degradation rates by peptidases or to differences in affinity for α-MSH or deacetylated α-MSH receptors. Recent studies demonstrate that brain peptidases degrade deacetylated α-MSH about ten times as fast as they degrade α-MSH (*6*). More rapid degradation of deacetylated α-MSH cannot, however, totally explain the differences in behavioral activity of the two peptides because deacetylated α-MSH is actually more potent than α-MSH in a number of behavioral actions. One behavioral action in which deacetylated α-MSH is more potent than α-MSH is in the ability to block opiate analgesia and opiate receptor binding. Deacetylated forms of α-MSH or structurally related ACTH analogs consistently block opiate-induced analgesia and opiate or β-endorphin receptor binding, whereas the *N*-acetylated α-MSH is devoid of activity (*11*). It is clear that in this case receptor affinity and not peptidase sensitivity is likely to be responsible for the differences in bioactivity and that the *N*-acetyl group may be an important determinant for receptor interaction. The data, therefore, indicate that there are different structural requirements for different behavioral actions of α-MSH, a finding that has been demonstrated previously (*12*), and that interactions with these different postsynaptic receptors could be regulated by acetylation reactions in the presynaptic opiomelanotropinergic neuron. It is interesting that β-endorphin also occurs in both acetylated and deacetylated forms in the opiomelanotropinergic neuron and that acetylation of the β-endorphin eliminates its potential to bind to the opiate receptor and elicit analgesia (*3*). The acetylation of both α-MSH and β-endorphin appears to result from enzymatic mechanisms, and recent results indicate that different enzymes acetylate each of these peptides (*13*). Furthermore, α-MSH acetylating enzymatic activity appears to be induced by physiological manipulations which induce α-MSH synthesis (*13*). Differential regulation of the two acetylating enzymes could alter the ratios of deacetylated and acetylated forms of α-MSH and β-endorphin and by doing so change both the composition of the secretory output of the opiomelanotropinergic neuron and the resulting postsynaptic actions. Such molecular regulatory processes are considerably more complex than the mechanism proposed by the classical model of the neurosecretory cell which releases one neurotransmitter or hormone. It is clear, however, that independent processing of individual neurotransmitters or hormones released from one multiple transmitter secretory cell adds another dimension of complexity and flexibility to intercellular communication. A question of importance is whether cells secreting multiple chemical signals are the exception or rule.

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Excretion of β-Phenethylamine Is Elevated in Humans After Profound Stress

Abstract. *The urinary excretion rate of the endogenous, amphetamine-like substance β-phenethylamine was markedly elevated in human subjects in association with an initial parachuting experience. The increases were delayed in most subjects and were not correlated with changes in urinary pH or creatinine excretion. The data suggest a stress-related role for β-phenethylamine.*

β-Phenethylamine (PEA), an endogenous amine that resembles amphetamine both structurally and pharmacologically (*1*), has been implicated as an etiological factor in paranoid schizophrenia. Patients with this disorder excrete large amounts of PEA in their urine (*2*). Stress may also play a role in paranoid schizophrenia, since in some patients it precipitates psychotic episodes (*3*). Amphetamines can produce a paranoid state (*4*), and stress can reinstate this psychosis in individuals who then are abstinent (*5*). It seems, therefore, that stress may play a role in changes in PEA excretion observed in paranoid schizophrenics. However, there has been no evidence that stress can alter PEA disposition in any species. We now report an elevation in the urinary excretion of PEA in humans following a parachute jump.

Our subjects were male and female college students 18 to 28 years of age. They were admonished to refrain from ingesting alcohol or other drugs during urine sampling periods. Urine was collected from each subject during a 24-hour control period (2100 to 2100 hours) 7 to 21 days before the jump and during the same 24-hour period encompassing the jump. In some subjects urine was collected for an additional 18 hours after the jump. Collected urine was stored at −70°C until being assayed for PEA by gas chromatography–mass spectroscopy (*6*). The urine from each micturition was assayed separately.

Stress during the parachuting experi-

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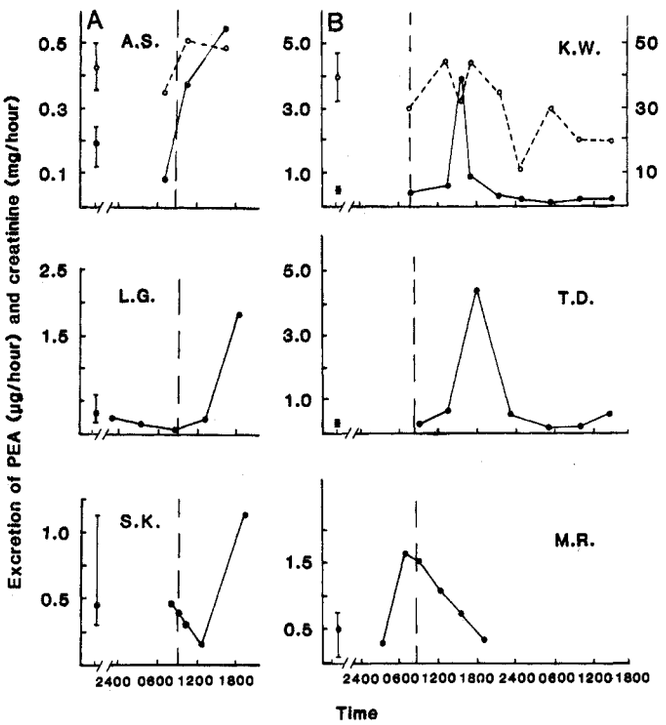

Fig. 1. Urinary excretion of PEA (●) and creatinine (○) in individual subjects before and after parachuting. Urinary excretion rate was calculated by dividing the PEA content in urine from a particular micturition by the time in hours since the previous micturition. Points are graphed at the temporal midpoint between each pair of micturitions. Symbols on the extreme left of each graph indicate mean excretion rates for four to seven micturitions (brackets on solid circles indicate the range and brackets on open circles indicate standard errors) for a 24-hour control period 7 to 21 days before the jump. Vertical dashed lines indicate the time of the jump.

ence was defined as an elevation in heart rate (*7*). Since an individual's perception of stress and his or her subsequent physiological reaction may be altered by denial (*8*), the subjects were administered the Minnesota Multiphasic Personality Inventory denial scale (*8*) before the jump.

Figure 1A presents the PEA excretion data for three of six subjects (two males, A.S. and L.G., and one female, S.K.). The rate of urinary excretion of PEA was markedly elevated in each subject after the jump; in L.G. and S.K. the increase did not occur until several hours after the jump. The post-jump values exceeded the control period means for A.S., L.G., and S.K. by factors of 2.9, 5.4, and 2.6, respectively. Similarly, in the remaining three subjects (two females, R.A. and D.G., and one male, J.H.), mean PEA excretion rates on control days were 0.43 (range, 0.19 to 0.76), 0.33 (0.21 to 0.48), and 0.36 (0.19 to 0.52) μg/hour, whereas the maximum rates on the jump day were 1.66, 0.86, and 0.96 μg/hour. Thus, excretion rates on the jump day were approximately three to four times higher than the control rates. In J.H. and D.G. these elevations occurred no earlier than 5 hours following the jump; the increase in R.A., however, occurred 1 hour before the jump.

In view of the apparent delay in the augmentation of PEA excretion, we added three more subjects to the study and collected their urine for longer periods. PEA excretion rates after the jump were elevated by factors of 8.6, 15.8, and 3.0 in K.W., T.D., and M.R., respectively (Fig. 1B). In K.W. and T.D. the increases were again delayed, occurring no earlier than 5.5 hours after the experience. Following the large increases the rates rapidly declined to low, stable values. In M.R., however, there was no delay. In fact, as with R.A., the maximum increase occurred before the jump. There was no statistically significant correlation between scores on the denial test and maximal increases in PEA excretion.

In spite of what may be considered a small sample size, these data indicate that the elevation (and the delay) in PEA excretion is reproducible under the conditions of the experiment. An important exception may have occurred in S.K. In this subject the maximum elevation in PEA was only slightly greater than the maximum that occurred during the control period. While this subject may not have demonstrated a jump-associated effect, it is appropriate to note that the control day or the day before it was not necessarily without stress. Consequently, the relatively large range in S.K.'s control PEA excretion rates, like the large range in her rates during the jump day, may reflect stress-related events.

The site or sites from which efflux of PEA occurs and the mechanisms responsible for these effects are unknown. PEA is a decarboxylation product of the amino acid phenylalanine and is metabolized primarily by monoamine oxidase B (MAO-B) to form phenylacetic acid (*9*). The excretion of free PEA per se in humans is apparently unaffected by the ingestion of PEA-containing foods and is independent of normal diurnal variations in urinary *p*H (*2*, *10*). The elevation in PEA excretion following stress may result in part from an increase in plasma phenylalanine or phenethylamine or from a decrease in MAO-B activity. Stress in animals, at least, has been shown to produce a decrease in MAO activity in the hypothalamus (*11*), a brain region with a higher PEA content than most other central nervous system tissues (*12*). Although correlations among stress, PEA, and paranoid schizophrenia are hypothetical, it is of interest that human platelet MAO activity is reduced in paranoid schizophrenics (*13*).

The delay in excess excretion of PEA until several hours after parachuting is even more difficult to account for. Because of the stress, changes in blood flow to the kidney or in other intrarenal processes could occur such that urinary excretion of many substances, including PEA, is delayed. Substances that normally are transferred from the plasma to urine might therefore accumulate in blood until the effects of stress dissipate. The return to normal kidney function would lead to elevated excretion of the accumulated substances. However, data on the effects of parachuting on the rate of creatinine excretion (Fig. 1) mitigate against an alteration in kidney function per se or a renally mediated accumulation of PEA in plasma. Creatinine is an end product of muscle metabolism (*14*). It enters the plasma at a relatively constant rate and is secreted and reabsorbed in the kidneys to such a small extent that its excretion is often used clinically as an indirect measure of kidney function in general and glomerular filtration in particular (*15*). Comparable rates of PEA and creatinine excretion, then, would support the above contentions. However, our data indicate that the excretion rates of PEA are not directly correlated with those of creatinine.

Another possibility is that the delay simply represents the normal time required for plasma PEA to be transferred to the urine. While we have no data on this, it should be noted that some of the effects of stress may be similar to those of the PEA analog amphetamine (*3*) and that amphetamine appears in human urine within minutes, even when administered orally (*16*). Since the chemical and pharmacological characteristics of PEA and amphetamine are similar (*1*), elevated excretion of PEA several hours after the stressful experience is unlikely

to represent a normal plasma-urine transfer time.

The most likely explanation for the delay is that some stress-related but delayed process occurred in several of our subjects some time after the parachuting experience. Although the process is unknown, the data suggest that stress can increase the amount of circulating PEA.

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6. Urine samples were collected and immediately frozen (−20°C) for transport to long-term storage (−70°C). In the analysis, 1-ml portions of each sample were mixed with 0.5 ml of 0.5*M* phosphate buffer. An internal standard (deuterated PEA) was added and the mixture was extracted into ether, then back-extracted into 0.1*N* HCl, which was blown off with argon. The PEA in the residue was derivatized with 5 percent pentafluoropropionyl imidazole in ethyl acetate (200 μl) by heating at 80°C for 10 minutes. The ethyl acetate and excess derivatizing agent were blown off with argon. The derivatized PEA was reconstituted with 50 μl of ethyl acetate. Gas chromatography was carried out on a 30 m by 0.32 mm (inside diameter) fused-silica capillary column. A quadrupole gas chromatograph–mass spectrometer (Ribermag R10-10) focused on ions of 104 and 107 amu to detect PEA and [$^2H_4$]PEA, respectively (*12*).
7. Heart rate and electrocardiogram (EKG) were recorded with a Holter monitor (Dynagram 5000 portable electrocardiograph), which was attached to each subject for continuous recording. The monitor was attached (three leads, modified V_4-V_5 recording) 1 hour before the jump, remained attached during the jump, and was removed 1 hour after the jump for a total of 2 hours of recordings. The EKG was recorded on cassette tape for later analysis by J. Schroeder, Department of Cardiology, Stanford University Medical Center. One hour before the jump, mean heart rate was 83 ± 5 beats per minute. Ten minutes after the jump, heart rate was maximally elevated (141 ± 7 beats per minute). One hour after the jump, heart rate had returned to 83 ± 5 beats per minute. No clear EKG abnormalities were noted.
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Mass Mortality of Harbor Seals: Pneumonia Associated with Influenza A Virus

Abstract. *More than 400 harbor seals, most of them immature, died along the New England coast between December 1979 and October 1980 of acute pneumonia associated with influenza virus, A/Seal/Mass/1/80 (H7N7). The virus has avian characteristics, replicates principally in mammals, and causes mild respiratory disease in experimentally infected seals. Concurrent infection with a previously undescribed mycoplasma or adverse environmental conditions may have triggered the epizootic. The similarities between this epizootic and other seal mortalities in the past suggest that these events may be linked by common biological and environmental factors.*

There have been three known incidents of unexplained large-scale mortalities which we think may be attributed to pneumonia in seals (*1–3*). While monitoring marine mammal strandings along the New England coast, we studied such an event in harbor seals (*Phoca vitulina*) that began in December 1979. The seals died of an acute and devastating pneumonia that we associate with a previously undescribed influenza virus and a mycoplasma. We suggest that the influenza virus, prompted by environmental conditions and the presence of the mycoplasma, may have been responsible for similar occurrences in the past (*1–3*).

The disease first appeared within a tight grouping of seals on Billingsgate Shoal, Cape Cod. It spread rapidly, killing at least 130 animals within a month. Thereafter, the mortality rate declined as the seals dispersed northward along the New England coast (Fig. 1). By October 1980, when the outbreak had run its course, at least 445 harbor seals had died. Ninety percent of the dead seals were under 3 years of age; males and females were equally affected.

The clinical signs of the disease were dramatic. Obviously well-nourished seals appeared weak, moving feebly and without coordination, and exhibited respiratory distress. Occasionally, one would thrash its head to clear a frothy white or bloody discharge from the airways. Otherwise the seals remained still, except for quivering of their muscles. Characteristically, their necks were swollen because of entrapped air that escaped from the lungs, through the thoracic inlet, and into the muscles and fascia of the neck and back. This caused some seals to be so buoyant that they drifted with the wind and tide. We estimate that the disease took 3 days or less to develop. The most acutely affected seals died within hours after feeding normally.

On postmortem examination of animals that died of the disease, we found pneumonia characterized by necrotizing bronchitis and bronchiolitis, and hemorrhagic alveolitis. We isolated an assortment of aerobic and anaerobic bacteria from the lungs (*4*) but could not implicate any as the cause of the disease. However, a mycoplasma was isolated from the lungs of all of the eight animals from which culture was attempted. The organism hydrolyzed arginine; it was also nonglycolytic, sensitive to digitonin, and resistant to penicillin, penicillin derivatives, and erythromycin. Thus far, the organism has been tested with antiserums to 55 known species and strains of mycoplasma and has shown no fluorescent antibody staining reaction or growth inhibition. Although mycoplasmas are not normally considered to be primary agents of such severe and widespread disease, we are nevertheless investigating the pathogenicity of this unidentified organism in seals.

The pathologic findings and the natural history of the epizootic pointed to a viral etiology. This assumption was strengthened by the isolation, in three independent laboratories, of an influenza A virus (A/Seal/Mass/1/80) from the lungs and brains of the dead seals (*5*,

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