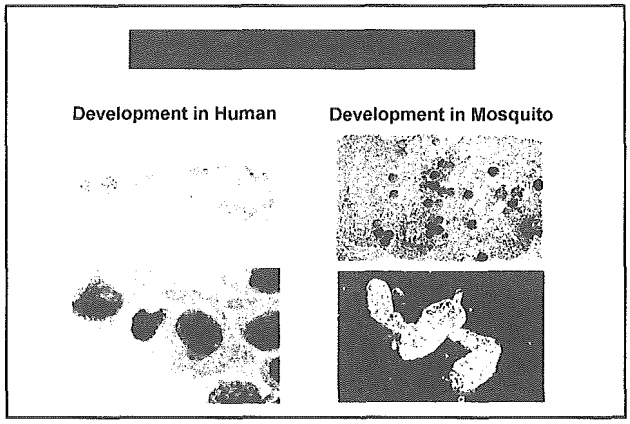


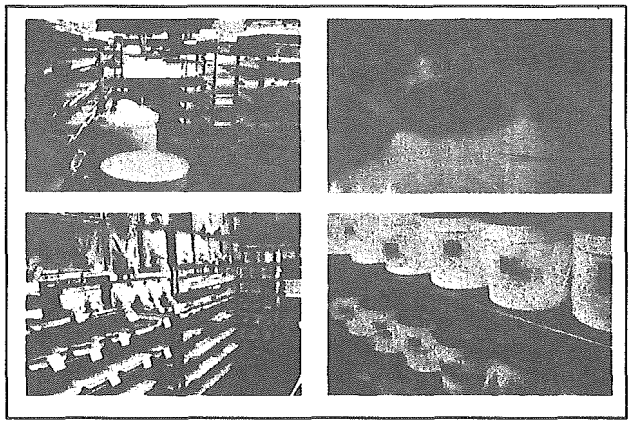


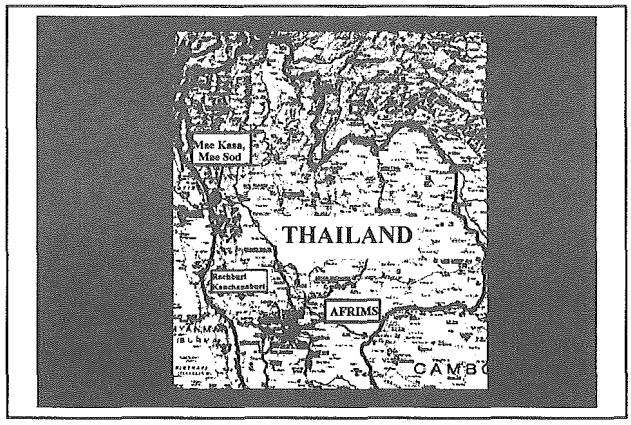

Monthly Thai malaria cases
Fiscal Year 1999-2005.
Number of cases
20,000
15,000
10,000
5,000
0
1999
2000
2001
2002
2003
2004
2005
Rainy Season
Dry Season
OCT NOV DEC JAN FEB MAR APR MAY JUN JUL AUG SEP
Month
Malaria Cluster, Department of Disease Control, MoPH


Malaria Cases in Thailand
Thai and Foreign National Cases , FY 1991 - 2004
300,000
250,000
200,000
150,000
100,000
50,000
0
FOREIGN NATI
THAIS
209,866
127,447
81,931
71,278
57,719
1991 1993 1995 1997 1999 2001 2003


MALARIA CASES BY SPECIES AMONG
FOREIGNERS (MIGRANTS)
IN THAILAND, FY 1992 - 2004
69.0%
Pf
56.6%
Pf
Pv
30.4%
Pf Pv
Pv
42.7%
There is also an increasing trend of vivax among foreigners


Development in Human
Development in Mosquito




Mae Kasa,
Mae Sod
THAILAND
Ratchburi
Kanchanaburi
AFRIMS

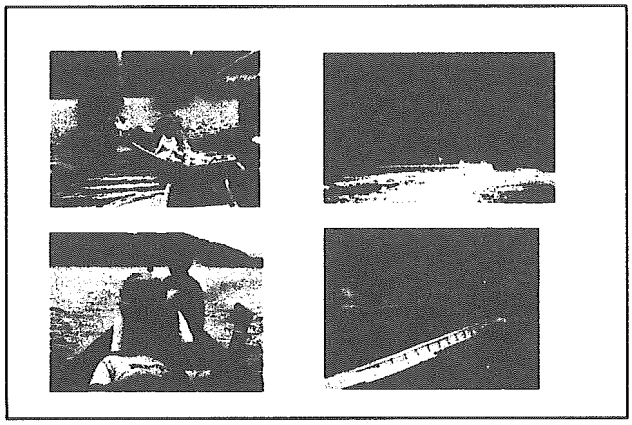
Naturally acquired malaria transmission blocking immunity in Thailand

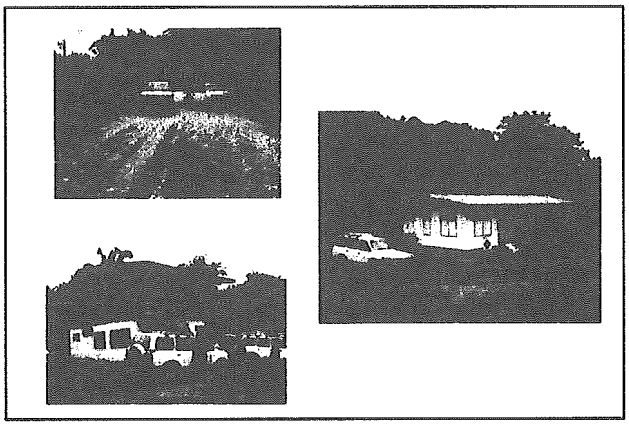
Objectives

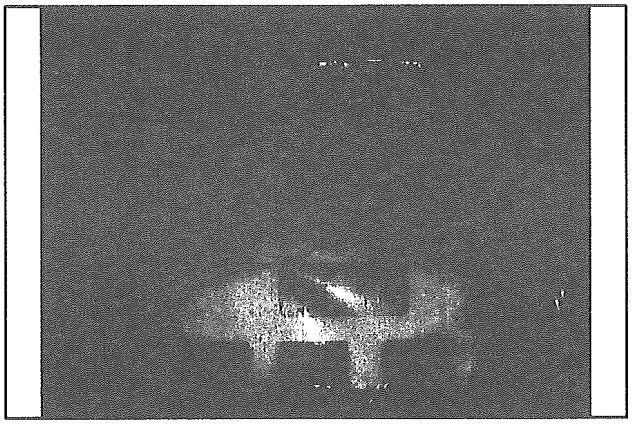
- Study naturally acquired immunity of population in malaria endemic village in western Thailand
- Identify parasite antigens leading to protective immunity

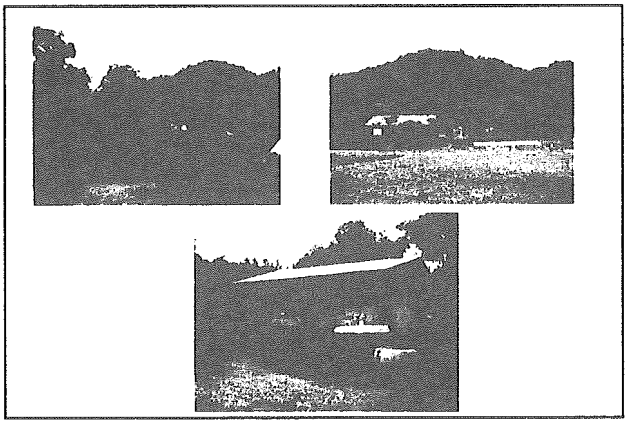
Background

- Population in malaria endemic areas
- Study sites:
  - Kong Mong Tha village, Kanchanaburi province
  - Malaria Clinics in Maesod, Tak province
- Volunteers:
  - Symptomatic population (patients)
  - Asymptomatic population: total of 717 persons

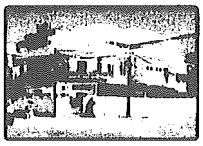








MALARIA SITUATION & MALARIA PROGRAMME IN CAMBODIA



Tokyo 31 Jan. – 01 Feb., 2006

Muth Sinuon

National Center for Parasitology, Entomology & Malaria Control

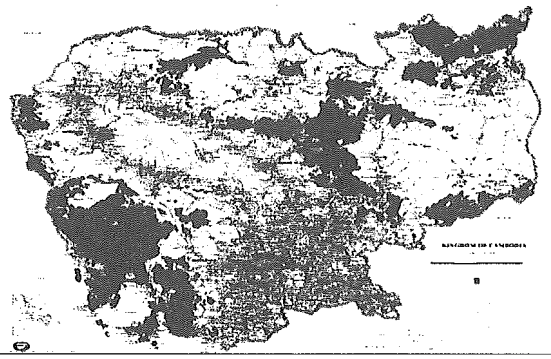
OUTLINE

- ❑Magnitude of malaria
- ❑Risk factors persist
- ❑Trends in disease incidence and mortality
- ❑Factors contributed to Malaria achievement
- ❑Factors contributing to Malaria deaths
- ❑National malaria control programme

Magnitude of Malaria in Cambodia

- 3rd most common cause of outpatient attendance (2.37% of patients in 2003)
- Fifth main health problem among inpatients (4.17% of in-patients in 2003)
- Second commonest cause of hospital mortality (3.89% of deaths in 2003)
- Mortality rate in 2003: 3.7 per 100,000 population

Total land area: 181,035 Km^2
More than 60% of Cambodia covered with forest, hills & mountains



Where about 1.6 millions (13% of total population 13.1 million) lives in scattered communities are at risk of malaria

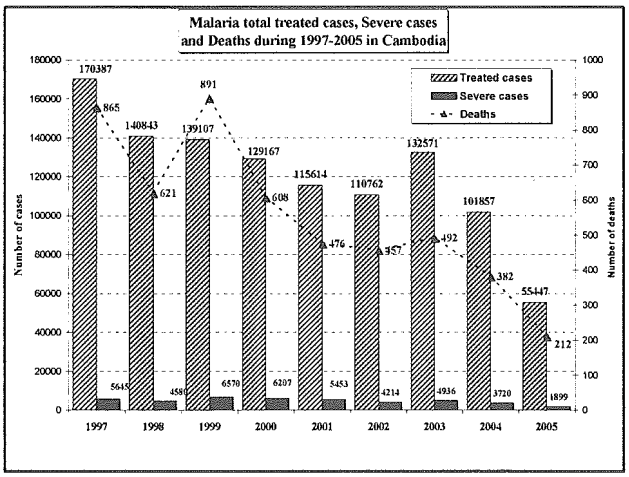
Vectors

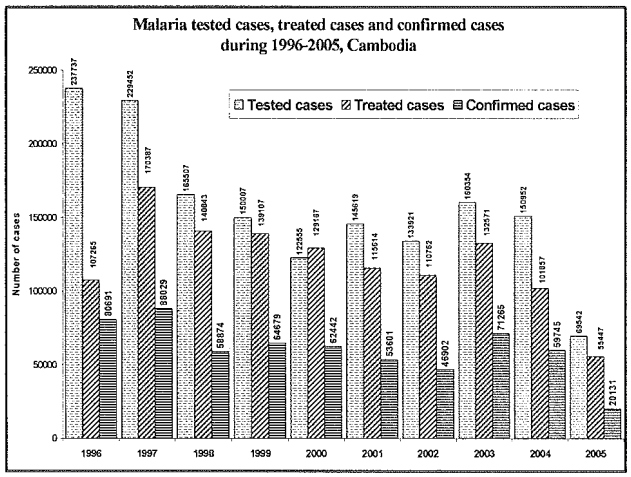
- Anopheles dirus (primary vector in forested area)
- Anopheles minimus (primary vector in forested area)
- Anopheles sundaicus (primary vector in coastal area)

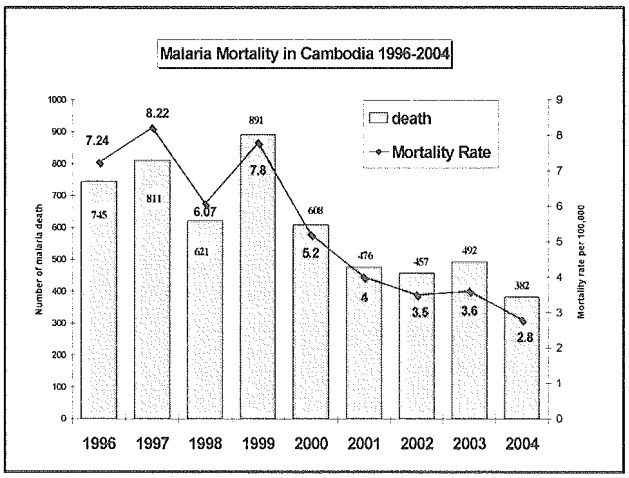
Risk factors persist

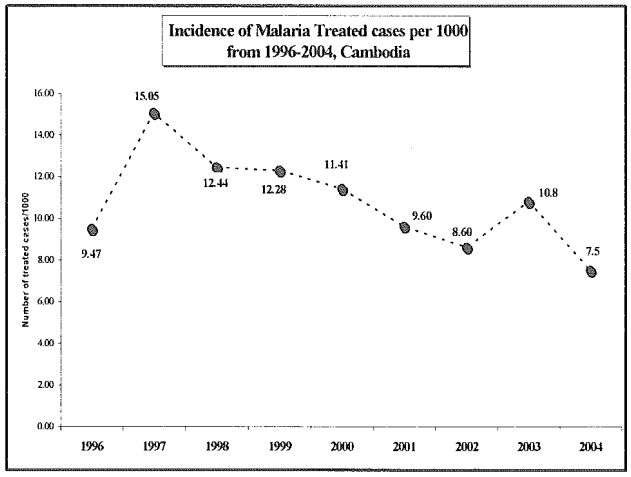
- Transmission area: 60% of land mass are thinly populated, forested and hilly areas
- Limited accessibility to public health services
- 337 of the 1,569 communes (13% of the population) are at medium to high risk to malaria infection (1.6 million)
- Traditional forest inhabitants, ethnic minorities
- Increasing migration of returnees and new forest settlers in the ex-Khmer Rouge areas (livelihood purpose)
- Trans-seasonal forest workers

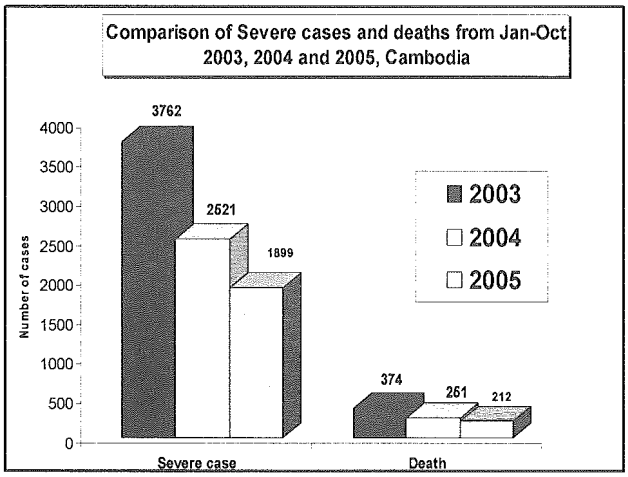
Trends in disease incidence and mortality

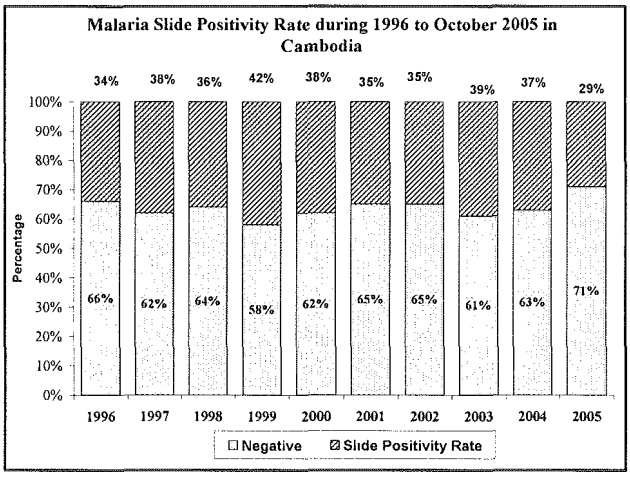


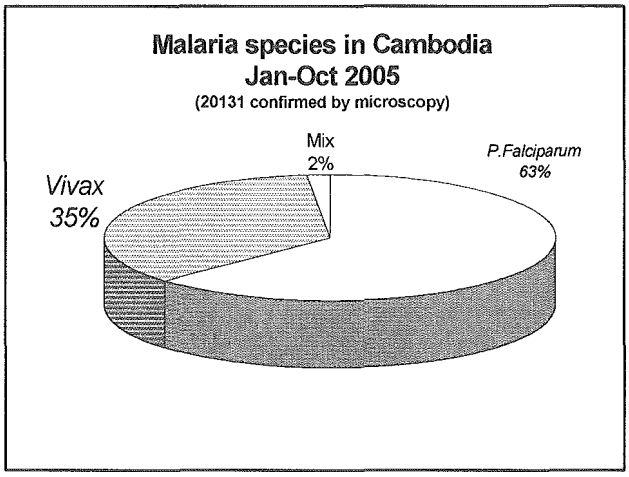


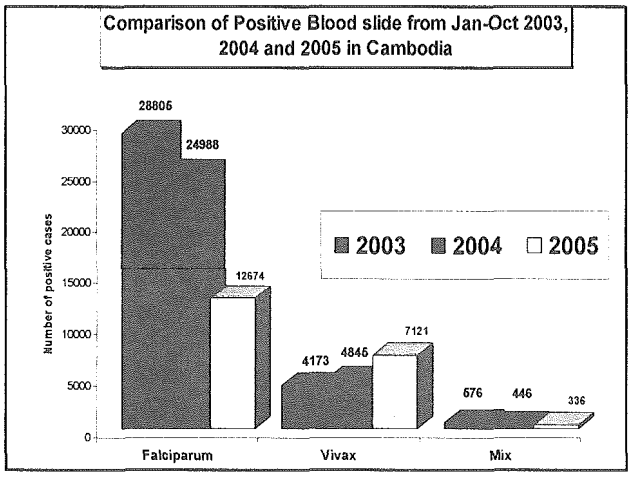


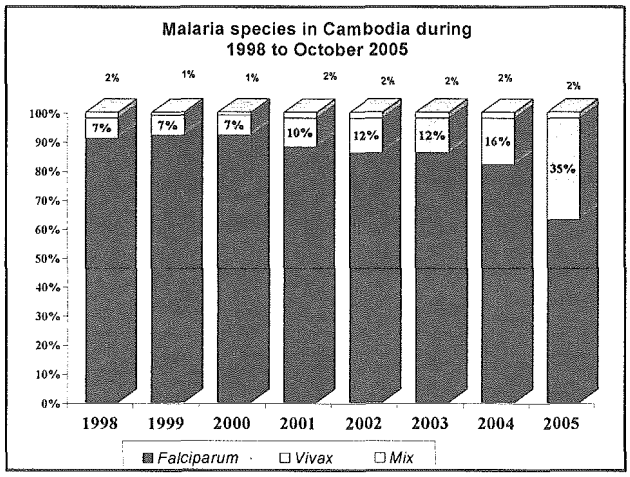


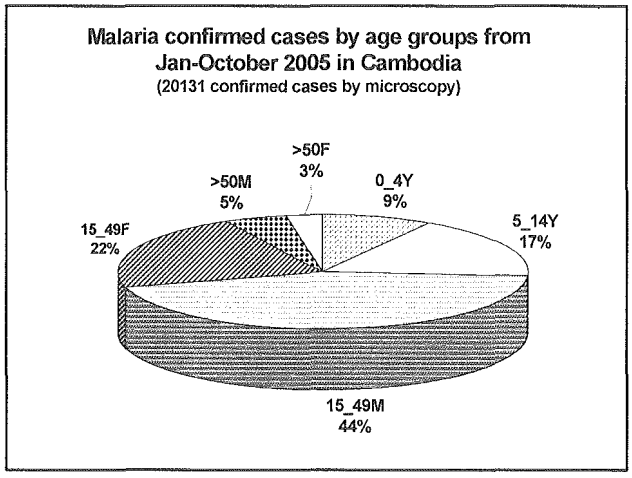


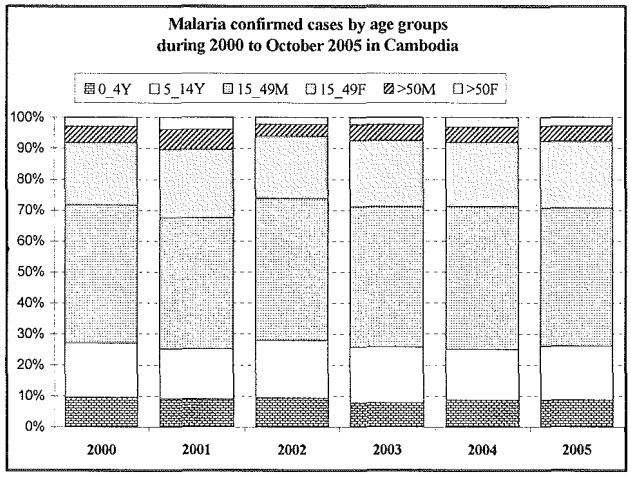

Malaria Slide Positivity Rate during 1996 to October 2005 in Cambodia
Percentage
34% 38% 36% 42% 38% 35% 35% 39% 37% 29%
66% 62% 64% 58% 62% 65% 65% 61% 63% 71%
1996 1997 1998 1999 2000 2001 2002 2003 2004 2005
Negative
Slide Positivity Rate


Malaria species in Cambodia
Jan-Oct 2005
(20131 confirmed by microscopy)
Mix
2%
P.Falciparum
63%
Vivax
35%

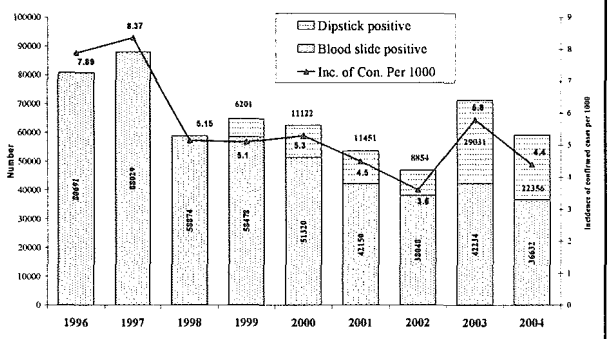

Comparison of Positive Blood slide from Jan-Oct 2003, 2004 and 2005 in Cambodia
Number of positive cases
28805
24988
12674
4173
4846
7121
676
446
336
2003
2004
2005
Falciparum
Vivax
Mix


Malaria species in Cambodia during 1998 to October 2005
2% 1% 1% 2% 2% 2% 2% 2%
7% 7% 7% 10% 12% 12% 16% 35%
1998 1999 2000 2001 2002 2003 2004 2005
Falciparum
Vivax
Mix


Malaria confirmed cases by age groups from Jan-October 2005 in Cambodia
(20131 confirmed cases by microscopy)
>50F
3%
>50M
5%
0_4Y
9%
15_49F
22%
5_14Y
17%
15_49M
44%


Malaria confirmed cases by age groups during 2000 to October 2005 in Cambodia
0_4Y
5_14Y
15_49M
15_49F
>50M
>50F
2000 2001 2002 2003 2004 2005

Number of Malaria Cases confirmed by blood slides and dipstick and Incidence of confirmed cases per 1,000 in Cambodia 1996-2004



Source: Epidemiology Department

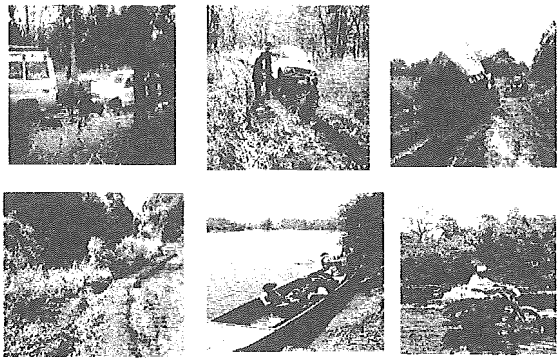
Factors contributing to improved malaria situation

- Efforts made by CNM to expand the programme activities to the maximum possible number of health centers.
- Enhanced malaria supervision and follow-up at provincial and health center level.
- Malaria training on malaria case management, dipstick use and laboratory diagnosis skills.
- Increasing conformity with the National Treatment Guidelines
- Wider usage of dipstick methodology has enhanced the scope for early diagnosis and prompt treatment.
- Active health education efforts coupled with the expansion in the distribution of IBNs to high and medium transmission zones through integrated approaches and outreach activities.

Factors contributing to Malaria deaths

- ❑ Predominance of the falciparum variety of malaria
- ❑ Poor health infrastructure especially in the high transmission areas
- ❑ Poor communication systems- poor road conditions, lack of suitable vehicles, lack of telephone facilities, etc.resulting in treatment delays
- ❑ Occupational hazard among the military personnel, forest workers, miners, etc.
- ❑ Remoteness & inaccessibility of affected areas
- ❑ Continuing shortages of diagnostic kits and appropriate drugs
- ❑ Large number of unqualified private practitioners and pharmacies who still do not follow national treatment guidelines
- ❑ Abundance of fake drugs available in the market
- ❑ Drug resistance in different pockets of the country

Difficult road communications



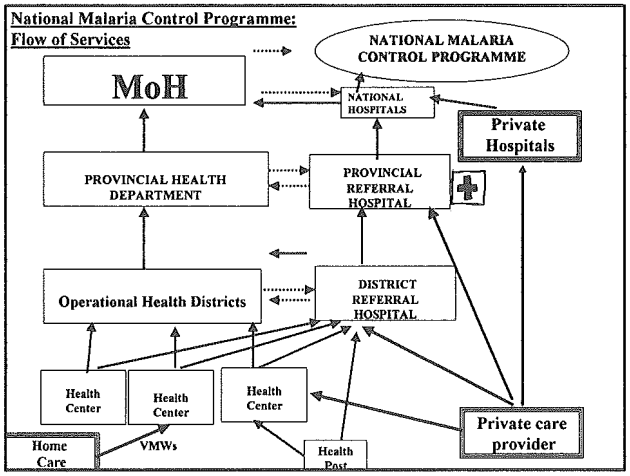
Malaria Control Programme in Cambodia

Goal

To reduce malaria related mortality by 50% and morbidity by 30% among the general population in the Kingdom of Cambodia within five years through the implementation of a comprehensive national malaria control strategy.

Strategic Objectives

1. To significantly increase community awareness and care-taking practices on malaria prevention and control with promotion of proper health seeking behavior in malaria endemic areas in Cambodia.
2. To improve access to preventive measures that protect the population with a focus on complete coverage for bed net distribution and re- impregnation in targeted malaria endemic areas, employing an effective community based approach.
3. To increase access to early diagnosis & treatment (EDAT) for Malaria for all the people in the country through the adoption of a three-pronged strategy
4. To strengthen the institutional capacity of the national malaria control program at central, provincial, operational district and commune levels

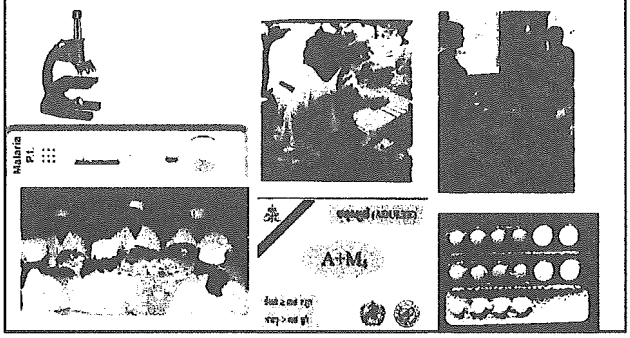


EARLY DIAGNOSIS AND PROMPT TREATMENT AT PUBLIC HEALTH FACILITIES

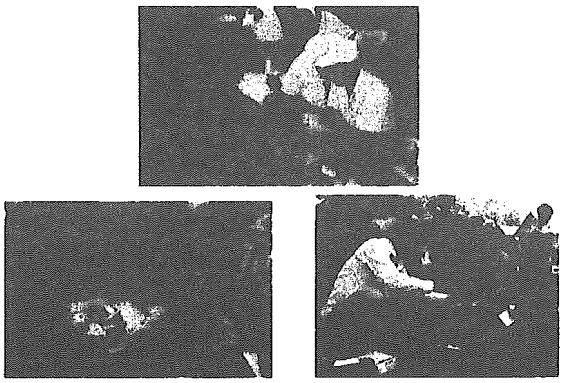
a. Diagnosis by Microscopy/Dipsticks

b. Treatment with prepackaged antimalarial drugs

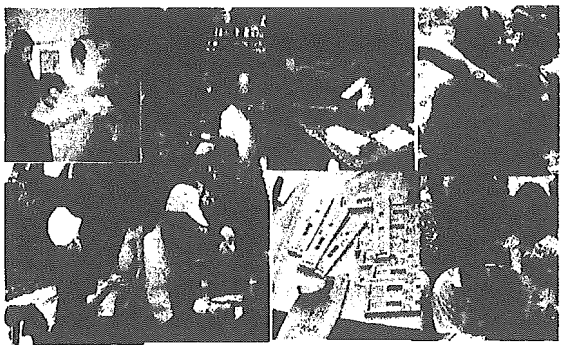
c. Hospital care for severe and complicated malaria cases



OUTREACH SERVICES



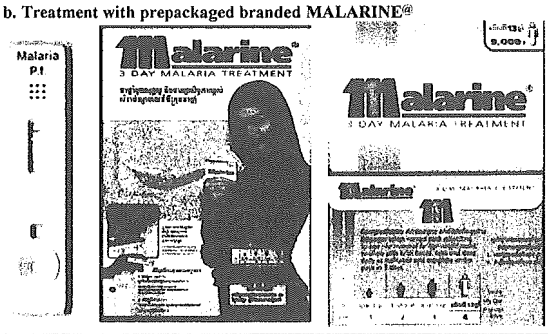
.....AND EDAT THROUGH VMWs



SOCIAL MARKETING FOR EARLY DIAGNOSIS AND PROMPT TREATMENT AT PRIVATE HEALTH FACILITIES

a. Diagnosis by Dipsticks

b. Treatment with prepackaged branded MALARINE®



COMMUNITY & PERSONAL PROTECTION THROUGH:
a. FREE DISTRIBUTION OF INSECTICIDE-TREATED BEDNETS

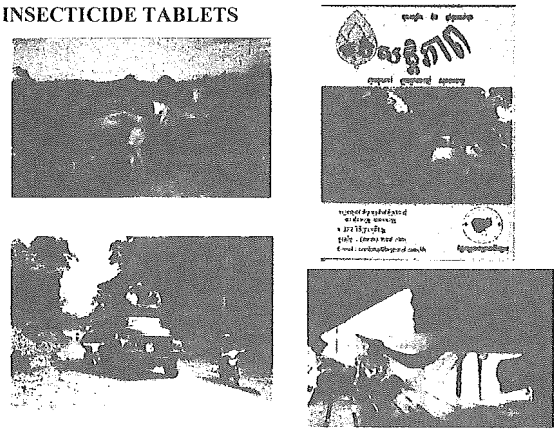
Classification of risk groups

- Category 1 defined as people reside in forested villages.
- Category 2 defined as people reside in the villages at 200 m distance from forest.
- Category 3 defined as people reside in the villages between 200 to 500 m distance from the forest.
- Category 4 defined as people reside in the villages between 500 to 1,000 m distance from the forest.

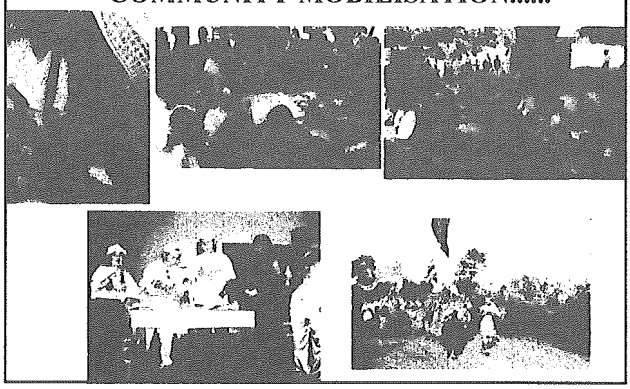
Strategy

- Impregnated bed net free distribution and implementation targeting to population at risk category 1 and 2.
- Social marketing of hammock net with insecticide K-O TAB targeting to population at risk category 3 and 4.

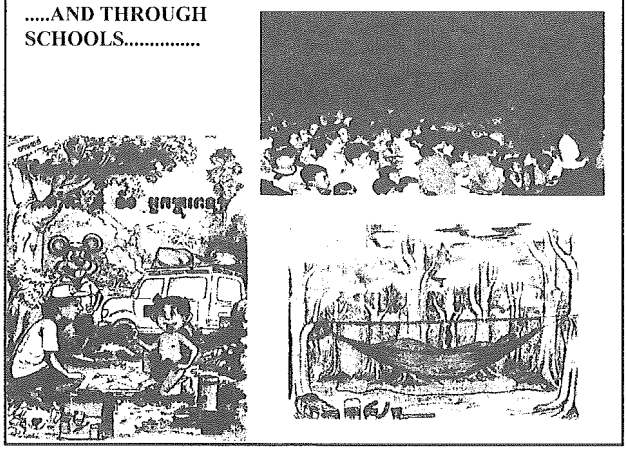
b. SOCIAL MARKETING OF HAMMOCK NETS AND INSECTICIDE TABLETS



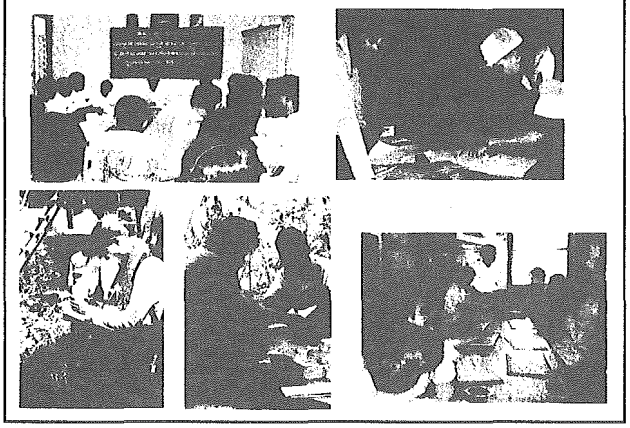
HEALTH EDUCATION AND COMMUNITY MOBILISATION.....



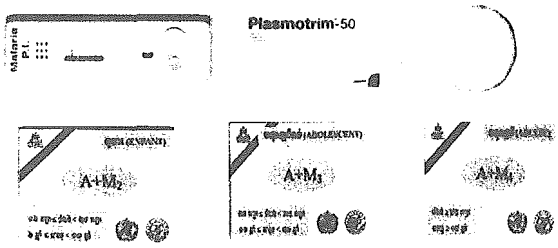
.....AND THROUGH SCHOOLS.....



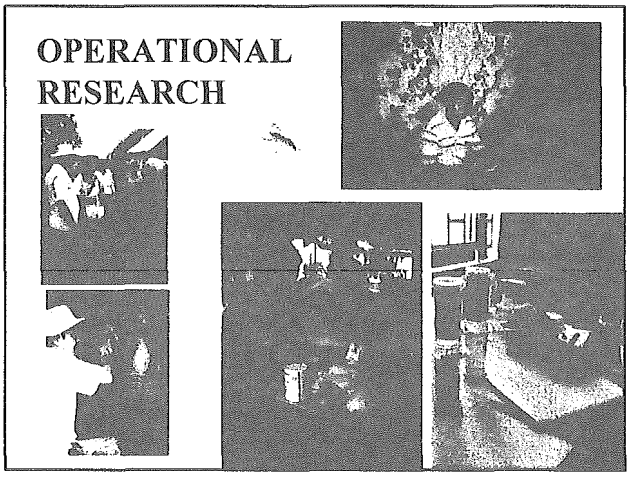
TRAINING, MONITORING AND SUPERVISION

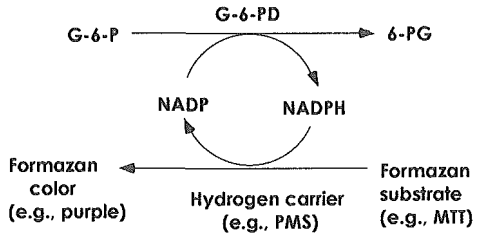


VMW approach



OPERATIONAL RESEARCH

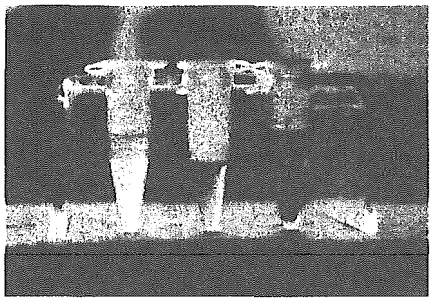




Principle of the chemical reactions for detection of G6PD activity

Major screening methods for detection of G6PD deficiency

1. **UV-spectrophotometric assay**
   Accurate, quantitative, require an expensive spectro-photometer, inadequate for field assay
2. **Spot test, using UV light**
   Simple & rapid, qualitative, require UV light and dark room or chamber, detection of only severe deficiency
3. **Formazan-ring (Fujii) method (MTT/PMS)**
   Accurate, time-consuming to make kit, qualitative, photo-sensitive, incubation at 37C for 8 hrs
4. **Sephadex-gel (Hirono) method (MTT/PMS)**
   Simple & rapid, qualitative, strong photo-sensitive, detection of only severe deficiency
   (difficult to identify heterozygous female)



Detection of G6PD-deficiency by the Sephadex-gel method

(Hirono method)

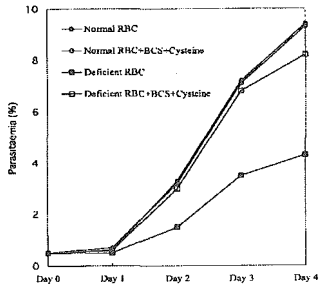
(reaction: 15 min at room temp.)

Useful for field surveys, but
(1) Strong photo-sensitive (eg., reacts with small light)
(2) Difficulty to stop the reaction
(3) Difficulty to detect heterozygous female

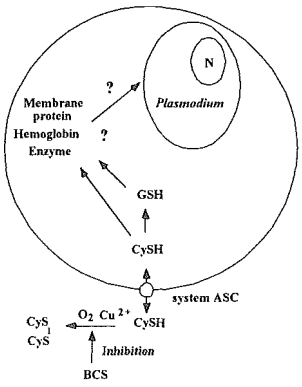
Table 4 Prevalence of malaria detected on-site in G6PD normal and severely deficient individuals

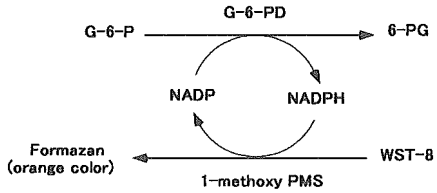
	No. of malaria patients detected on-site (%)	Malaria species identified on-site† (%)							Total	
		F	V	M	F + V	F + M	V + M	F + V + M	F	V
Indonesia										
Normal G6PD*	237/1084 (21.7)	202 (18.6)	18 (1.7)	0	34 (3.1)	1 (0.1)	0	2 (0.2)	239 (22.1)	54 (5.0)
Severe deficiency	16/ 42 (38.1)	14 (33.3)	2 (4.8)	0	0	0	0	0	14 (33.3)	2 (4.8)
Myanmar										
Normal G6PD*	263/1032 (25.3)	194 (18.8)	27 (2.6)	1 (0.1)	27 (2.6)	2 (0.2)	8 (0.8)	6 (0.6)	229 (22.2)	68 (6.6)
Severe deficiency	16/ 47 (34.0)	11 (23.4)	3 (6.4)	0	2 (4.3)	0	0	0	13 (27.7)	5 (10.6)

*Including mild deficiency (>10% activity of the control). †F, *Plasmodium falciparum*; V, *P. vivax*; M, *P. malariae*



Suppression of asexual growth of *P. falciparum* in G6PD-deficient RBC and synergistic enhancement of growth by a copper chelator (BCS, bathocuproine disulfonate) and cycteine





A new method using WST-8 and 1-methoxy PMS:
- **Photo-resistant**
- **Quantitative (ELISA reader)**
- **Stop reaction by 1N-HCl**

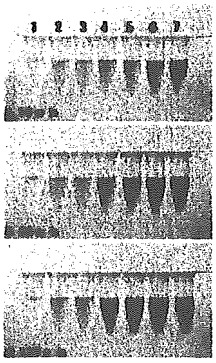
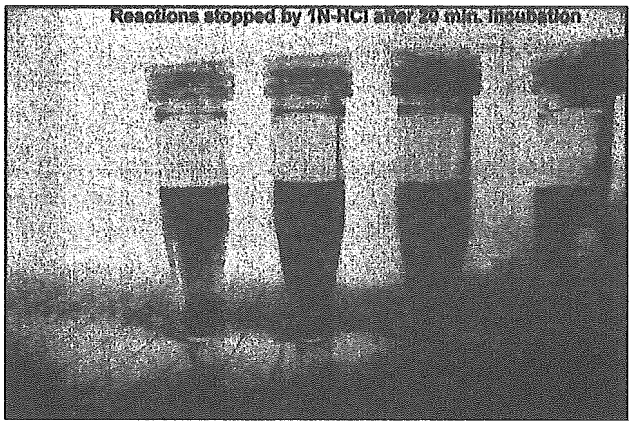
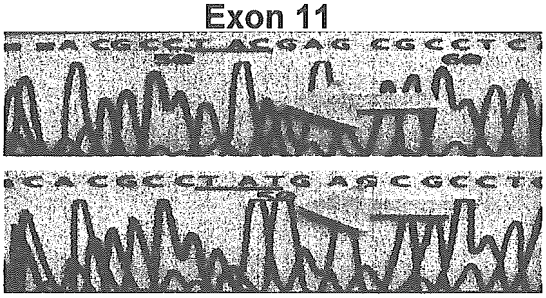


Figure 2. Orange color development with blood samples of different G6PD activities by mixing normal blood (8.0 IU/g Hb) and G6PD-deficient blood (<0.1 IU/g Hb). 5 µl blood is loaded in each tube of 2-7.

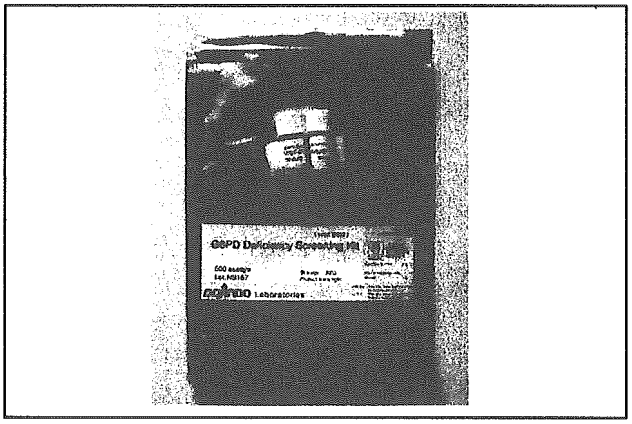
1, Reaction mixtures without blood;
2, Normal blood without the substrates;
3, G6PD-deficient blood;
4-6, G6PD-deficient blood mixed with normal blood at ratio of 3:1, 1:1 and 1:3, respectively;
7, Normal blood.

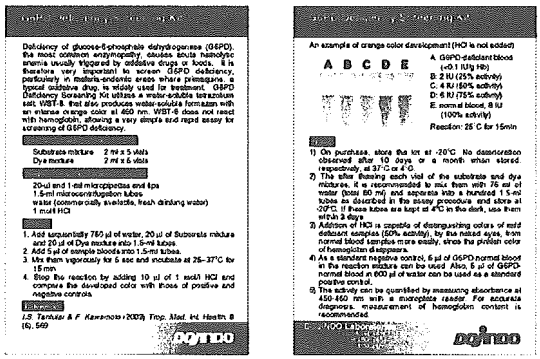


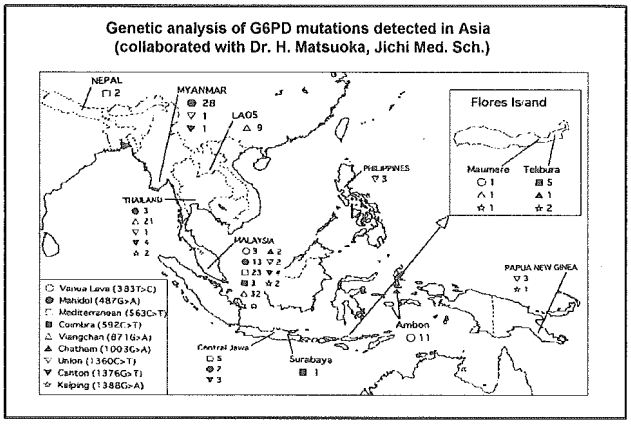
Exon 11



Upper : normal
Lower : Viangchan type (TAC→TAT)







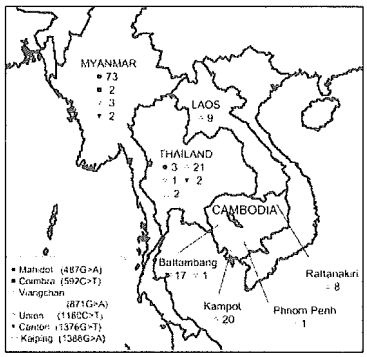


Fig. 1 G6PD variants in Southeast Asian countries

Border malaria characters of Korean *Plasmodium vivax* and its control

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Division of Malaria and Parasitic diseases, National Institute of Health, Center for Disease Control and Prevention, Seoul, Korea

Malaria Situation in Korea

20 century

Early of 20 century

Number of malaria patients treated at the hospitals in 1910 (Hasegawa, 1913)

	Locality	Korean civilians	Japanese soldiers
North	Uiju	63	-
	Pyongyang	36	-
	Hamhung	328	16
	Onjong	-	40
	Wonsan	-	31
Middle	Chunchon	294	-
	Seoul	114	47
	Kongju	66	-
	Taejon	-	13
South	Chungju	93	-
	Kwangju	198	-
	Chinju	136	-
	Total	1,328	147

Ree HI. 2000. Korean J. of Parasitology. 38: 119-138

Malaria cases admitted in the Severance Hospital, Seoul,1926-1935 (Choi, 1936)

Year	No. of Patients	No. of Malaria cases	%
1926	2,179	13	0.6
1927	2,349	7	0.3
1928	2,530	20	0.8
1929	2,641	19	0.7
1930	2,435	2	0.1
1931	2,661	2	0.1
1932	2,774	3	0.1
1933	3,140	3	0.1
1934	3,062	7	0.2
1935	3,034	8	0.3
Total	26,775	84	0.31

Ree HI. 2000. Korean J. of Parasitology. 38: 119-138

Middle of 20 century

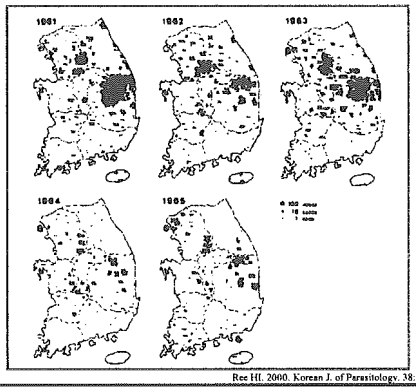
- 1948, 584/3,983 (14.7%), Middle school, (Seoul) (Chun, 1959)
- 1952, 1,032 malaria cases reported in Yangyang-gun, Kangwon-do (Kim and Han, 1953)
- Military cases (400,000)
  - 1953; 8,855 cases (2.2%)
  - 1954; 5,741 cases (1.4%)
- US army
  - 1951-1952; 1,513 cases (Hanky et al., 1953)
- Canadian army
  - 1952; 152/1,350 (11.3%) (Hale and Halpenny, 1953)

Ree HI. 2000. Korean J. of Parasitology. 38: 119-138

Number of malaria cases by province as reported by passive case detection (PCD) program in 1961-1965 (NMES, 1966)

Locality	No. of Exam.	No. of Positives	%
Seoul	1,092	56	5.1
Kyonggido	12,501	3,831	30.6
Kangwondo	2,077	680	32.7
Chungchongbukdo	1,717	353	20.6
Chungchongnamdo	2,939	169	5.8
Kyongsangbukdo	20,526	8,522	41.5
Kyongsangnamdo	1,279	60	4.7
Chollabukdo	1,823	153	8.4
Chollanamdo	1,220	81	6.6
Chejudo	221	24	10.9
Total	45,395	13,929	30.7%

Distribution of the malaria cases detected by PCD in 1961-1965 (NMES, 1966)



Ree HI. 2000. Korean J. of Parasitology. 38: 119-138

Number of reported malaria cases by PCD in north Kyonggido in 1966-1969 (Shim and Kim, 1999)

Area	Population	No. of Exam.	No. of Positive	%	API[a]
Paju	182,804	11,032	638	6.2	0.35
Koyang	86,151	366	41	11.2	0.12
Yonchon	68,638	58	10	17.2	0.04
Pochon	134,684	647	24	3.7	0.05
Kimpo	84,928	478	55	11.5	0.16
Kangwha	119,129	749	72	9.6	0.15
Yangju	210,470	890	10	1.1	0.01
Uijongbu	69,969	698	5	0.7	0.02
Total	956,773	14,918	900	6.0	0.24

[a]Annual parasite incidence per 1,000 population

Ree HI. 2000. Korean J. of Parasitology. 38: 119-138

Late of 20 Century

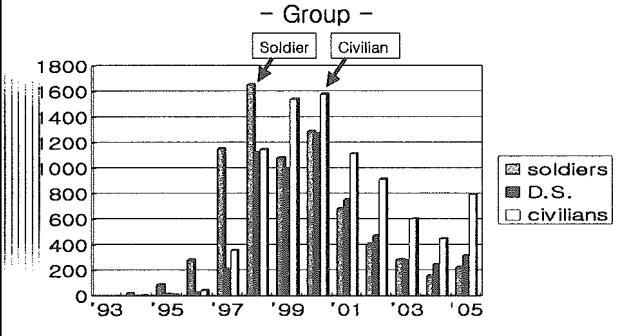
Annual incidence

	'93 – '95	'96	'97	'98	'99	'00	'01	'02	'03	'04	'05
Civilians	9	46	361	1,148	1,541	1,580	1,115	897	597	449	796
D.S.	13	285	1,156	1,657	1,048	1,289	685	430	274	249	319
Soldiers	107	25	207	1,127	9 96	1,273	756	472	286	159	223
Total	129	356	1,724	3,932	3,621	4,142	2,556	1,799	1,157	857	1,338

•D.S: Discharged soldier

Center for Disease Control and Prevention, Korea

Annual incidence of malaria - Group -



* D.S: Discharged soldier

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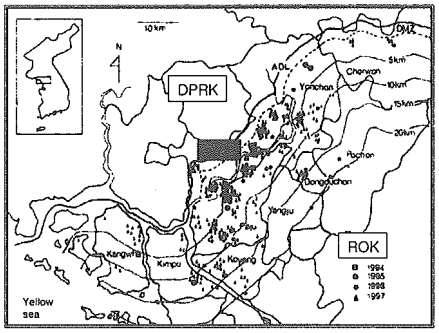
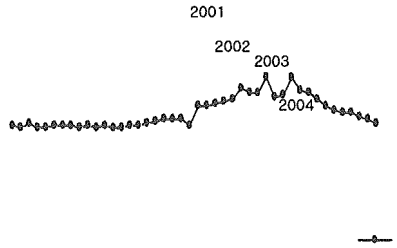
Distribution of 278 civilian inhabitant cases of vivax malaria in 1994-1997 (Kho et al., 1999)



Malaria cases within 10 km: 232/278 (80.8%)

Kho et al., 1999. Korean J. of Parasitology. 37: 71-76

Weekly reported malaria cases (2001-2004)



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Classification of risk areas

◎ 20 cities or counties

	Risk area (2004)		
	High (>100/100,000)	Middle (>10/100,000)	Low (1-10/100,000)
Gyeonggido (Province)	-	Gimpo-si, Dongducheon-si, Paju-si, Yeoncheon-gun	Yangju-si, Uijeongbu-si, Gapyung-gun, Goyang-si (Ilsan-gu, Deokyang-gu)
Incheon metropolitan city	-	Gangwha-gun	Seo-gu, Dong-gu, Jung-gu, Ongjin-gun
Gangwondo (Province)	-	Gosung-gun, Cheorwon-gun	Whacheon-gun, Chuncheon-si, Yanggu-gun, Inje-gun

* This classification was deduced by the number of civilian and discharged soldier group only

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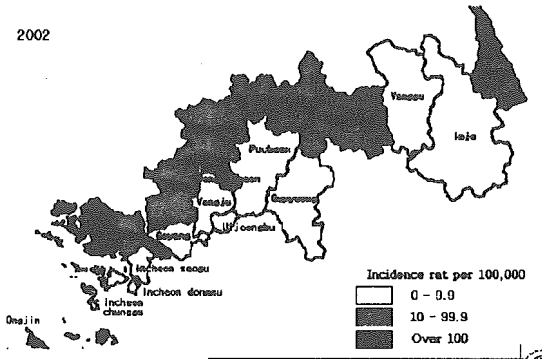
Mapping of malaria cases in risk areas (1997-2004)



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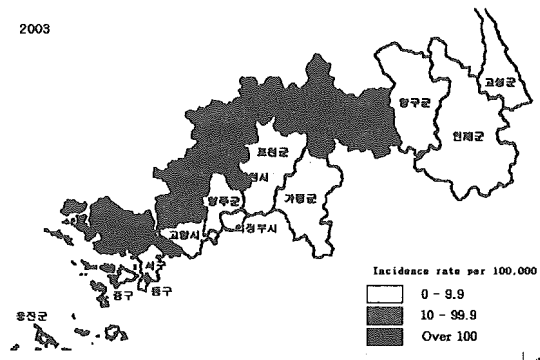

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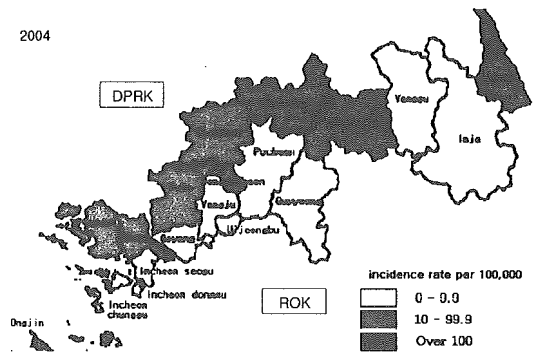

Strengthen the management abilities
- Soldier group -

◎ Chemoprophylaxis in risk areas (May~Sep.)
- Chloroquine: 400 mg/week (20 weeks)
- Primaquine: 15 mg/day (14 days)

◎ Mosquito control together with local Public Health Center
- Larva control: exclusion of weeds and puddles
- Adult control: space and residual spray

◎ Individual protection: permethrin treated clothes

◎ Early detection of patients and treatment

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New trial for the control of malaria in temperate zone

Antibody detection method

- Indirect Immunofluorescent Antibody Test (IFAT)
  - Pv, Pf, Pm detection
- Enzyme linked immunosorbent assay (ELISA)
  1. Circumsporozotite protein (CSP) – Pv247, Pv210 detection
  2. Merozoite surface protein (MSP) – Pv detection
  3. Liver stage specific antigen (LSA) – Pf detection
- Western blot
  - Pv, Pf detection

Application of malaria antibody test

- Case detection & determination of malaria species
- Screening of blood donor
- Determination of malaria endemicity rate
- Evaluation of malaria transmission after malaria eradication or control program
- Determination of malaria foci
- Determination the areas where need malaria control program

Indirect Immunofluorescent Antibody Test (IFAT)

- Malaria antigen preparation for the IFAT

1. Collect 5 ml of blood preferably from a patient with a parasitemia>3% (minimum 1%).
2. Centrifuge the blood at 2,500 rpm for 5 min.
3. Discard the supernatant and suspend the pellet in PBS.
4. Repeat steps 1 & 2, i.e. wash pellet three times with PBS.
5. Resuspend the packed cells in a small volume of PBS, approximately half the original volume.
6. Add a drop of the above preparation to each well of a Teflon coated slide.
7. Air dry the slide overnight and freeze at – 70℃ until required.

Measurement of antibody using IFAT

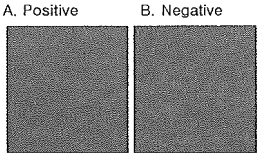
1. Remove antigen slides from the deep-freezer, fix in aceton, dry, and label appropriately (each slide must contain a positive and a negative control).
2. Dilute the test sera in PBS, and add 20 ㎕ diluted sera to the appropriate wells, and incubate in moisture chamber for 30 min. at 37℃.
3. Wash the slides in PBS for 6 min. then air dry.
4. Dilute FITC conjugated anti-human IgG, 1:32 in PBS. Add 20 ㎕ to each well.
5. Incubate the slides in moisture chamber at 37 ℃ for 30 min.
6. Wash the slides in PBS for 6 min. then air dry.
7. Add one drop of buffered glycerol to each well. Attach a coverslip and view the slides under x 40 fluorescence objective.

The power of IFAT

Sensitivity: more than 96%

Specificity: more than 97%

A. Positive B. Negative



Positive and negative control of IFAT. A; *P. vivax* infected patient, B; Normal person.

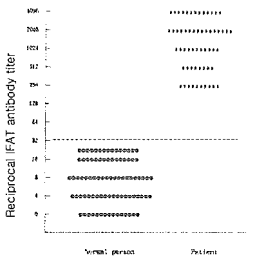


Fig. . IFAT antibody titers of normal people and malaria patients.

Evaluation of antibody test as useful tool for the detection of potential patients and application of selective chemoprophylaxis to reduce the possible appearance of drug resistance

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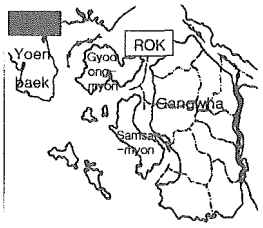
Study 1

Efficacy of selective chemoprophylaxis using antibody test



Study area: Gyodong-myon, Gangwha-gun, Gyonggi-do, South Korea (2000)

- No. of test: 1,475
- Positive rate: 3.7% (n=54)

Treatment: Chloroquine, 2,000mg (3 days)
Primaquine, 15mg x14 days

	2000 (before)	2001.9 (after)	Reduction rate
Gyodong-myon	43	18	58.1%
Samsan-myon	15	11	26.7%
Gangwha-gun (Total)	183	106	42.1%

Table 1. Comparison the situation of malaria patient before (2000) and after (2001) selective chemoprophylaxis through the antibody test against malaria.

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Study 2

Follow up study of non-drug treatment group of antibody positive cases (2001-2002)

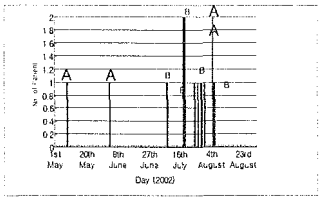
Method: Examine the total malaria patients who reported in 2002 (n=86) and checked their antibody history of 2001

A: Malaria patients (n=4) who showed antibody positive in winter season, 2002(8th May, 3rd June, 5th Aug, 5th Aug).

B: Malaria patients (n=10) who showed antibody negative in winter season, 2002(8th Jul, 18th Jul, 18th Jul, 19th Jul, 19th Jul, 25th Jul, 27th Jul, 29th Jul, 31st Jul, 5th Aug)

Note: The other malaria patients (n=72) did not involved in antibody test in 2001.

Study area: Gangwha-gun, Gyonggido, South Korea



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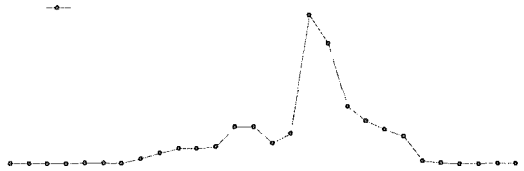
Seasonal population density of mosquito, 2002



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Study 3

1. Study area: Gympo-si, Gyonggido, South Korea

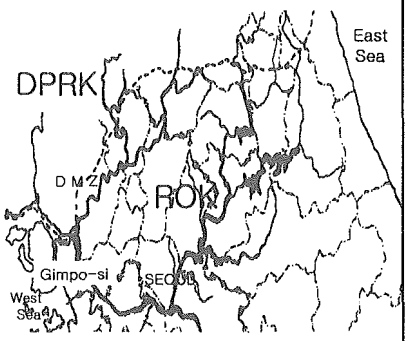
Blood collection: Nov-Dec, 1998

2. No. of Test: 845 inhabitants

No. of Positive: 24

(Non-treatment group)

3. No. of patient who fell ill in 1999 with long incubation period among sero-positive inhabitants (n=24) ; 4 (16.7%)



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Study 4

Effect of selective chemoprophylaxis on reduction of malaria patients in follow year

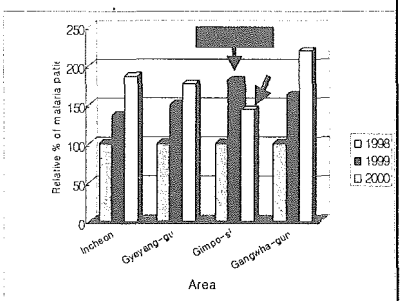
1. Study area: Gympo-si, Gyonggido, South Korea

2. Blood collection: Nov-Dec, 1999

3. No. of Test: 10,864 inhabitants

4. No. of Positive: 275 (2.54%)

5. Treatment: Primaquine, 14 mg x 14 days



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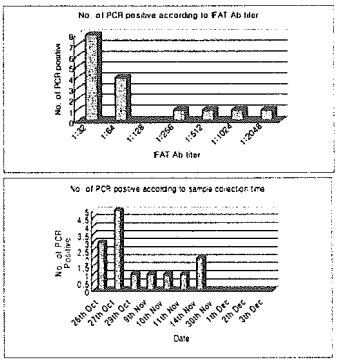
Study 5

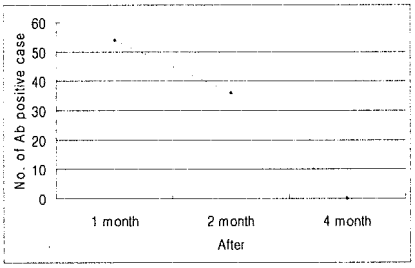
Relationship between IFAT and PCR

- Blood collection; Oct – Dec, 1998
- Study area: Gyonggido, Gangwondo (Province)
- No. of test: 3,422
- Method: IFAT, PCR
- Positive rate
  - Gyonggido: 7.09% (151/2,128)
  - Gangwondo: 3.71% (48/1,294)
- PCR positive rate among IFAT positive cases: 8.04% (16/199)



Study 6

Longevity of IFAT antibody



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Limitation of application of IFAT

- It might be used to detect the long incubation period patients in temperate zone only, that is long winter should be there.
- To apply the IFAT, it must be considered whether the level of antibodies against malaria in inhabitants who live in risk areas can be down below the negative criteria before malaria epidemic seasons coming.
- It has to be considered the false positive of examinees through paperweight individually.

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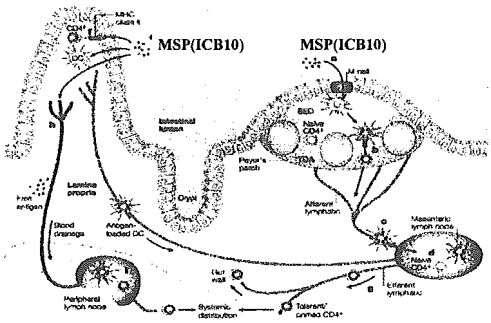
Mucosal immune response in Mice intestine

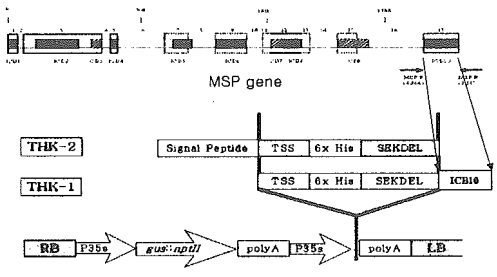
(Merozoite surface protein)

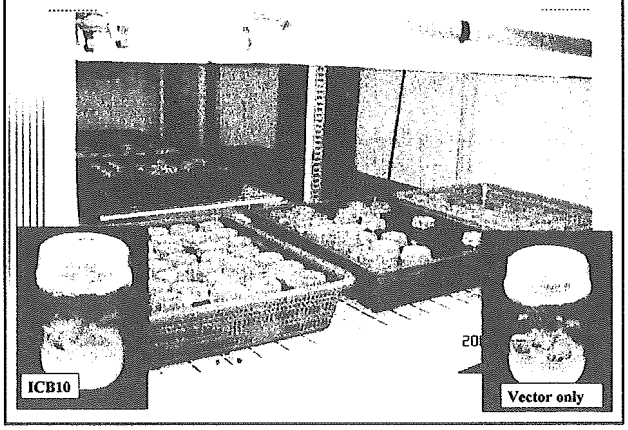


Mowat, A. M. 2003. Anatomical basis of tolerance and immunity to intestinal antigens. Nat. Rev. 3:331-341

Transgenic plant (edible vaccine trial)

merozoite surface protein gene (ICB10)





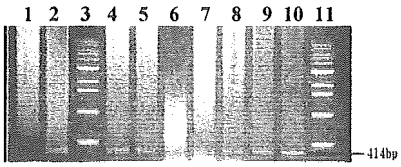
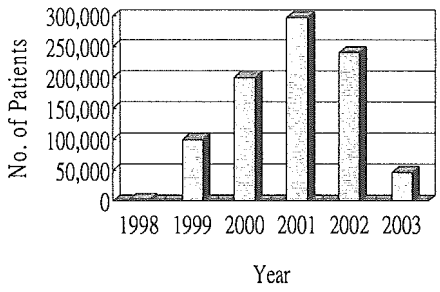


Fig. 1. Presence of the recombinant sequence in representative transgenic *Brassica carinata* plants detected by RT-PCR with THK-1 primer set.

Lane 1; Negative control (THK-1 vector DNA)
Lane 2; Positive control (GV3101/THK-1:ICB10 plasmid DNA)
Lane 4~10; *B. carinata* [pHS737-THK-1:ICB10], transgenic plants
Lane 3, 11; 1kb DNA Ladder

Vivax malaria in North Korea

Malaria cases in North Korea



P. vivax malaria epidemic during past 3 years in North Korea

	2002	2003	2004
No. of smear positive	98,852	16,538	15,827
No. of clinical cases	142,338	29,713	17,850
Total	241,190	46,251	33,677

P. Vivax Malaria incidence by year

(per 1,000 populations)

- *P. Vivax* Malaria incidence in 2002: 10.7
- *P. Vivax* Malaria incidence in 2003: 2.7
- *P. Vivax* Malaria incidence in 2004: 1.4