A Comparison of the Pathological Effects of 2-Acetylamino-fluorene, 4-Acetylaminobiphenyl, and 2-Acetylaminobiphenyl in the Dog*

ANNE G. JABARA

(Department of Pathology, University of Melbourne, Melbourne, Australia)

SUMMARY

The oral administration of 2-acetylamino-fluorene (2-AAF) to seven bitches was followed by epithelial hyperplasia in the urinary bladders of two dogs surviving 40 months, and one of these animals also developed multiple hepatomas. The oral administration of 4-acetylaminobiphenyl (4-AAB) to six bitches induced metastasizing urinary bladder carcinomas in three dogs surviving 36 months or longer and multiple hepatomas in one of those animals. The oral administration of 2-AAB to two males and six bitches resulted in urinary bladder epithelial hyperplasia in one dog surviving 21 months of treatment, and dysplastic proliferation in the urinary bladders of six animals surviving 39–41 months.

4-AAB was a more potent bladder carcinogen in the dog than 2-AAF but was not so active as its nonacetylated homolog, taking into account the dose levels and latent periods of the three amides. Hepatoma formation was not directly related to the amount of 2-AAF or 4-AAB consumed but appeared to depend mainly on the duration of administration of amide and possibly partly on constitutional factors. Hepatomas induced with both 2-AAF and 4-AAB were histologically similar and resembled those arising spontaneously in dogs.

The non-neoplastic hepatic changes induced with all three amides probably represented a nonspecific toxic reaction either to the compounds themselves or to their metabolites, rather than a precancerous state.

The aromatic amines 2-acetylamino-fluorene (2-AAF) and two compounds structurally related to it (4-acetylaminobiphenyl [4-AAB] and 2-acetylaminobiphenyl [2-AAB]) have induced cancer in the rat, 2-AAF and 4-AAB being particularly effective in this regard (15–17).

Several aromatic amines (Table 4) have been reported to induce tumors in dogs, but of the three mentioned above only 2-AAF has been previously administered to this species. In this experiment, 2-AAF was used to confirm previous findings (1, 18) and to determine whether any differences were produced by feeding the amine at a different dose level from that previously used.

MATERIALS AND METHODS

Each dog was weighed every 5 weeks, and the dosage of amine per dog per day was calculated and multiplied by the number of days in a 5-week period. The total dose for each animal was then weighed out and loaded into gelatin capsules sizes 0 and 1 (Parke, Davis & Co. Ltd.); the capsules were embedded in a ball of dough and fed manually to each dog.

2-Acetylamino-fluorene was fed to seven bitches at the rate of 33 mg/kg body weight/day 3 times a week. 4-Acetylaminobiphenyl was administered to six bitches, and 2-AAB was given to two males and six bitches, both compounds being fed at the rate of 28 mg/kg body weight/day 3 times a week.

2-Acetylamino-fluorene was obtained from L. Light and Co., England; 4-AAB was prepared from commercial diphenyl by nitration with fuming nitric acid, reduction with stannous chloride dissolved in concentrated hydrochloric acid, and acetylation with acetic acid and acetic anhydride; 2-AAB was prepared by acetylation of 2-AB (obtained from L. Light and Co., England) with acetic anhydride and sodium acetate.

*This work was carried out during the tenure of a grant from the Anti-Cancer Council of Victoria, Australia.

Received for publication January 29, 1963.
Most of the dogs in the three groups were mongrels of the spaniel, terrier, and heeler types, and ranged in age from 2 months to 5 years at the start of the experiments (Tables 1-3).

The conditions under which the animals were housed, their diet, and the precautions taken against infections prior to, and during, the experiments have been reported previously (11).

During the course of the experiments, each of the three groups of dogs suffered casualties from fights, but the surviving members of the 2-AAF and 2-AAB series remained well and active and without any clinical symptoms up to the time of being killed. However, at a late stage, dogs of the 4-AAB group exhibited gradual wasting and development of icterus, anemia, and hematuria, two animals dying from these symptoms.

TABLE 1

<table>
<thead>
<tr>
<th>No.</th>
<th>Age at start of experiment</th>
<th>Breed of dog*</th>
<th>Period of survival (months)</th>
<th>Total dosage (gm.)</th>
<th>Variation in weight (lb.)</th>
<th>Microscopic findings</th>
<th>Cause of death</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2 mo.</td>
<td>Spaniel</td>
<td>1</td>
<td>1</td>
<td>3-4</td>
<td>Slight hepatic damage</td>
<td>Dog fight</td>
</tr>
<tr>
<td>2</td>
<td>11 mo.</td>
<td>Fox terrier</td>
<td>2</td>
<td>5</td>
<td>11-16</td>
<td>&quot; &quot; &quot; &quot; &quot; &quot; &quot; &quot; &quot; &quot;</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>4 yr.</td>
<td>Australian terrier</td>
<td>6</td>
<td>17</td>
<td>9-11</td>
<td>&quot; &quot; &quot; &quot; &quot; &quot; &quot; &quot; &quot; &quot;</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>2 yr.</td>
<td>Spaniel</td>
<td>10</td>
<td>50</td>
<td>16-19</td>
<td>&quot; &quot; &quot; &quot; &quot; &quot; &quot; &quot; &quot; &quot;</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>4 mo.</td>
<td>Wirehaired terrier</td>
<td>13</td>
<td>42</td>
<td>9-13</td>
<td>Multiple hepatomas; epithelial hyperplasia urinary bladder</td>
<td>Overdose pentobarbitone</td>
</tr>
<tr>
<td>6</td>
<td>4 mo.</td>
<td>Wirehaired terrier</td>
<td>40</td>
<td>220</td>
<td>10-15</td>
<td>Moderate hepatic damage; epithelial hyperplasia urinary bladder</td>
<td>&quot; &quot; &quot; &quot; &quot; &quot; &quot; &quot;</td>
</tr>
<tr>
<td>7</td>
<td>6 mo.</td>
<td>Wirehaired terrier</td>
<td>40</td>
<td>300</td>
<td>15-20</td>
<td>&quot; &quot; &quot; &quot; &quot; &quot; &quot; &quot; &quot; &quot;</td>
<td></td>
</tr>
</tbody>
</table>

* Predominantly of this dog type.

The average hepatic cell volume was obtained by the method used by Christie and LePage (4) for measuring nuclear volumes.

Fluorescein-globulin stain, prepared by Louis’s method (14) was applied to fresh, unfixed, frozen sections of normal and abnormal tissues, and the preparations were examined with the fluorescence microscope. Normal or hyperplastic canine tissues stained by the fluorescein-globulin complex showed a bright green fluorescence of the epithelial cytoplasm, but the cytoplasmic component of malignant epithelial tissues showed very weak fluorescence only.

RESULTS

2-ACETYLAMINOPHLORENE GROUP

Macroscopic findings.—Five dogs (Nos. 1–6) died from trauma received in fights within the first 13 months of the experiment (Table 1).

The two remaining animals (Nos. 6 and 7) were killed with an overdose of pentobarbitone sodium (by intravenous injection) after 40 months of treatment, and at necropsy each had an enlarged liver which weighed 432 gm. (a gross increase in weight) and 297 gm. (a slight increase), respectively. Both livers appeared paler and felt firmer than normal; the liver from No. 6, in addition, contained several large, firm, cream-colored tumors with areas of congestion and hemorrhage and multiple smaller nodules. The largest growth was situated in the left lateral lobe and measured 8.0 × 6.5 cm.; the next in size were located in the right central lobe and measured 2.5 cm. and 1.5 cm. in diameter, respectively (Fig. 1).

Microscopic findings.—

Liver: No disruption of the lobular architecture was apparent in the five earlier cases or in No. 7, although atrophy of many of the central and midzonal hepatic cells and large and small vacuoles in the cytoplasm due to fat and glycogen (sudan red and PAS stains) were present in all cases. The liver cells of No. 7 showed considerable variation in size, a moderate increase in cytoplasmic bulk resulting in an average cellular volume of 8,171 cμ (normal value, 4,064 cμ), decrease in cytoplasmic granularity, and a reduction in the staining densities of RNA and of mitochondria (pyronin and phosphotungstic acid-hematoxylin stains).

Assays on the liver from No. 7 by Christie and LePage revealed 26 mg. of nitrogen/gm wet weight of liver (a 20 per cent reduction) compared with 32.5 mg. in the control liver, and 3.8 mg. mitochondrial nitrogen/gm wet weight of liver tissue (a 24 per cent reduction) against 5.0 mg/gm wet weight of control liver.

1 G. S. Christie and R. N. LePage, personal communication.
Dog 6 showed histological changes in the non-neoplastic areas similar to those seen in No. 7. The neoplastic areas showed complete loss of a lobular pattern, areas of closely packed hepatic cells alternated with areas where the cells were more widely separated and contained large fatty vacuoles (Fig. 2), and frequently in the latter regions there were numerous large irregular blood sinuses. Cytological alterations included a gross enlargement of hepatic cells, especially in the fatty areas; decrease in cytoplasmic granularity; considerable variation in nuclear size, number, and shape; and a low frequency of mitoses. Occasional small collections of tumor cells were observed within blood vessels.

Other organs: Hyperplastic foci were present throughout the mucosa of the urinary bladders from dogs 6 and 7, pyelonephritis in the kidneys of Nos. 4-7, and bronchopneumonia in the lungs of all dogs, except Nos. 3 and 5.

4-ACETYLAMINOBIPHENYL GROUP

**Macroscopic findings.**—Within the first 4 months of the experiment, dogs 8-10 died from trauma received in fights (Table 2). Dogs 11 and 12 died from hemolytic anemia after 36 months of treatment. Both dogs showed considerable wasting during the 36th month; within the last few days before death their mucous and nictitating membranes and conjunctivae appeared icteric and anemic, and at autopsy the skin and fat of No. 12 were also pale yellow (Table 2). Both animals developed hematuria in the last few days of life, and the urine was frothy, cloudy, and very dark.

Dog 13 developed hematuria and difficulty in micturating in the 41st month of the experiment and was killed with pentobarbitone in the 42d month (Table 2).

At autopsy, examination of the urinary bladders from Nos. 11-13, revealed multiple hemorrhagic, cream- or greyish white-colored, sessile or pedunculated papillomas (2-8 mm. in diameter) situated predominantly in the fundi, and the bladder walls were increased in thickness up to 8 mm. (Figs. 3, 4). The livers from these three dogs showed coarse nodular cirrhosis, and that from No. 13 also contained multiple tumor nodules and weighed 363 gm. (a moderate increase in weight) (Fig. 5). The two largest hepatomas were situated in the left central lobe and measured 2.5 x 2.5 cm. and 2.5 x 2.0 cm.; the two next in size (2.0 cm. in diameter) were situated in the right central and the right lateral lobes; multiple smaller nodules (6 mm. or less in diameter) were situated in the right and left lateral lobes. The cut surfaces of the growths showed nodular tissue, white or hemorrhagic, sometimes containing small cystic spaces filled with colorless gelatinous material. The lungs from the three dogs were broncho-pneumonic and contained multiple white metastatic nodules 1 mm. in diameter. Bilateral pyelonephrosis and hydroureter (Fig. 4) were present in one dog (No. 11); the kidneys and ureters in Nos. 12 and 13 appeared normal. Multiple pe++techial hemorrhages were observed in the bowel in Nos. 11 and 12, and in the latter dog a duodenal ulcer (1 cm. in diameter) was situated just distal to the pylorus.

**Microscopic findings.**—

Urinary bladder: The mucosa was normal in dogs 8 and 9. Desquamation (probably due to post-mortem change) had occurred in No. 10.

Dogs 11 and 12 showed mucosal hyperplasia, ulceration, and neoplastic sessile papillomas (Fig. 6). The tumors consisted of sheets, cords, or clumps of epithelial cells mainly of the transitional
type showing considerable variation in cell and nuclear size and shape, a low frequency of mitoses, necrotic foci, metaplastic squamous cells (Fig. 7), invasion of the submucosa and muscle (Fig. 8), permeation of small blood vessels and lymphatics by tumor cells, and absence of bright green fluorescence with fluorescein-globulin stain, indicative of malignancy.

The neoplastic areas in dog 13 showed a predominance of metaplastic squamous cells, large and small droplets of mucin in many of the squamous cells (alcian blue stain), and areas containing glandular cellular arrangements (Fig. 9).

Lungs: All cases showed bronchopneumonia, and cases 11–13 contained, in addition, multiple irregular metastatic tumor foci of various sizes. The secondary tumors in Nos. 11 and 12 showed a predominance of transitional epithelial cells, occasional grouped or isolated metaplastic squamous cells (Fig. 10), and a low frequency of mitoses. In dog 13 most foci showed a predominance of metaplastic squamous cells. The neoplastic cells were sometimes arranged in a single row lining the alveoli or entirely filling alveoli, or, in the case of the larger nodules, replacing the parenchyma completely.

Liver: The three short-term cases showed diffuse cloudy swelling of the parenchyma and a few small necrotic foci which were mainly situated centrilobularly.

The livers from the three dogs which survived longer showed thickened capsules and replacement of the normal lobular architecture by numerous irregular nodules of "regenerated" hepatic cells. These nodules were surrounded by relatively vascular connective tissue which contained an increased number of relatively small bile ducts. The cytological changes included an increase in hepatic cell size, resulting in an average cellular volume of 4,450 cμ and 6,704 cμ in dogs 11 and 13, respectively, reduction in cytoplasmic granularity, reduction in the staining densities of DNA, RNA, and mitochondria (Feulgen, pyronin, and phosphotungstic acid-hematoxylin stains), considerable fatty vacuolation and a low frequency of mitoses.

The neoplastic areas in dog 13 showed partial encapsulation, absence of lobular pattern, and consisted of closely packed hepatic cells. Areas of fatty vacuolation (Fig. 11) were often associated with numerous large, irregular sinuses containing hemosiderin (prussian blue stain). The cytological alterations in viable areas included considerable variation of cell size, reduction in the densities of Feulgen, pyronin, and phosphotungstic acid-hematoxylin staining, considerable variation in nuclear size and shape, a low frequency of mitoses, and tumor giant cells.

Christie and LePage⁴ assayed the total nitrogen/gm wet weight of cirrhotic liver and tumor liver tissues in dog 13. They found these tissues contained, respectively, 21.4 mg. (a 24 per cent reduction) and 25.4 mg. of nitrogen/gm wet weight of liver (a 9 per cent reduction) compared with 28 mg. in the control liver.

Other organs: Pyelonephritis was present in five dogs and pyonephrosis in the sixth. Prussian blue staining showed relatively abundant hemosiderin in the proximal tubular epithelium of the later cases. A large subacute duodenal ulcer which had penetrated into the serosa and mesentery was found in No. 12.

2-Acetylaminobiphenyl Group

**Macroscopic findings.**—Except for dogs 14 and 15, which died from trauma received in fights after 23 months of treatment, the other six dogs survived for almost 3½ years and were still well and were maintaining a steady body weight when killed with pentobarbitone (Table 3). At autopsy only the urinary bladders and livers from the six dogs which survived longest showed alteration. In these the bladder mucosa was hyperplastic, and the wall increased in thickness up to 7 mm.; the livers appeared paler and felt firmer than normal, and, except that from No. 20, all were slightly increased in weight.

**Microscopic findings.**—

Urinary bladder: The bladder of dog 15 was normal, but that of No. 14 showed mucosal hyperplasia. The epithelium in the other six cases showed foci of dysplasia (Fig. 12); fluorescein-globulin staining in dog 20 indicated nonmalignancy.

Ureter and urethra: The ureteral and urethral mucosae appeared hyperplastic in the six animals which survived longest.

Liver: No case showed disruption of lobular pattern, but, particularly in the later cases, an obvious increase in hepatic cell cytoplasmic bulk was apparent, resulting in an average cellular volume of 8,590 cμ, 9,847 cμ, and 10,057 cμ in dogs 16, 19, and 21, respectively. A decrease in cytoplasmic granularity and a slight to moderate reduction in the densities of Feulgen, pyronin, and phosphotungstic acid-hematoxylin staining were observed.

Other organs: All dogs were pyelonephritic, and pelvic epithelial dysplasia was present in the six later cases. The lungs in all animals showed bronchopneumonia and occasionally foci of squamous epithelium in the alveolae.
Considerable changes were observed in the hemopoietic and lymphatic system in every group and will be separately reported.

**DISCUSSION**

Morris and Eyestone (18) fed 2-AAF at the rate of 8.3–10.9 mg/day/dog (total dosage ranged from 90 to 198 gm.) to three male and two female dogs, and four animals developed hepatomas 5½–7½ years after the start of the experiment. Allison et al. (1), on the other hand, administered 0.03 per cent 2-AAF in a synthetic diet to twelve dogs and stated that three animals developed liver tumors in 30–34 weeks. The discrepancy in the latent period is considerable, and unfortunately in the latter case the authors gave no indication of the multiple hepatomas were found only in dog 6, which had received the smaller total dose. Similarly, in the 4-AAB group No. 13 developed multiple hepatomas after consuming 274 gm. of the chemical in 42 months, whereas Nos. 11 and 12 both survived 36 months, received a total of 312 and 335 gm. of 4-AAB, respectively, and neither showed hepatoma formation. Thus, it appears that the length of treatment may be a more important factor in canine carcinogenesis than the total dose of a carcinogen, and possibly constitutional factors, as in rats and mice, may also play a part.

The only other aromatic amine which has induced liver tumors in dogs is o-aminoazotoluene (20), though Hueper, Wiley, and Wolfe (9) reported “adenomatoid” proliferation and some

---

**TABLE 3**

<table>
<thead>
<tr>
<th>No.</th>
<th>Age at start of experiment</th>
<th>Breed of dog*</th>
<th>Period of survival (months)</th>
<th>Total dosage (gm.)</th>
<th>Variation in weight (lb.)</th>
<th>Microscopic findings</th>
<th>Cause of death</th>
</tr>
</thead>
<tbody>
<tr>
<td>14</td>
<td>2 mo.</td>
<td>Australian terrier</td>
<td>21</td>
<td>94</td>
<td>13–15</td>
<td>Epithelial hyperplasia urinary bladder; slight hepatic damage</td>
<td>Dog fight</td>
</tr>
<tr>
<td>15</td>
<td>3 yr.</td>
<td>Australian terrier</td>
<td>23</td>
<td>108</td>
<td>13–16</td>
<td>Slight hepatic damage</td>
<td>&quot;</td>
</tr>
<tr>
<td>16</td>
<td>2 mo.</td>
<td>Heeler</td>
<td>39</td>
<td>290</td>
<td>15–21</td>
<td>Epithelial dysplasia renal pelvis, urinary bladder; epithelial hyperplasia ureter, urethra; slight hepatic damage</td>
<td>Overdose pen-tobarbitone</td>
</tr>
<tr>
<td>17</td>
<td>2 mo.</td>
<td>Heeler</td>
<td>39</td>
<td>245</td>
<td>16–22</td>
<td>&quot;</td>
<td>&quot;</td>
</tr>
<tr>
<td>18</td>
<td>2 yr.</td>
<td>Heeler</td>
<td>39</td>
<td>230</td>
<td>16–22</td>
<td>&quot;</td>
<td>&quot;</td>
</tr>
<tr>
<td>19</td>
<td>5 yr.</td>
<td>Heeler</td>
<td>40</td>
<td>220</td>
<td>12–16</td>
<td>&quot;</td>
<td>&quot;</td>
</tr>
<tr>
<td>20†</td>
<td>5 mo.</td>
<td>Terrier</td>
<td>40</td>
<td>215</td>
<td>21–24</td>
<td>&quot;</td>
<td>&quot;</td>
</tr>
<tr>
<td>21†</td>
<td>2 mo.</td>
<td>Spaniel</td>
<td>41</td>
<td>199</td>
<td>15–15</td>
<td>&quot;</td>
<td>&quot;</td>
</tr>
</tbody>
</table>

* Predominantly of this dog type.
† Male.

daily consumption of 2-AAF nor of the total dosage the dogs received. There is no doubt that nodular livers developed in the dogs treated by Allison et al. (1), but it appears more likely either that these were actually hyperplasias or that their 2-AAF was impure. Morris and Eyestone (18) administered a total of 32–37 gm. of 2-AAF in 2½ years to another group of four dogs, and none developed neoplasms. In the present experiment the animals were fed this compound at a considerably higher dose rate than that used by Morris and Eyestone (18) in their group of five dogs, and this reduced the latent period for carcinogenesis from 5½ to 3½ years.

Morris and Eyestone (18) concluded that the extent of hepatic tumor formation in the dog was directly related to the amount of the carcinogen consumed. In the present experiment, however, both Nos. 6 and 7 survived 40 months, but minor hepatic atrophy in dogs following β-naphthylamine administration.

Kirby (13) stated that in the rat the majority of 2-AAF-induced hepatic tumors occurred in the right lateral lobes and only later appeared in the central and left lobes. Hahn, Donald, and Grier (cited in [13]) injected radioactive phosphorus into the mesenteric vein of several dogs and found that a large excess of isotope was localized on the right side of the liver; similar injections into the splenic vein produced an excess of isotope on the left side. In the present experiments dogs 6 (2-AAF group) and 13 (4-AAB series) both exhibited the largest hepatomas on the left side of the liver; but, disregarding size, the number of tumors on the right and left sides of the liver was approximately equal (Figs. 1, 5).

Histologically, the hepatomas induced in both the 2-AAF and 4-AAB series were similar and also
closely resembled those induced by Morris and Eyestone (18), and also those arising spontaneously in dogs (18).

Prior to neoplasia, the severe generalized hepatic cell enlargement observed with all three aromatic amines was accompanied by a decrease in the DNA staining density of the liver cell nuclei, of the cytoplasmic RNA, and of the mitochondria. Furthermore, Christie and LePage observed considerable decreases in the total nitrogen/gm wet weight of cirrhotic liver and tumor.

Both the transitional epithelium of the canine urinary tract (particularly the portion lining the urinary bladder) appeared susceptible to the action of all three aromatic amines. In the 2-AAF group, two dogs which survived longest exhibited hyperplastic foci in the urinary bladder; Morris and Eyestone (18), after a longer period of administration, induced canine bladder tumors with this compound. All the urinary bladders in the 2-AAF group exhibited dysplastic epithelial proliferation, and the epithelium of the ureters and urethras appeared hyperplastic; the dysplastic epithelial proliferation seen in the renal pelvis probably resulted

TABLE 4
ADDITIONAL CANINE BLADDER CARCINOGENS AND THEIR LATENT PERIODS FOR TOTAL DOSAGES GIVEN

<table>
<thead>
<tr>
<th>Chemical</th>
<th>Method of administration</th>
<th>Total dosage (gm.)</th>
<th>No. dogs used</th>
<th>No. dogs with bladder tumors</th>
<th>Time of tumor appearance (years)</th>
<th>Investigators</th>
</tr>
</thead>
<tbody>
<tr>
<td>2-Acetylaminofluorene</td>
<td>Oral</td>
<td>90–108</td>
<td>5</td>
<td>4</td>
<td>54–74</td>
<td>Morris and Eyestone (18)</td>
</tr>
<tr>
<td>4-Aminobiphenyl</td>
<td>Oral</td>
<td>30–34</td>
<td>2</td>
<td>2</td>
<td>54–74</td>
<td>Walpole et al. (23)</td>
</tr>
<tr>
<td>4-Aminobiphenyl</td>
<td>Oral</td>
<td>87–144</td>
<td>4</td>
<td>4</td>
<td>13–21</td>
<td>Deichmann et al. (6, 8)</td>
</tr>
<tr>
<td>β-Naphthylamine</td>
<td>Oral</td>
<td>Large</td>
<td>4</td>
<td>3</td>
<td>38–5</td>
<td>Bonser (4)</td>
</tr>
<tr>
<td>β-Naphthylamine</td>
<td>Subcut. inj.</td>
<td>15</td>
<td>1</td>
<td>1</td>
<td>6</td>
<td>Gehrmann et al. (cited in [23])</td>
</tr>
<tr>
<td>β-Naphthylamine</td>
<td>Subcut. inj. and oral</td>
<td>55–180</td>
<td>16</td>
<td>4</td>
<td>3</td>
<td>Hueper and Wolfe (10)</td>
</tr>
<tr>
<td>Benzidine</td>
<td>Oral</td>
<td>325</td>
<td>7</td>
<td>1</td>
<td>8</td>
<td>Spitz et al. (22)</td>
</tr>
<tr>
<td>3,4,5,6-Dibenzcarbazole</td>
<td>Intravesical inj.</td>
<td>1</td>
<td>1</td>
<td>34</td>
<td>Bonser et al. (3)</td>
<td></td>
</tr>
<tr>
<td>o-Aminoazotoluene</td>
<td>Oral</td>
<td>1</td>
<td>1</td>
<td>34</td>
<td>Nelson and Woodard (20)</td>
<td></td>
</tr>
<tr>
<td>p-Dimethylanilinazoobenzene</td>
<td>Oral</td>
<td>98–198</td>
<td>19</td>
<td>3</td>
<td>2–4</td>
<td>Nelson and Woodard (20)</td>
</tr>
<tr>
<td>p-Nitrobiphenyl</td>
<td>Oral</td>
<td>325</td>
<td>7</td>
<td>1</td>
<td>8</td>
<td>Spitz et al. (22)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

It could be postulated that these hepatic changes, which always preceded neoplastic transformation, were indicative of a precancerous state, but it is more likely that they were coincidental and simply represented a nonspecific hepatic reaction to a toxic agent (either the compound itself or metabolites).

FIG. 1.—Dog No. 6; macroscopic view of liver containing multiple, cream-colored, malignant hepatomas. X1.

FIG. 2.—Dog No. 6; photomicrograph of one area of a malignant hepatoma, showing considerably fatty vacuolation in some pleomorphic cells and relatively little in others. X630.

FIG. 3.—Dog No. 15; macroscopic view of papillary carcinomas of urinary bladder. X1.

FIG. 4.—Dog No. 11; macroscopic view showing bilateral pyonephrosis, bilateral hydronephrosis and papillary carcinomas of urinary bladder. X4.
Fig. 5.—Dog No. 13; macroscopic view of liver showing coarse nodular cirrhosis and multiple, cream-colored, malignant hepatomas. X4.

Fig. 6.—Dog No. 11; photomicrograph showing papillary carcinoma of urinary bladder containing cellular clumps and glandular structures, and reactive small round-cell infiltration. X240.

Fig. 7.—Dog No. 11, photomicrograph of portion of a papillary carcinoma of urinary bladder, showing squamous metaplasia and multinucleate giant cells. X1410.

Fig. 8.—Dog No. 11; photomicrograph of urinary bladder wall, showing neoplastic invasion of muscle and permeation of lymphatics and small blood vessels. X240.
one exception, these neoplasms were similar to those observed in the 4-AAB group. Nelson and Woodard (20), however, reported that in their series one urinary bladder carcinoma infiltrated through the organ wall into the serosa and prostate gland.

The microscopic appearances of the neoplasms arising in dogs 11 and 12 were similar to the majority of tumors induced with the other carcinogens (Table 4). However, the neoplasms induced with 4-aminobiphenyl (4-AB) by Deichmann et al. (6, 8) were predominantly squamous-cell carcinomas similar to those seen in No. 18. Secondary tumors were not reported in any case, unlike the present series where multiple pulmonary metastases occurred in all tumor-bearing animals.

If one relates the average latent periods for the induction of canine bladder tumors with the total administered dose of several aromatic amines (Table 4), it appears that 4-AB, β-naphthylamine, 3, 4, 5, 6-dibenzcarbazole, o-aminozotoluene, p-nitroaniline, and p-dimethylaminoazobenzene are considerably more potent bladder carcinogens in the dog than either benzidine or 2-AAF. In the present experiment, the 3-year latent period for bladder tumor induction with 4-AAB was comparable to that of the six more potent amines (the long latent period Gehrmann, Foulger, and Fleming—cited in [23]—found following β-naphthylamine administration [Table 4] was due to these investigators’ using only a single small dose of this amine), though the total dose was higher (274—335 gm.). Therefore, 4-AAB appears to be a considerably more active carcinogen than benzidine in the dog, though not so potent as its nonacetylated homolog.

The kidney changes observed in all three groups, particularly in the dogs which survived a relatively long period of treatment, were similar and resembled the alterations described in canine kidneys following β-naphthylamine and 4-AB administration (8, 6, 9); it is considered that these modifications represent a nonspecific reaction in this organ to the toxic action of the amines or their metabolites which predisposed to their infection.

The changes in the lungs of the dogs from all three groups and in the small bowel of dog No. 12 were probably unrelated to the administration of the three compounds.

REFERENCES


A Comparison of the Pathological Effects of 2-Acetylaminofluorene, 4-Acetylaminobiphenyl, and 2-Acetylaminobiphenyl in the Dog

Anne G. Jabara


Updated version
Access the most recent version of this article at:
http://cancerres.aacrjournals.org/content/23/6_Part_1/921

E-mail alerts
Sign up to receive free email-alerts related to this article or journal.

Reprints and Subscriptions
To order reprints of this article or to subscribe to the journal, contact the AACR Publications Department at pubs@aacr.org.

Permissions
To request permission to re-use all or part of this article, use this link http://cancerres.aacrjournals.org/content/23/6_Part_1/921.
Click on "Request Permissions" which will take you to the Copyright Clearance Center's (CCC) Rightslink site.