Onderstepoort J. vet Res., 55, 5–9 (1988)

BROILER PULMONARY HYPERTENSION SYNDROME. III. COMMERCIAL BROILER STRAINS DIFFER IN THEIR SUSCEPTIBILITY

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ABSTRACT

HUCHZERMEYER, F. W., DE RUYCK, AN M. C. & VAN ARK, H., 1988. Broiler pulmonary hypertension syndrome. III. Commercial broiler strains differ in their susceptibility. *Onderstepoort Journal of Veterinary Research*, 55, 5–9 (1988)

Broilers of 4 different commercial strains were kept during winter at an altitude of 1 350 m. The birds were slaughtered at weekly intervals and their relative right ventricular mass (pulmonary arterial pressure index = API values) determined. In addition, the incidence of ascites was recorded. Two of the strains suffered high losses from ascites, while the other 2 showed a certain degree of resistance. The former 2 groups had a higher mean API and a greater percentage of high API values in clinically normal birds than the other 2 groups. The group with the lowest incidence of ascites achieved the highest mean live mass at 51 days. There was also a decrease in mean API with age in clinically normal birds. There was no difference in the incidence of ascites between males and females.

INTRODUCTION

A high altitude disease of broiler chickens with right ventricular (RV) hypertrophy, caused by pulmonary hypertension, and ascites has been described by Burton, Besch & Smith (1968), Burton & Smith (1967, 1969), Cueva, Sillau, Valenzuela, Ploog & Cardenas (1970), Cueva, Sillau, Valenzuela & Ploog (1974), Sillau, Cueva & Morales (1980) and Tellez Isaias, Paasch Martinez, Lopez Coello & Esperanza Garcia (1986). Dilation of the right ventricle found in broilers with ascites was suspected of having been caused by a combination of furazolidone and salt toxicity (Orr, Little, Schoenderwoerd & Rehmtulla, 1986). Julian & Wilson (1986) found RV hypertrophy in ascites at sea level, while Huchzermeyer & De Ruyck (1986) found apparently clinically normal broilers with various degrees of RV hypertrophy both at sea level and at altitude.

In cattle the genetic basis for high altitude adaptation was demonstrated by the selection of susceptible and resistant lines (Will, Hicks, Card & Alexander, 1970, 1975; Weir, Tucker, Reeves, Will & Grover, 1974; Grover, Will, Reeves, Weir, McMurtry & Alexander, 1975).

The susceptibility and resistance to altitude disease of the Hilltop and Madison strains, respectively, of Sprague Dawley rats were described by Ou & Smith (1983, 1984) and Ou, Hill & Tenney (1984).

By breeding from survivors at an altitude of 3 093 m it was possible to reduce rearing mortality in White Leghorn chicks from 80 % to 25 % within 6 generations (Smith & Abbot, 1961). Broilers were found to be more susceptible to altitude disease than laying breeds, while village fowls were completely resistant (Pizarro, Salas & Paredes, 1970).

Apparent strain differences in the susceptibility of broilers to ascites have been reported by Julian & Wilson (1984) and Lopez Coello, Odom & Wideman (1985). Sillau & Montalvo (1981) reported on the genetical adaptation to high altitude in broilers after exposure for several generations.

In order to investigate further the possible involvement of genetic factors in the pathogenesis of the pulmonary hypertension syndrome, a trial was carried out to study the susceptibility of commercial broiler strains available at present in South Africa.

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Received 16 November 1987—Editor

MATERIALS AND METHODS

Broiler strains

Fertile eggs were obtained from different broiler breeders representing 4 commercial strains, but originating from parents of different ages and held at different locations and altitudes.

Incubation and rearing

These eggs were set simultaneously, incubated and hatched at one commercial hatchery operating at an altitude of 1 585 m. After hatching, the chicks were transferred to the trial site at the Animal and Dairy Science Research Institute, Irene, at an altitude of 1 350 m. Four hundred and forty chicks of each strain were divided into 4 groups of 110 birds each. The pens housing the groups were randomly distributed in a controlled environment house, to provide equal conditions for all 4 strains, and normal broiler vaccination and feeding programs were followed.

Because of the reported higher incidence of ascites in winter than in summer (Villaseñor & Rivera-Cruz, 1980), the trial was conducted during winter from 16 July–4 September 1984.

Collection and specimens

On Days 9 and 16, 48 birds per strain (12 per pen) were mass-measured and slaughtered and the hearts collected for dissection. On Days 23, 30, 37 and 44 each, a further 24 birds per strain (6 per pen) were sacrificed following the same procedure.

All the birds that died during the trial were examined. Birds that died from ascites were also mass-measured and their hearts collected for dissection. At 51 days the remaining birds were mass-measured and slaughtered and their hearts collected.

Dissection of the hearts

All the hearts were dissected as soon as possible after death or after collection to obviate mass changes through drying. Dissection and determination of the API followed the previously described procedure (Huchzermeyer & De Ruyck, 1986).

Statistical analyses

Frequencies between groups, ages or sexes of birds were compared by means of the chi-squared test, with a test level of $\alpha = 0{,}05$. If multiple comparisons between groups, ages or sexes, were carried out, the 2×2 chi-squared test was used, for which the test level was divided by the number of comparisons made ($\alpha = 0{,}05/x$) (Type I error protection).

Differences between mean API-values were tested by means of an analysis of variance for a completely random design. Multiple comparisons between means were done, using the Bonferroni method. If heterogeneous

TABLE 1 Incidence of ascites in the 4 groups throughout the trial

	Group				Test values
	A	B	C	D	
Ascites birds Normal birds	43 374 a	53 386 a	19 388 b	19 404 b	$\chi^2_{(3)} = 26,285$ P = 0,000008
% ascites	10,3	12,1	4,7	4,5	

Values followed by the same letter are not significantly different

TABLE 2 Influence of age on the incidence of ascites for all 4 groups combined

Age (days)	Cases of ascites	Normal	% ascitic
0–9	0(+2)*	1552	0,13
10–16	5	1363	0,37
17–23	10	1172	0,85
24–30	25(+1)	1079	2,41
31–37	37	983	3,76
38–44	28(+1)	890	3,26
45–51	25	769	3,25
Total	130(+4)		

$\chi^2_{(5)} = 4,013$

P = 0,547546

Regression details of per cent mortality on age

$\hat{Y} = 3,292/[1 + 2998,4 (0,7383)^t]$

$F_{(3,4)} = 44,84$; P = 0,001548; Ajd.$R^2 \times 0,9567$

* Values in brackets indicate unsexed birds

variances (non-normality) were present according to Bartlett's test, the Kruskal-Wallis non-parametric method was used and the multiple comparisons made using Dunn's method.

A logistic curve was fitted to the percentage mortality on the ages of birds. The equation was $\hat{Y} = a/1 + br^t$

where a = saturation value for Y
b = potential growth
r = change in growth rate
and t = age

Simple linear regression calculations were made to describe the increase in mean API values on age and the decrease in the associated standard deviations.

Appropriate computer programmes from the programme library of the Directorate of Biometric and Datametric Services[1], were used for the analyses. The programmes were run on a Burroughs B7900 computer.

RESULTS

The incidence of ascites in the 4 groups throughout the trial is shown in Table 1. It is evident that groups A and B had significantly more ascites than groups C and D.

The influence of age on the incidence of ascites for all 4 groups combined is shown in Table 2. A curvilinear relationship was evident for percentage ascites mortality on age. A logistic (growth) curve fitted better than a Gompertz curve (based on probability). More values over age, however, are preferred to describe the possible decrease of percentage ascites mortality after approximately 40 days. Because of the relatively large deviations of the observed values from the logistic curve, the 95 % confidence intervals are large. To obtain the exact relationship with more age groups may require too big an experiment to justify the costs involved.

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The influence of sex on the incidence of ascites for all groups combined is presented in Table 3. Assuming equal numbers of male and female birds in each age group (in fact 50,92 % males), it is evident, that sex apparently has no influence on the incidence of ascites.

The percentages of incidence of high API values (> 0,29) of clinically normal birds of the 4 groups of all ages are given in Table 4. It is evident that significant differences in the totals exist between groups. Although some overlapping occurs, the general trend is that Groups C and D have fewer birds with high API values than Groups A and B. This trend corresponds closely with the percentage ascites birds in each group and points to a relationship between the incidence of higher API values and the frequency of ascites mortality in a particular group. This is, of course, supported by the mean API values per group for 51-day-old normal birds (Table 5).

The details of API values for the 4 groups at 51 days are demonstrated in Table 5. It was possible to single out this age group because of the large number of birds per group sacrificed at this age (ranging from 184 to 215), which allowed for meaningful statistical analysis.

An indication of bimodality of API-values is given by the fact, that heterogenous variances among the 4 groups existed ($\chi^2_{(3)} = 30,172$; P = 0,000 000). The results of Dunn's multiple comparison test indicate that Group B birds had significantly higher API values than Groups C and D. Again evidence of a relation between API values and ascites frequencies is present. Compare mean ranks with ascites frequencies in Table 1.

The distribution of percentages of API values of the 4 groups at 51 days is also presented in Fig. 1, together with that of the ascites cases of all groups and ages combined. Higher peaks in the ranges from 0,18–0,22 characterize the groups with lower incidence of ascites (C and D), whereas Groups A and B have lower peaks in this range. The peak of the ascites cases ranges from 0,38–0,46 with a total range from 0,30–0,64.

A comparison of the mean API values of clinically normal birds over age for all groups combined is given in Table 6. This shows a significant (linear) decrease of mean API with age, while the standard deviation increases significantly with age, an indication of increased variability of the API with age.

A further comparison of mean live mass at 51 days, percentage incidence of ascites, percentage of high API values of clinically normal birds at 51 days and mean API values of clinically normal birds at 51 days of the 4 groups is presented in Table 7.

DISCUSSION

Mortality from ascites reportedly occurs as early as 1 week of age (Buys & Barnes, 1981; Lopez Colleo *et al.*, 1985). In the present trial the 1st cases of ascites were recorded on Day 6. Mortality subsequently increased until Day 37 and then gradually declined. It is impossible to say, however, whether this decline after Day 37 is a true reflection of the field situation or an anomaly due to

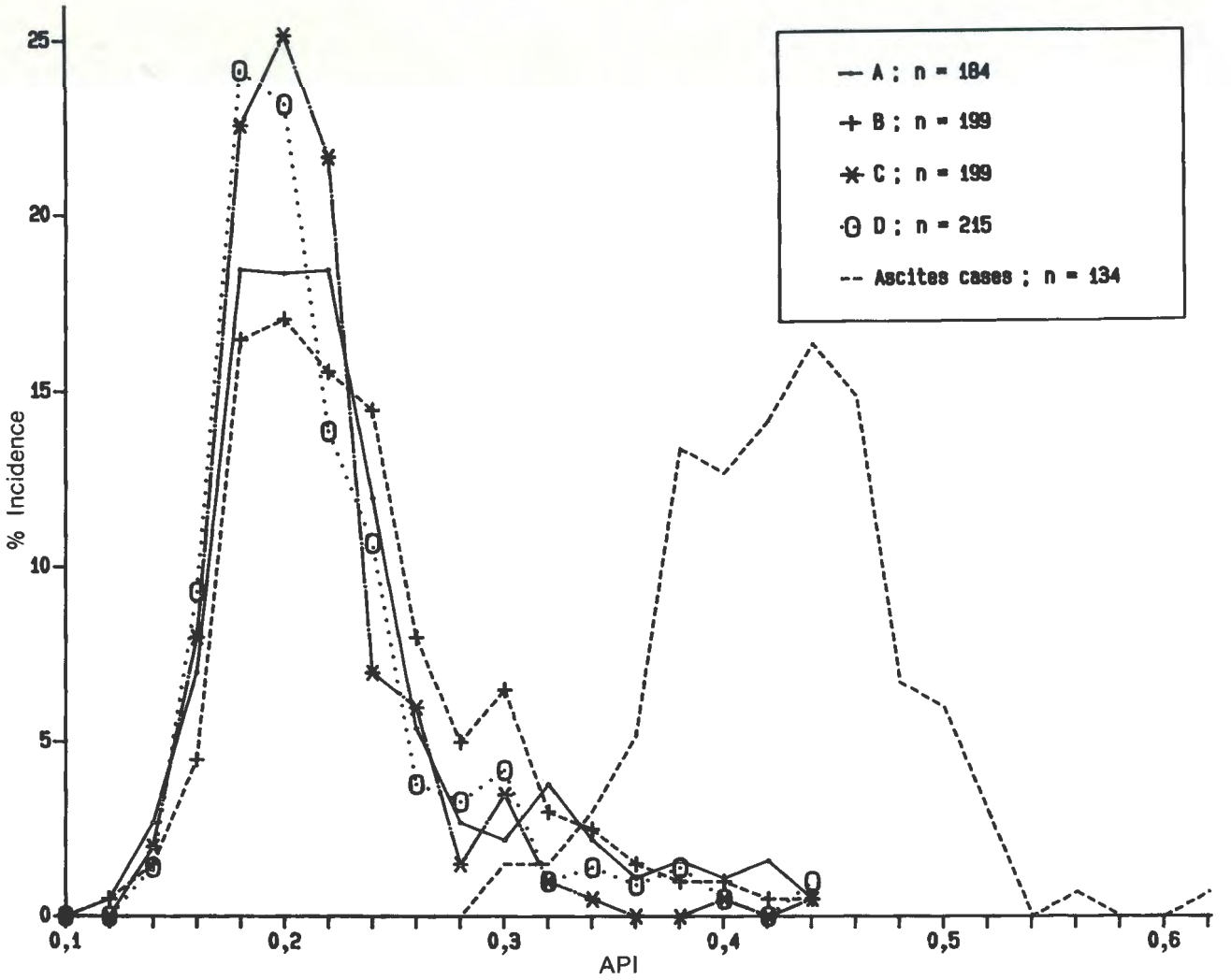

FIG. 1 Frequency distribution in per cent of the API values of clinically normal broilers of the 4 different groups at 51 days (A: n = 184, B: n = 199, C: n = 199, D: n = 215) and of ascites cases of all ages and groups combined (n = 134)

TABLE 3 Differences in incidence of ascites between sexes

Age	Male		Female		$\chi^2_{(1)}$	Probability
	Ascitic	Normal	Ascitic	Normal		
0–9	0	120	0	69	—	—
10–16	1	99	4	92	0,9072	0,340850
17–23	5	40	5	53	0,0077	0,929924
24–30	13	62	12	84	0,8523	0,355905
31–37	17	41	20	52	0,00001	0,997600
38–44	14	49	14	45	0,0003	0,985913
45–51	16	380	9	417	1,9738	0,160045
Total	66	791	64	762	0,0755	0,783457

TABLE 4 Percentages of incidence of high API values in clinically normal birds of all ages by group

Group	A	B	C	D
API % >0,29	17,91a	18,39a	8,50b	12,13ab

$\chi^2_{(3)} = 21,589$; P = 0,000079

Values followed by the same letter are not significantly different

the relatively small numbers involved. Conversely, mean API values of clinically normal birds decreased with age (Table 6).

Apparent differences of commercial broiler strains in their susceptibility to ascites have been observed in the past (Julian & Wilson, 1984; Lopez Coello *et al.*, 1985). Two of the strains, A and B, used in this study suffered high losses from ascites, while the other 2 strains, C and D, showed a certain degree of resistance.

There were practically no differences in the averages of API values of the ascites cases from the different groups. Their API values ranged from 0,30–0,64 (Fig. 1). This range and the dip in the incidence curves of API values of clinically normal birds killed on Day 51 at 0,28 were taken as the cut-off point between low (<0,30) and high (>0,29) API values (Table 4). It is interesting to note that the groups with a higher incidence of high API values also suffered higher ascites mortality. There was a wide range to the right of the normal distribution curve

TABLE 5 Details of API values for the 4 groups at 51 days

	Group			
	A	B	C	D
Mean API	0,232	0,238	0,216	0,222
Range	0,13–0,44	0,13–0,44	0,14–0,45	0,14–0,47
n	184	199	199	215
S.D.	0,0618	0,0572	0,0419	0,0537
Mean rank	412,57 ab	451,24 b	361,40 a	373,83 a
Kruska-Wallis $\chi^2(_3)$ = 18,891; P = 0,000288				

Values followed by the same letter are not significantly different

TABLE 6 Comparison of mean API-values of clinically normal birds over age for all groups combined

Age (days)	$\bar{x}$ API	n	SD
9	0,261	184	0,038
16	0,269	191	0,042
23	0,258	93	0,050
30	0,250	96	0,047
37	0,244	93	0,046
44	0,231	94	0,058
51	0,227	797	0,055
Regression equation	$\hat{Y}$ = 0,27769–0,00098x		$\hat{Y}$ = 0,03591 + 0,00040X
F;P	46,64; 0,00103		15,67; 0,01075

TABLE 7 Comparison of mean live mass at 51 days, percentage incidence of ascites (Table 1), percentage of high API values of clinically normal birds at 51 days (Table 4) and mean API values of clinically normal birds at 51 days (Table 5) of the 4 groups

Groups	$\bar{x}$ live mass	% ascites	% API >0,29	$\bar{x}$ API
A	2327,3	10,3	17,91	0,232
B	2328,7	12,1	18,39	0,238
C	2329,8	4,7	8,50	0,216
D	2478,6	4,5	12,13	0,222

(Fig. 1) and possible bimodality of distribution of the API values. Consequently, their separation into low and high offered an alternative way to present the results.

In spite of this wide range of distribution there were still significant differences between some of the average API values of the different groups coinciding with the ascites mortality pattern. Consequently, the determination and analysis of API values of all birds in a group can be regarded as an important tool in the investigation of broiler ascites.

The finding in this trial that sex had no influence on the incidence of ascites (Table 3), is in contrast to the results reported by Velasco (1973), who in a trial involving 6 331 broilers at an altitude of 3 320 m found an incidence of ascites of 12,4 % in males and 3,7 % in females. At present there is no explanation for this discrepancy.

It is interesting to observe that the group with the highest mean live mass at 51 days showed the lowest incidence of ascites cases (Table 7).

Differences in the susceptibility to ascites in different commercial broiler strains as seen in this study can only be explained as genetic. The practical solution of the broiler ascites problem therefore rests with the selection for resistance to ascites in broiler breeding stock.

ACKNOWLEDGEMENTS

The authors wish to thank the Director, Animal and Dairy Science Research Institute, Irene, for allowing us the use of the facilities for this trial and, in particular, Messrs J. J. Gregorowski and M. J. Runchiman for their practical assistance. They also wish to express their gratitude to Festive Farms, Farm Fare, Gayman's Poultry, Gambier Poultry, Earlybird Farm and Ora et Labora for the donation of hatching eggs for this trial, as well as to the De Ruyck Hatcheries for incubating and hatching the eggs.

REFERENCES

BURTON, R. R., BESCH, E. L. & SMITH, A. H., 1968. Effect of chronic hypoxia on the pulmonary arterial bloodpressure of the chicken. *American Journal of Physiology*, 214, 1438–1442.

BURTON, R. R. & SMITH, A. H., 1967. Effect of polycythemia and chronic hypoxia on heart mass in the chicken. *Journal of Applied Physiology*, 22, 782–785.

BUYS, S. B. & BARNES, P., 1981. Ascites in broilers. *Veterinary Record*, 108, 266.

CUEVA, S., SILLAU, H., VALENZUELA, ANTONIETA, PLOOG, H. & CARDENAS, W., 1970. Hipertension pulmonar, hipertrofia cardiaca derecha y mal de altura en pollos parilleros. *Instituto Veterinario de Investigaciones Tropicales y de Altura Cuarto Boletin Extraordinario*, 142–146.

CUEVA, S., SILLAU, H., VALENZUELA, ANTONIETA & PLOOG, H., 1974. High altitude induced pulmonary hypertension and right heart failure in broiler chickens. *Research in Veterinary Science*, 16, 370–374.

GROVER, R. F. WILL, D. H., REEVES, J. T., WEIR, E. K., MCMURTRY, I. F. & ALEXANDER, A. F., 1975. Genetic transmission of susceptibility to hypoxic pulmonary hypertension. *Progress in Respiration Research*, 9, 112–117.

HUCHZERMEYER, F. W. & DE RUYCK, AN M. C., 1986. Pulmonary hypertension syndrome associated with ascites in broilers. *Veterinary Record*, 119, 94.

JULIAN, R. J. & WILSON, J. B., 1984. Ascites in broiler chickens caused by high levels of carbon monoxide. *Proceedings of 56th Annual Meeting of the Northeastern Conference of Avian Diseases, University Park*, June 1984.

JULIAN, R. J. & WILSON, J. B., 1986. Right ventricular failure as cause of ascites in broiler and roaster chickens. *Proceedings of 4th International Symposium of Veterinary Laboratory Diagnosticians, Amsterdam*, June 1986.

LOPEZ COELLO, C., ODOM, T. W. & WIDEMAN, R. F., 1985. Ascites major cause of mortality in broilers. *Poultry Digest*, 44, 284–286 & 288.

ORR, J. P., LITTLE, K. S., SCHOENDERWOERD, M. & REHMTULLA, A. J., 1986. Ascites in broiler chickens. *Canadian Veterinary Journal*, 27, 99–100.

OU, L. C. & SMITH, R. P., 1983. Probable strain differences of rats in susceptibilities and cardiopulmonary responses to chronic hypoxia. *Respiration Physiology*, 53, 367–377.

OU, L. C., HILL, N. S. & TENNEY, S. M., 1984. Ventilatory responses in blood gases in susceptible and resistant rats to high altitude. *Respiration Physiology*, 58, 161–170.

OU, L. C. & SMITH, R. P., 1984. Strain and sex differences in the cardio-pulmonary adaptation of rats to high altitude. *Proceedings of the Society for Experimental Biology and Medicine*, 177, 308–311.

PIZARRO, B., SALAS, A. & PAREDES, J., 1970. Mal de altura en aves. *Instituto Veterinario de Investigaciones Tropicales y de Altura, Cuarto Boletin Extraordinario*, 147–151.

SILLAU, A. H., CUEVA, S. & MORALES, P., 1980. Pulmonary arterial hypertension in male and female chicks at 3 300 m. *Pflügers Archiv*, 386, 269–275.

SILLAU, A. H. & MONTALVO, C., 1981. Pulmonary hypertension and the smooth muscle of the pulmonary arterioles in chickens at high altitude. *Comparative Biochemistry and Physiology*, A71, 125–130.

SMITH, A. H. & ABBOT, h0006.5w0006.5RSULA K., 1961. Adaptation fowl to high altitude. *Poultry Science*, 40, 1459.

TELLEZ ISAIAS, Q., PAASCH MARTINEZ, L., LOPEZ COELLO, C. & ESPERANZA GARCIA C., 1986. Hipertension pulmonar en la patogenia del sindrome ascitico. *Proceedings of the 35th Western Poultry Disease Conference*, Puerto Vallarta, 192–193.

VELASCO, T., 1973. Correlation fenotipica entre mortalidad por mal de altura y peso a la quinta semana en parrilleros. *Revista de Investigaciones pecuarias (Instituto Veterinario de Investigaciones Tropicales y de Altura) Universidad Nacional de San Marcos*, 2, 43–48.

VILLASEÑOR, J. & RIVERA-CRUZ, E., 1980. Que esta pasando con la ascitis? *Proceedings of the 29th Western Poultry Disease Conference*, 89–92.

WEIR, E. K., TUCKER, A., REEVES, J. T., WILL, D. H. & GROVER, R. F., 1974. The genetic factor influencing pulmonary hypertension in cattle at high altitude. *Cardiovascular Research*, 8, 745–749.

WILL, D. H., HICKS, J. L., CARD, C. S. & ALEXANDER, A. F., 1970. Genetic basis for high altitude adaptation in cattle. *Federation Proceedings, Federation of American Societies of Experimental Biology*, 29, 591.

WILL, D. H., HICKS, J. L., CARD, C. S. & ALEXANDER, A. F., 1975. Inherited susceptibility of cattle to high-altitude pulmonary hypertension. *Journal of Applied Physiology*, 38, 491–494.