cell surface, RCA_I-Au_{50} reacted very poorly. Similar results were obtained with human erythrocytes using RCA_I and SBA gold markers. However, the large size ConA-Au_{50} was bound by hepatocytes (but not ConA-Au_{64}) contrary to erythrocytes where the ConA marker size had to be below 13 nm in size to obtain a binding reaction (unpublished observations). These differences are attributed to the more or less narrow spacing between the brushes of ektoprotein or glycoprotein of the cell surface[13] which does not allow the marker to reach its receptor.

Although WGA and anti-H lectins in the presence of L-fucose have similar specificities, the cells bound 5 times less anti-H-Au_{50} than WGA-Au_{50}. This could indicate that these lectins do not share a common receptor. SBA competed very little with WGA-Au_{50} which suggested that their receptors are not close to each other. Since RCA_I is a glycoprotein[20] and is precipitated by ConA, binding of RCA_I-Au_{32} increased when the cells were prelabelled with ConA. It has been claimed that ConA and WGA bind to separate sites on liver cell membranes[7]. This was confirmed by competitive experiment with WGA-Au_{32}, whose binding was inhibited only by 10% when the cells were saturated with ConA (table).

Rat hepatocytes were successively marked with WGA-Au_{17} and ConA-Au_5 (figure 1). As reported by Virtanen and Wartiovaara using ConA conjugated with ferritin[4], the cells had a microvillous surface continuously marked with ConA-Au_5. However, the cells were marked with WGA-Au_{17} preferentially where they were in contact with each other. No significant difference was found when the cells were labelled in the reverse order.

Hepatocytes marked with WGA-Au_{75}, WGA-Au_{50} and ConA-Au_{50} were examined by SEM (figures 2–4). The markers were distributed in clusters, the density of WGA-Au_{50} being greater than that of ConA-Au_{50} (figures 3 and 4) in agreement with the data of the table. Calculations on micrographs indicated that 35,000 WGA-Au_{50} granules were bound per cell. This figure, lower than that estimated by spectrophotometric measurements (44,000), is explained by a small loss of particules during the preparation of the specimen for SEM. A similar distribution of the markers-Au_{50} was found with the other lectins by SEM examination, the density of marking corresponding to the data of the table.

In conclusion, lectin-labelled gold markers having different sizes are useful for determining simultaneously the distribution of 2 lectin receptors but also for estimating the spacing between the glycoprotein brushes. While spacing must be below 32, 50 and 64 nm before the receptors can bind SBA-, RCA_I and ConA-Au markers, respectively, all sizes of WGA-Au markers bind to the cell surface. This indicates that WGA-receptors extend from the cell surface and are not masked by other glycoproteins.

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