

**Evaluation of a Simple  
Immunological Test (sIgA) During  
the RAAF Survival Course**

J.E. Carins and C.K. Booth

DSTO-RR-0224

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# Evaluation of a Simple Immunological Test (sIgA) During the RAAF Survival Course

*J. E Carins, and C. K. Booth*

**Combatant Protection and Nutrition Branch  
Aeronautical and Maritime Research Laboratory**

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## **ABSTRACT**

A simple marker of immune function, salivary immunoglobulin A (sIgA), was evaluated as a potential indicator of stress during the RAAF Survival School courses conducted in November 1999. Twenty-seven males and two females with an average age of 26 years participated in the study by keeping a food diary, collecting saliva samples and recording their health problems (daily checklist) and level of anxiety (State-Trait Anxiety Inventory). Participants height and weight changes were also recorded. Dietary restriction, consumption of alcohol, loss of weight and negative emotions were all shown to have a negative effect on sIgA. Salivary IgA was shown to be a useful marker of the severity of stresses encountered during the survival course.

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# Evaluation of a Simple Immunological Test (sIgA) During the RAAF Survival Course

## Executive Summary

Maintaining the health of Defence personnel is integral to ensuring a high level of performance in the field. To adjust training programs or duties in order to maintain optimal health of personnel, an indicator is required that predicts an increasing vulnerability to infection or illness. Because decreased concentration of immunoglobulin A in saliva (sIgA) is a potential indicator of increased disease risk, it was evaluated as a marker of the severity of stress during a RAAF Survival School course. The RAAF Combat Survival Training School (RAAF Base Townsville) is the only Australian military unit wholly dedicated to conducting survival training, and students experience the stresses of hunger, boredom, loneliness, extremes of thirst, heat and cold combined with demanding physical effort. The course deals with survival in three types of terrain, namely sea, arid land and jungle. This report details the results of a study conducted during the November 1999 survival course.

Twenty-seven males and two females agreed to participate (mean age 26 years, range 18 – 39). Subjects were instructed how to keep a daily food diary, from which daily intake of energy and macronutrients (carbohydrate, protein, fat and alcohol) were calculated. Saliva samples were collected at the beginning of the course, and on days 2, 4, 6, 9, 13, 15, 17, and 18. These were used for the measurement of the ratio of sIgA (mg/L) to albumin (mg/L). Subjects completed a checklist each time they donated a saliva sample, to indicate any health problems they had experienced. The Profile of Mood States questionnaire was used to assess changes in mood, and the State Anxiety Inventory was used to assess changes in anxiety during the survival course.

The study found significant changes in sIgA:Alb, with measurements taken at baseline increasing during the early part of the course, then decreasing after each of the arid and jungle phases of the course. There were also significant changes in dietary intake, weight, health problems, mood factors and anxiety.

Dietary intake in barracks provided more than sufficient protein and fat, but for most students, inadequate carbohydrate and energy, and inadvisable levels of alcohol. Despite these less-than-optimal intakes, sIgA:Alb levels appeared to recover in barracks, and might have recovered further with better diet, including less alcohol.

Factors that may have contributed to decreased sIgA:Alb were inadequate intake of energy and macronutrients, consumption of alcohol, loss of body mass and the negative emotions of anger and depression. Multi-regression analysis found inadequate carbohydrate consumption to most clearly predict the decline in sIgA:Alb. Decreased sIgA:Alb was associated with increased reporting of health problems.

The results presented here show that sIgA:Alb is a useful marker of the severity of stresses encountered during the survival course. This simple saliva test may be used as a tool for evaluating the effectiveness of interventions to boost immune function such as improved diet and measures designed to reduce fatigue.

It is recommended that evaluation of sIgA as a marker of overtraining continue and that consideration be given to the provision of nutritional education in situations where overtraining may occur, such as the RAAF survival school.

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# 1. Introduction

## 1.1 Background

Maintaining the health of Defence personnel is integral to ensuring optimal performance in the field. Prolonged physical activity, exposure to the elements, periods of inadequate nutrition, and psychological challenges are all factors that could have an impact on the health of personnel. To be able to alter training programs or duties to maintain optimal health of personnel, an indicator is required that predicts an increasing vulnerability to infection or illness.

Decreased concentration of secretory immunoglobulin A (IgA) is a potential indicator of increased disease risk. Secretory IgA is found at the mucosal membranes, and represents the first line of defence against viruses and bacteria. As most infectious agents enter the host via the mucosal membranes, a secretory IgA deficiency represents a reduced level of protection for the body, and an increased risk of infection.

Saliva, tears, nasal associated fluids, bronchial, intestinal and genitourinary secretions contain immunoglobulins, predominantly secretory IgA [1]. Secretory IgA protects the mucosal surfaces by several mechanisms, which result in the inability of pathogens to attach to mucosal surfaces, thereby preventing entry into the body, limiting viral replication or aiding in elimination [2]. The levels of secretory IgA are correlated with resistance to certain viruses responsible for upper respiratory tract infections [3].

In a study of US army recruits performing an 8-week physical training program about one third were classified as overtrained [4]. Overtraining is typified by a decrease in performance, but other symptoms can include fatigue, mood disturbances including apathy, irritability, anger, and depression, frequent upper respiratory tract infections, injury, muscle soreness and joint pain [3].

Athletes undergoing high intensity training have a higher incidence of upper respiratory tract infection [5, 6]. It has been found that concentrations of secretory IgA in saliva (sIgA) have been suppressed after intense exercise in elite swimmers [7], in moderately active runners [8], cross country skiers, [9], distance runners [10], cyclists [11] and kayakers [12].

There is also an indication that exposure to psychosocial stressors and negative mood leads to immune suppression [13, 14]. A negative relationship has been found between the number of daily hassles and sIgA levels [15] and the secretion rate of sIgA was lower in nurses who reported that they were "frequently anxious" [16]. Salivary sIgA concentration was also lower in students during high stress periods [17] and positive associations have been found between sIgA concentration and positive mood states [18, 19].



Malnutrition has negative effects on the overall health of the individual, and it has been found that protein – calorie malnourished children have suppressed sIgA levels [20, 21, 22]. Recently it has been postulated that carbohydrates could affect the delicate balance of immunoglobulins, micro-organisms and lectins existing in the alimentary canal, with possible benefits for health [23].

A single diagnostic assay such as a disposable saliva test stick, which could indicate overtraining by estimating sIgA, would be ideal, and could be used by commanding officers to predict overwork, particularly in training situations. Due to the ease of saliva collection, measurement in the field may be feasible, if a robust and cheap method of analysis can be developed. It is first necessary, however, to verify that sIgA is a sensitive marker of the overtraining problems that can occur during ADF military training.

## 1.2 RAAF Survival Training Course

The RAAF Combat Survival Training School (RAAF Base Townsville) is the only Australian military unit wholly dedicated to conducting survival training. Their focus is on training crew from downed aircraft to survive in a hostile environment until rescued. During the survival courses, personnel experience the stresses of hunger, boredom, loneliness, extremes of thirst, heat and cold, combined with demanding physical effort.

This study was conducted during the November survival course, from 2 November 1999 to 20 November 1999 (19 days). Upon arrival the students were required to complete physically challenging activities to determine if they had the minimum physical and mental fitness needed to attempt the course. This initial (baseline) phase was conducted over 3 days (days 0 to 2). The students were then required to demonstrate survival skills in three different environments, a sea phase (days 3 to 4.5), an arid phase (days 7 to 9.5) and a jungle phase (Days 13 to 18).

The final jungle phase, which involves an escape and evasion exercise, is the most demanding. Students evade an enemy for two days, during which time they cover up to 25km over rough terrain. After that they are required to employ their survival skills for another five days in the jungle, the last two of which are spent in a solitary situation. When they are rescued they are totally exhausted.

While at the Base, (between phases) students are fed well in the dining hall, but their physical activity levels are high. They are required to complete at least two bouts of strenuous activity each day, such as a 1000ft climb in 2km, over rocky terrain. There are time limits on these activities, and those who fail to complete them within the time are eliminated from the course.

During the survival phases, the students complete survival activities in groups. They create shelters from the materials they would have in the event of a plane crash, (parachutes) and from the surrounding area (eg. branches). They are exposed to the

elements. They disguise their location, or promote it for rescue purposes, depending on the simulated hostility of the area. They are provided with very little food, so they search for native foods, and water sources.

Against this adversity, they try to maintain a sense of mental and physical well being. While in teams the members help each other, but in the final few days, they are also required to complete two days of solo survival. This is after having already spent five days in the jungle, two of which were spent evading from an enemy. In a weakened physical and mental condition, this solo survival presents a formidable challenge.

### **1.3 Salivary IgA Study**

In a twelve-day nutrition study conducted by DNRC during Exercise Northern Awakening (RAAF Base Scherger, April-May 1999) Airfield Defence Guards displayed significantly reduced sIgA concentrations. Subjects receiving combat ration packs had significantly lower levels than control subjects, who received freshly prepared meals [24].

This Survival School course provided ideal conditions for the Defence Nutrition Research Centre (DNRC) to further evaluate the use of this immunological test. The subjects were under physical stress, combined with malnutrition and psychological stress. Most students lose body mass during the survival course, some as much as 10 kg. Their 'ration' during the survival phases provides between 1,000 - 2,000kJ/day, while their estimated energy expenditure can be as high as 19,000kJ/day. The hypothesis tested during this study was that the combination of exercise stress, malnutrition, adverse environmental conditions, fatigue and psychological stress would result in decreased secretion of IgA into saliva.

## **2. Methods**

### **2.1 Subjects**

All students in the November 1999 RAAF Survival course were briefed about the study and were invited to participate. Twenty-nine subjects agreed to participate. Only two of the subjects were female. The average age of the group was 26 years (range 18 - 39). Most subjects were in the normal body mass range with an average BMI (Body Mass Index) of 24 (range 20 - 31). Subjects were Air Force personnel from around Australia, except for three British Air Force personnel and one Navy clearance diver. The Australian Defence Human Research Ethics Committee approved the experimental procedures. Written consent was obtained from each participant after the details of the study were explained to him or her.

## 2.2 Dietary intake

Subjects were instructed on how to keep a food intake diary for the duration of the course. From these diaries the daily intake of energy and macronutrients (carbohydrate, protein, fat and alcohol) was calculated by use of the Foodworks software (Version 2.05, 1999, Xyris Software, QLD), which used the NUTTAB 95 food composition database. The daily energy intake for each subject was compared with their recommended energy requirement as determined by the Foodworks software. In this way the daily energy deficit was calculated for each subject.

## 2.3 Saliva analysis

The subjects donated three 'baseline' saliva samples during the initial classroom session on day zero of the course. Further saliva samples were collected immediately before and after each of the survival phases and twice during the final jungle phase (days 2, 4, 6, 9, 13, 15, 17, 18).

Subjects were asked to rinse their mouth then place a cotton swab in their mouth, leaving it in place without chewing till they had to swallow. The subject then returned the cotton swab into a Salivette tube (Starstedt, Germany). Samples were transported cold to the barracks where they were stored frozen. Albumin (Alb) and salivary immunoglobulin A (sIgA) were measured by nephelometric assay (Behring BNA II) using manufacturer-supplied reagents (antisera to Human IgA  $\alpha$  chain and human albumin). The results were presented as the ratio of sIgA (mg/L) to Alb (mg/L).

## 2.4 Body Composition

Height (m) and body mass (kg) were measured at baseline (day 2) and body mass was measured again on days 13 (pre-jungle phase) and 18 (post-jungle phase). Body composition was presented as the BMI (body mass/(ht)<sup>2</sup>). Bioelectrical impedance measurements were also recorded in an effort to calculate body fat composition, but because of a technical problem with the instrument, the results were unreliable and are not presented here.

## 2.5 Health

Subjects filled out a health questionnaire, at the beginning of the study, to identify any subjects with existing health problems. No subjects were excluded on the basis of health problems recorded in the health questionnaire.

Subjects were asked to complete a simple health checklist ('physical condition report') when saliva samples were collected (baseline, days 2, 4, 6, 9, 13, 15, 17, 18). The checklist included the following health problems: cough, cold, runny nose or sore throat (suspected upper respiratory tract infection – URTI), skin complaints (rashes or sores), dehydration, insect bites, headaches, allergic reactions, gut problems (stomach

upset, nausea or vomiting), exhaustion, muscle or joint problems (fracture, dislocation or muscle strain), sunburn, cuts or bruises and other physical symptoms. Each symptom was scored as *present* or *absent* at each time point to give a physical condition score of between 0 and 12.

## 2.6 Psychology testing

The Profile of Mood States questionnaire (POMS, McNair, Lorr and Droppleman, Edits., CA USA) was used to assess changes in mood and the State Anxiety Inventory, Form Y (STAI, Spielberger C.D., 1983, Mind Garden Inc., Redwood City, CA, USA) was used to assess changes in anxiety during the survival course. The STAI was administered at the same time as the health checklist (baseline, days 2, 4, 6, 9, 13, 15, 17, 18) and the POMS, at the same time each day, on days 2 (baseline), 13 (pre-jungle) and 18 (post-jungle). The STAI results were presented as a single (weighted) score and the POMS was scored for each of the factors, tension-anxiety, depression-dejection, anger-hostility, vigour-activity, fatigue-inertia, confusion-bewilderment, friendliness and total mood disturbance.

## 2.7 Statistical analyses

Statistical analyses were performed with SPSS (Statistical Package for the Social Sciences, version 9.0, 1999, SPSS, Inc., Chicago, IL). Descriptive statistics were obtained to establish a measure of central tendency and are presented as means, standard deviations and range. Data were checked for outliers and non-homogeneity of the population by use of pair wise scatter plots, box plots and Q-Q plots.

Significance was accepted at  $p < 0.05$  and in linear regression analyses an  $r^2$  value  $\geq 0.25$  was accepted as significant in prediction models. Multiple linear regression analyses were used to assess associations between variables. Repeated measures analysis of variance, which was based on covariance-adjusted post-baseline responses when appropriate, was used to determine significant change in responses for tests with serial measurements. Only when these tests provided evidence of difference were pairwise comparisons employed. Pairwise comparisons were obtained as contrasts specified in the repeated measures analysis of variance performed by SPSS.

# 3. Results

## 3.1 Dietary intake

Figure 1 shows the group mean daily intake of energy and macronutrients during each of the survival course phases. The mean daily requirements at base were determined as energy  $13,600 \pm 1,100$  kJ, carbohydrate  $450 \pm 40$  g, protein  $90 \pm 7$  g and fat  $110 \pm 9$  g and during survival phases as, energy  $15,800 \pm 1,500$  kJ, carbohydrate  $550 \pm 50$  g, protein 90

$\pm 7$  g and fat  $130 \pm 12$  g. Table 1 and Figure 1 illustrate the magnitude of the dietary deficits.

The changes in energy ( $F = 60.82$ ,  $p < 0.01$ ), carbohydrate ( $F = 65.28$ ,  $p < 0.01$ ), protein ( $F = 61.75$ ,  $p < 0.01$ ) and fat ( $F = 46.80$ ,  $p < 0.01$ ) recorded over the survival course were significant. Although the energy consumption in-barracks was similar on each occasion, the consumption of protein was significantly less for the post-sea phase ( $F = 7.24$ ,  $p = 0.01$ ). This coincided with the period of greatest consumption of alcohol. There was a trend to lower consumption of carbohydrates during the post-sea phase ( $F = 3.32$ ,  $p = 0.08$ ).

Table 1 Mean daily intake of energy and macronutrients by subjects for each of the survival school phases.

	Baseline In barracks	Sea phase	Post-sea In barracks	Arid phase	Post-arid In barracks	Jungle phase
Energy (kJ/day)	12500 $\pm$ 3600	2200 $\pm$ 1200	11800 $\pm$ 5400	1900 $\pm$ 750	12000 $\pm$ 4100	1400 $\pm$ 400
Protein (g/day)	140 $\pm$ 30	20 $\pm$ 10	110 $\pm$ 60	30 $\pm$ 8	120 $\pm$ 40	10 $\pm$ 4
CHO (g/day)	360 $\pm$ 130	70 $\pm$ 50	310 $\pm$ 170	50 $\pm$ 30	330 $\pm$ 110	44 $\pm$ 10
Fat (g/day)	110 $\pm$ 30	20 $\pm$ 8	100 $\pm$ 50	20 $\pm$ 7	100 $\pm$ 40	10 $\pm$ 3
Alcohol (g/day)	5 $\pm$ 12		34 $\pm$ 43		27 $\pm$ 27	
No. drinking alcohol	6		15		22	

### 3.2 Body mass

Body mass loss over the survival course was significant ( $F = 748.38$ ,  $p < 0.01$ ). Body mass recorded at baseline were greater than the pre-jungle phase body mass ( $p = 0.04$ ) and greater than the post-jungle phase body mass ( $p < 0.01$ ). The greatest body mass loss occurred during the jungle phase ( $p < 0.01$ ). All subjects lost body mass during this phase of the survival course. The mean body mass loss over the course was 6.5 kg or an average of 9% of initial body mass (range 3.4 kg – 10 kg). Changes in BMI for each phase were also significant. Mean changes in body mass and BMI are shown in Table 2.

Table 2 Mean changes in body mass and BMI

	Initial	Pre-jungle	Post-jungle	Change from Initial to Pre-jungle	Change from Pre-jungle to Post-jungle	Change over whole course
Body mass (kg)	77.1	76.8	70.7	0.4	6.1	6.5
BMI	24.4	24.3	22.4	0.1	1.9	2.0

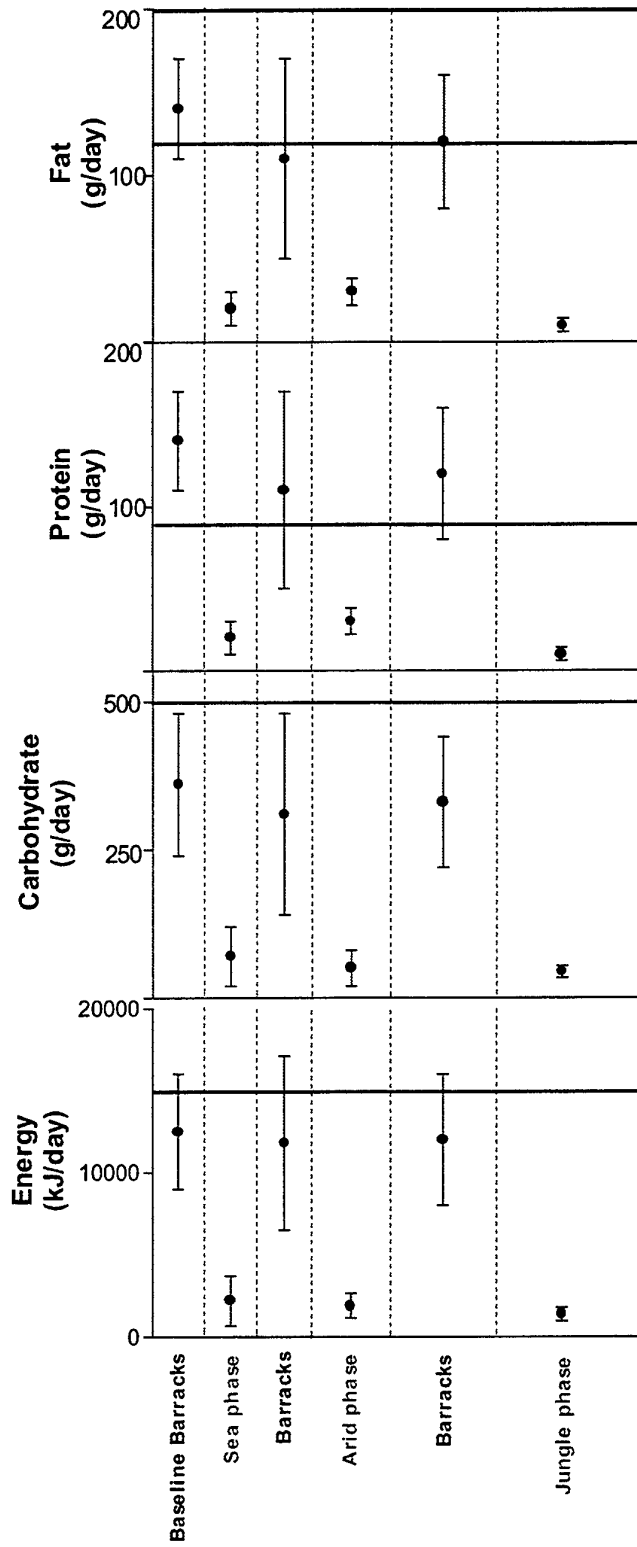


Figure 1 Average daily intake of energy and macronutrients by Survival School subjects. The red/dark line represents the average estimated requirement of energy or macronutrient.

### 3.3 Salivary IgA

Changes in sIgA:Alb over the survival course were significant ( $F = 21.88$ ,  $p < 0.01$ ). Mean values recorded on days 2 (pre-sea,  $p < 0.01$ ), 9 (post-arid,  $p = 0.02$ ), 17 (mid-jungle,  $p = 0.02$ ) and 18 (post-jungle,  $p = 0.00$ ) were significantly different from the baseline. Values reached the lowest point on days 9 (post-arid) and 18 (post-jungle) with the post-jungle values being lower than all other time points ( $p < 0.01$ ). Significant drops in sIgA:Alb were recorded after the arid ( $p < 0.01$ ) and jungle phases ( $p < 0.01$ ). There was a trend to decreased sIgA:Alb over the sea phase ( $p = 0.08$ ). Figure 2 shows the average % sIgA:Alb concentrations at each collection point.

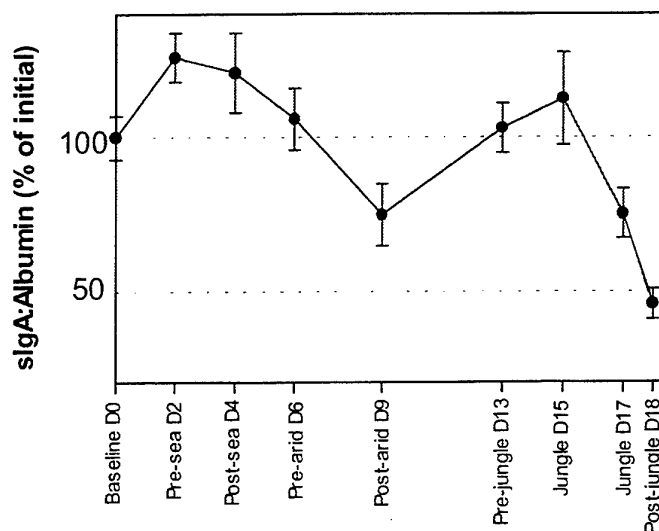


Figure 2 Average & SEM % sIgA:Alb measurements taken during each phase of the survival school course.

### 3.4 Psychological data

The recorded changes in the STAI scores were significant ( $F = 9.59$ ,  $p < 0.01$ ). The baseline (day 0) measurements were quite high with a significant decrease during the pre-sea (in barracks) phase ( $F = 20.80$ ,  $p < 0.01$ ). There were also significant increases during the pre-jungle phase ( $F = 13.15$ ,  $p < 0.01$ ) and the early part of the jungle phase ( $F = 16.22$ ,  $p < 0.01$ ). Significant decreases were then seen in the latter parts of the jungle phase ( $F = 13.83$ ,  $p < 0.01$ ;  $F = 31.61$ ,  $p < 0.01$ ). Figure 3 shows the STAI scores for each phase of the survival course.

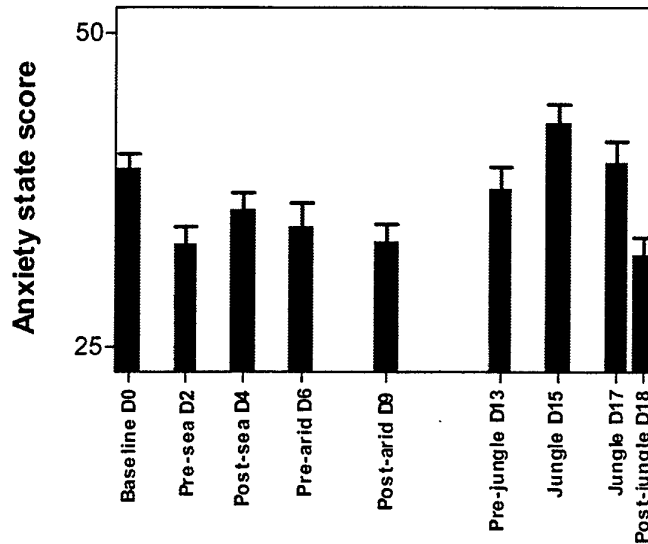


Figure 3 Average and SEM for STAI scores recorded during each phase of the survival school course.

Changes in the mood factors (Table 3) tension-anxiety ( $F = 19.56$ ,  $p < 0.01$ ), vigour-activity ( $F = 7.35$ ,  $p = 0.01$ ), fatigue-inertia ( $F = 58.69$ ,  $p < 0.01$ ) and total mood disturbance ( $F = 4.88$ ,  $p = 0.04$ ) were significant. The increase recorded for depression-dejection was of borderline significance ( $F = 4.15$ ,  $p = 0.05$ ).

Mood changes during the early phases of the course were not significant. Both vigour-activity and tension-anxiety tended to decrease from the initial baseline measurement to the pre-jungle measurement and the decrease after the jungle phase was significant ( $p = 0.01$ ,  $p < 0.01$ , respectively). Similarly for the factors fatigue-inertia and total mood disturbance the scores tended to increase across the early part of the course but only the changes during the jungle phase were significant ( $p = 0.00$ ,  $p = 0.05$ , respectively).

Table 3 Average and SD for POMS factors recorded during the survival school course

Day	Tension	Depression	Anger	Vigour	Fatigue	Confusion	Friendliness	Total mood disorder
Baseline	10 ± 5	4 ± 4	3 ± 3	17 ± 4	6 ± 3	6 ± 3	18 ± 3	11 ± 17
Pre-jungle	9 ± 6	3 ± 4	4 ± 4	17 ± 6	6 ± 5	5 ± 3	16 ± 6	10 ± 19
Post-jungle	5 ± 5	5 ± 8	4 ± 6	14 ± 6	16 ± 6	5 ± 3	17 ± 5	21 ± 28



### 3.5 Health Problems

Changes in the health problems scores (maximum possible in any one day = 12) were significant ( $F = 26.32, p < 0.01$ ). Health problems peaked on days 4 (post-sea,  $p < 0.01$ ), day 9 (post-arid,  $p = 0.04$ ) and day 15 (mid-jungle,  $p < 0.01$ ). The increase in health problems attributed to each survival phase were significant; sea-phase ( $p < 0.01$ ), arid phase ( $p < 0.01$ ) and jungle phase ( $p < 0.01$ ). Figure 4 shows the average total health problem scores recorded during each phase of the survival course in relation to sIgA:Alb and recorded frequency of URTIs. Table 4 lists the frequency of health problems experienced during each phase of the survival course.

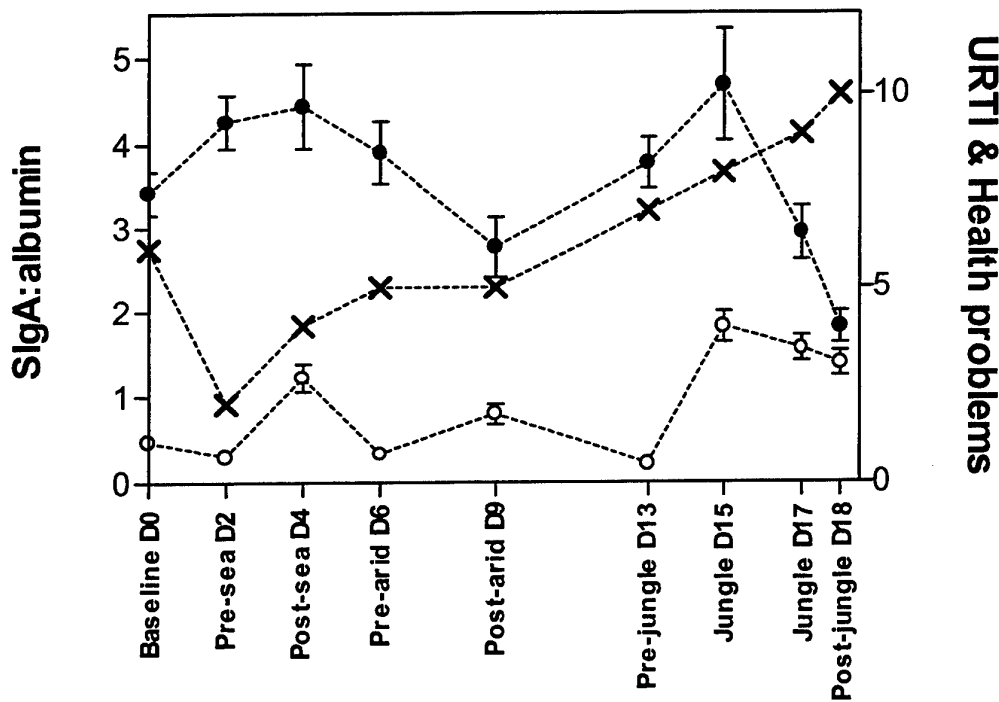


Figure 4 Average health problems scores (o) and total frequency of URTI (X) in relation to sIgA:Alb (•).

Table 4 Frequency of common health problems<sup>a</sup> reported during the Survival School course.

	URTI	Skin rash	Dehydration	Insect bites	Headache	Exhaustion	Cuts	Sun burn
Initial	6	5	0	4	2	0	1	8
Pre-sea	2	4	0	5	0	1	0	2
Post-sea <sup>c</sup>	4	6	15	9	8	13	6	4
Pre-arid	5	4	1	4	0	1	2	1
Post-arid	5	5	11	5	2	12	4	3
Pre-jungle	7	4	0	1	0	0	1	0
Mid-jungle	8	12	19	18	10	23	13	3
Mid-jungle	9	14	6	21	3	23	17	1
Post-jungle	10	17	6	24	1	14	15	0

<sup>a</sup> Nausea, which is not reported here was recorded by 12 subjects during the Sea Phase.

<sup>b</sup> Bolded and coloured text refers to complaints arising during the survival phases.

### 3.6 Correlations

#### 3.6.1 sIgA:Alb, dietary intake, body mass change, health and mood

Dietary intake, body mass change and the POMS factor fatigue-inertia were associated with sIgA:Alb. Multi linear regression analysis, which included these variables, revealed carbohydrate to be predictive of sIgA:Alb (adjusted  $r^2 = 0.50$ ). The average intake of carbohydrate by students over the survival course correlated positively with their average sIgA:Alb measured over the same period.

The mood factors depression-dejection ( $p = 0.01$ ), anger-hostility ( $p < 0.01$ ), and alcohol intake ( $p = 0.01$ ) were negatively correlated with the % change in sIgA:Alb.

Multinomial logistic regression analysis revealed a negative association between sIgA:Alb and health problem score (chi-square = 218,  $df = 180$ ,  $p = 0.03$ ). When the frequency of the various health complaints during each survival phase were compared with the average sIgA:Alb recorded by all subjects during each survival phase, URTI was shown to be negatively correlated with sIgA ( $r = -0.62$ ,  $p = 0.04$ ).

BMI (and body mass) was found to be negatively associated with the health problems score ( $r = 0.40$ ,  $p = 0.02$ ). Health problems were also associated with the STAI score ( $r = 0.39$ ,  $p = 0.02$ ) and the mood factors, tension-anxiety ( $r = 0.43$ ,  $p = 0.01$ ), depression-dejection ( $r = 0.44$ ,  $p = 0.01$ ), anger-hostility ( $r = 0.46$ ,  $p = 0.01$ ), fatigue-inertia ( $r = 0.61$ ,  $p < 0.01$ ), confusion-bewilderment ( $r = 0.48$ ,  $p < 0.01$ ) and total mood disturbance ( $r = 0.49$ ,  $p < 0.01$ ). Table 4 shows some significant correlations between the above factors.

Table 3 Pearson correlation coefficients for significant correlations between sIgA, diet, body mass, health problems and psychological factors

	sIgA:Alb	% change in sIgA:Alb <sup>a</sup>	Average sIgA over 18 days for each subject
Energy (kJ/day)	0.692 (p < 0.001) <sup>b</sup>	-	-
Carbohydrate (g/day)	0.703 (p < 0.001)	-	0.381 (p = 0.023)
Protein (g/day)	0.613 (p < 0.001)	-	-
Fat (f/day)	0.649 (p < 0.001)	-	-
Alcohol/kg body mass (g/day)	-0.317 (p = 0.003)	-0.427 (p = 0.012)	-
BMI	0.642 (p < 0.001)	-	-
Depression-dejection	-	-0.415 (p = 0.014)	-
Anger-hostility	-	-0.532 (p = 0.002)	-
Fatigue-inertia	-0.324 (p = 0.010)	-	-

<sup>a</sup>% Change in sIgA:Alb was calculated by subtracting the baseline value from the final value and dividing by the baseline value and the other variables in this column are the average values over the 18 days for each subject.

<sup>b</sup> Significance of the correlation is presented in parentheses.

## 4. Discussion

Upon arrival at the Survival School the students appeared well nourished and physically fit. It was noted that five of the students were cigarette smokers, although the level of smoking was not monitored during the course. While the general health of the group was good, six of the students arrived with an URTI, and 16 students had experienced an URTI in the previous month. The correlation between the reported incidence of URTI in the previous month and the initial sIgA:Alb level approached significance (p = 0.07).

The first few days of the course which were spent in barracks resulted in positive health effects for the students. For example, the students' sIgA:Alb significantly improved during these first few days (Fig 2) and the number of URTI reported by the subjects declined. On the first day of the course students may have been feeling anxious about the course as evidenced by their scores for the POMS mood factor - tension-anxiety, and anxiety as measured by the STAI [25]. The level of anxiety significantly decreased as students became involved in the course (Fig 3).

While in-barracks, students had access to meals served in the RAAF mess. It is noteworthy that the students' choice of foods provided more than sufficient protein and fat, but for most students inadequate carbohydrate and energy. More than half the students drank in excess of the four standard drinks advised by the National and Medical Research Council as the maximum safe level on the days between the arid and jungle phases [26].

Levels of high physical activity and malnutrition were experienced during the survival phases of the course. The survival phases were characterised by increased reporting of health problems, particularly dehydration, insect bites and exhaustion, and decreased sIgA:Alb. Twelve students experienced nausea during the sea-phase. Significant increases in Anxiety, Fatigue and decreases in Vigour were recorded during the jungle phase.

Despite the less-than-optimal food consumption whilst in barracks, sIgA:Alb levels appeared to recover after the arid phase when the students were in barracks for four days. With better diet, including less alcohol, most students' immune function (as measured by sIgA) may have been able to fully recover and the impact of the arid and jungle phases may not have been so severe.

There are many factors that may have contributed to the changes in students' sIgA:Alb during the survival course. These were shown to have a negative relationship with sIgA:Alb included inadequate intake of energy and macronutrients, consumption of alcohol, loss of body body mass and the negative emotions of anger and depression. Increased fatigue-inertia and decreased vigour-activity were also correlated with declining sIgA:Alb levels.

A multi-regression analysis including all of the variables significantly associated with changes in sIgA:Alb found carbohydrate consumption to most clearly predict the decline in sIgA:Alb. Inadequate consumption of carbohydrate was shown to explain 50% of the observed change in sIgA:Alb. Decreased sIgA:Alb was also associated with increased reporting of URTI and with total health problems. The incidence of URTI steadily increased over the course.

Possible factors that may influence sIgA:Alb levels but were not examined during this study included cigarette smoking, sleep quality and heat strain. It was not practical to monitor students' mood and state of anxiety closely during the survival phases, so changes in the POMS mood factors may have been missed. The same applies to for students' state of anxiety. Dramatic increases in anxiety during the sea and arid phases and even on alternate days during the jungle phase may have been missed. Shortened versions of both the POMS and the STAI are available and with hindsight, it may have been preferable to use these during the present study. It is likely that a lag exists between an acute change in mood and the immune function response. Although the present study indicated an interaction between the negative emotions of anger-hostility and depression-dejection and declining sIgA:Alb levels, further research is required to determine the interaction between mood states and immune function. In particular, more sensitive measures of fatigue, including sleep quality, need to be used in future studies.

The periods spent in-barracks suggest that sIgA:Alb may be a marker of recovery from intense physical activity and food deprivation. It would have been useful (but not practical) to continue monitoring the health, diet, immune function and mood state of

the students for the month following the course. Future studies are needed to assess the value of sIgA:Alb in gauging recovery from stressful military exercises.

## 5. Conclusions

The RAAF Survival School course presented a unique opportunity to evaluate sIgA as a marker of overtraining in physically, mentally, nutritionally and emotionally stressed subjects. The results presented here show that sIgA:Alb is a useful marker of the severity of stresses encountered during the Survival School course. This simple saliva test may be used as a tool for evaluating the effectiveness of interventions such as improved diet and measures designed to reduce fatigue. Further research is needed before sIgA:Alb can be recommended as a marker in the evaluation of positive psychological interventions.

It is recommended that future Survival School courses provide some simple nutritional advice, which emphasises the need to eat well while in barracks and to avoid alcohol. In particular students should be advised to eat more high carbohydrate foods such as breakfast cereals, breads, pasta, rice and starchy vegetables than they would normally eat at home when their level of physical activity is less. Students should be informed about the need to drink plenty of water and avoid getting sunburnt.

## 6. Recommendations

It is recommended that:

- evaluation of sIgA as a marker of overtraining continue.
- Salivary sIgA be used as a marker for the success of nutritional interventions designed to improve immune function.
- consideration be given to the provision of nutrition education in situations where overtraining may occur, such as the Survival School.

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19. ABSTRACT A simple marker of immune function, salivary immunoglobulin A (sIgA), was evaluated as a potential indicator of stress during the RAAF Survival School courses conducted in November 1999. Twenty-seven males and two females with an average age of 26 years participated in the study by keeping a food diary, collecting saliva samples and recording their health problems (daily checklist) and level of anxiety (State-Trait Anxiety Inventory). Participants height and weight changes were also recorded. Dietary restriction, consumption of alcohol, loss of weight and negative emotions were all shown to have a negative effect on sIgA. Salivary IgA was shown to be a useful marker of the severity of stresses encountered during the survival course.					

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