ASPARTAME: REVIEW OF RECENT EXPERIMENTAL AND OBSERVATIONAL DATA*

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(Received October 23rd, 1987)

SUMMARY

In this report the neurotoxicity of aspartame and its constituent amino acids aspartic acid and phenylalanine is reviewed. The adverse reactions ascribed to the consumption of aspartame-containing products, as reported in the U.S.A., are discussed and placed in perspective with the results of recent behavioural studies in humans and animals. The issue of common intake levels associated with proposed uses of aspartame is addressed.

In brief, the following conclusions can be drawn: When aspartame is consumed at levels within the ADI-limit of 40 mg/kg body wt, there is no significant risk for an aspartate-induced neurotoxic effect in the brain. When aspartame is consumed at levels within the ADI-limit by normal subjects or persons heterozygous for phenylketonuria (PKU) the resultant plasma phenylalanine concentrations are practically always within the normal post-prandial range; elevation to plasma concentrations commonly associated with adverse effects has not been observed. Persons suffering from phenylketonuria (PKU-homozygotes) on a phenylalanine-restricted diet should avoid consumption of aspartame. PKU-homozygotes on the (less strict) phenylalanine-liberalised diet should be made aware of the phenylalanine content of aspartame. In the available behavioural studies in humans with acute dosing, no adverse effects were observed. Long-term studies on behaviour and cognitive function in (sensitive) humans are lacking. Analyses

*Project No. 86/678606/005 and 86/678608/010.
Abbreviations: ADI, acceptable daily intake; DKP, the diketopiperazine of aspartame; FDA, food and drug administration; 5-HIAA, 5-hydroxyindole acetic acid; HVA, homovanillic acid; IGTC, International Glutamate Technical Committee; JECFA, Joint Expert Committee on Food Additives; LNAA, large neutral amino acids; MRCA Information Inc., Marketing Research Corporation of America Information Inc., PKA, phenylketonuria.
of adverse reaction reports made by consumers in the U.S.A. have not yielded a specific constellation of symptoms clearly related to aspartame that would suggest a widespread public health hazard associated with aspartame use. Focussed clinical studies are now being carried out in the U.S.A.; the results should provide additional evidence concerning the interpretation of the reports on adverse reactions ascribed to aspartame. In the regulation of admitted uses for aspartame the possibility of intake levels exceeding the ADI-limit in some groups of consumers should be a point of attention.

**Key words:** Aspartame; Toxicity/tolerance studies; Hazards; Humans; Animals; Behavioural studies

1. INTRODUCTION

Aspartame (L-aspartyl-L-phenylalanine methyl ester) is an artificial sweetener, composed of substances normally found in the diet and the body, i.e. the amino acids aspartic acid and phenylalanine and the alcohol methanol.

The sweetness of aspartame relative to sucrose is inversely related to the concentration of sucrose. At 3% sucrose, aspartame has 215 times the sweetness of sucrose, but it has only 133 times the sweetness potency at 10% sucrose concentration [1]. The present report will briefly review the results obtained in toxicity/tolerance studies performed with aspartame and will subsequently address the possible hazards associated with the 3 constituents and metabolites of aspartame: methanol, aspartic acid and phenylalanine. In addition, the adverse reactions associated with the consumption of aspartame-containing products, which have been reported in the U.S.A., are discussed and compared with the (mostly) recent results of behavioural studies in humans and animals. The last section deals with the question of common human intake levels associated with admitted or proposed uses of aspartame in the light of the current state of knowledge.

The possible implications of the decomposition of aspartame — at lower pH-levels (e.g. in soft drinks) such decomposition occurs to a significant degree when storage is prolonged — are not reviewed in this report. Neither is this the case for the toxicological data on the aspartame decomposition products in food DKP (diketopiperazine of aspartame) and β-aspartame.

2. STANDARD TOXICITY STUDIES IN ANIMALS AND TOLERANCE STUDIES IN HUMANS

The toxicity and metabolism of aspartame have been studied in many experiments. A comprehensive review of standard animal toxicity studies is given by the Joint Expert Committee on Food Additives (JECFA, [2]). The animal experiments included several chronic toxicity studies in rats. The JECFA estimated the level causing no effect in the rat to be 4 g/kg body wt/
day and proposed an ADI (safety factor 100) of 40 mg/kg body wt/day for humans.

Several tolerance studies were performed in humans of various population types (normal adults, obese adults, normal children, persons heterozygotic for phenylketonuria). Both acute large dose loading and prolonged reasonable dietary dosing up to 90 days were carried out. No indications for significant toxicological problems were found within the limitations of the studies [2].

3. POSSIBLE EFFECTS OF ASPARTAME METABOLITES

After oral uptake aspartame is rapidly and extensively metabolised to its constituent amino acids (aspartate and phenylalanine) and methanol. The possible effects of the 3 metabolites after aspartame consumption will be discussed in the following paragraphs.

3.1. The methanol moiety

The metabolism and toxicity of methanol is reviewed by Tephly and McMartin [3].

Stegink [4] and Pardridge [5] address the issue of a possible toxic reaction caused by methanol when aspartame is consumed. Both authors conclude that no hazard exists on this point. The central consideration to support this conclusion is that use of aspartame in soft drinks provides theoretical methanol concentrations not exceeding the methanol levels normally present in fruit juices. Another line of reasoning can support this conclusion. A bolus intake of aspartame of 40 mg/kg body wt (equal to the ADI) implicates a methanol intake of 4.4 mg/kg body wt; this ingestion level is well below the methanol exposure levels required to achieve toxic reactions in humans — (as indicated by Pardridge [5] such levels are > 1 g/kg body wt).

3.2 The aspartate moiety

Aspartic acid has been shown to induce neuronal necrosis in the arcuate nucleus of the hypothalamus in the brain of the neonatal mouse after a single application [6]. Just the same lesions are produced by single doses of glutamate, a widely used food additive. Glutamate and aspartate are additive with regard to this effect in neonatal mice [7]. The crucial factor determining the occurrence of these lesions is the sum-concentration of glutamate and aspartate reached in blood plasma and brain. The transport of glutamate and aspartate from the blood into the brain depends on the presence of a blood-brain barrier. Normally the blood-brain barrier precludes influx of glutamate and aspartate into the brain but there are several circumventricular organs of the brain that lack such a barrier. The neuronal necrosis that occurs when large doses of glutamate or aspartate are administered is typically present in areas contiguous with the circumventricular organs, e.g. the arcuate nucleus or the pre-optic area. At
very high plasma concentrations of glutamate/aspartate the blood-brain barriers breaks down and even non-circumventricular organs may be affected. Young animals are more susceptible to glutamate-aspartate-induced neuronal damage primarily because the blood-brain barrier does not function completely yet [8] and because the metabolism of glutamate (and aspartate) is slower than in adults, on account of which higher plasma concentrations are reached [9]. A great number of studies, performed in mice, rats, monkeys and humans, has focussed on measuring concentrations of glutamate and aspartate in blood plasma after oral administration of these amino acids under various conditions. Several factors were found to have considerable influence on the plasma concentrations, the most important of which is the vehicle used. When glutamate (or aspartate) is consumed as part of a high-protein meal (or an amino acid mixture) the concentrations reached in plasma are markedly lower in comparison to consumption with water as vehicle [10]. Simultaneous administration of carbohydrates also reduces the glutamate/aspartate levels reached in plasma [4,11]. Another point of note is the considerable intraspecies variation in glutamate (and aspartate) metabolism which has been demonstrated in mice, monkeys and humans [9,10,12].

The potency of glutamate and aspartate to induce the characteristic hypothalamic lesions in neonatal animals after single application, has been investigated in a large number of studies (review by Garattini [13] and IGTC [14]). In neonatal mice the lesions were consistently found in all studies both with glutamate and aspartate. For monosodium glutamate 500 mg/kg body wt is the lowest effective oral dose, whereas at 250 mg/kg body wt there is no effect [13,14]. For aspartate Applebaum et al. [6] found 650 mg/kg body wt as lowest effective dose. Applebaum et al. [6] conclude that in neonatal mice plasma concentrations of glutamate and aspartate of 600—1000 μmol/l (sum-concentration glutamate + aspartate) must be reached before neuronal necrosis in the hypothalamus occurs. In neonatal rats the lowest effective doses for monosodium glutamate and potassium aspartate were 2000 mg/kg body wt and 440 mg/kg body wt, respectively [13,15]. In neonatal monkeys original positive findings for monosodium glutamate made by Olney et al. [16] could not be reproduced by 4 other research groups [17—22]. In one of the negative studies plasma glutamate and aspartate concentrations in the neonatal monkeys were measured and at sum-concentrations (glutamate + aspartate) as high as 4400 μmol/l no hypothalamic lesions were found [12]. Reynolds et al. [23] were also unable to produce the characteristic hypothalamic lesion in monkey fetuses via directly injecting sodium glutamate in the fetal circulation. Reynolds et al. [24], Pardridge [8] and Olney (personal communication-letter to Dr. C.A. van der Heijden from J.W. Olney, August 20th, 1986) give possible reasons for the discrepancy in the results in neonatal monkeys; a generally accepted explanation however, seems as yet not to have been found. In conclusion, the available data do not provide convincing evidence for the characteristic hypothalamic lesions, observed in rodents, to occur also in primates.

Aspartame has been tested in single-dose studies in neonatal mice and
neonatal monkeys. In mice aspartame produced the hypothalamic lesions at dose levels ≥1000 mg/kg body wt. In neonatal monkeys, again, the effect was not found (tested dose levels: 2000 mg/kg body wt and 2000 mg/kg body wt aspartame + 1000 mg/kg body wt monosodium glutamate) [25].

The concentrations of aspartate and glutamate in plasma of humans after ingestion of aspartame have been measured in adults and in infants. In adults single doses of 34, 50, 100, 150 or 200 mg/kg body wt aspartame were given (vehicle: orange juice). Afterwards the concentrations of glutamate and aspartate were found to be only slightly elevated. The peak concentrations did not exceed the ones normally found after a high protein meal [26]. Similar investigations were performed in human infants. Single doses of 0, 34, 50, or 100 mg/kg body wt aspartame (vehicle: carbohydrate-free cherry flavoured beverages) were administered orally to 1-year-olds. The zero-time concentrations of glutamate and aspartate in plasma were higher than the ones in adults. The aspartame treatment at 34 and 50 mg/kg body wt did not elevate the plasma concentrations and at 100 mg/kg body wt the observed increase was small (sum-concentration: 86 μmol/l at zero-time vs. 106 μmol/l after 30 min [27,28].

The above results show that even at bolus doses of aspartame exceeding the ADI-limit, in both adults and infants the plasma concentrations of glutamate and aspartate remain well below the toxic threshold for neonatal mice of 600—1000 μmol/l. The negative results of the neurotoxicity studies in neonatal monkeys provide further evidence that consumption of aspartame does not provide a significant risk for aspartate-induced neuronal necrosis in the brain.

The possible risk for human fetuses and sucklings associated with the aspartate moiety of aspartame can be evaluated on the basis of the results reported by Pitkin et al. [29] and Baker [30]. The placental transfer of aspartate and glutamate was measured in rhesus monkeys using radiolabelled aspartate and glutamate. Up to concentrations of 2000 μmol glutamate/l and 1000 μmol aspartate/l no placental transfer for these amino acids was found [29,31]. Baker reports the absence of a significant effect (in comparison to a lactose-control) on breast milk amino acid concentrations in lactating women after a single oral application of 50 mg/kg body wt aspartame [30]. These results indicate that the aspartate moiety does not pose a risk for the unborn fetus or the suckling in case of maternal aspartame consumption.

3.3 The phenylalanine moiety

The neutral amino acid phenylalanine is essential for humans. The initial step in phenylalanine degradation is hydroxylation to tyrosine. Thus, beyond this step, the catabolic pathway of phenylalanine is that of tyrosine. Tyrosine is the immediate precursor of the 2 catecholamine brain neurotransmitters dopamine and norepinephrine. Via tyrosine as intermediary phenylalanine may affect brain levels of dopamine and norepinephrine and thus influence brain functions [32,33].
For the uptake of large neutral amino acids (LNAA) from the blood into the brain, there is a stereospecific, saturable transport system. Via this system phenylalanine, tyrosine, leucine, iso-leucine, valine, tryptophan, histidine as well as methionine are transported into the brain. These neutral amino acids must compete with each other for available transfer sites for entry into the brain. Imbalances in the plasma amino acid concentrations are reflected in the uptake levels into the brain. When, for instance, the phenylalanine concentration in blood plasma is elevated, the uptake into the brain increases at the expense of that of the other LNAA [32,34]. Thus, an increased phenylalanine (or tyrosine) uptake might reduce the uptake of tryptophan. As tryptophan is the precursor of the brain neurotransmitter serotonin, increased plasma phenylalanine concentrations could indirectly influence the biosynthesis level of serotonin in the brain, and thus affect brain function [5,35].

An important genetic defect of phenylalanine metabolism is the hereditary absence or defect in the ability to hydroxylate phenylalanine to tyrosine. This disorder is called phenylketonuria (PKU). Kaufman distinguishes three forms of PKU, each corresponding with the dysfunctioning of 1 of the 3 (co-)enzymes participating in the phenylalanine hydroxylase system [36]. The classical form of PKU is caused by the total or near-total absence of hepatic phenylalanine hydroxylase activity. Marked phenylalaninemia occurs (concentrations ≥1200 μmol/l) and if the disease remains untreated, it results in delayed development, electroencephalic abnormalities, mental retardation and, in some cases, in seizures and eczema [32]. When the disease is detected early in life the patient is placed on a phenylalanine-restricted diet; normal development is then possible. In most hospitals newborn infants are tested for this defect, which can be detected by blood phenylalanine concentrations of 1200 μmol/l or more. The genetic defect is due to the homozygous state of a single autosomal recessive gene that is carried by about 1 in 60 individuals [32].

According to Kaufman the PKU-experience shows that it is the great increase in plasma phenylalanine levels unmatched by increases in the levels of other amino acids that is harmful to the developing brain, the reason for this being that high phenylalanine levels reduce the uptake into the brain of the other neutral amino acids [36]. In concurrence with this Caballero and Wurtman argue that the balance between the relative intake of phenylalanine and the other neutral amino acids is the main factor limiting phenylalanine’s access to the brain [37]. In normal food-proteins the phenylalanine content compared to the other neutral amino acids is relatively low. Given this information, it is important to determine to what degree consumption of phenylalanine through aspartame might mimic the amino acid imbalance seen in PKU.

Along the lines of the above theoretical considerations about possible effects on concentrations of neurotransmitters in the brain on the one hand and the experience with PKU on the other — (note there is a relation between the 2: PKU-neuropathology is probably, at least in part, due to
neurotransmitter deficiencies [36]) — 2 research approaches have developed. One line of research focussed on the measuring of the plasma phenylalanine concentrations in humans after consumption of aspartame and comparing these with the toxic concentrations found in PKU-patients. The alternative approach was the study of concentrations of phenylalanine, tyrosine, tryptophan and neurotransmitters in plasma and/or brain in experimental animals and, recently, the effect on (neurotransmitter-related) seizure thresholds in experimental animals. Below, the results of the 2 research approaches will be reviewed separately in the sections 3.3.1 and 3.3.2. The studies on behaviour and learning ability in humans and animals, although modeled for detection of abnormal functioning caused by the phenylalanine moiety of aspartame, will be reviewed, not in this section, but in the section on consumer complaints and clinical studies (section 4).

3.3.1. Plasma phenylalanine levels in humans and PKU

In the evaluation of plasma phenylalanine concentrations after aspartame consumption distinction should be made between the general population (persons not carrying the recessive PKU-gene), persons heterozygous for PKU and persons suffering from PKU (homozygotes). Given the 1:2 concentration gradient for phenylalanine between maternal and fetal blood [31], the use of aspartame during pregnancy represents a situation requiring special attention.

3.3.1.1. PKU-homozygotes. In PKU-homozygotes, not placed on a phenylalanine-restricted diet, plasma concentrations rise to 1200—3600 μmol/l and mental retardation occurs. In Western Europe a phenylalanine concentration in plasma of 480 μmol/l is regarded as the upper limit permissible for children on a phenylalanine-restricted diet. The restriction should be aimed at maintaining concentrations below this limit [38].

Koch and Wenz [39] discuss the potential impact of aspartame ingestion on the dietary phenylalanine regime. The same issue is addressed by Gütlinger and Lou [40]. For the young PKU-homozygote consumption of aspartame at the ADI-level would contribute significantly to the phenylalanine content of the diet (dietary phenylalanine-intake 20—30 mg/kg body wt; aspartame-related phenylalanine-intake at ADI-level: 22 mg/kg body wt). Gütlinger and Lou [40] indeed have found marked increases in plasma phenylalanine levels (peak level 850 μmol/l) in three 9-year-old PKU-homozygous children after a single dose of 34 mg/kg body wt aspartame. Also in older PKU-patients maintaining the phenylalanine-restricted diet, aspartame consumption at the ADI-level would significantly elevate the phenylalanine intake [39]. This is noteworthy as there is an increased tendency to maintain PKU-patients on the restricted diet also beyond the age of 6 or 7 [36,38]. Thus, ingestion of aspartame might easily derange the dietary phenylalanine restriction and consequently all PKU-patients on the phenylalanine-restricted diet should avoid consumption of aspartame. In addition Koch and Wenz [39] recommend that other PKU-patients on the so-called phenylalanine-liberalised diet (dietary phenylalanine intake 50—100 mg/kg body wt; phenylalanine intake
in a normal diet is 50—200 mg/kg body wt) should be made aware of the phenylalanine-content of aspartame and its use by these people be discouraged.

During pregnancy of PKU-homozygotes the maternal phenylalanine concentrations must be controlled since there is the risk of phenylalaninemia in fetal blood, causing mental retardation in the offspring (even when the offspring does not suffer from PKU). Several studies have focussed on this so-called maternal PKU-syndrome. Waisbren and Levy [41] report the result of an international survey which indicated an increased risk of mental retardation in the offspring, in case of severe maternal hyperphenylalaninemia (>1200 μmol/l), but also when the maternal hyperphenylalaninemia was only of a mild nature (200—600 μmol). As this study was of a retrospective nature, so Waisbren and Levy note, the possibility of an ascertainment bias exists. In a more direct study, the same authors [42] identified 22 untreated pregnant PKU-women (through routine umbilical cord blood screening) and evaluated the outcome of 59 pregnancies in these women. The IQ of offspring was significantly correlated with both maternal IQ and maternal blood phenylalanine level. The correlation between maternal IQ and offspring IQ is a complicating factor in the interpretation of the correlation between offspring and maternal phenylalanine level, especially at the intermediate phenylalanine concentration range. The authors present as their main finding that concentrations above 1100 μmol/l were consistently associated with mental retardation and concentrations below 600 μmol/l were consistently associated with normal intelligence. Levy and Waisbren take this result as suggestive evidence for the existence of a threshold concentration for the adverse effect of phenylalanine at 600 μmol/l. Pardridge [5,43] has contested this view and concluded that the issue of whether or not the data of Levy and Waisbren demonstrate that the deleterious effects on brain function caused by hyperphenylalaninemia follow a linear or a threshold pattern, is as yet unsettled.

In the U.S.A. the National Collaborative Study for Maternal PKU has recommended that during pregnancy (of PKU-homozygotes) blood phenylalanine-levels should not exceed 380 μmol/l [44].

3.3.1.2 Normal subjects. Persons not carrying the recessive PKU-gene normally have fasting plasma phenylalanine concentrations in the range of 46—67 μmol/l [26,37,45]. As the mean fasting concentration in fetal plasma 105 μmol/l has been reported; in newborns fasting levels up to 77 μmol/l occur [37]. In adults after a high-protein meal levels of 90—120 μmol/l are reached [26,45].

A number of acute dosing studies has been performed with aspartame in normal adults. The peak plasma phenylalanine concentrations found in these studies are presented in the appended Table I. Interpolation from the data of Filer and Stegink [46] indicates that a single bolus intake of aspartame at the ADI-level (40 mg/kg body wt) by a normal adult would result in a plasma phenylalanine level of about 130 μmol/l.
From their data, Filer and Stegink have calculated several pharmacokinetic parameters and with the use of these, they calculated the average steady state concentrations of plasma phenylalanine after oral doses of aspartame of 34, 100 or 200 mg/kg body wt repeated at intervals of 1–8 h. Repeated administration of 34 mg/kg body wt at 8-h intervals — (such a dosing regimen would result in an intake exceeding the ADI) — would bring about steady state plasma concentrations of 60 µmol/l. The same dose given at intervals of 4, 3, 2 or 1 h would result in steady state concentrations of 90, 100, 120 and 180 µmol/l respectively [46].

Two-multiple-dose studies were carried out in normal adults. Three doses of 10 mg/kg body wt aspartame, given at inter-dose intervals of 2 h, resulted in a high mean plasma phenylalanine concentration of 81 µmol/l. After 8 doses of about 8.5 mg/kg body wt given at inter-dose intervals of 1 h, the high mean plasma phenylalanine level was 109 µmol/l [46]. Matalon et al. [44]

### TABLE I

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<th>Dose (mg/kg body wt)</th>
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*Determined after 60 min (and 180 min) only.

bDetermined after 30 and 180 min only.

*Not reported.
report the result of a 12-week study in 22 normal adults, with a daily intake of 100 mg/kg body wt (administration and sampling regimen not reported). The plasma phenylalanine concentrations were determined on 4 occasions throughout the study. Most subjects (16) consistently showed plasma phenylalanine concentrations in the 0–354 μmol/l-range; 5 subjects had concentrations between 364 and 480 μmol/l on 1 occasion; in 1 subject the concentrations exceeded 600 μmol/l (not further specified). Stegink and co-workers have also examined plasma phenylalanine concentration after ingestion of aspartame as part of a meal. After a protein-rich meal to which 23 mg/kg body wt aspartame was added, the peak plasma level was 102 μmol/l (vs. 97 after the meal alone). An identical study with an aspartame dose level of 34 mg/kg body wt showed a peak level of 93 μmol/l (vs. 71 μmol/l after meal alone). Ingestion of 34 mg/kg body wt aspartame as part of a low-protein meal produced a peak of 145 μmol/l [46]. Apparently the presence of protein (amino acids) moderates the increase in plasma phenylalanine concentrations.

Several investigators have suggested that, given the competition for entry into the brain among large neutral amino acids (LNAA), one should evaluate the degree of imbalance in the plasma concentrations of these amino acids occurring after aspartame ingestion, rather than focussing on plasma phenylalanine concentrations only. Filer and Stegink [46] have calculated the ratio of plasma phenylalanine concentrations to the sum of the plasma concentrations of the LNAA (the phe/LNAA-ratio) for some of their studies. Not surprisingly, the plasma phe/LNAA-ratio is lower when aspartame is consumed as a part of a meal rich in proteins than when aspartame is consumed without proteins. Thus, the phe/LNAA-ratio adequately denotes that the potency of aspartame in producing imbalance in plasma amino acid levels is moderated by simultaneous ingestion of proteins. When aspartame is consumed without protein or amino acid mixtures, the phe/LNAA-approach will not provide extra information since the LNAA-concentrations are not affected in these cases, and consequently the plasma phe/LNAA-ratio will parallel the phenylalanine levels.

The results of the above studies demonstrate that aspartame consumption by normal adults at the ADI-level, even when such is done in the form of a single bolus dose, entails no risk of toxic levels of phenylalanine in blood plasma.

A study in normal infants has been reported by Filer and Stegink [46]. In 1-year-olds plasma phenylalanine concentrations were measured after a single dose of aspartame (vehicle: cherry-flavoured beverage). At the tested dose levels of 34, 50 and 100 mg/kg body wt the respective peak plasma phenylalanine concentrations were 94, 116, and 233 μmol/l (vs. 49 μmol/l in controls) [46]. Visek [47] reports the results of a 13-week-study in groups of normal children ranging in age from 2 to 21 years. Aspartame was given as a part of the normal diet at dose levels of 30–35 mg/kg body wt/day (children < 6 years old) or 30–77 mg/kg body wt (older children). Controls received sucrose. The incomplete report states that no increases in blood
phenylalanine concentrations (determined once/2 weeks) were found [47]. Thus, the studies provide no evidence that in normal infants, consumption of aspartame at the ADI-level produces phenylalanine concentrations in the toxic range.

Since even when a normal adult consumes the entire ADI as a single bolus dose — note that is a situation not representing the normal use of aspartame as a sweetener; the bolus model overestimates the impact on plasma phenylalanine concentrations — so, because even under these unusual conditions the plasma phenylalanine concentrations practically remain within the normal post-prandial range, there is no reason to expect a risk for the unborn in normal pregnant women.

Use of aspartame during lactation was demonstrated by Baker [30] to represent no special risk. A single bolus intake of 50 mg/kg body wt aspartame by lactating females did not produce significant elevations in phenylalanine content of breast milk in comparison with a control treatment with lactose.

3.3.1.3. PKU-heterozygotes. Persons heterozygous for PKU have a reduced capacity to metabolise phenylalanine because of a reduction in phenylalanine hydroxylase activity. The recessive gene for PKU is carried by about 1 in every 60 individuals. The PKU-heterozygotes do not have overt signs of PKU. In the heterozygotes the plasma phenylalanine concentrations are usually slightly elevated and the time required for clearance of phenylalanine is longer than in normal subjects [32].

Several studies have focussed on measuring phenylalanine concentrations in plasma of PKU-heterozygotes after ingestion of aspartame. The results of the single-dose studies are presented in Table II. Interpolation from the data

<table>
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<th>Dose (mg/kg body wt)</th>
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* Determined after 60 min (and 180 min) only.
of Filer and Stegink [46] indicates that a single bolus intake of aspartame at the ADI-level (40 mg/kg body wt) by a PKU-heterozygote would result in a plasma phenylalanine level of about 200 μmol/l.

Three multiple dose studies were carried out in PKU-heterozygous adults. Three doses of 10 mg/kg body wt aspartame, given at inter-dose intervals of 2 h, resulted in a high mean plasma phenylalanine concentration of 139 μmol/l. After 8 doses of about 8.5 mg/kg body wt, given at inter-dose intervals of 1 h, the high mean plasma phenylalanine level was 165 μmol/l [46]. Matalon et al. [44] report the result of a 12 week-study in 18 adult PKU-heterozygotes, with a daily intake of 100 mg/kg body wt (administration and sampling regimen not reported). The plasma phenylalanine levels were determined on 4 occasions throughout the study. Most subjects (11) consistently showed plasma phenylalanine concentrations in the 0—354 μmol/l range; 4 subjects mostly had phenylalanine concentrations in the 0—354 μmol/l range but showed concentrations of 364—594 μmol/l on 1 or more occasions; in 2 subjects concentrations >600 μmol/l (not further specified) were observed.

Visek [47] reports that the fasting plasma phenylalanine concentrations in adult PKU-heterozygotes consuming aspartame capsules at meal times (total daily intake 600—8100 mg) were not elevated during a treatment period of 27 weeks.

The results of the above studies show that after aspartame consumption at the ADI-level by adult PKU-heterozygotes, even when the entire ADI is consumed as single bolus dose, the plasma phenylalanine concentrations will not rise to levels commonly associated with adverse effects.

There are no data indicating a higher risk for PKU-heterozygous infants (compared to similar adults). A priori, there is no reason to assume such a elevated risk.

Kang and Paine [48] have reported the plasma phenylalanine levels in pregnant PKU-heterozygotes to be higher than the ones in non-pregnant female PKU-heterozygotes when normal meals are consumed (mean concentrations of 137 μmol/l vs. 102 μmol/l). No studies have been performed concerning the possible extra-increment in plasma phenylalanine concentrations after aspartame consumption by pregnant adult heterozygotes. The data obtained for non-pregnant adult heterozygotes indicate levels of about 200 μmol/l when the entire ADI is consumed as one bolus dose (see above). Making use of the data for non-pregnant heterozygotes underestimates the increments in phenylalanine concentrations expected in pregnant heterozygotes to an unknown extent. Probably such an underestimation is compensated by the overestimation of the phenylalanine concentrations implied by the assumption of the single bolus consumption of the ADI. So probably there is no significant risk for the pregnant PKU-heterozygote when aspartame is used at normal levels. A definite conclusion on this point, however, is only possible after adequate measurements of the phenylalanine concentrations in pregnant PKU-heterozygotes. A special case in heterozygote-pregnancy possibly is
represented by the homozygous fetus. Notably this homozygous state usually remains unnoticed before birth. The relevant question here is whether the homozygous fetus is more at risk at a given phenylalanine concentration than is the heterozygous or normal fetus. No publications specifically addressing this point were found.

3.3.2 Phenylalanine and brain neurotransmitters in experimental animals

The effect of aspartame on the serotonin-pathway has been studied in rats and mice. Fernstrom [35] determined the concentrations of tryptophan, serotonin and 5-hydroxyindole acetic acid (5-HIAA; this is the principal metabolite of serotonin) in serum and in whole brain of fasted adult rats after single oral doses of 0, 50, 100 or 200 mg/kg body wt aspartame and found no significant changes (the levels of phenylalanine and tyrosine, also measured, were increased). Also in fasted adult rats there was no effect on the tryptophan-hydroxylation rate in the brain [35]. Coulombe and Sharma [49] found the concentration of tryptophan and 5-HIAA in 6 brains regions in unfasted mice did not change significantly after a single oral dose of 0, 13, 130 or 650 mg/kg body wt aspartame.

Other studies have focussed on measuring the effect of apartame on the increases in brain tryptophan levels commonly observed after ingestion of carbohydrates by rats. This increase in brain tryptophan concentration is brought about via insulin, which lowers the blood levels of most LNAA but not that of tryptophan. Thus, tryptophan gains a competitive advantage for access to the brain, as a result of which elevated tryptophan concentrations in the brain occur. Wurtman [50] intubated fasted adult rats with 200 mg/kg body wt aspartame with or without glucose and found that aspartame markedly reduced the glucose-mediated increases in brain concentrations of tryptophan, serotonin and 5-HIAA. In addition he observed an extra elevation of the brain phenylalanine and tyrosine concentrations after intubation of aspartame and glucose simultaneously in comparison with aspartame alone. Fernstrom [35] administered a single meal of carbohydrates with or without aspartame to fasting adult rats and measured the brain tryptophan concentration. The reduction of the carbohydrate-induced increment of tryptophan concentration was observed at 881 and 530 mg/kg body wt but not at 656, 386, 315 and 267 mg/kg body wt. Fernstrom points out that his results fit in with the data obtained by Carlsson and Lindqvist [51] who observed that an intraperitoneal dose of 300 mg/kg body wt phenylalanine (or tyrosine) did reduce brain tryptophan levels in rats, whereas an intraperitoneal dose of 100 mg/kg body wt phenylalanine (or tyrosine) did not. Garattini et al. [52] attempted to reproduce the result of Wurtman [50] but found no effect whatever on brain concentrations of tryptophan after gavage-dosing glucose and/or 250 mg/kg body wt aspartame (the usual glucose-induced increase in brain tryptophan was not observed). Neither did they observe the extra, (i.e. beyond levels bound after intake of aspartame alone), increase in phenylalanine concentration in the brain after simultaneous administration of glucose and aspartame (250 mg/kg body wt).
Via tyrosine as intermediary the phenylalanine-moiety of aspartame may influence the formation of catecholamine neurotransmitters in the brain (dopamine, norepinephrine and their metabolites). Fernstrom [53] observed no effect on the whole brain concentrations of dopamine, norepinephrine or their metabolites dihydroxyphenylacetic acid and homovanillic acid (HVA) in rats after a single dose of 200 mg/kg body wt aspartame. The same author reports no significant effect on the tyrosine hydroxylation rate (or dopamine production rate) in the corpus striatum in the brain of adult fasted rats given single oral doses of 0, 50, 100 or 200 mg/kg body wt aspartame [35,54]. Garattini et al. [52] report the absence of a significant effect on the release of dopamine, dihydroxyphenylacetic acid and HVA in the striatum of the brain in rats given an aspartame dose as high as 100 mg/kg body wt Fernstrom [54] mentions some other studies in the same field in rats, the most significant result of which he considers to be an increased tyrosine hydroxylation rate in the pre-frontal and cingulate cortices, observed only at aspartame doses approaching 400 mg/kg body wt. Coulombe and Sharma [49] measured concentrations of norepinephrine, dopamine and several of their metabolites in 6 different brain regions after intubation of 0, 13, 130 or 650 mg/kg body wt aspartame in unfasted mice. They observed significant increases in some brain regions (most marked increases in hypothalamus; in the striatum no effect) at all dose levels. The value of these studies concerning the catecholamine pathway in rodents for the evaluation of such an effect in humans, is reduced by the marked difference in hepatic phenylalanine hydroxylase activity between the 2 species. The activity of this enzyme system is much greater in rodents than in humans [54,55] and consequently the tyrosine concentration will increase significantly less (and phenylalanine concentration will increase more) in humans after aspartame uptake. The effect of selectively incrementing brain phenylalanine levels on dopamine synthesis has not been studied in rats or humans.

There is a substantial body of evidence showing that both dopamine and norepinephrine have the potential to regulate seizure threshold, seizure severity and other manifestations of seizure activity in normal animals. Serotonin is an anti-convulsant neurotransmitter. Decreasing its concentration produces a pro-convulsant effect [56,57]. By inducing alterations in the release of these neurotransmitters aspartame possibly influences seizure activity. That high doses of aspartame are active was already found in an early limited study in neonatal monkeys in which seizures were observed at dose levels ≥3000 mg/kg body wt/day [2]. Recently, several studies have focussed on the effect of aspartame on seizure thresholds in animals. Garattini [58] reviews most of the studies. When metrazol was used as convulsant agent doses ≥750 mg/kg body wt aspartame were effective in lowering the seizure threshold in rats. An aspartame dose of 1000 mg/kg body wt was ineffective towards quinolinic-acid-induced convulsions in rats [58]. With pentylene tetrazole as convulsant drug in mice, doses of 50, 500 or 750 mg/kg body wt aspartame showed no effect whereas 1000, 1500 and 2000 mg/kg did [56,57,59]. Aspartame doses ≥1000 mg/kg body...
wt lowered the threshold for seizures induced by fluorothyl in mature mice; 500 and 750 mg/kg body wt were ineffective in these animals. In immature mice the effect was found at doses \( \geq 500 \) mg/kg body wt \cite{59}. The threshold for electroshock-induced convulsions in rats was lowered at 1000 mg/kg body wt aspartame but not at 500 mg/kg body wt \cite{60}.

The relevance of the findings in animal models for the representation of seizures in humans is discussed by Schomer \cite{61}. He points out that there is not one animal model that accurately represents all aspects of human epilepsy. In the human condition epilepsy there are many variables and it is, so Schomer argues, not surprising that there are conflicting reports on altering seizure susceptibility with various manipulations of the monoaminergic systems (serotonin, dopamine, norepinephrine and their metabolites). Apparently clear relations have yet to be established in this field.

The limited observational and clinical data in humans concerning the possible effect of aspartame on seizure activity will be discussed in section 4.

4. CONSUMER COMPLAINTS AND COGNITIVE AND BEHAVIOURAL OBSERVATIONS

In the U.S.A. consumer complaints on aspartame use have been frequent since the compound was admitted for use. Adverse reaction reports made to several institutions or to the manufacturer have been evaluated by the Center of Disease Control (Atlanta, Georgia) on behalf of the FDA in 1984 and by the Center for Food Safety and Applied Nutrition of the FDA in 1986. The initial analysis of 517 complaints from 1984 was updated and extended to 2800 complaints in 1986. The reported symptoms were grouped by nature and severity of the reaction and by the frequency and consistency of the association with ingestion of the product of interest. Additional analyses focussed on demographics, specific product or lot association and on consistency of dose, time and response relationships within an individual and within a case-defined reporting group. Complainants reporting severe reactions were interviewed and their medical records were obtained. The reported symptoms included the categories neurological seizures (100 complaints), headache (832), dizziness (383), change in mood quality (310), gastrointestinal (387) and allergic (239). The main conclusion was that no specific constellation of symptoms could be identified as being clearly related to aspartame consumption that would suggest a widespread public health hazard associated with aspartame use. Most of the frequently reported symptoms were mild and are common in the general population. In most cases a causal relation between aspartame consumption and the complaints was considered to be questionable. Some of the reports, however, possibly may be attributable to some as yet undefined sensitivity of some individuals to aspartame in commonly consumed amounts. Through focussed clinical studies a more thorough evaluation of the attributability of the complaints to aspartame consumption would be possible \cite{62-64}.

Roberts \cite{65} reviews 505 aspartame-related complaints and identifies
pregnant women, lactating mothers, young children and older persons with memory impairment as high-risk groups. Walton [66] reports cases of seizure after consumption of aspartame-sweetened beverages and considers these cases to be suggestive for a lowering of the seizure threshold by aspartame and the triggering of seizures in certain vulnerable individuals. He urges that appropriate double-blind studies, utilising populations at risk, be done.

Actuated by the above FDA-evaluation of consumer complaints controlled clinical investigations are now being initiated and carried out. The currently available results of these studies will be discussed below, as will be other relevant studies on the effect of aspartame on cognitive function and behaviour (also animal studies).

Animal experiments were carried out in rats and monkeys. In an early behavioural study in rats, diets containing 0, 2, 4 or 6% aspartame were administered from day 14 before mating continuously throughout breeding, gestation and lactation. An additional group was given a diet containing 3% phenylalanine. A number of effects was noted at 6% aspartame and 3% phenylalanine: increased offspring mortality, delayed physical and reflex development (including impaired eye opening, surface righting and swimming development), hypoactivity in the open field, delayed auditory startle and forward quadripal locomotor development. In the 4 and 2% aspartame groups delay in swimming development was the only effect [67,68]. During a 13-week-period of dietary feeding of aspartame (concentrations of 4.5 and 9%) the following behavioural effects were found in male rats (but not in female rats): increased motor activity (at 9% only), decreased conditioned avoidance response in shuttle box task (both dose levels) and impaired performance in the acquisition and performance of a non-cued continuous avoidance response in a two-way shuttle box (both dose levels). With a diet containing 5% phenylalanine similar results were obtained (results cited by Tilson et al. [60]). Torii et al. [69] observed no effect on spontaneous motor activity in rats given a diet containing 5% aspartame, with or without added sucrose, for 3 weeks. In a test in rats for operant learning behaviour in a Skinner-box, after a single oral dose of 250 mg/kg body wt aspartame, with or without added glucose, no effect was observed [70]. Tilson et al. [60] studied the effect of a single aspartame dose (250, 500 or 1000 mg/kg body wt) on the acoustic startle reflex, pre-pulse inhibition of the startle reflex, motor activity, the acquisition or retention of a passive avoidance task and a 2-way shuttle box response. They observed no effect. Learning ability was studied in juvenile stumptail macaques who had been exposed to dietary aspartame (dose levels: 1000, 2000 or 3000 mg/kg body wt/day) or phenylalanine (dose level: 1650 mg/kg body wt/day) for 270 days during infancy. When the animals were 1.5 years of age tests on learning ability, (i.e. object discrimination, pattern discrimination, object discrimination learning set, oddity learning set) and hearing ability were performed. No effect was observed [71]. Spiers et al. [72] have criticised this study for its long withdrawal period between dosing and actual behavioural testing.

Adequate controlled studies in humans are relatively scarce. (Note: the double blind design, a minimum requirement for behavioural testing, was
used in the studies below.) Krause et al. [73] have found a prolonged performance time on a computerised choice reaction in adult treated PKU-patients after induction of plasma phenylalanine concentrations of 2300 μmol/l (baseline value: 600 ± 400 μmol/l) via suppletion of the diet with phenylalanine. Another study with a phenylalanine-supplemented diet was performed by Elsas and Trotter [74]. A group of 8 adults (6 PKU-heterozygotes and 2 normals) received a phenylalanine-supplemented diet (100 mg/kg body wt/day added) for two 2-week periods over a period of 8 weeks. On the final day of each 2-week treatment period plasma phenylalanine concentrations were determined and electroencephalography and tests for higher cognitive function (10 tests, not specified) were carried out. There was considerable inter-individual variation in plasma phenylalanine concentration: maximum concentration: 539 μmol/l; mean 239 μmol/l (baseline values: maximum approx. 150 μmol/l and mean approx. 99 μmol/l). When changes in plasma phenylalanine concentrations were correlated to observed EEG-changes, it appeared that the mean power frequency of the EEG was slowed when phenylalanine concentrations increased and was accelerated when plasma concentrations decreased. This reversible, inverse relationship was found to hold with plasma phenylalanine changes as low as 7 μmol/l. An effect on cognitive function is also reported, but the actual results of the cognitive tests are not presented [74]. Clearly, the result of this limited study should be corroborated and extended and, as yet, cannot be regarded as convincing evidence for an adverse effect on brain function at plasma phenylalanine concentrations of 200–500 μmol/l. Lieberman et al. [75] tested aspartame itself. They administered a single dose of 20 or 60 mg/kg body wt aspartame (administration in capsules) with or without added carbohydrates to 20 male volunteers and found no effect on behaviour in the hours after treatment (administered behavioural tests: short-duration reaction time, 4-choice reaction time, Wilkinson auditory vigilance test, digit symbol substitution test, tapping, profile of mood states, visual analogue mood scales, Stanford sleepiness scale). In another experiment no effects on subjective feeling of hunger, mood and arousal (scoring on stomach sensations, nausea, headache, faintness, nervousness, tension, alertness) were found after consuming single doses of about 70 and 140 mg/kg body wt aspartame (and neither at 12–140 mg/kg body wt phenylalanine) by normal adult males. In this study cognitive functions were not tested [76]. Wolraich [77] reviews the available behavioural studies in children. Two studies were performed in groups of 16 hyperactive boys (age 7–12 years), one focussing on cognitive and laboratory tests (including playroom observations, examiner rating, learning and memory tasks and measures of impulsivity) and the other studying the behaviour in the children’s natural setting. The treatment consisted of a single ingestion of flavoured beverage containing aspartame (6.4 to 8.8 mg/kg body wt) or sucrose (1.8 mg/kg body wt). No effect was observed. A further 2 studies were carried out in normal pre-school children (3–5 years old) who were challenged with aspartame-sweetened beverage (total dose about 167 mg).
No adverse effects were noted in developmental assessment (including drawings, peg boards, coordination tasks), actometer readings and parent and teacher ratings for behaviour [77]. Kruesi et al. [78] report the results of a study in which higher dose levels were used. They tested the effect of acute doses of sucrose, glucose, saccharin or aspartame (30 mg/kg body wt) on the behaviour of 30 pre-school boys on 8 challenge days. Among the test subjects were 11 boys with presumed sucrose-related behaviour disorders. Challenges occurred in 2 settings: at home and in a playroom. The children were observed (at home by parents and in the playroom by trained personnel) for hyperactivity, aggression and emotional reactions. The general activity level was measured with an actometer. The observers would not detect any behavioural effects. The actometer measurements showed a significantly lower activity level after the aspartame treatment. The authors consider it doubtful whether this finding is of clinical significance since observers do not detect a difference [78]. A study in children (4–15 years) with seizure disorders, in which aspartame is given at a level of 34 mg/kg body wt for 2-week-periods, is still in progress [79].

Actuated by the headache complaints about aspartame several target-directed clinical studies were implemented and are now in progress [80]. Koehler et al. [81] have recently published the result of one of these studies. They found an increased incidence of migraine in 11 migraineurs receiving 1200 mg aspartame/day (as 300 mg capsules) for 4 weeks. The result of other studies are not available yet.

In conclusion, in rats no effects on behaviour were observed in single-dose studies. In studies of longer duration in this species, however, behavioural effects were noted at dose levels of 1000 mg/kg body wt and higher. In the available single dose studies in humans (both adults and children tested; dose levels up to 140 mg/kg body wt) there were no significant effects on cognitive function and behaviour. Studies of longer duration are scarce. At permanently elevated phenylalanine concentrations of 2300 μmol/l effects occur; the limited experimental data concerning more moderate permanent elevations (up to 500 μmol/l) are inconclusive. Full value clinical studies on behaviour and cognitive function in (sensitive) humans more closely representing the situation of long-term aspartame use close to the ADI-limit, are lacking.

The result of the clinical studies in humans, now in progress in the U.S.A., will provide additional evidence concerning the interpretation of the adverse reaction reports of aspartame.

5. INTAKE LEVELS OF ASPARTAME

One approach in evaluating the intake levels of phenylalanine and aspartate via aspartame has been the comparison of the extra uptake of these amino acids via aspartame with the normal uptake via dietary proteins, (e.g. [26]). Such a comparison, however, disregards the crucial factor of the competition for uptake between amino acids and the notion that it is
the degree of imbalance between amino acids which is the foremost determining factor for uptake of aspartame-derived phenylalanine and aspartate. Notably, aspartame is also intended for use in low-protein products like coffee, tea and soft drinks. Thus, the approach of comparing aspartame-derived amino acid intake with intake via dietary protein is of limited relevance only.

For estimating the common intake levels of food additives several techniques are available including the per capita disappearance data, food purchase survey (total diet study) and food consumption survey (via questionnaires) (reviews by Heybach and Allen, [82]; Abrams, [83]).

There is some variation in the published estimates of aspartame intake. In 1981, based on the MRCA Information Inc.-survey of food and beverage preparation by households and individuals (2000 households involved in the survey) for 14-day periods, the 99th percentile of the projected aspartame intake was estimated to be 34 mg/kg body wt [26,83]. Glinsmann et al. [64] based an estimate on the same MRCA-survey, assuming total aspartame substitution in all relevant products (products not specified). His findings are outlined in Table III.

Kirkpatrick and Lauer [84], in discussing the possible approaches for estimating the aspartame intake in Canada after the 1981-approval for several food items, point out the importance of considering whether the mean for eaters-only or the mean for all persons is used for the different food items. The data of Kirkpatrick and Lauer are summarized in Table IV. The eaters-only data show that for young children the 40 mg/kg body wt-limit may be exceeded when aspartame replaces sucrose in all products and the person in question is an eater of all the products listed. This combination of total replacement of sucrose and consumption of all products listed probably is an unusual situation.

Contrary to the above data, which suggest intakes within the ADI-limit, several authors have pointed out that intake levels exceeding the ADI-limit are possible, especially in 7–12-year-olds, when consumers have free access to aspartame-containing products [5,85]. Matalon et al. [44] suggest than in the U.S.A. the consumption levels for aspartame may triple in the next
### TABLE IV

**ESTIMATED INTAKE OF ASPARTAME, DATA FROM KIRKPATRICK AND LAUER [84]**

<table>
<thead>
<tr>
<th>Product</th>
<th>Intake of food (g/day) and intake of aspartame (mg)</th>
<th>All persons mean intake</th>
<th>Eaters-only intake</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Food (g)</td>
<td>Aspartame (mg)</td>
</tr>
<tr>
<td><strong>Age 1—4 years</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Breakfast cereals</td>
<td></td>
<td>3.7</td>
<td>5</td>
</tr>
<tr>
<td>Soft drinks</td>
<td></td>
<td>40.0</td>
<td>19</td>
</tr>
<tr>
<td>Hot chocolate mix</td>
<td></td>
<td>7.2</td>
<td>3</td>
</tr>
<tr>
<td>Iced tea mix</td>
<td></td>
<td>9.3</td>
<td>2</td>
</tr>
<tr>
<td>Choc. flav. dairy</td>
<td></td>
<td>10.7</td>
<td>3</td>
</tr>
<tr>
<td>drink</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Drink mixes</td>
<td></td>
<td>31.0</td>
<td>8</td>
</tr>
<tr>
<td>Yoghurt</td>
<td></td>
<td>0.9</td>
<td>0.5</td>
</tr>
<tr>
<td>Ice cream analogs</td>
<td></td>
<td>16.5</td>
<td>13</td>
</tr>
<tr>
<td>Puddings</td>
<td></td>
<td>11.7</td>
<td>6</td>
</tr>
<tr>
<td>Gelatins</td>
<td></td>
<td>5.0</td>
<td>2</td>
</tr>
<tr>
<td>Mints</td>
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<td>4.0</td>
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</tr>
<tr>
<td><strong>Total</strong></td>
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</tr>
<tr>
<td><strong>Age 12—19 years</strong></td>
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<td></td>
</tr>
<tr>
<td>Breakfast cereals</td>
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<td>5.4</td>
<td>7</td>
</tr>
<tr>
<td>Soft drinks</td>
<td></td>
<td>200.5</td>
<td>96</td>
</tr>
<tr>
<td>Hot chocolate mix</td>
<td></td>
<td>14.2</td>
<td>6</td>
</tr>
<tr>
<td>Iced tea mix</td>
<td></td>
<td>79.3</td>
<td>20</td>
</tr>
<tr>
<td>Choc. flav. dairy</td>
<td></td>
<td>61.1</td>
<td>18</td>
</tr>
<tr>
<td>drink</td>
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<td></td>
</tr>
<tr>
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<td>41.0</td>
<td>10</td>
</tr>
<tr>
<td>Yoghurt</td>
<td></td>
<td>1.5</td>
<td>0.9</td>
</tr>
<tr>
<td>Ice cream analogs</td>
<td></td>
<td>29.4</td>
<td>24</td>
</tr>
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<td>Puddings</td>
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<td>6</td>
</tr>
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</tr>
<tr>
<td>Mints</td>
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<td>70</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td></td>
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<td></td>
</tr>
<tr>
<td><strong>All ages</strong></td>
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<td></td>
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<tr>
<td>Breakfast cereals</td>
<td></td>
<td>2.5</td>
<td>3</td>
</tr>
<tr>
<td>Soft drinks</td>
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<td>51</td>
</tr>
<tr>
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<td>221.6</td>
<td>55</td>
</tr>
<tr>
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<td></td>
<td>No data</td>
<td>—</td>
</tr>
<tr>
<td>drink</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Drink mixes</td>
<td></td>
<td>24</td>
<td>6</td>
</tr>
<tr>
<td>Yoghurt</td>
<td></td>
<td>1.3</td>
<td>0.8</td>
</tr>
<tr>
<td>Ice cream analogs</td>
<td></td>
<td>17.6</td>
<td>14</td>
</tr>
<tr>
<td>Puddings</td>
<td></td>
<td>9.5</td>
<td>5</td>
</tr>
<tr>
<td>Gelatins</td>
<td></td>
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<td>—</td>
</tr>
<tr>
<td>Mints</td>
<td></td>
<td>4.3</td>
<td>43</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td></td>
<td>177.8</td>
<td></td>
</tr>
</tbody>
</table>
decade. Intake levels of 50–100 mg/kg body wt would then not be uncommon.

6. CONCLUSIONS

When aspartame is consumed at levels within the ADI-limit there is no significant risk for an aspartate-induced neurotoxic effect in the brain. The experimental evidence indicates that this conclusion holds for adults, infants, sucklings as well as fetuses.

The phenylalanine-moiety of aspartame potentially exerts adverse effects on brain function in humans. Plasma phenylalanine concentrations of ≥1200 μmol/l are toxic in this respect. For the lower concentration range no firm conclusion can be drawn. Controversy exists on the question of whether the deleterious effects on brain function caused by hyperphenylalaninemia follows linear or a threshold pattern. Controlled studies in this field are virtually lacking. Clearly, as yet there are no observational data supporting the view that adverse effects occur at moderately elevated plasma phenylalanine concentrations (200–400 μmol/l).

In humans there is considerable intra-species variation in plasma concentration reached after ingestion of a given aspartame-dose. Nevertheles the evidence shows that after aspartame ingestion at the ADI-level, even when the ADI is consumed as a bolus dose, the plasma phenylalanine concentrations are practically always within the normal postprandial range; elevation to plasma concentrations commonly associated with adverse effects has not been observed. This statement holds for normal subjects (adults, infants), for PKU-heterozygous adults and probably also for PKU-heterozygous infants and pregnant subjects (normal, PKU-heterozygous). For the consumption of aspartame within the ADI-limit under practical conditions, the more gradual intake pattern and the competitive effect of simultaneously consumed other amino acids mostly will provide a moderating effect on plasma phenylalanine concentrations reached.

Ingestion of aspartame at the ADI-level would significantly increase the phenylalanine intake of PKU-homozygotes, thus deranging the phenylalanine restriction maintained by these persons. The PKU-homozygotes on the phenylalanine-restricted diet should therefore avoid aspartame consumption. PKU-homozygotes on the phenylalanine-liberalised diet should be made aware of the phenylalanine content of aspartame and its use by these people be discouraged.

Consumer complaints have raised the point of a possible effect of aspartame on behaviour. The available behavioural studies in animals showed effects after long-term administration at dose levels of 1000 mg/kg body wt or higher. In the available acute dosing studies in humans no effects were observed. Long-term studies on behaviour and cognitive function in (sensitive) humans are lacking.

Analyses of adverse reaction reports made by consumers in the U.S.A. have not yielded a specific constellation of symptoms clearly related to aspartame that would suggest a widespread public health hazard associated
with aspartame use. The possibility of an as yet undefined sensitivity to aspartame in at least some of the complainants could, however, not be ruled out. Focussed clinical studies are now being carried out in the U.S.A.; the results should provide additional evidence concerning the interpretation of the adverse reaction reports of aspartame.

In the regulation of admitted uses for aspartame the possibility of intake levels exceeding the ADI-limit in some groups of consumers should be a point of attention.

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