Early- and late-onset startle modulation in unipolar depression

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Abstract

In two experimental sessions, we assessed early- and late-onset acoustic startle eyeblink modulation and subjective ratings of emotional pictures by nondepressed participants and by unipolar depressed participants. Depressed participants were assessed before and after treatment with the antidepressant medication Bupropion SR. Both depressed and nondepressed participants exhibited arousal-dependent startle modulation to early probes occurring 300 ms after picture onset. Nondepressed participants demonstrated the expected valence-dependent startle modulation to late probes (3,500–4,500 ms post-onset). In contrast, the late-probe blink magnitudes of depressed patients were unrelated to picture valence. This pattern of group differences was not moderated by treatment. There were no between-group differences in self-report ratings to pictures. These results suggest that depression may be characterized by anomalous responses to affective stimuli and that startle modulation can be a more sensitive index of affective response deficits linked to depression than self-report measures.

Descriptors: Depression, Startle modulation, Emotion, Valence, Arousal

Many theorists have argued that deficits in emotional and motivational responses to stimuli are core feature of unipolar depression (e.g., American Psychiatric Association, 1994; Fowles, 1988; Tomarken & Keener, 1998). Surprisingly, however, there have been relatively few experimental studies and even fewer psychophysiological studies that have directly compared how depressed and nondepressed individuals actually respond to hedonic stimuli (for exceptions, see, e.g., Allen, Trinder, & Brennan, 1999; Schwartz et al., 1978: Schwartz, Fair, Salt, Mandel, & Klerman, 1976a, 1976b; Sloan, Bradley, Dimoulas, & Lang, 2002). We assessed whether depressed and nondepressed individuals differ in affective modulation of the startle eyeblink reflex. We used the startle-blink paradigm because, on theoretical grounds, it would appear ideally suited for assessing the affective response deficits hypothesized to be characteristic of depression.

Startle modulation induced by emotional stimuli is a well-established phenomenon among predominantly nondepressed samples. When nondepressed individuals view affective pictures and the latency between picture onset and startle probe onset is relatively long (3,500–4,500 ms), response magnitude is modulated by the emotional content of the picture. Unpleasant pictures potentiate and pleasant pictures attenuate the amplitude of the startle blink relative to neutral pictures (Bradley, Cuthbert, & Lang, 1993). This linear pattern of affective startle modulation is thought to reflect the priming of neurobiologically based defensive and appetitive systems by unpleasant and pleasant foreground stimuli, respectively (Lang, Bradley, & Cuthbert, 1998). Correspondingly, several theorists have argued that: (a) core features of unipolar depression are exaggerated responses to stressful or other aversive stimuli and/or deficient responses to positive hedonic stimuli, and (b) such responses are attributable to dysfunction in neurobiologically based defensive and appetitive systems that organize responses to motivationally significant stimuli (e.g., Fowles, 1988; Klein, 1974; Tomarken & Keener, 1998). Thus, both theoretical accounts of the motivational dysfunction characteristic of depression and models of affective startle modulation highlight the critical role of higher order appetitive and defensive motivational systems. If depression is characterized by deficits in the mobilization and/or operation of such systems, one would expect a pattern of startle modulation among depressed individuals that deviates from the classic linear pattern commonly observed among nondepressed samples.

In an initial study, Allen et al. (1999) evaluated affective startle modulation to probes occurring between 4,000 and 6,000 ms after picture onset in depressed individuals. They failed to find...
the linear pattern observed among nondepressed participants. If anything, startle blinks elicited while viewing both pleasant and unpleasant pictures appeared potentiated relative to neutral pictures, although this trend was not significant. Such anomalous responses to pleasant pictures are consistent with prior empirical studies on the responses of depressed individuals to pleasant stimuli (e.g., Sloan, Strauss, Quirk, & Sajatovic, 1997; Sloan, Strauss, & Wisner, 2001).

We intended to extend the initial findings of Allen et al. (1999) in several ways. First, a significant limitation of the Allen et al. study was that all depressed participants were on antidepressant medication at the time of assessment. Therefore, it is unclear to what degree the anomalous startle responses observed were attributable to clinical status versus the effects of antidepressants. In addition, their findings leave unclear whether an atypical pattern of startle modulation is a state or trait marker of unipolar depression. To address these issues, we assessed depressed participants both before and after antidepressant treatment and, perhaps most importantly, insured that all participants were medication-free during the initial experimental session.

Second, we examined startle eyeblink modulation to both the arousal and valence dimensions of pictorial stimuli by presenting startle probes at two latencies (300 ms and 3,500–4,500 ms) after picture onset. Using predominantly normal college student participants, Bradley et al. (1993) found that, when startle probes were presented soon after the onset of foreground stimuli (300 ms), both unpleasant and pleasant foreground stimuli attenuated blink magnitudes relative to neutral pictures. They proposed that this pattern of startle modulation reflected differential allocation of attentional resources, with heightened allocation to those pictures that elicit high levels of arousal. Thus, by manipulating probe latency (early vs. late), we were able to compare depressed and nondepressed participants’ startle modulation to both the arousal (early probe) and valence (late probe) dimensions of the pictorial foreground stimuli.

Concerning predictions, based on theoretical accounts of the motivational deficits linked to depression and the initial results of Allen et al. (1999), we expected that depressed patients would not demonstrate the linear pattern of responses to late-onset probes that is characteristic of nondepressed individuals. Our hypotheses concerning group differences in responses to early probes were more cautious. Several sources of evidence indicate that depression is often associated with deficits in attentional, orienting, or arousal processes that appear linked to the arousal dimension of emotion (e.g., Heller, Nitschke, Etienne, & Miller, 1997; Yee & Miller, 1994). Based on these results, we predicted that depressed individuals would demonstrate an attenuated pattern of responding to the arousal properties of pictures in the early-probe condition relative to nondepressed individuals. Finally, we expected that both the early- and late-probe startle responses of depressed patients would normalize with effective antidepressant treatment.

Method

Participants
Participants provided written informed consent after all procedures were explained. Seventeen depressed and 16 nondepressed adults participated. Outpatient depressed participants (9 women, 8 men; age range: 23.0–61.1 years, mean = 39.13, SD = 8.97) were recruited from the greater Nashville community by advertisements placed in local newspapers. Respondents qualified for the study if they: (1) were at least 18 years of age; (2) met criteria for DSM-IV Major Depression; (3) had scores on the 17-item version of the Hamilton Rating Scale for Depression (HRSD-17; Hamilton, 1960) that were greater than 17; (4) were free of fluoxetine, antipsychotics, lithium, anticonvulsants, and mood stabilizers for at least 4 weeks prior to their baseline assessment and free of all other psychotropic medications for at least 2 weeks prior to their baseline assessment; (5) did not meet diagnostic criteria for atypical depression or a history of bipolar disorder; (6) did not have a recent history of drug abuse; and (7) did not have a history of seizure disorders, other central nervous system disorders, head trauma, or antisocial, borderline, or schizotypal personality disorders. Three depressed participants, all male, did not return for their second startle sessions because they discontinued their participation in the treatment study. The data from only those patients who participated in both startle sessions are included in the present analyses. Of the final sample, no participants met criteria for atypical depression and 9 met criteria for melancholic depression, 3 of whom also met criteria for an anxiety disorder (i.e., social phobia, generalized anxiety disorder, and panic disorder with agoraphobia). None met criteria for any other Axis I disorder.

Nondepressed participants were recruited from control participant pools maintained through the Vanderbilt University Medical Center General Clinical Research Center. Nondepressed participants (12 women, 4 men; age range: 30.0–69.1 years, mean = 46.49, SD = 8.29) had a BDI score of five or less at the time of both their startle sessions (session 1 mean [SD] = 1.9 [2.1], session 2 mean [SD] = 1.3 [1.8]) and did not meet lifetime criteria for major depressive disorder or any other Axis I disorder, assessed by semistructured clinical interview (Frist, Spitzer, Gibbon, & Williams, 1996). Participant groups did not differ with respect to age, t(28) = 1.71, p > .05 or gender distribution, χ² (1) = 0.41, p > .50.

Experimental Design

In addition to the between-subjects variable of depression status (depressed/nondepressed), three within-subjects variables were varied: picture valence (pleasant/neutral/unpleasant), probe onset time (early/late), and session (1/2).

Stimulus Materials

In each of the two experimental sessions, each participant viewed two sets of color pictures (one habituation set and one experimental set) chosen from the International Affective Picture System (IAPS; Center for the Study of Emotion and Attention, 1999) on the basis of their published affective valence and arousal ratings (see Appendix for a complete list of IAPS pictures used in this study). The habituation set consisted of six neutral pictures (two unprobed, two each with early and late probes). The experimental set consisted of 42 pictures (14 pleasant, 14 neutral, and 14 unpleasant). The experimental pictures were presented in seven blocks of six pictures. Every block contained two pictures of each picture category (pleasant/neural/unpleasant). To increase the generalizability of the results beyond the specific features of a limited set of experimental stimuli, three sets of 42 pictures were selected for male and female participants with minimal picture overlap between sets. All sets were invariant with respect to valence and probe order and were matched with respect to published IAPS ratings of valence and arousal. Each participant viewed different stimulus sets in the two experimental
sessions. The particular sets chosen for each participant and their
order of presentation across the two sessions were randomly
determined. Pictures were displayed on a 17-in. color monitor
approximately 1.3 m in front of the participant.

Procedure
Depressed participants received Bupropion SR for at least 6
weeks and were titrated to dosages of at least 300 mg/day by the
time of their second startle session. Depressed participants came
to the clinic at least every 2 weeks for symptom assessments and
received study medication and psychopharmacologic consulta-
tion. Participants completed the Beck Anxiety Inventory
Depression (BAI; Beck, Epstein, Brown, & Steer, 1988), the
Beck Depression Inventory, second version (BDI; Beck, Steer, &
Brown, 1996), the Hamilton Rating Scale for Anxiety (HRSA;
Hamilton, 1960), and the 17-item version of the Hamilton
Rating Scale for Depression (HRSD-17; Hamilton, 1960) at
each of the two startle sessions.

The two startle sessions were separated by approximately 12
weeks for both groups (depressed mean [SD] = 12.2 [3.3]; control
mean [SD] = 12.0 [0.8]). Depressed participants were medica-
tion-free at the first startle session, began treatment with
Bupropion SR the day after the first startle session, and were
still taking Bupropion SR at the second startle session 12 weeks
later. During each startle session, EMG was recorded while
participants first viewed the habituation pictures and then viewed
the experimental pictures. During both the habituation and
experimental phases, each picture was presented for 6 s, with a
16–24 s (mean = 20 s) interstimulus interval. Next, the same two
sets of pictures were presented again, without startle probes,
while participants controlled picture exposure duration and rated
each picture with respect to emotional pleasure (i.e., extremely
positive–extremely negative). The digitized EMG was digitally
rectified and low-pass filtered by integrating values in 32-ms time
windows. Consistent with other research groups (Bradley et al.,
1993), we computed the peak EMG amplitudes in search
windows between 10 and 250 ms after the onset of the startle
probes. Trials with artifact were detected by EMGART software
from the James Long Company. This software computes the
standard deviation and mean of each trial’s baseline (defined as
the 50 ms prior to probe onset) and rejects trials with baselines
that exceed at any time either the baseline mean plus two
standard deviations of that mean or four times the mean.

After deletion of trials with artifact (less than 1.0% of the
trials), we averaged amplitude measures across each of the six
levels of the Valence (pleasant/neural/unpleasant) × Probe
Presentation (early/late) conditions for each participant.

Data Analysis
The six habituation trials were not included in analyses. Omnibus
repeated measures effects were evaluated using multivariate test
criteria that do not require the assumption of sphericity (Vasey
& Thayer, 1987). The primary omnibus analyses of interest were
Group (depressed, control) × Valence (pleasant, neutral, un-
pleasant) × Session (1, 2) repeated measures ANOVAs per-
formed separately on startle amplitudes during the early and late
probe conditions. In the early probe condition, the primary focus
of interest was testing for a quadratic pattern of startle
modulation across the pleasant, neutral, and unpleasant pictures.
Therefore, in addition to the omnibus analyses, we specified a set
of contrasts. One main effect contrast tested the overall quadratic
trend averaging across groups and sessions (trend coefficients =
−1, +2, and −1, for the pleasant, neutral, and unpleasant
marginal means). Three other contrasts tested for the effects of
group, session, and the Group × Session interaction on the
quadratic trend. In the late probe conditions, the primary focus
of interest was the linear valence contrast across the pleasant,
neutral, and unpleasant pictures (trend coefficients = −1, 0, and
+1, respectively). Therefore, in addition to the omnibus
analyses, we specified a set of contrasts that tested the overall
linear trend (pooling across groups and sessions) and the effects
of group, session, and their interaction on this trend. Below, to
delineate more completely the nature of the effects across picture
categories, we report the results of both linear and quadratic
trend contrasts for the early- and late-probe data.

Results
Symptom Measures
Table 1 indicates clinician-rated and self-report depression and
anxiety symptom measures at both startle sessions for depressed
participants. Overall, depressed participants attained statistically
significant improvement on measures of both depression (i.e., the
BDI and HRSD-17) and anxiety (i.e., the BAI and HRSA).

We should note that although we are presenting results based on raw
startle magnitudes, analyses using z-transformed startle magnitudes
yielded essentially the same results. Other researchers have noted highly
similar results obtained with both raw and standardized scores (Grillon
& Ameli, 2001). Raw startle magnitudes are reported to be consistent with
studies in the startle literature we are addressing (e.g., the core diagnostic
analyses of Allen et al., 1999; Bradley et al., 1993).
Table 1. Depression and Anxiety Scores at Both Picture-Viewing Sessions for Depressed Participants

<table>
<thead>
<tr>
<th>Measure</th>
<th>Session 1</th>
<th>Session 2</th>
<th>t(13)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>BDI</td>
<td>23.0 (12.9)</td>
<td>9.3 (14.2)</td>
<td>5.05</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>HRS-D17</td>
<td>18.7 (4.7)</td>
<td>7.7 (7.4)</td>
<td>3.41</td>
<td>0.005</td>
</tr>
<tr>
<td>BAI</td>
<td>12.9 (11.0)</td>
<td>5.4 (6.9)</td>
<td>5.91</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>HRSA</td>
<td>11.4 (4.3)</td>
<td>5.3 (6.0)</td>
<td>4.12</td>
<td>0.001</td>
</tr>
</tbody>
</table>

Standard deviations are in parentheses. All t tests are two-tailed. *BDI: Beck Depression Inventory--II; HRS-D17: 17-item version of the Hamilton Rating Scale for Depression; BAI: Beck Anxiety Inventory; HRSA: Hamilton Rating Scale for Anxiety.

Startle Modulation

When gender, age, and picture set were entered into analyses of affective startle modulation as covariates, there were no main effects or interactions involving these factors on the primary dependent measures. Thus, they were excluded from all the analyses reported below.

Early startle probes. Figure 1 shows the mean blink magnitude in response to early probes for the depressed and nondepressed groups. The only significant effect yielded was a main effect of valence, \(F(2,27) = 4.12, p < .03\) (all other \(p > .05\)). Of prime importance was the set of contrasts testing for quadratic effects across the valence categories. The only significant effect yielded by these contrasts was a significant overall quadratic trend averaging across groups and sessions, \(F(1,28) = 8.28, p < .01\) (all other \(p > .35\)). The nonsignificant contrast testing for between-group differences on the quadratic trend, \(F(1,28) = 0.06, p > .80\), indicates that groups did not differ in startle modulation as a function of picture valence to early probes. No significant linear contrast effects were observed, \(p > .80\).

Late startle probes. Figure 2 shows the mean blink magnitudes during the late probe conditions for the depressed and nondepressed groups. The omnibus Group \(\times\) Valence \(\times\) Session ANOVA revealed a main effect of valence, \(F(2,27) = 7.63, p < .005\) and trends toward a main effect of session, \(F(1,28) = 4.03, p < .10\), and a Group \(\times\) Valence interaction, \(F(2,27) = 2.75, p < .10\) (all other \(p > .40\)). More centrally relevant were the results of the planned contrasts testing for linear effects across the valence categories. These revealed stronger evidence for group differences in responding (see Figure 2). We observed a significant overall linear trend, \(F(1,28) = 15.83, p < .001\), but also significant between-group differences in linearity, Group \(\times\) Linear \(F(1,28) = 5.35, p < .03\). The latter effect indicates that depressed and nondepressed participants responded differently to the affective pictures during the late probe condition. Separate analyses conducted within each of the two diagnostic groups, averaged over sessions, indicated that there was a significant linear trend within the control group, \(F(1,15) = 15.69, p < .005\), but not within the depressed group, \(F(1,13) = 2.19, p > .15\). There were no significant effects involving the session factor on linear trends computed either across or within groups (\(p > .45\)). No quadratic contrasts were significant on late-probe startle measures (\(p > .40\)).

Exploratory follow-up analyses were conducted on late probe data averaged over sessions to test whether there was evidence of startle potentiation (i.e., a significant difference in responses to unpleasant and neutral pictures) and startle inhibition (i.e., a significant difference in responses to pleasant and neutral pictures) and whether diagnostic groups differed with respect to startle potentiation and inhibition. There was no evidence of startle potentiation or startle inhibition in the control group alone, \((p > .10)\), in the depressed group alone, \((p > .30)\), or in analyses pooling across groups (\(p > .05\)). There were also no significant interactions between group and startle potentiation, \(p > .50\), or startle inhibition, \(p > .35\).

Self-Report Responses to Pictures

Table 2 shows the mean picture ratings for each diagnostic group. The Group (depressed, control) \(\times\) Valence (pleasant, neutral, unpleasant) \(\times\) Session (1, 2) ANOVA on pleasure ratings revealed only a significant main effect for valence, \(F(2,27) = 253.91, p < .0001\) (all other \(p > .20\)). A planned linear valence contrast analysis was highly significant, \(F(1,28) = 446.33, p < .0001\). A similar analysis on arousal ratings indicated only a significant main effect for valence, \(F(2,27) = 97.14, p < .0001\) (all other \(p > .25\)), and a significant quadratic trend, \(F(1,28) = 142.92, p < .0001\). There were no significant effects involving group (\(p > .20\)).

Discussion

Startle responses to early-onset probes replicated the arousal-modulated startle findings of Bradley et al. (1993). Of particular importance is the evidence that this effect was not moderated by clinical status: There was only a significant main effect for the overall quadratic trend and no interactions involving depression status. Bradley et al. suggested that the early-onset effect is an example of prepulse inhibition (Graham & Hackley, 1991). Prepulse inhibition is observed when startle probe onset occurs soon after the onset of a nonstartle eliciting stimulus (the prepulse stimulus). The attenuation of startle amplitude that occurs under these circumstances has been attributed to nonvolitional attentional processes that serve to protect the processing of the prepulse stimulus from disruption. The failure to find depressed versus nondepressed differences in early-onset startle responding suggests that attentional mechanisms during initial processing of affective stimuli may be preserved in depression—at least in the context of the experimental stimuli and procedures used in the present study.

Nondepressed participants exhibited the well-known linear increase in startle amplitude across the pleasant, neutral, and unpleasant picture categories, whereas depressed participants failed to show evidence of valence modulation. Although there were significant differences between depressed and nondepressed participants in responses to the late probes, there were no significant main effects or interactions involving session. In particular, we did not find evidence that depressed patients’ patterns of startle modulation normalized after treatment. Such null findings were observed despite the fact that the depressed group demonstrated significant declines in depressive symptoms over the course of treatment.

There are two possible explanations for the failure to find changes across time in startle responding among depressed patients. The first is that the core motivational processes indexed by startle modulation do not change despite changes in clinical symptoms. If so, than an anomalous pattern of affective startle modulation may be a trait marker of depression that reflects a persistent vulnerability.
The second explanation is that several factors may have compromised the statistical power of our analyses to detect across-time changes in startle responding. First, the sample size in the depressed group is rather small for a study assessing treatment effects. Second, as indicated by the means and standard deviations shown in Table 1, although the depressed group showed notable changes in mean levels of depression, not all patients were clearly in the nondepressed range at the posttreatment assessment. Future studies assessing changes in startle modulation over the course of treatment should use especially large sample sizes and/or select from a large pool of participants only those formerly depressed individuals who are clearly euthymic at the time of assessment.

A third factor is the stability of startle modulation over time. A glance at Figure 2 reveals that the pattern of means across the three valence categories was stable over time among both control and depressed participants.

**Figure 1.** Within- and aggregated across-session mean blink magnitudes for the early probe condition (300 ms after picture onset). Top: Control participants. Middle: Depressed participants. Bottom: Quadratic trend scores for depressed and control participants. Quadratic trend scores were generated for each participant by weighting startle magnitude values as follows: \((-1) \times \text{Pleasant} + (2) \times \text{Neutral} - (1) \times \text{Unpleasant}\). The endpoints of error bars represent values one standard error above and below the mean.
depressed and nondepressed participants. At the individual level, overall startle amplitudes were also stable over time. For example, the test–retest correlation of late-probe amplitude values aggregated across the three valence categories was .75 ($p < .01$) for depressed participants and .79 ($p < .001$) for nondepressed participants. However, at the individual level, the linear pattern of startle modulation was markedly less stable across sessions. Indeed, when we computed a linear contrast value for each participant and computed test–retest correlations across sessions, negative correlations were observed ($r = -.39$ and $-.65$ for control and depressed participants, respectively). Other studies have also found low test–retest stability values for the linear component of late-onset startle modulation (Larson, Ruffalo, Nietert, & Davidson, 2000). Stability values that are negative or nearly zero will also serve to lower statistical power to detect differences between time periods because they will inflate the error term for tests of main effects and interactions involving the time factor (Zimmerman, Williams, & Zumbo, 1993). Future studies designed to address these issues should use larger sample sizes.

Figure 2. Within- and aggregated across-session mean blink magnitudes for the late probe condition (3,500–4,500 ms after picture onset). Top: Control participants. Middle: Depressed participants. Bottom: Linear trend scores for depressed and control participants. Linear trend scores were generated for each participant by weighting startle magnitude values as follows: $(-1) \times$ Pleasant $+(0) \times$ Neutral $+(+1) \times$ Unpleasant. The endpoints of error bars represent values one standard error above and below the mean.
and several other methodological changes that might serve to increase the reliability and/or stability of estimates of affective startle modulation.

For all groups and sessions, pleasure ratings followed the pattern of the a priori valence categories, and pleasant and unpleasant pictures were judged more arousing than neutral pictures. We failed to observe any significant between-group differences on these measures or changes over time. The absence of between-group differences in self-report ratings are particularly unexpected given the evidence from previous studies that depressed individuals rate IAPS pictures as less pleasant than nondepressed controls (Allen et al., 1999; Sloan et al., 1997, 2001). Discrepancies in the procedures used to assess self-reports might have been one factor that contributed to the differences between our results and those of prior studies. For example, in the current study, we assessed self-reports using 1-to-9 Likert rating scales. Such scales were administered in a separate phase after the initial set of exposures during which startle responses were assessed. In prior studies, self-report responses were assessed during the initial viewing phase (Allen et al., 1999; Sloan et al., 1997, 2001) and via the computerized version of the Self-Assessment Manikin (Allen et al., 1999; Sloan et al., 1997). Whatever the reasons for our failure to find group differences in self-reported emotion, there is a notable contrast between our null findings in this domain and the significant differences between depressed and nondepressed groups in late-probe startle responding that we observed. This disparity suggests that startle modulation may be a more sensitive index of the affective response deficits linked to depression than self-report measures.

In summary, depressed and nondepressed participants responded similarly to early-onset startle probes but differed in responses to late-onset probes. In the latter case, depressed participants failed to exhibit the typical valence-based startle modulation. Although these findings warrant replication with a larger sample size, such dissociation indicates that unipolar major depressive disorder may not compromise early attentional processing of affective stimuli but may be associated with anomalies in later-onset processes that subserve responses to the valence properties of affective and motivational stimuli. Future research is necessary to assess more thoroughly whether or not the affective response deficits that we observed are trait or state markers of depression. More broadly, our results add to the growing body of literature indicating that various forms of psychopathology may be linked to unique patterns of startle responding to affective stimuli (e.g., Grillon & Morgan, 1999; Hamm, Cuthbert, Globisch, & Vaitl, 1997; Patrick, Bradley, & Lang, 1993).

### REFERENCES


APPENDIX: IAPS PICTURES USED IN THIS STUDY

For clarity, pictures are presented here grouped by valence condition (pleasant, neutral, unpleasant) and probe condition (early, late).

**Male Picture Set A**
- Pleasant, early: 8501, 8470, 1650, 8180, 8340, 4210.
- Pleasant, late: 5629, 4659, 8300, 4320, 8260, 8170.
- Neutral, early: 7491, 7006, 2630, 7025, 7187, 5030.
- Neutral, late: 7217, 7950, 7010, 2570, 5020, 7224.
- Unpleasant, early: 6570, 6250, 9810, 3080, 3053, 6313.
- Unpleasant, late: 9410, 3530, 9630, 9252, 3400, 6260.

**Male Picture Set B**
- Pleasant, early: 4652, 2030, 8034, 8030, 5460, 4664.
- Pleasant, late: 8380, 4232, 5600, 4240, 8200, 8080.
- Neutral, early: 7150, 7080, 2440, 7050, 7020, 7185.
- Neutral, late: 5740, 2190, 7175, 2850, 7490, 7009.
- Unpleasant, early: 9570, 3150, 6230, 3060, 6550, 3030.
- Unpleasant, late: 3110, 3000, 9921, 3100, 6510, 3130.

**Male Picture Set C**
- Pleasant, early: 8400, 8370, 5260, 8190, 5470, 4670.
- Pleasant, late: 5950, 4660, 5700, 5621, 4235, 4608.
- Neutral, early: 7035, 7004, 5731, 7233, 7090, 7040.
- Neutral, late: 2200, 2840, 7110, 2320, 7140, 2880.
- Unpleasant, early: 6560, 9910, 3071, 3170, 3500, 6540.
- Unpleasant, late: 3120, 3010, 9800, 9250, 3102, 6350.

**Female Picture Set A**
- Pleasant, early: 8080, 5910, 5460, 8180, 4670, 8370.
- Pleasant, late: 7330, 8200, 8090, 4660, 8400, 4572.
- Neutral, early: 5740, 7491, 7150, 7185, 7031, 7110.
- Neutral, late: 2840, 7187, 7175, 7035, 7002, 5530.
- Unpleasant, early: 9433, 3500, 3010, 3000, 3030, 9571.
- Unpleasant, late: 6230, 3120, 6821, 6510, 9921, 3064.

**Female Picture Set B**
- Pleasant, early: 7502, 4640, 4608, 8034, 7270, 5621.
- Pleasant, late: 8170, 8490, 5450, 8501, 8041, 8210.
- Neutral, early: 9360, 7000, 2480, 7080, 7490, 7009.
- Neutral, late: 7217, 2381, 7004, 7140, 2890, 6150.
- Unpleasant, early: 2730, 6560, 3102, 3170, 6313, 3140.
- Unpleasant, late: 9410, 6360, 3530, 6540, 3100, 6350.

**Female Picture Set C**
- Pleasant, early: 8190, 8502, 5623, 8470, 4510, 8030.
- Pleasant, late: 1710, 8496, 8300, 4680, 8161, 5629.
- Neutral, early: 2440, 7020, 7006, 7100, 7950, 5130.
- Neutral, late: 2190, 7060, 7010, 7030, 7050, 5510.
- Unpleasant, early: 3400, 9405, 3064, 3053, 3060, 9252.
- Unpleasant, late: 3110, 3080, 9571, 6550, 3500, 3130.