One-Hour Postload Plasma Glucose in Obese Indian Adults with Nonalcoholic Fatty Liver Disease: An Observational Study from North India

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Abstract

Background: Metabolic perturbations and hyperglycemia are increasingly identified as causal factors for nonalcoholic fatty liver disease (NAFLD). Insulin resistance, an indirect marker for initiation of NAFLD can be promptly diagnosed with standard oral glucose tolerance test (OGTT). One-hour postOGTT plasma glucose measurement can have a significant impact on early identification of dysglycemia with NAFLD and may be superior to fasting and 2-hour plasma glucose. Objective: To assess 1-hour post OGTT plasma glucose levels and presence of NAFLD in obese adults. Materials and Methods: In this observational study, we included 101 consecutive obese (body mass index >25 kg/m²) participants of age 20–50 years without known illness of diabetes mellitus. Their anthropometric and laboratory characteristics were recorded and a standard OGTT was performed. Plasma glucose (PG) levels were measured during fasting, 1-hour (1-hour-PPG), and 2-hour (2-hour-PPG) intervals. All participants underwent ultrasound of the abdomen by a single, experienced observer for fatty liver (FL) grade assessment. Comparison of the PG and FL was done by the Chi-square test and a P value <0.05 was considered statistically significant with a 95% confidence interval. Data analysis was done using SPSS version 24.0 (IBM® SPSS Statistics Inc., Chicago, Illinois, USA).

Results: The result demonstrated that 53 adults had 1-hour-PPG values above the cutoff (≥155 mg/dl), whereas only 20 individuals had raised PG at 2 hours (≥140 mg/dl). All study subjects (100%) had an evidence of FL disease on ultrasound scan. Among these, 33 had grade I and 68 participants had grade II or III FL. About 41.6% of individuals had deranged 1-hour-PPG levels and higher FL grades as compared to 11.9% individuals with raised 2-hour-PPG values and FL of same grades. The relationship between 1-hour-PPG and FL grades was also statistically significant (P value <0.05). Conclusions: 1-hour-PPG levels were more deranged in obese adults without diabetes, and had more consistent and significant relationship with higher FL grades than the 2-hour-PPG levels.

Keywords: Fasting plasma glucose, hyperglycemia, nonalcoholic fatty liver disease, 1-hour post-load plasma glucose, 2-hour post-load plasma glucose

INTRODUCTION

Nonalcoholic fatty liver disease (NAFLD) comprises an array of the liver conditions, denoted by hepatic fat accumulation (steatosis) in the absence of insults like alcohol abuse and viral hepatitis, and is usually associated with features of the metabolic syndrome like abnormal glucose tolerance or diabetes mellitus (DM), visceral obesity, hypertension, and dyslipidemia. The disease spectrum ranges from simple steatosis to an inflammatory nonalcoholic steatohepatitis (NASH) that may progress to cirrhosis, and finally to hepatocellular carcinoma.[¹,²] The disease not only affects the liver, but also has deleterious effect on cardiovascular (CV) and renal systems through increased insulin resistance and accelerated atherosclerosis.[³,⁴] NAFLD was associated with two-fold risk of CV disease in a study comprising 2000 subjects with diabetes, and the risk was not attributable to variables like sex, age, smoking, diabetes...
duration, HbA1c, and LDL cholesterol (LDL-C) levels indicating that NAFLD can involve CV system even in the absence of traditional risk factors. Insulin resistance plays a crucial role in the development of NAFLD, which further perpetuates insulin resistance within the liver and the other organs, and therefore, type 2 DM (T2DM) and development of NAFLD has an invariably strong association. It has been estimated that NAFLD prevalence was greater than 70% in individuals with DM.

Prediabetes or intermediate hyperglycemia delineates an increment of plasma glucose (PG) levels over the normal range, but below the diagnostic cutoff for DM. The condition describes the biochemical starting of insulin resistance and discernible alterations in lipid metabolism. Elevated 1-hour post-load PG (1-hour-PPG) is an emerging marker for insulin resistance and onset of lipid alteration in various metabolically active organs like liver. Insulin resistance provides unrestrained fatty acids to the hepatocytes, which leads to triglyceride (TG) generation, impaired fatty acid oxidation and accumulation of fat globules. Therefore, a paramount stimulus in NAFLD pathogenesis is insulin resistance, besides its constitutional role in obesity and diabetes. This insulin secretion and action derangement can be visualized by elevated 1-hour-PPG levels and OGTT is one simple tool applied for this PG elevation recognition.

Conventional dysglycemic markers like HbA1c and 2-hour post-load PG (2-hour-PPG) are current defining tools for diabetes and prediabetes, but research has demonstrated that these markers can be normal in the early disease course and cannot identify ongoing target organ damage like NAFLD. In an observation by Fiorentino et al., hepatic steatosis was identified in higher frequency among individuals with HbA1c levels of 5.7–6.4% and 1-hour-PPG of >155 mg/dl in comparison to individuals with same HbA1c levels, but 1-hour-PPG levels of <155 mg/dl. Similarly, the integration of HbA1c <5.7% and 1-hour-PPG >155 mg/dl revealed a higher risk of hepatic steatosis as compared with integration of HbA1c <5.7% and 1-hour-PPG <155 mg/dl. Also, in individuals with 1-hour-PPG >155 mg/dl having or not prediabetic HbA1c levels, exhibit significantly higher serum insulin concentrations as compared with individuals with 1-hour-PPG <155 mg/dl. Further, it was observed that the association between 1-hour-PPG >155 mg/dl and hepatic steatosis was significant both in obese (Body mass index, BMI >30 kg/m²) and in non-obese individuals (BMI <30 kg/m²). These observations indicate that post-load PG and abnormal insulin levels synergistically induce lipogenic metabolic state, leading to increased hepatic fat storage. Taken together, these data suggest that elevation in 1-hour-PPG can be a major driver of hepatic steatosis whereas 2-hour-PPG appears to play a smaller role.

**Objective**

To assess 1-hour post OGTT plasma glucose levels and presence of NAFLD in obese adults.

**Materials and Methods**

**Design and study population**

The study was conducted from January 2019 to December 2019 in our tertiary healthcare center. Obese individuals of age 20–50 years, BMI ≥25 kg/m² and without known illness of DM were selected. OGTT parameters and FL grades were assessed among these and compared for any possible relationship. Individuals suffering from any chronic illness of major organ system like congestive heart failure, chronic kidney disease, chronic liver disease, and cerebrovascular accidents were not included. Similarly, known endocrine causes like acromegaly, Cushing’s syndrome, thyroid disorders, pheochromocytoma, chronic pancreatitis and patients on chronic medications like atypical anti-psychotics, beta blockers, corticosteroids, and thiazide diuretics were excluded due to their potential to cause dysglycemia. Individuals with BMI ≥40 kg/m² were also excluded as excess fatty tissue and abdominal circumference leads to poor visibility of hepatic tissue by ultrasound (USG), and also to prevent the skewness of data.

As per the recent study, the prevalence of prediabetes and diabetes was 85% and non-diabetes was 30% in NAFLD patients, and for 95% power of the study (alpha error 0.05), a minimum of 72 individuals were required in this study. A total of 130 individuals fulfilled the above stated eligibility criteria, but 21 subjects had raised FPG and HbA1c levels and eight individuals didn’t undergo the USG abdomen, so they were not included in the study. Thus, 101 out of a total of 130 subjects completed the required investigations and were assigned to this study.

**Ethical approval and consent to participate**

The study was ethically approved form local (institutional) ethical committee with letter no. GMC/IEC/2018/167. The participants, who gave written informed consent after explaining the study and objectives, were included for further assessment. The study participants were assured that confidentiality of their personal data would be maintained.

**Data collection**

The relevant history and complete physical examination, including anthropometry was carried out initially. As per the study protocol, selected individuals were subjected to blood biochemistry investigations and in addition, underwent standard 75-gm OGTT as per the standard protocol described by WHO after a fasting period of 8–12 hours. A zero-hour (0-hour, base line) blood sample was collected for FPG. The individual was then given 75 gm oral glucose solution to drink within a 5-min time frame and blood was drawn at 1-hour and 2-hour intervals.

**Distribution according to OGTT**

We categorized 1-hour-PPG and 2-hour-PPG values to assess distribution of FL grades within these categories. There were two 1-hour-PPG categories, that is blood glucose <155 mg/dl and blood glucose ≥155 mg/dl, which were based on recent literature depicting the harmful effect of 1-hour-PPG above
155 mg/dl.[7] A cutoff value of 140 mg/dl and above this was considered significant for 2-hour-PPG, according to the American Diabetes Association, in which 140 mg/dl cutoff is used to define pre-diabetes and one study objectifying this value unfavorable.[11,12]

**Diagnosis of NAFLD**
Assessment of FL was done by USG abdomen in fasting state, prior to OGTT or on the next day, by a single experienced observer to avoid inter-observer variability and for uniform result acquisition. Participants were assessed in supine position with a high resolution B-mode USG scanner (Philips® IU 22 Unit) using convex curved array transducer of 1–5 MHz frequency by a consultant radiologist. The grades were recorded from 0-III. Grade 0 was taken as normal liver echogenic pattern and grade I to III were taken as abnormal, that is, fatty infiltration as mild, moderate, and severe, respectively. Grade I fatty infiltration was given when the liver echogenicity was raised with normal visualization of intrahepatic vessel outlines, grade II was given when echogenicity of the liver was increased with mild impairment of visualization of diaphragm and intrahepatic vessel outlines,[13] Grade III fatty infiltration was established when liver parenchymal echogenicity was increased markedly with poor visualization of diaphragm and intrahepatic vessel borders.

**Statistical analysis**
Obese individuals having a BMI ≥25 kg/m² underwent an OGTT, and their anthropometric, metabolic and laboratory characteristics were classified and compared by using the Student’s t-test and One-Way ANOVA analysis. All continuous data were expressed as mean and standard deviation (±SD). Using the OGTT as a first stage screening, individuals were categorized according to their PG value at fasting, 1-hour, and 2-hour. Population distribution significance for FPG and FL grades was assessed by the Z-test and Chi-square test was applied for comparison of FL, 1-hour-PPG, and 2-hour-PPG. For 95% confidence interval, a P value <0.05 was considered statistically significant. Data analysis was done using SPSS version 24.0 (IBM® SPSS Statistics Inc., Chicago, Illinois, USA).

**RESULTS**
**Description of study population**
The study comprised 101 obese participants. The mean age was 38.8 (±8.05) years and the majority (45.5%) belonged to 41–50 years’ age group. There were 59 (58.4%) females and 42 (41.6%) males. Region-wise distribution was Chandigarh (53.5%), followed by Punjab (34.7%) and Haryana (10.9%). Anthropometric analysis depicted higher BMI values in female population (30.3 ± 3.59 kg/m²). In biochemical analysis of the study participants, the mean SGOT levels were slightly higher for both male (50.2 ± 27.8 IU/L, normal <50) and female population (46.5 ± 24.1 IU/L, normal <45), but for SGPT (Normal <56 IU/L) levels, male population had more derangement (59.3 vs 50.0 IU/L). This population also had abnormal lipid profile. Total cholesterol (TC) mean levels were similar in the male and female group (222.2 and 222.9 mg/dl) but higher than the upper cutoff value (<200 mg/dl). Moreover, the average triglyceride (TG) levels among the male participants were significantly higher than the female participants (224.6 mg/dl vs 193.8 mg/dl), and the average values for both the groups were notably raised (Normal <150 mg/dl). For high density lipoprotein (HDL) levels, female population had unfavorable levels than the counterpart male population as in both gender groups, the cutoff value is different (desirable HDL levels for male >40 mg/dl and >50 for female population). The detailed data is depicted in Table 1.

In metabolic parameter evaluation, individuals with high 1-hour-PPG had more deranged lipid profile and raised mean HbA1c levels [Table 2], but anthropometric measurements were not significantly discrete between the two groups. Similarly, these parameters were also compared in different FL grade groups, and the results demonstrated an adverse metabolic profile (except TG levels) in association with higher FL grades [Table 3].

**Comparison between FPG, 1-hour-PPG, 2-hour-PPG, and NAFLD**
A striking finding of the present study was that all the study individuals had some grade of fatty infiltration within the hepatic parenchyma, so the prevalence of FL was 100% in our study population. But to discriminate the relationship of different OGTT parameters with FL grades, grade I FL was considered as less significant and FL grade II and III as more significant. More than half of the total study cohort (53 out of 101 individuals) had raised 1-hour-PPG values (≥155 mg/dl), and out of these, 79.25% had higher FL grades. The calculation also demonstrated that the finding of raised 1-hour-PPG levels with higher FL grades was statistically significant (P value 0.007) [Table 4]. Thus, 1-hour-PPG levels were markedly deranged among the study participants, and majority of them demonstrated high grades of FL.

When 2-hour-PPG was compared to FL grades, 69.14% individuals with PG <140 mg/dl had higher liver grades (II, III), in contrast to 60% with glucose levels of ≥140 mg/dl and the same FL grades. So, elevated 2-hour-PPG probably had a negative relationship with FL grades as majority of individuals with high FL grades had low PG at 2 hours. However, the P value for this comparison was 0.43, which was insignificant to conclude this relationship valid.

In the present study, all of the individuals had FPG <100 mg/dl but USG abdomen demonstrated presence of higher FL in more than two-thirds of them (67.33%). This describes that FPG levels cannot be correlated with high FL grades, and this inverse distribution had a significant P value (<0.05) [Figure 1].

**DISCUSSION**
This observational study was done among 101 obese adults without diabetes and OGTT based glycemic parameters were
assessed for any derangement, and compared with different FL grades. The unique strength of this study was that it measured all the three OGTT values—FPG, 1-hour-PPG, and 2-hour-PPG simultaneously for concealed hyperglycemia, and correlated with different FL grades separately.

Females had a higher prevalence of obesity in the present study population. Indian population statistics on obesity (BMI >25 kg/m$^2$) has shown a higher prevalence in the females; 38.7% in female and 24.2% in male population in a North Indian region (Chandigarh).\[14\] Our population was younger and the reason for targeting this population is justified keeping in mind the trends in onset of diabetes in Asian Indians. An Indian research has illustrated the mean age of T2DM onset to be 51 years.\[15\] The age group for inclusion of 20-50 years and the mean age of 38.8 (±8.05) years was particularly important as early intervention can halt the progression to diabetes complications due to hyperglycemia including progressive liver disease. However, all individuals are susceptible as previous research has suggested that age

### Table 1: Anthropometric and laboratory characteristics of the study population

| Variables       | Total population Mean±SD (range) | Male Mean±SD (range) | Female Mean±SD (range) | $P^*$  |
|-----------------|---------------------------------|----------------------|------------------------|--------|
| Age (years)     | 38.8±8.05 (20-50)               | 37.6±7.97 (20-50)    | 39.6±8.06 (20-50)      | 0.47 (NS) |
| BMI (kg/m$^2$)  | 29.7±3.34 (25.0-39.9)           | 28.7±2.73 (25.1-35.4) | 30.3±3.59 (25.0-39.9)  | 0.06 (NS) |
| WC (cm)         | 90.9±5.63 (81.2-104)            | 89.6±5.43 (81.2-102) | 91.9±5.63 (82.0-104)   | 0.14090 (NS) |
| SGOT (IU/L)     | 48.1±25.6 (12-112)              | 50.2±27.8 (15-112)   | 46.5±24.1 (12-112)     | 0.77539 (NS) |
| SGPT (IU/L)     | 53.9±33.8 (10-181)              | 59.3±34.0 (10-33)    | 50.0±33.5 (10-181)     | 0.39735 (NS) |
| TC (mg/dl)      | 222.7±54.4 (125-399)            | 222.2±48.6 (136-302) | 222.9±58.7 (125-399)   | 0.99741 (NS) |
| TG (mg/dl)      | 206.6±83.4 (65-475)             | 224.6±93.7 (82-475)  | 193.8±73.4 (65-454)    | 0.18728 (NS) |
| HDL (mg/dl)     | 39.9±7.50 (26-73)               | 39.1±8.10 (26-73)    | 40.4±7.05 (28-60)      | 0.67079 (NS) |
| HbA1c (%)       | 5.62±0.39 (4.3-6.1)             | 5.46±0.44 (4.3-6.1)  | 5.73±0.30 (4.7-6.0)    | 0.003 (S) |

*One-Way ANOVA, (S)- Significant, (NS)- Non-significant. BMI: Body mass index; WC: Waist circumference; SGOT (AST): Serum Glutamic Oxaloacetic Transaminase; SGPT (ALT): Serum Glutamic Pyruvic Transaminase; TC: Total Cholesterol; TG: Triglyceride; HDL: High density lipoprotein, HbA1c: Glycated hemoglobin

| Metabolic parameter | Mean±SD (Range) | 1-h PPG (n=48) | 2-hour PPG (n=53) | $P^*$ |
|---------------------|-----------------|---------------|-------------------|-------|
| BMI (kg/m$^2$)      | 29.53±3.27 (25.0-39.5) | 29.84±3.42 (25.1-39.9) | 0.32 (NS) |
| WC (cm)             | 91.0±5.50 (81.2-104)  | 90.8±5.79 (81.2-104) | 0.41 (NS) |
| SGOT (IU/L)         | 50.2±27.8 (15-112)   | 46.5±24.1 (12-112)  | 0.22 (NS) |
| SGPT (IU/L)         | 59.3±34.0 (10-33)    | 50.0±33.5 (10-181)  | 0.21 (NS) |
| TC (mg/dl)          | 233.0±61.25 (125-399) | 212.15±88.02 (82-454) | 0.24 (NS) |
| TG (mg/dl)          | 37.5±5.80 (26-73)    | 5.69±0.38 (4.6-6.0)  | 0.03 (S) |

*Student’s t-Test, (S)- Significant, (NS)- Non-significant. BMI: Body mass index; WC: Waist circumference; SGOT (AST): Serum Glutamic Oxaloacetic Transaminase; SGPT (ALT): Serum Glutamic Pyruvic Transaminase; TC: Total Cholesterol; TG: Triglyceride; HDL: High density lipoprotein, HbA1c: Glycated hemoglobin

| Metabolic parameter | Total population Mean±SD (range) | Fatty liver grade n=101 (%) | $P^*$ |
|---------------------|---------------------------------|-----------------------------|-------|
|                      | I Mean±SD (Range)               | II, III Mean±SD (Range)     |       |
| BMI (kg/m$^2$)       | 28.7±2.73 (25.2-35.4)           | 30.15±3.53 (25.0-39.9)      | 0.02 (S) |
| WC (cm)              | 90.49±4.86 (81.2-100)           | 91.21±5.99 (81.2-104)       | 0.27 (NS) |
| SGOT (IU/L)          | 44.5±23.21 (12-92)              | 49.8±26.76 (17-112)         | 0.16 (NS) |
| SGPT (IU/L)          | 45.42±35.93 (10-181)            | 58.06±32.27 (11-140)        | 0.03 (S) |
| TC (mg/dl)           | 209.39±57.89 (125-360)          | 229.11±51.97 (130-399)      | 0.04 (S) |
| TG (mg/dl)           | 207.55±101.21 (65-475)          | 206.19±74.18 (85-454)       | 0.46 (NS) |
| HDL (mg/dl)          | 40.73±5.47 (32-54)              | 39.51±8.31 (26-73)          | 0.22 (NS) |
| HbA1c (%)            | 5.54±0.38 (4.3-6.0)             | 5.67±0.38 (4.6-6.1)         | 0.05 (NS) |

*Student’s t-Test, (S)- Significant, (NS)- Non-significant. BMI: Body mass index; WC: Waist circumference; SGOT (AST): Serum Glutamic Oxaloacetic Transaminase; SGPT (ALT): Serum Glutamic Pyruvic Transaminase; TC: Total Cholesterol; TG: Triglyceride; HDL: High density lipoprotein, HbA1c: Glycated hemoglobin
is not an independent factor for insulin sensitivity and β-cell dysfunction.\[^{[16]}\]

One striking factor of the present study was 100% prevalence of FL disease in obese individuals without known disease of DM. This result can be due to inclusion criteria of BMI >25 kg/m\(^2\), but the result is alarming and indicates the NAFLD-prone body composition of North Indian adults. This is far more than the previously described prevalence in obese Asians (25%), and describes either the swiftly increasing burden of obesity and insulin resistance or a finding unique to this region based on the dietary and other factors.\[^{[17]}\] As an epidemiologic study on 500,000 Chinese adults observed that T2DM and hyperglycemia are associated with higher chances of chronic liver disease and liver malignancy; the presence of large numbers of fatty liver cases in our study population raises serious concerns.\[^{[18]}\] As both these complications are irreversible and prevention is the only sustainable approach, the importance of timely identification and management of hyperglycemia cannot be ignored.

When OGTT-based parameters were compared with different FL grades, elevated 1-hour-PPG levels had a more consistent and statistically significant relationship with higher FL grades as individuals with raised 1-hour-PPG were more in numbers with a higher FL grade than in low 1-hour-PPG group. In contrast to this, majority of the participants had lower PG at 2 hours, but demonstrated higher burden of obesity and insulin resistance or a finding unique to this region based on the dietary and other factors.\[^{[19]}\] As an epidemiologic study on 500,000 Chinese adults observed that T2DM and hyperglycemia are associated with higher chances of chronic liver disease and liver malignancy; the presence of large numbers of fatty liver cases in our study population raises serious concerns.\[^{[18]}\] As both these complications are irreversible and prevention is the only sustainable approach, the importance of timely identification and management of hyperglycemia cannot be ignored.

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As both these complications are irreversible and prevention is the only sustainable approach, the importance of timely identification and management of hyperglycemia cannot be ignored. Instead of having FPG <100 mg/dl, majority of the study individuals had higher FL grades. These observations, along with the results of 2-hour-PPG, define that traditional dysglycemic markers (i.e., FPG and 2-hour-PPG) have poor relationship with higher FL grades. A research by Zou et al.\[^{[21]}\] observed that raised FPG was associated with increased risk of NAFLD, but this study was done on non-obese individuals having a normal lipid profile. Opposite to this, our study population was composed of obese individuals and had relatively raised lipid parameters. So, constitutional chances for having FL in our population were higher, but the results for different OGTT categories were assessed in the same population cohort. This indicates a clear direct relation between raised PG and FL development. The results of this research were similar to an analysis done by Sesti et al.\[^{[22]}\] which revealed that PG at one-hour value above 155 mg/dl is strongly associated with hepatic steatosis. All these findings conclude the substantial importance of OGTT-based parameters in intermediate hyperglycemia and FL delineation, and the superiority of 1-hour-PPG over the FPG and 2-hour-PPG in this context. Hence, early recognition of this raised 1-hour-PPG in obese individuals without DM can provide a time-frame, in which interventions can halt the hepatic damage.

**Strength and limitations**

The present study included all three glycemic parameters simultaneously in obese individuals with no known disease of DM, whereas previously described researches included only a single OGTT parameter at one time. This study focused on...
the age group of 20–50 years, which is the usual time period when hyperglycemia sets in due to sedentary lifestyle, and intervention at this stage can prevent the progression to diabetes. One major limitation of our study was the non-availability of fibro-scan to confirm the findings of FL grades. Likewise, the participants were selected of BMI >25 kg/m², so the study lacks the comparison of FL grades and OGTT-based parameters in the lower BMI category.

**Recommendation**

We recommend the measurement of 1-hour-PPG as this is less time-consuming than the traditional 2-hour-PPG estimation. Single measurement of PG at 1-hour reduces the overall testing cost, provides rapid results, and increases the patient compliance. Further, the research proposes that 1-hour-PPG should be included as a routine additional glycemic marker in standard OGTT as it can identify individuals with intermediate hyperglycemia with higher sensitivity. Our study also provides a base for future studies in obese and non-obese individuals for association of 1-hour-PPG with FL disease. As the incidence of FL in obese individuals without diabetes was high in this study, routine USG evaluation of these patient is also desired for prompt recognition and timely intervention.

**Conclusions**

This study revealed that 1-hour-PPG is a superior marker in accurately identifying the dysglycemic events in obese adults without DM, and had more consistent and significant relationship with higher FL grades as compared to FPG and 2-hour-PPG. The outcome also favors the hypothesis that 1-hour-PPG is an early marker for dysglycemia in NAFLD, while the other traditional markers are still within the normal range.

**Declaration of patient consent**

The authors certify that they have obtained all appropriate patient consent forms. In the form the patient(s) has/have given his/her/their consent for his/her/their images and other clinical information to be reported in the journal. The patients understand that their names and initials will not be published and due efforts will be made to conceal their identity, but anonymity cannot be guaranteed.

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**Conflicts of interest**

There are no conflicts of interest.

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